Supplemental Computational Analysis

Design and *in vitro* realization of carbon-conserving photorespiration

Devin L. Trudeau[1‡], Christian Edlich-Muth[2‡], Jan Zarzycki[3], Marieke Scheffen[3], Moshe Goldsmith[1], Olga Khersonsky[1], Ziv Avizemer[1], Sarel J. Fleishman[1], Charles A.R. Cotton[2], Tobias J. Erb[3], Dan S. Tawfik[1*], Arren Bar-Even[2*]

[1]Department of Biomolecular Sciences, Weizmann Institute of Science, 234 Herzl Street, Rehovot 7610001, Israel
[2]Max Planck Institute of Molecular Plant Physiology, Am Mühlenberg 1, 14476 Potsdam-Golm, Germany
[3]Max Planck Institute for Terrestrial Microbiology, Karl-von-Frisch-Straße 10, D-35043 Marburg, Germany

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Contents

1 pathSeekR **5**

1.1 Introduction . . . 5

1.2 Compounds . . . 5

1.2.1 Repertoire of chemical groups . . . 6

1.2.2 Combining groups into molecules . . . 6

1.2.3 Disallowed compounds . . . 6

1.3 Reactions . . . 7

1.4 Network . . . 9

1.5 Multi-substrate reactions . . . 9

1.6 pathSeekR algorithm . . . 9

1.7 Categorisation . . . 10

1.8 Results . . . 10

1.9 Reactive C2 intermediates . . . 11

1.10 Pathway architectures . . . 15

2 The stoichiometric-kinetic model **16**

2.1 Theory . . . 16

2.1.1 Pathways . . . 16

2.1.2 The stoichiometric consumer model of photosynthesis . . . 19

2.1.3 Stoichiometric-kinetic model . . . 23

2.1.4 Physicochemical parameters . . . 34

2.2 Results . . . 37

2.2.1 Parameters and variables . . . 37

2.2.2 Rubisco kinetics . . . 38

2.2.3 Limitation by light . . . 39

2.2.4 Limitation by enzyme activities other than Rubisco . . . 40
2.2.5 Limitation by a Calvin cycle enzyme . . . 41
2.2.6 Limitation by a photorespiratory enzyme . . . 44
2.2.7 Limitation by a photorespiration carboxylase . . . 45

3 Kinetic models of photorespiration shunts 45

3.1 Introduction . . . 45
3.2 Generic form of a random order rate reaction with one substrate, one products and a variable number of co-factors . . . 46
3.2.1 Abbreviations . . . 46
3.2.2 Linear reactions . . . 47
3.2.3 Generic example of a linear reaction . . . 47
3.2.4 Canonical form of the equation . . . 47
3.2.5 Linear relation between the concentrations of substrate and product . . . 47
3.2.6 Minimal required activity . . . 48
3.2.7 Thermodynamic limitations . . . 48
3.3 Rate laws of all photorespiration shunt reactions . . . 48
3.3.1 Compound abbreviations . . . 49
3.3.2 Glycolate 2-phosphatase . . . 49
3.3.3 Glycolyl-CoA synthetase . . . 50
3.3.4 Glycolyl-CoA transferase . . . 50
3.3.5 Glycolaldehyde dehydrogenase, CoA-acylating (Glycolyl-CoA reductase) . . . 50
3.3.6 Ar5P aldolase . . . 51
3.3.7 Ru1P aldolase . . . 51
3.3.8 Transketolase . . . 52
3.3.9 Transaldolase . . . 52
3.3.10 Erythrulose 4-kinase . . . 53

3.3.11 Xylulose 5-kinase . . . 53
3.3.12 Ribulose-1-phosphate 5-kinase . . . 53
3.3.13 Glycolyl-CoA carboxylase . . . 54
3.3.14 Tartronyl-CoA reductase . . . 54
3.3.15 Glycerate 3-kinase . . . 55
3.4 Parameters . . . 55
3.4.1 Imputing unknown parameters . . . 55
3.4.2 Kinetic parameters . . . 55
3.4.3 Thermodynamic parameters . . . 56
3.4.4 Kinetic and thermodynamic parameters of the simulation . . . 57
3.4.5 Steady-state concentrations of co-factors and Calvin cycle intermediates . . . 58
3.4.6 Sampling . . . 58
3.4.7 Lower and upper limits on pathway metabolites . . . 58
3.4.8 Summary . . . 60
3.4.9 Calculating fluxes . . . 60
3.5 Pareto analysis . . . 60
3.5.1 Assumptions . . . 60
3.5.2 Solving the rate equations . . . 61
3.5.3 Metabolite distributions . . . 61
3.5.4 Carrying out the Pareto analysis . . . 61
3.5.5 Results for all photorespiratory shunts . . . 62

4 pathSeekR pathway architectures 67

4.1 Glycolyl-CoA pathways . . . 67
4.2 Glycolaldehyde 2-phosphate pathways . . . 90
4.3 Glycolaldehyde pathways . . . 111
4.4 Glyoxylate pathways . . . 176

4.5 Glycine pathways 202

4.6 Aminoethanol pathways 206

4.7 Aminoacetaldehyde pathways 220

5 References **226**

1 pathSeekR

1.1 Introduction

pathSeekR is an algorithm that looks for pathways in a reaction network, starting with a source compound and ending in any of a set of sink compounds. In the context of photorespiration shunts, the source is glycolate 2-phosphate (G2P), and the sink comprises all Calvin cycle intermediates.

pathSeekR creates a reaction network where the nodes are compounds and the (directed) edges reactions. Compounds are assembled from a small repertoire of chemical groups (Section 1.2). A universe of compounds is created by combining all permutations of chemical groups in the repertoire up to a maximal number of groups. The universe is pruned by removing compounds that contravene rules about biological compounds (Section 1.2.3). Importantly, the chemical groups of pathSeekR are achiral which means that all stereoisomers of a compound are represented by a single achiral one.

A chemical reaction maps one compound (the substrate) to another (the product), which is represented as a directed edge between the two nodes in the network (Section 1.4). Co-factors and inorganic substrates are ignored. If the reaction is reversible, two edges, corresponding to the two directions of the reaction are created. If there is more than one substrate (or product), an edge for each of these is created (Section 1.5) and the co-substrate (co-product) becomes an attribute of the edge.

Each reaction is implemented as a simple rule that captures a naturally occurring reaction mechanism (Section 1.3). The rule is defined as a pair of chemical patterns - a sequence of chemical groups - one for the substrate, one for the product. The reaction is implemented by substituting the substrate pattern with the product pattern.

pathSeekR will search the network until a certain number of pathways have been found or to a certain depth (number of reactions). Here we restrict the solutions to a maximal length of 9 nodes (including source and sink), which is equivalent to 8 reactions.

1.2 Compounds

In pathSeekR, compounds (or molecules) are represented as a sequence of chemical groups, much like a DNA or protein sequence, but allowing branching. A chemical group in pathSeekR is a carbon atom with or without heavy atom substituents (Table 1), for example aldehyde and hydroxycarbon groups. We differentiate between primary, secondary and tertiary hydroxycarbons, and the aldehyde group is not identical to the keto group. The repertoire of groups can be amended or restricted arbitrarily. We have chosen our set of groups such that most compounds of central metabolism can be generated. CoA-thioesters, phosphate esters and anhydrides have a shorthand notation, effectively representing them as pseudoatoms. To give two examples, in pathSeekR glycerate 3-phosphate is (CH_2O-PO_3^{2-},CHOH,COOH) and glycolyl-CoA is (CH_2OH,COS-CoA).

With this approach chirality and cyclical compounds cannot be represented. For the purpose of finding photorespiration shunts, we chose to also exclude compounds with double bonds, branched carbon chains and bridging non-carbon atoms (e.g. an oxygen atom between two carbons). The rationale for this is that both the source (G2P) and the sink (Calvin cycle intermediates) are devoid of these features, and a pathways that contained e.g. a double bond, would first have to create one only to then remove it again. Clearly this would lead to much longer pathways. For the same reason, fully

reduced carbon groups (CH_2, CH_3) were excluded. These are practical decisions (heuristics) that can in principle be reversed.

1.2.1 Repertoire of chemical groups

Group	Description	Position
CH_2OH	hydroxycarbon	terminal
CHOH	hydroxycarbon	non-terminal
CHO	aldehyde	terminal
CO	carbonyl	non-terminal
COOH	carboxylate	terminal
COO-PO_3^{2-}	acyl phosphate anhydride	terminal
COS-CoA	acyl CoA thioester	terminal
CH_2O-PO_3^{2-}	phosphate ester	terminal
CHO-PO_3^{2-}	phosphate ester	non-terminal
CH_2NH_2	aminocarbon	terminal
$CHNH_2$	aminocarbon	non-terminal

Table 1: Repertoire of chemical groups for finding photorespiration shunts. The repertoire can be expanded to include many other chemistries e.g. double bonds, branched carbon chains.

1.2.2 Combining groups into molecules

pathSeekR makes all possible permutations of the repertoire of groups up to a certain size and making sure that terminal groups are present in the correct places. With the repertoire we have chosen, all compounds that are generated are linear (non-branched, non-cyclical). After creating all permutations duplicates (where the order of groups is exactly reversed) are removed.

1.2.3 Disallowed compounds

- No more than 7 carbons
- No double bonds, no cycles and no tertiary/quaternary carbons (no branches)
- No bridging heteroatoms in chain
- No methyl or methylene groups
- No more than one carbonyl group (either keto or aldehyde) per molecule
- No more than one thioester per molecule, no phosphate groups in CoA compounds
- No more than one phosphate anhydride per molecule
- No more than two phosphate groups per molecule, only one of which can be non-terminal
- No more than one amine group per molecule
- No imines

Compounds that contravene the above rules are removed. The rules are either motivated by chemistry (e.g. two carbonyl groups next to each other are unstable) or by what is known about metabolism (e.g. no compound with both a CoA and phosphate group, or more than two phosphate groups in central metabolism).

None of these rules are essential. It would therefore be possible to add, amend or remove the rules to define a different set of allowed and disallowed compounds.

1.3 Reactions

We defined 27 rules that encode the vast majority of reaction mechanisms in central metabolism. A rule defines the chemical groups the substrate must have, and what the molecule looks like after the reaction has taken place. We call this the substrate and the product pattern respectively. In addition, we classify the reactions as reversible or irreversible and whether or not the enzyme catalysing the reaction is oxygen sensitive. It would also be possible to record other types of information about the reaction, e.g. co-factor requirements. For the purpose of finding new photorespiratory shunts we excluded all oxygen sensitive reactions because the chloroplast is fully oxic, and all energy-dissipating reactions such as phosphatases and acyl-CoA hydrolyases with the exception of 2-phosphoglycolate phosphatase.

Class	Reaction	Substrate	Product	Co-sub	Co-P	R
Oxidoreductase	Alcohol dehydrogenase	C(O)	C(=O)			Y
Oxidoreductase	Aldehyde dehydrogenase	C(=O)	C(=O)(O)			N
Oxidoreductase	Aldehyde dehydrogenase, CoA-acylating	C(=O)	C(=O)(S[CoA])			Y
Oxidoreductase	Aldehyde dehydrogenase, phosphorylating	C(=O)	C(=O)(O[PO3])			Y
Oxidoreductase	Amine dehydrogenase[a]	C(N)	C(=O)			Y
Activase	Kinase hydroxyl	C(O)	C(O[PO3])			N
Activase	Kinase carboxyl	C(=O)(O)	C(=O)(O[PO3])			N
Activase	CoA synthetase[b]	C(=O)(O)	C(=O)(S[CoA])			N
Activase	Phosphate CoA-acetyltransferase	C(=O)(O[PO3])	C(=O)(S[CoA])			Y
Isomerase	Aldo/keto isomerase	C(=O)C(O)	C(O)C(=O)			Y
Isomerase	Phosphoester isomerase*	C(O[PO3])C(O)	C(O)C(O[PO3])			Y
Isomerase	Phosphoanhydride isomerase*	C(=O)(O[PO3])C(=O)(O)	C(=O)(O)C(=O)(O[PO3])			Y
Isomerase	Thioester isomerase*	C(=O)(O[PO3])C(=O)(O)	C(=O)(O)C(=O)(O[PO3])			Y
Carboxylase	α-carboxylation (thioester)[c]	C(=O)(S[CoA])C(O)	C(=O)(S[CoA])C(O)C(=O)(O)			N
Carboxylase	α-carboxylation (carbonyl)	C(=O)C(O)	C(=O)C(O)C(=O)(O)			N
Carboxylase	Reductive α-carboxylation (carbonyl)	C(=O)C(O)	C(O)C(O)C(=O)(O)			N
Transferase	Transketolase	C(=O)	C(O)C(=O)C(O)	C(O)C(=O)C(O)	C(=O)	Y
Transferase	Transaldolase	C(=O)	C(O)C(=O)C(O)C(O)	C(O)C(=O)C(O)C(O)	C(=O)	Y
Carbo-lyase	Aldolase (carbonyl)	C(=O)C(O)	C(=O)C(O)C(O)	C(=O)		Y
Carbo-lyase	Aldolase (amine)	C(N)	C(N)C(O)	C(=O)		Y
Carbo-lyase	Ketolase	C(=O)	C(=O)C(O)	C(=O)		Y
Carbo-lyase	Aldolase (thioester)	C(=O)(S[CoA])C(O)	C(=O)(S[CoA])C(O)C(O)	C(=O)		Y
Carbo-lyase	Aldolase (thioester, hydrolysing)	C(=O)(S[CoA])C(O)	C(=O)(O)C(O)C(O)	C(=O)		Y
Carbo-lyase	Aldolase (carboxyl)	C(=O)(O])C(O)	C(=O)(O)C(O)C(O)	C(=O)		Y
Carbo-lyase	Acyl-CoA C-transferase (thioester)	C(=O)(S[CoA])C(O)	C(=O)(S[CoA])C(=O)	C(=O)(S[CoA])		Y
Carbo-lyase	Acyl-CoA C-transferase (amine)	C(N)	C(N)C(=O)	C(=O)(S[CoA])		Y

Table 2: pathSeekR reactions. Substrate, product, co-substrate and co-product ('Co-P') are in SMILES notation. CoA and phosphate are represented as pseudo-atoms, [CoA] and [PO3] respectively. 'R' stands for reversibility. All kinase and CoA transferase reactions are irreversible, with one exception: dephosphorylation of 2-phosphoglycolate is explicitly allowed. * Phosphoester, phosphoanhydride and thioester isomerases also work on non-adjacent groups. Some of the reactions in the table can be carried out by different classes of enzymes: [a] transaminase, [b] CoA transferase, [b] transcarboxylase.

1.4 Network

A reaction network consists of nodes (compounds) and edges (reactions). In pathSeekR, every edge is simple, i.e. it connects exactly two nodes. The edges are also directed, which is important for irreversible reactions. Reversible reactions are represented as two irreversible reactions. Edges of a node to itself are forbidden (this could happen in an isomerisation reaction).

The network starts off as nodes without edges. By applying all reaction rules to all compounds, edges are added. Multiple edges in the same direction are combined into one.

1.5 Multi-substrate reactions

If a reaction has more than one substrate, neither of which fall into the class of co-factors (which are ignored), special treatment is necessary. We connect both substrate nodes to the product (or products) and record the co-substrate (and co-product if any) as a property of the edge. In this manner, all edges can remain simple but the information on the co-substrate (co-product) is not lost and can be used or reported as appropriate.

In our pathSeekR runs we only allow compounds as co-substrates and co-products that zero or one reaction removed from the Calvin cycle.

1.6 pathSeekR algorithm

The algorithm has three steps: creation of compounds, creation of the reaction network and the actual pathway search.

For the actual search, the network is represented as a neighbourhood list: the neighbourhood of a node (compound) consists of all nodes (compounds) that can be reached from the node via an edge (reaction). The search is initialised with the source node as the only proto-path (by this we mean a path that is not terminated). In the first iteration, all nodes from the neighbourhood of the source are added to the proto-path, resulting in as many proto-paths of length 2 as there are nodes in the neighbourhood of the source. In the next and all following iterations, this procedure is repeated with every proto-path and the respective neighbourhood of the terminal node.

This generic path search leads to super-exponential growth in the number of proto-paths. To make the search efficient, it is necessary to kill off as many proto-paths as possible as quickly as possible. In pathSeekR, a proto-path is terminated if

(i) It ends in a sink (a pathway has been found)

(ii) It ends in a node that has been visited before (a cycle)

(iii) There are no nodes in the neighbourhood of the terminal node left that have not been visited before (dead end)

In addition we use some rules and heuristics that accelerate the search.

(iv) A proto-path is terminated if it ends in a node that has been defined as a forbidden node (this helps to restrict the scope of the search).

(v) A proto-path is terminated if any other proto-path has visited the node before.

This second rule is a heuristic that reduces the computational complexity very considerably. The reason why it makes sense to exclude nodes that have been visited by other paths is that a) there must be a faster or equally fast reaction path to this node and b) one can later use the solutions from the 'faster' path from that node on and paste them onto the 'slower' proto-path. It is in theory possible to miss out on some solutions (pathways) because the nodes the faster path has visited cannot be visited again but may not have been visited by the slower path and therefore could lead to additional solutions. In practice, if the pathways is meant to be short, and if it has a clear 'directionality' (e.g. from smaller to larger and/or from more oxidised to more reduced compounds) it is very unlikely that interesting candidates are lost.

Finally, any number of rules can be stated as exit criteria

(vi) Do not extend beyond a certain length

(vii) Terminate if a certain reaction or type of reaction has taken place more than n times (e.g. forbid dephosphorylation)

It would also be possible to limit the number of ATPs consumed (this would require that the relevant information be defined in the reactions table). All in all there are many possibilities to fine-tune the search - not by adjusting fudge parameters, but by defining explicit rules.

1.7 Categorisation

We define two levels of categorisation, class and superclass. To this end we introduce two new concepts: firstly, a 'key reaction' is a reaction that changes the number of carbons from substrate to product, and secondly the 'carbon signature' of a key reaction is a pair of integers representing the number of carbons before and after the reaction. The carbon signature of a pathway is simply the concatenation of the carbon signatures of its key reactions.

Two pathways belong to the same superclass if they have the same carbon signature. They belong to the same class, if, in addition, the source and sink of the pathway, as well as the substrate and product of each key reaction are the same.

1.8 Results

Pathway architectures that were discovered with the pathSeekR algorithm are shown in order of ascending length, with a maximum of 8 reactions in the pathway (9 compounds) - which is the number of reactions of native photorespiration (not counting re-fixation of ammonia). The pathway diagrams display phosphate groups as "O-Po" and CoA-thioesters as "S-Co". All co-factors such as ATP and CoA-SH are not shown. All molecules are assumed to be achiral, therefore generic names are given, e.g. C4-ketose is a ketose with 4 carbons, which could be D- or L-erythrose. Most sugars, sugar phosphates etc are named and more unusual compounds are simply numbered in the pathway diagram

and given as SMILES string in the table below - "(O-[PO3])" and "(S-[CoA])" representing phosphate and CoA-thioester pseudo-groups.

Every pathway shown in the results section is the **shortest representative** of a pathway class. A **pathway class** is defined by three metabolites: the last C2 intermediate before the key reaction, the compound that is the product of the key reaction and the final compound of the pathway, a Calvin cycle intermediate. A **key reaction** is a reaction that changes the the number of carbons from substrate to product and is either a carboxylase, carbo-ligase or a transferase type reaction. In all pathways shown below, there is only one key reaction (pathways with more than one key reaction can of course also be generated with pathSeekR). A **pathway superclass** is defined by the number of carbons of the molecules before and after the key reaction(s). As the name suggests, many pathway classes fall into one superclass.

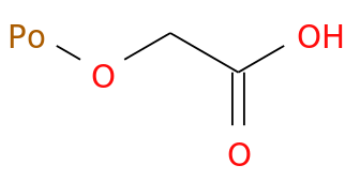
Co-products and **co-substrates** of the key reaction (excluding co-factors) are listed in the results section. It is assumed here that for transferase-type reactions such as transketolase, donor-acceptor pairs exist in the Calvin cycle that can regenerate the acylated enzyme intermediate. For all aldolase reactions, the co-substrate is either a Calvin cycle intermediate or a compound that is one reaction removed from the Calvin cycle (excluding aldolase and acyl-transferase type reactions).

1.9 Reactive C2 intermediates

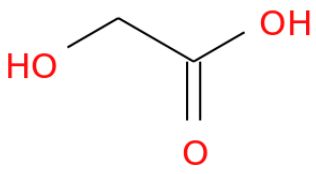
We define a **reactive C2 intermediate** as a two-carbon compound with either an aldehyde or amine group. Reactive C2 intermediates are the substrates of key reactions and the first defining compound of a pathway class. Within the restricted set of compounds that are allowed in our analysis (most notably, at least one heteroatom per carbon), reactive C2 intermediates are glycolyl-CoA, glycolaldehdye, glyoxylate, glycine, ethanolamine and aminoacetaldehyde. Other reactive C2 compounds, e.g. glyoxal, are excluded by our rules for compounds (only one carbonyl per molecule).

In the following we enumerate the pathways that generate reactive C2 intermediates from glycolate 2-phosphate. For example, there are several possibilities for producing glycolaldehyde that are all listed here. In the results section, wherever glcyolaldehyde is the reactive C2 intermediate, only one variant of the glycolaldehyde-generating pathways will be shown for reasons of clarity.

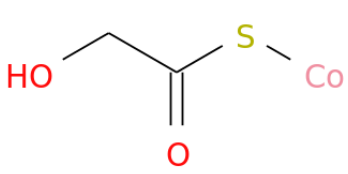
C2 reactive intermediate Pathway 1: Glycolyl-CoA



1: glycolate 2-phosphate

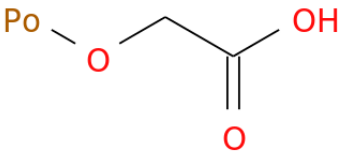


2: glycolate

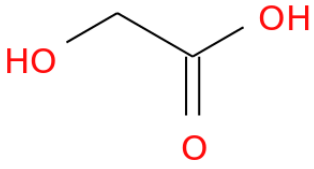


3: glycolyl-CoA

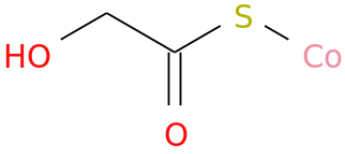
C2 reactive intermediate Pathway 2: Glycolaldehyde



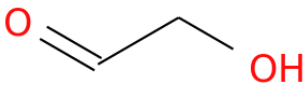
1: glycolate 2-phosphate



2: glycolate

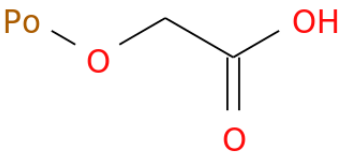


3: glycolyl-CoA

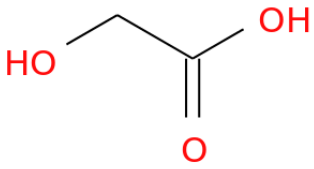


4: glycolaldehyde

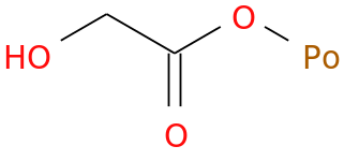
C2 reactive intermediate Pathway 3: Glycolaldehyde



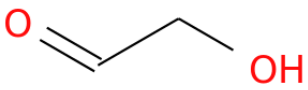
1: glycolate 2-phosphate



2: glycolate

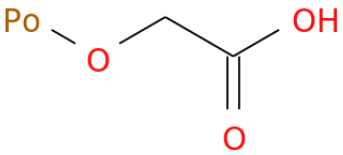


3: glycoloyl phosphate

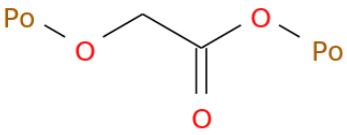


4: glycolaldehyde

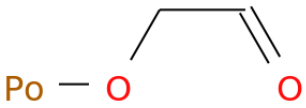
C2 reactive intermediate Pathway 4: Glycolaldehyde phosphate



1: glycolate 2-phosphate

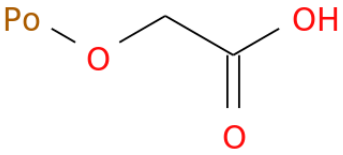


2: glycolate bisphosphate

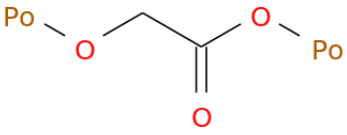


3: glycolaldehyde 2-phosphate

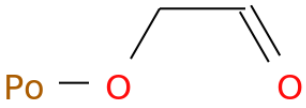
C2 reactive intermediate Pathway 5: Glycolaldehyde



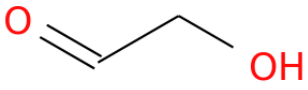
1: glycolate 2-phosphate



2: glycolate bisphosphate

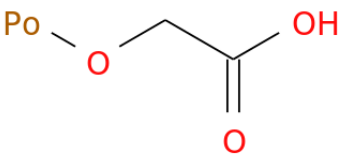


3: glycolaldehyde 2-phosphate

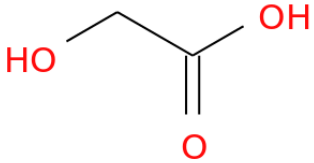


4: glycolaldehyde

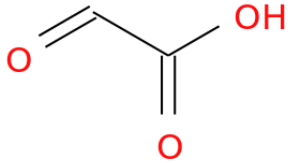
C2 reactive intermediate Pathway 6: Glyoxylate



1: glycolate 2-phosphate

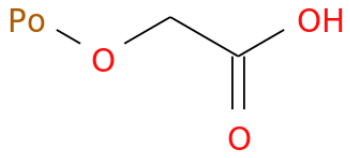


2: glycolate

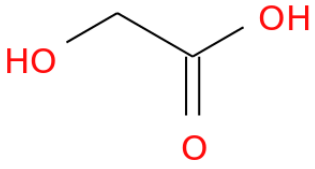


3: glyoxylate

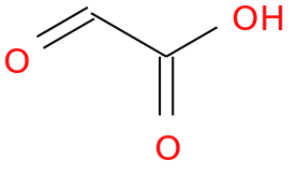
C2 reactive intermediate Pathway 7: Glycine



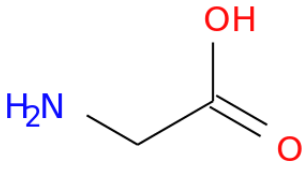
1: glycolate 2-phosphate



2: glycolate

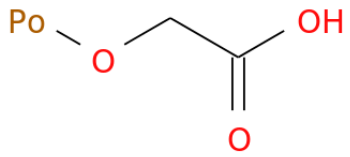


3: glyoxylate

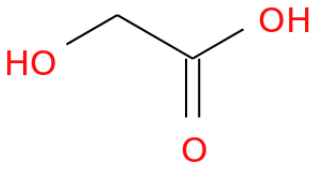


4: glycine

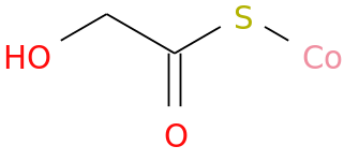
C2 reactive intermediate Pathway 8: Ethanolamine



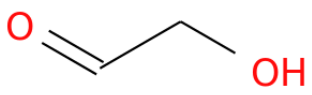
1: glycolate 2-phosphate



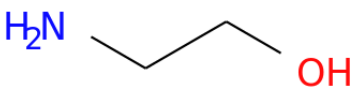
2: glycolate



3: glycolyl-CoA

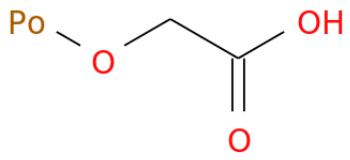


4: glycolaldehyde

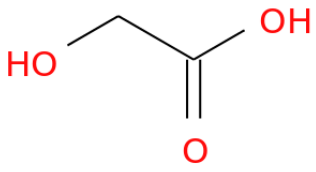


5: aminoethanol

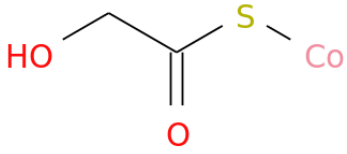
C2 reactive intermediate Pathway 9: Aminoacetaldehyde



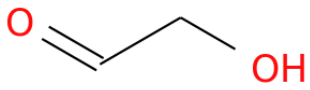
1: glycolate 2-phosphate



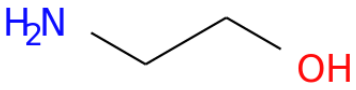
2: glycolate



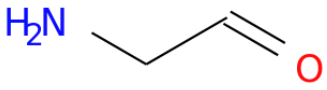
3: glycolyl-CoA



4: glycolaldehyde

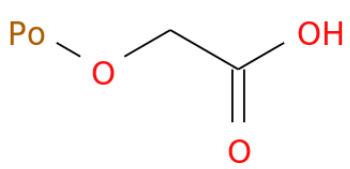


5: aminoethanol

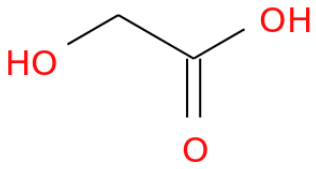


6: aminoacetaldehyde

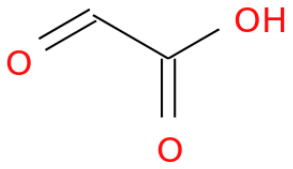
C2 reactive intermediate Pathway 10: Aminoacetaldehyde



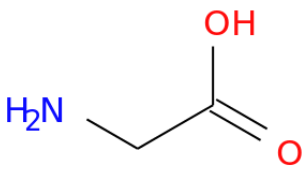
1: glycolate 2-phosphate



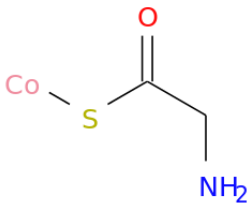
2: glycolate



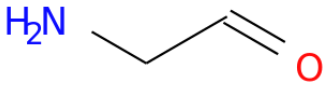
3: glyoxylate



4: glycine



5: glycyl-CoA



6: aminoacetaldehyde

1.10 Pathway architectures

The following results combine the output of several pathSeekR runs (159 pathway classes):

1. Glycolyl-CoA as source. 23 pathway classes
2. Glycolaldehyde 2-phosphate as source. Cosubstrate(s): glycolaldehyde. 21 pathway classes
3. Glycolaldehyde as source. Cosubstrate(s): glycolyl-CoA. 65 pathway classes
4. Glyoxylate as source. 26 pathway classes
5. Glycine as source. Cosubstrate(s): glyoxylate. 4 pathway classes
6. Aminoethanol as source. Cosubstrate(s): glycolyl-CoA, glycolaldehyde. 14 pathway classes
7. Aminoacetaldehyde as source. Cosubstrate(s): glycolyl-CoA, glycolaldehyde, aminoethanol. 6 pathway classes

The rationale behind this is that intermediates of the same pathway can react with each other, while cross-talk between different reactive C2-generating pathways, e.g glycine with glycolaldehyde are not allowed.

All pathway architectures are listed at the end of this document (Section 4). For each pathway class one example is shown that has the minimal number of reactions; however, there may be other representatives of that class with the same number of reactions and slightly different chemistry (e.g. carboxyl activation with CoA instead of phosphoanhydride prior to reduction to an aldehyde; e.g. Pathway 1) or with a permutation in the reaction sequence (e.g. phosphorylation before reduction instead of the other way round; see Pathway 3, where phosphorylation first would prevent ring closure of the sugar). Further selection of optimal representative of a the most promising pathway classes was carried out manually (see main text).

2 The stoichiometric-kinetic model

2.1 Theory

Our aim is to develop a framework for modeling C3 photosynthesis (PS) in mesophyll cells that allows us to compare native photorespiration (PR) with engineered photosynthetic shunts. In particular, we want to model conditions that are most relevant to agricultural crops, i.e. a range of light intensities and both ambient and low CO_2 intercellular airspace concentrations. The latter can for example occur in drought when the plant closes its stomata.

2.1.1 Pathways

The pathways we are concerned with are the Calvin cycle, native photorespiration, the published tartronic glycerate pathway [1] and the synthetic ribulose 1-phosphate (Ru1P), arabinose 5-phosphate (Ar5P), erythrulose (Eu), xylulose (Xu) and tartronyl CoA (TrCoA) shunts.

The analysis of energy requirements of photorespiration shunts is not straight-forward. Simply counting the number of ATPs and NADPHs that are consumed in a pathway can be misleading. For example, native photorespiration converts one molecule of G2P into half a molecule of PGA, consuming 1 ATP and 0.5 NADPHs in the process (0.5 ATP for re-fixing 0.5 units of ammonia, 0.5 ATP for phosphorylating 0.5 units of glycerate and 0.5 NADPH for reducing 0.5 units of hydroxypyruvate; 0.5

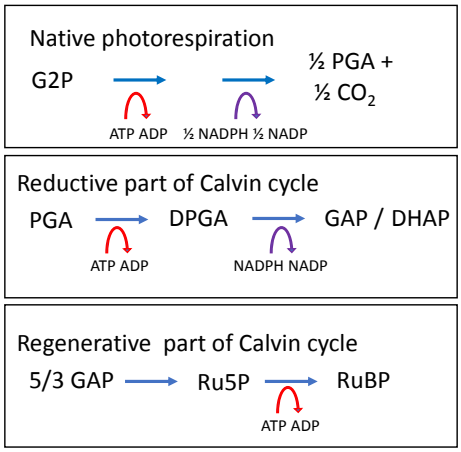


Figure 1: Energy requirements of native photorespiration and the Calvin cycle. Summary of energy consumption of native photorespiration, the reductive and regenerative Calvin cycle.

NADH are generated in the glycine cleavage complex but cancel with the 0.5 units of NADH that are required to regenerate the donor of the transaminase reaction, e.g. glutamate from 2-oxoglutarate). The difficulty arises if we try to compare this number to e.g. a carbon-neutral pathway like the Ru1P pathway. We can count the number of ATPs and NADPHs easily enough but it does not make any sense to compare them to native photorespiration because the product of the Ru1P pathway is RuBP, not PGA.

The solution is to calculate the number of ATPs and NADPHs that are required to regenerate one molecule of RuBP from the products of RuBP oxygenase, PGA and G2P. However, this raises the issue of how to deal with the fact that e.g. native photorespiration is 'carbon negative' - 0.5 units of CO_2 (i.e. 1/6 of a molecule of GAP) are lost, so we cannot regenerate a full molecule of RuBP. We deal with this by making the reasonable assumption that the missing amount of GAP is supplied by the Calvin cycle.

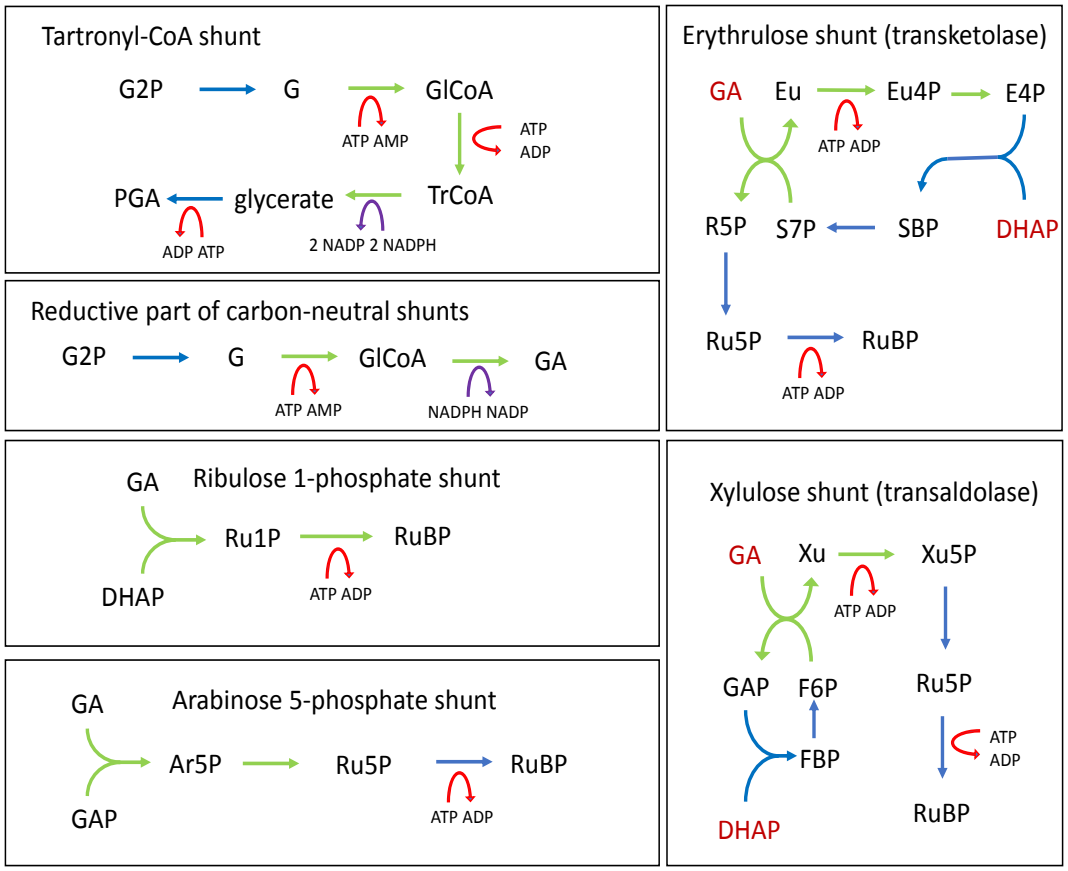


Figure 2: Energy requirements of synthetic photorespiration shunts. The carbon-positive tartronyl-CoA shunt converts G2P to PGA. The regenerative phase (from PGA to RuBP) is identical to the Calvin cycle. The four carbon-neutral shunts consist of a generic reductive phase, followed by individual regenerative phases. In the reductive phase, glycolaldehyde (GA) is made from glycolate 2-phosphate (G2P) and in the regenerative phase, GA is combined with either GAP or DHAP to regenerate RuBP. Green arrows indicate non-native reactions, and blue arrows native Calvin cycle or photorespiration reactions.

This description is extremely useful as it allows the comparison of any photorespiration shunt both with one another and native photorespiration. The energy requirements of native photorespiration are calculated in the following manner. G2P is converted to 0.5 PGA consuming 1 ATP and 0.5 NADPH in the process. Together with the molecule of PGA that is also produced by RuBP oxygenase, we have 1.5 PGA which require 1.5 ATP and 1.5 NADPH to form 1.5 molecules of GAP. Now the Calvin cycle supplies the missing 1/6 of a molecule of GAP so that 5/3 GAP + 1 ATP can be converted back to RuBP. All in all 3.5 ATPs and 2 NADPHs have been consumed to achieve this.

The same analysis for the tartronyl-CoA pathway is as follows: G2P is converted to PGA consuming 4 ATPs (2 by CoA synthetase, 1 by the carboxylase and 1 by glycerate kinase) and 2 NADPHs. Together with the molecule of PGA that is also produced by RuBP oxygenase, we have 2 PGA which require 2 ATP and 2 NADPH to form 2 molecules of GAP. The excess 1/3 of a GAP (equivalent to one fixed and reduced CO_2) is exported from the chloroplast so that 5/3 GAP + 1 ATP can be converted back to RuBP. All in all 7 ATPs and 4 NADPHs have been consumed to achieve this.

For the Ru1P, Ar5P, Erythrulose and Xylulose shunts the narrative is different: in the reductive part that all carbon-neutral shunts share, G2P is converted to GA consuming 2 ATPs and 1 NADPH. The molecule of PGA that is also produced by RuBP oxygenase, consumes 1 ATP and 1 NADPH to form

1 molecules of GAP or DHAP in the reductive part of the Calvin cycle. The two molecules are then combined, to form a pentose phosphate that is ultimately converted to RuBP. In the Ru1P shunt, DHAP and GAP form Ru1P in an aldolase reaction; Ru1P is phosphorylated to RuBP. All in all we have consumed 4 ATPs and 2 NADPHs. The Ar5P shunt is a variation on this theme, also consuming 4 ATPs and 2 NADPHs. In contrast, the Erythrulose shunt forms erythrulose in a transketolase reaction. Erythrulose is phosphorylated and isomerised to E4P. E4P is then condensed with DHAP to form SBP which is dephosphorylated to S7P, the donor of the transketolase reaction. Ultimately, what remains is one molecule of ribose 5-phosphate (R5P) which is converted to Ru5P and finally RuBP. Since an additional phosphorylation (erythrulose kinase) / dephosphorylation (SBPase) occurs, one more unit of ATP is consumed than in the Ru1P/Ar5P shunts. The Xylulose shunt also requires the additional expense of one ATP compared to Ru1P/Ar5P because the regeneration of F6P as the substrate of the transaldolase reaction proceeds via FBP.

2.1.2 The stoichiometric consumer model of photosynthesis

Thus, native photorespiration, each of the synthetic photorespiration shunts, and the Calvin Cycle proper, can be understood as isolated cycles that regenerate RuBP and either fix or release CO_2 in the process and thereby produce or consume triose phosphates at the same time (Fig. 3). From this viewpoint, native photorespiration is the process that converts RuBP and one sixth of a molecule of GAP back into one molecule of RuBP plus 0.5 molecules of CO_2, consuming 3.5 ATPs and 2 NADPHs in the process. The intermediates, e.g. PGA and glycolate-2-phosphate, are irrelevant in this description because they are made and consumed in equal amounts. Moreover, by balancing CO_2 with GAP in each of the pathways, we obtain catalytic cycles for photosynthesis, native photorespiration and synthetic photorespiration shunts that do not depend on one another to supply intermediates.

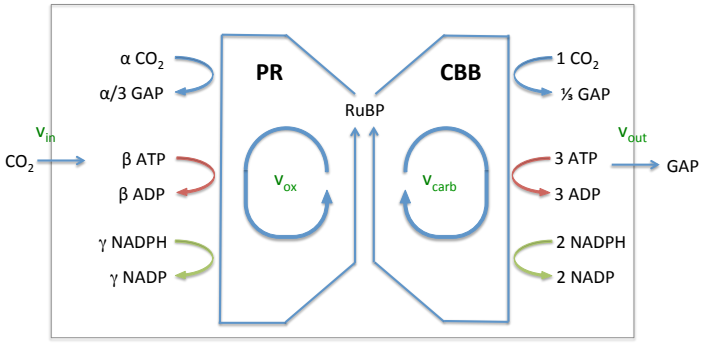


Figure 3: The consumer model of photosynthesis. The Calvin cycle (CBB, right) and photorespiration (PR, left) are conceptualised as catalytic loops that regenerate RuBP independently of each other. Three stoichiometric coefficients, (α, β and γ) fully define the properties of a pathway. α (can be negative) is the number of CO_2 molecules, β is the number of ATP molecules and γ is the number of NADPH molecules that are consumed per turn of the cycle (see Table 3). The total amount of CO_2, ATP or NADPH consumed can be calculated by multiplying the appropriate stoichiometric coefficient with the flux through RuBP oxygenase in photorespiration (v_{ox}) or RuBP carboxylase in the Calvin Cycle (v_{carb}). The sum of these two processes describes the total amount of CO_2, ATP or NADPH consumed. For example, total ATP consumption is $3\,v_{carb} + \beta\,v_{ox} = 3\,v_{carb} + 3.5\,v_{ox}$ in native photorespiration.

The most important consequence of this description is that despite the fact that native/synthetic photorespiration on the one hand and photosynthesis on the other share some enzymes and metabolic intermediates, the two processes can now be separated and treated in isolation.

Each catalytic pathway that regenerates RuBP is characterised by just three numbers: the number of CO_2 molecules that are consumed (α, can be negative) per turn of the cycle, the number of ATP molecules that are consumed (β) per turn of the cycle and the number of NADPH molecules that are consumed (γ) per turn of the cycle (Table 3). These numbers have to be obtained from the detailed description of the actual pathways(Fig. 1 and 2).

We call this model the consumer model of photosynthesis, since it omits a description of the supply of CO_2 (via diffusion), ATP and NADPH (from the light reactions). The consumer model assumes that supply rates of CO_2, ATP and NADPH are infinite and therefore not limiting. A supply-demand steady-state model is presented in Section 2.1.3.

When only the consumption (but not the supply) of CO_2 is taken into account, the net carbon fixation rate is defined as the sum of the activities of Rubisco carboxylase ($v^\dagger_{carb}$) and oxygenase ($v^\dagger_{ox}$), weighted by the parameters α_0 and α respectively.

$$A^\dagger = \alpha_0\, v^\dagger_{carb} + \alpha\, v^\dagger_{ox} = v^\dagger_{carb} + \alpha\, v^\dagger_{ox} \tag{1}$$

The number of carbons that are consumed per turn of the Calvin Cycle is $\alpha_0 = 1$, while per turn of the native photorespiration cycle, 0.5 units of CO_2 are released, which is accounted for in our notation as $\alpha = -0.5$. In contrast a carbon-neutral shunt, as the name suggest, has $\alpha = 0$, and a carbon-fixing shunt has $\alpha = 1$. We use the † superscript here and in the following to signify that Eq. 1 refers to the consumer model only. The corresponding rate A without superscript is described in the full model that also takes into account limitations imposed by the supply of CO_2, ATP and NADPH.

The overall number of ATP or NADPH molecules that are consumed is calculated in a similar way, simply by replacing α_0 and α with β_0 and β or γ_0 and γ in Eq 1, respectively.

$$N^\dagger_{atp} = \beta_0\, v^\dagger_{carb} + \beta\, v^\dagger_{ox} = 3\, v^\dagger_{carb} + \beta\, v^\dagger_{ox} \tag{2}$$

$$N^\dagger_{nadp} = \gamma_0\, v^\dagger_{carb} + \gamma\, v^\dagger_{ox} = 2\, v^\dagger_{carb} + \gamma\, v^\dagger_{ox} \tag{3}$$

The two numbers $N^\dagger_{atp}$ and $N^\dagger_{nadp}$ therefore describe the number of ATP and NADPH molecules that are consumed by the Calvin Cycle and a given photorespiration pathway, be it native or a synthetic shunt. These two numbers, like $A^\dagger$, depend on the rates $v^\dagger_{carb}$ and $v^\dagger_{ox}$ that depend in turn on the kinetic parameters of Rubisco and the concentrations of CO_2 and O_2 in the chloroplast.

Energy balance at a fixed oxygenation to carboxylation ratio

We describe three performance measures that allow the comparison of a synthetic shunt with native photorespiration when one or the other operates alongside the Calvin Cycle.

The first performance measure defines the number of units of CO_2 that are fixed on average per reaction of Rubisco. The other two describe the number of units of CO_2 that are fixed on average per molecule of ATP or NADPH consumed. These averages can be quickly derived if one assumes a fixed oxygenation proportion, $\rho = v^\dagger_{ox}/(v^\dagger_{carb} + v^\dagger_{ox})$, for example $\rho = 0.25$. This is a representative value

Pathway	α	β	γ
Native photorespiration	-0.5	3.5	2
Tartronic semialdehyde shunt	-0.5	3	1
Ribulose 1-P shunt	0	4	2
Arabinose 5-P shunt	0	4	2
Erythrulose shunt	0	5	2
Xylulose shunt	0	5	2
Tartronyl-CoA shunt	1	7	4
Calvin cycle	1	3	2

Table 3: Bypass coefficients. The coefficient α describes how many units of CO_2 are fixed per cycle, β and γ describe how many ATPs and NADPHs respectively are consumed in each cycle. The values of the Calvin cycle, denoted with the subscript '0', are $\alpha_0 = 1$, $\beta_0 = 3$ and $\gamma_0 = 2$.

at ambient CO_2 partial pressure, open stomata and saturating light. The value of $\rho = 0.25$ can be interpreted as meaning that, on average, every fourth reaction of Rubisco is oxygenating, while the three remaining ones are carboxylating (the ratio $v^\dagger_{ox}/v^\dagger_{carb} = 0.33$). First we calculate the number of CO_2 molecules that are fixed on average per Rubisco reaction. By 'average' we mean the expected number of fixed CO_2 molecules per reaction of Rubisco given that the probability of oxygenation is equal to ρ and the probability of carboxylation is equal to $1 - \rho$. To this end we divide $A^\dagger$ (Eq. 1) by the total activity of Rubisco, $v^\dagger_{carb} + v^\dagger_{ox}$, and use the definition of ρ:

$$\varepsilon^\dagger_{CO_2} = \frac{A^\dagger}{v^\dagger_{carb} + v^\dagger_{ox}} = \alpha_0 \cdot \frac{v^\dagger_{carb}}{v^\dagger_{carb} + v^\dagger_{ox}} + \alpha \cdot \frac{v^\dagger_{ox}}{v^\dagger_{carb} + v^\dagger_{ox}} = \alpha_0 \, (1 - \rho) + \alpha \, \rho \tag{4}$$

We call the performance measure $\varepsilon^\dagger_{CO_2}$ the carbon efficiency of the consumer model. It is positively correlated with α. For $\rho = 0.25$, $\varepsilon^\dagger_{CO_2}$ is 0.63 for native photorespiration and the glycerate shunt, 0.75 for the carbon-neutral and 1.0 for the carbon-fixing shunts. The carbon-neutral and carbon-fixing shunts are therefore at least 20 and 60% better, respectively, than native photorespiration with respect to carbon efficiency of the consumer model, i.e. given a fixed ratio ρ of oxygenation and no limitation by the supply of CO_2, ATP of NADPH.

Next we calculate the average number of ATPs per reaction of Rubisco that are consumed in photorespiration and the Calvin Cycle to regenerate RuBP. This is achieved by dividing $N^\dagger_{atp}$ (Eq. 2) by the total activity of Rubisco, $v^\dagger_{carb} + v^\dagger_{ox}$:

$$\nu^\dagger_{atp} = \frac{\beta_0 \, v^\dagger_{carb} + \beta \, v^\dagger_{ox}}{v^\dagger_{carb} + v^\dagger_{ox}} = \beta_0 \, (1 - \rho) + \beta \, \rho \tag{5}$$

This average in itself it not very useful because it is only meaningful in relation to how many units of CO_2 are fixed on average per reaction of Rubisco. Therefore, to arrive at ATP efficiency, i.e. the number of CO_2 fixed per ATP, we need to divide the carbon efficiency $\varepsilon^\dagger_{CO_2}$ by $\nu^\dagger_{atp}$:

$$\varepsilon^\dagger_{atp} = \frac{\varepsilon^\dagger_{CO_2}}{\nu^\dagger_{atp}} = \frac{\alpha_0 \, (1 - \rho) + \alpha \, \rho}{\beta_0 \, (1 - \rho) + \beta \, \rho} \tag{6}$$

ATP efficiency is a function of α, β and ρ. For $\rho = 0.25$, the ATP efficiencies of native photorespiration and glycerate shunt are 0.2 and 0.21 (i.e. 5.0 and 4.8 ATPs are needed to fix one CO_2) respectively. All synthetic shunts are more ATP efficient, the Ru1P/Ar5P shunts by 15% ($\varepsilon^{\dagger}_{atp} = 0.23$, 4.33 ATPs per CO_2 fixed) and the tartronyl-CoA shunt by 25% ($\varepsilon^{\dagger}_{atp} = 0.25$, 4.0 ATPs per CO_2 fixed). The Erythrulose and Xylulose shunts are as ATP efficient as the glycerate shunt. This shows that a higher ATP cost in the photorespiration shunts (i.e. $\beta \cdot \rho$) is more than compensated by the higher carbon efficiency.

If we follow the same procedure for NADPH instead of ATP, we arrive at

$$\nu^{\dagger}_{nadp} = \frac{\gamma_0\, v^{\dagger}_{carb} + \gamma\, v^{\dagger}_{ox}}{v^{\dagger}_{carb} + v^{\dagger}_{ox}} = \gamma_0\,(1-\rho) + \gamma\,\rho \tag{7}$$

and

$$\varepsilon^{\dagger}_{nadp} = \frac{\varepsilon^{\dagger}_{CO_2}}{\nu^{\dagger}_{nadp}} = \frac{\alpha_0\,(1-\rho) + \alpha\,\rho}{\gamma_0\,(1-\rho) + \gamma\,\rho} \tag{8}$$

For native photorespiration, glycerate, all carbon-neutral and tartronyl-CoA shunts $\varepsilon^{\dagger}_{nadp} = 0.31$, 0.36, 0.38 and 0.40. The synthetic shunts are 14, 20 and 28% more NADPH efficient than native photorespiration. In summary the consumer model shows that we can expect the synthetic shunts operating alongside the Calvin Cycle to be more energy and carbon efficient than native photorespiration/photosynthesis. Moreover, the carbon-neutral and carbon-fixing shunts are also more efficient than the glycerate shunt.

Rubisco kinetics

In all of the following, we assume that Rubisco is always saturated with RuBP and that the concentration of RuBP exceeds the concentration of Rubisco. Then

$$v^{\dagger}_{carb} = v_{carb_max}\,\frac{x}{x + k_3} \tag{9}$$

$$v^{\dagger}_{ox} = v_{ox_max}\,\frac{k_2}{x + k_3} \tag{10}$$

with the following definition for k_2 and k_3:

$$k_2 = k_c\,\frac{O}{k_o} \tag{11}$$

$$k_3 = k_c\,(1 + \frac{O}{k_o}) \tag{12}$$

The concentration of CO_2 at the site of carboxylation in the stroma (C_c), is here given the symbol x.

The parametrisation of Rubisco's carboxylase ($v^{\dagger}_{carb}$) and oxygenase ($v^{\dagger}_{ox}$) rates in the consumer model is as follows: k_c and k_o are the Michaelis constants for CO_2 and O_2 respectively, v_{carb_max} and v_{ox_max} are the maximal rates of carboxylase and oxygenase respectively. O is chloroplast O_2 concentration and is assumed to be constant. It is calculated with Henri's law at atmospheric oxygen partial pressure (see Section 2.1.4).

Since $v^{\dagger}_{carb}$ and $v^{\dagger}_{ox}$ have the same denominator, it is possible to express one in terms of the other.

$$v^{\dagger}_{ox} = v_{ox_max}\,\frac{k_2}{x+k_3}\cdot\frac{v_{carb_max}\cdot x}{v_{carb_max}\cdot x} = v_{carb_max}\,\frac{x}{x+k_3}\cdot\frac{v_{ox_max}\cdot k_2}{v_{carb_max}\cdot x} = v^{\dagger}_{carb}\cdot(\Lambda/x) \tag{13}$$

with

$$\Lambda = \frac{v_{ox_max}}{v_{carb_max}}\,k_2 = \frac{v_{ox_max}}{v_{carb_max}}\,\frac{k_c}{k_o}\,O \tag{14}$$

The † superscript indicates that we are still referring to the consumer model of photosynthesis.

2.1.3 Stoichiometric-kinetic model

We now extend the consumer model to a full steady state model that also takes the supply of CO_2, ATP and NADPH into account. At steady-state, for all three of these, the supply must equal the demand (consumption). This model is in many features very similar to the Farquhar, von Caemmerer and Berry (FvCB) model [2] that has been the cornerstone of most photosynthetic models in the past 30 years. However, we extend and generalise the model, both to our new synthetic shunts and to previously neglected limitations, namely limitations by the enzymatic activities of Calvin Cycle and photorespiration enzymes.

The overall carbon fixation rate A is a function of the environmentally controlled variables light irradiance (I) and intercellular carbon dioxide concentration (C_i). The latter is the CO_2 concentration in the intercellular airspaces inside the leaf that are connected to the outside via stomata (pores) on the leaf's surface. CO_2 diffuses from the atmosphere into intercellular airspaces and from there into the photosynthetic (mesophyll) cells. Plants can regulate the amount of CO_2 that diffuses into the intercellular airspaces (which is correlated to water loss) according to their needs (which can change with e.g. temperature and water status), by opening or closing their stomata. Thus, intercellular CO_2 concentration can vary considerably. At any given combination of I and C_i, A can be described as the minimum of six rates (we ignore limitation by product removal since this only occurs at very high carbon concentrations, well above physiological levels):

$$A = \min\{A_{rbc},\, A_{atp},\, A_{nadp},\, A_{cbb},\, A_{pr},\, A_{cbx}\} \tag{15}$$

The carbon fixation rate is either limited by Rubisco (A_{rbc}), light, or a second enzyme in either the Calvin Cycle, native photorespiration or the shunt replacing native photorespiration. Light limits both NADPH (A_{nadp}) and ATP (A_{atp}) production and in different circumstances one or the other

can become limiting. As to a second limiting enzyme, we differentiate between enzymes in the Calvin Cycle proper (A_{cbb}) or in native photorespiration / a photorespiration shunt (A_{pr}). Moreover, when a second carboxylating enzyme is present, as in the carbon-fixing shunt, we treat this enzyme separately (A_{cbx}) because its rate, unlike the others, depends on the concentration of CO_2 in the chloroplast.

In contrast to the FvCB model [2, 3], which is routinely applied to experimental data on photosynthesis, we prefer a 'bottom-up' approach, i.e. we determine chloroplast CO_2 concentration from quadratic equations and A is then calculated from the diffusion equation (Eq. 16; in the FvCB model this sequence is reversed). Since chloroplast CO_2 (C_c) has such a central role in our models we give it the symbol x.

$$\begin{aligned} A &= g_i\,(C_i - x) \\ x &= \max\{x_{rbc},\, x_{atp},\, x_{nadp},\, x_{cbb},\, x_{pr},\, x_{cbx}\} \end{aligned} \tag{16}$$

We omit the day respiration rate from our description since it only adds an offset to the apparent C_i. The indices of x have the same meaning as for A. In each case it implies a steady state where only the corresponding limitation is in force.

What is not explicitly stated in Eq. 15 is that CO_2 diffusion itself is co-limiting no matter what other limitation we impose because we will never assume (except earlier in the consumer model) that CO_2 supply is unlimited. This is the consequence of the finite and limiting value of g_i (in relation to v_{carb_max}) which is small enough to lead to a non-negligible difference between C_i and x, and x needs to be positive. Therefore, in the following, any specific limitation defined in Eq. 15 by implication also means CO_2 diffusion co-limitation.

Calculating steady-state chloroplast CO_2 concentration

At steady state, CO_2 diffusion into the chloroplast has to be equal to CO_2 fixation in the chloroplast.

$$A = g_i\,(C_i - x) = v_{carb} + \alpha\, v_{ox} \tag{17}$$

Equally,

$$v_{carb} = v_{ox} \cdot (x/\Lambda) \tag{18}$$

and the fundamental diffusion equation (Eq. 17) can be succinctly written as

$$A = g_i\,(C_i - x) = v_{carb}\,(1 + \alpha\,\Lambda/x) \tag{19}$$

or

$$A = g_i\,(C_i - x) = v_{ox}\,(x/\Lambda + \alpha) \tag{20}$$

Rubisco rate parametrisation

We have been careful to distinguish between A and $A^\dagger$, and also between v_{carb}, v_{ox} and its counterparts $v^\dagger_{carb}$, $v^\dagger_{ox}$, indicating either the supply-demand (no superscript) or the consumer model (with †) respectively. The reason we do this is to address a problem that becomes apparent if one expresses the steady-state model in terms of x, the chloroplast CO_2 concentration, and not in terms of A, as in the FvCB model. If we solve Eq. 17 assuming $v_{carb} = v^\dagger_{carb}$ and $v_{ox} = v^\dagger_{ox}$, i.e. the Rubisco parametrisation introduced earlier (Eqs 9 and 10)), we obtain a single solution for $x = x_{rbc}$ and fixed values of g_i and C_i as well as Rubisco kinetic constants. This implies that there can be no other steady state than $x = x_{rbc}$ if none of of these parameters change. On the other it is clear that other steady states must exist, e.g. when light is limiting. The FvCB model does not propose that the aforementioned parameters change (this is a justifiable simplification), and as a matter of fact it does not explain how other steady states are possible that simultaneously fullfil the diffusion equation Eq. 17 and the Rubisco rate equations Eq. 9 and 10. The conundrum is hidden rather than resolved in the FvCB model. The focus on A instead of x avoids the issue we describe here.

We, too, do not propose that parameters such as g_i and C_i need to change to make other steady states possible. Rather, we introduce a new parameter, the attenuation factor τ that scales down both the oxygenase and carboxylase activity of Rubisco such that steady states at lower carbon fixation rates (e.g. light limitation) become feasible. This can be easily understood as the down-regulation, inhibition or inactivation of Rubisco at low light. From this perspective, τ is the proportion of active enzyme remaining. Thus we define the oxygenase and carboxylase rate as

$$v_{carb} = \tau \cdot v_{carb_max} \frac{x}{x + k_3} = \tau \cdot v^\dagger_{carb} \tag{21}$$

$$v_{ox} = \tau \cdot v_{ox_max} \frac{k_2}{x + k_3} = \tau \cdot v^\dagger_{ox} \tag{22}$$

and

$$\begin{aligned} \tau = \frac{A}{A^\dagger} &= \frac{g_i\,(C_i - x)}{v^\dagger_{carb} + \alpha \cdot v^\dagger_{ox}} \\ &= \frac{g_i\,(C_i - x)}{v_{carb_max}\,\frac{x+\alpha\,\Lambda}{x+k_3}} \\ 0 \le \tau &\le 1 \end{aligned} \tag{23}$$

The introduction of τ makes steady states at net carbon fixation rates less than $A^\dagger$ possible. Importantly, this is not the frivolous introduction of a new parameter, but rather the necessary amendment of the FvCB model in which the light limited chloroplast CO_2 concentration cannot simultaneously fulfil the diffusion and the Rubisco rate equations. Far from constituting an additional degree of freedom, the the new parameter τ is fully determined in every limitation that the FvCB model deals with, as we shall see in the following. For each limitation there is a (x, τ) pair that fulfils all the equations.

Rubisco limited rate A_{rbc}

To begin with, we look at the situation where Rubisco is limiting. By this we mean the situation where there is unlimited supply of NADPH and ATP (e.g. at saturating light) and no other enzyme that is limiting. Therefore the steady state is a trade-off between CO_2 diffusion (supply) and Rubisco-dependent carbon fixation (demand) only. We assume under these conditions that Rubisco is fully activated i.e. $\tau = 1$ and $A = A^{\dagger}$. With the definitions of Eqs 17, 21, and 22, we obtain

$$\begin{aligned} A_{rbc} = g_i\,(C_i - x_{rbc}) &= \tau \cdot v_{carb_max}\,\frac{x_{rbc}}{x_{rbc}+k_3} + \tau \cdot \alpha\, v_{ox_max}\,\frac{k_2}{x_{rbc}+k_3} \\ &= v_{carb_max}\,\frac{x_{rbc}}{x_{rbc}+k_3} + \alpha\, v_{ox_max}\,\frac{k_2}{x_{rbc}+k_3} \\ &= v_{carb_max}\,\frac{x_{rbc}+\alpha\,\Lambda}{x_{rbc}+k_3} \end{aligned} \tag{24}$$

After rearranging Eq. 24 we obtain a quadratic equation that has a single positive solution (assuming all parameters other than x are fixed). In other words, the value $x = x_{rbc}$ is the only (positive) value of x that fulfils the above equation given that $\tau = 1$. The generic solution is

$$x = \frac{-b + \sqrt{b^2 - 4ac}}{2a} \tag{25}$$

with the following coefficients when solving for x_{rbc}.

$$\begin{aligned} a &= 1 \\ b &= \frac{v_{carb_max}}{g_i} + k_3 - C_i \\ c &= \alpha\,\frac{v_{carb_max}\,\Lambda}{g_i} - C_i\,k_3 \end{aligned} \tag{26}$$

The rate A_{rbc} is then calculated by substituting x_{rbc} into Eq. 17.

NADPH limitation

Light irradiance (I) is converted to electron flux (J) and this in turn is used to generate ATP and NADPH. The equations for the first part of this process [4], i.e. conversion of I to J, are described in the Section 2.1.4.

We now make the assumption that when NADPH is limiting a stoichiometric amount of NADPH is made from the available electron flux J, i.e. that the rate of supply of NADPH equals $J/2$ (two electrons make one NADPH). Since we know how many NADPHs are needed to sustain one turn of the Calvin Cycle (i.e. $\gamma_0 = 2$) and equally how many NADPHs are needed for one turn of a photorespiration cycle (depending on the pathway, e.g. $\gamma = 2$ in native photorespiration), we can calculate the rate of demand for NADPH as a function of v_{ox} and v_{carb}. In this manner we obtain a relationship between the light-dependent supply and the Rubisco-dependent demand for NADPH.

$$0.5\,J = \gamma_0\,v_{carb} + \gamma\,v_{ox} = v_{carb}\,(\gamma_0 + \gamma\,\Lambda/x_{nadp}) \tag{27}$$

On the left side of the equation we have the supply of NADPH. The factor of 0.5 reflects that two electrons are needed for the production of one NADPH. On the right side of the equation, the demand for NADPH is expressed in terms of the flux through the carboxylating and oxygenating cycle. $\gamma_0 = 2$ is the number of NADPHs consumed in the Calvin Cycle and γ is the number NADPHs consumed in the photorespiration shunt.

Next, we arrange Eq. 27, such that we can substitute v_{carb} with an expression containing only J, x_{nadp} and fixed parameters.

$$v_{carb} = 0.5\,J\,\frac{x_{nadp}}{\gamma_0\,x_{nadp} + \gamma\,\Lambda} \tag{28}$$

Clearly there is an upper limit of v_{carb} that can be expressed in terms of J:

$$\lim_{x_{nadp}\to\infty} v_{carb} = \frac{0.5\,J}{\gamma_0} = \frac{J}{4} \tag{29}$$

On substituting Eq. 28 into the diffusion equation (Eq. 17), we obtain

$$\begin{aligned} A_{nadp} = g_i\,(C_i - x_{nadp}) &= v_{carb} + \alpha \cdot v_{ox} \\ &= v_{carb} \cdot (1 + \alpha/x_{nadp}) \\ &= 0.5\,J\,\frac{x_{nadp}}{\gamma_0\,x_{nadp} + \gamma\,\Lambda} \cdot (1 + \alpha/x_{nadp}) \\ &= 0.5\,J\,\frac{x_{nadp} + \alpha\,\Lambda}{\gamma_0\,x_{nadp} + \gamma\,\Lambda} \\ &= \frac{J}{2\,\gamma_0} \cdot \frac{x_{nadp} + \alpha\,\Lambda}{x_{nadp} + \frac{\gamma}{\gamma_0}\,\Lambda} \end{aligned} \tag{30}$$

The similarities to Eq. 24 are evident. This also leads to a quadratic equation in x_{nadp} with the generic solution given in Eq. 25. Here the coefficients are

$$\begin{aligned} a &= \gamma_0 \\ b &= \frac{0.5\,J}{g_i} + \gamma\,\Lambda - \gamma_0\,C_i \\ c &= \alpha\,\frac{0.5\,J\,\Lambda}{g_i} - C_i\,\gamma\,\Lambda \end{aligned} \tag{31}$$

The rearrangement of Eq. 27 and the subsequent substitution of v_{carb} (Eq. 30), which is analogously done in the FvCB model, effectively makes τ disappear: The final equation does no longer contain the maximal rates of Rubisco, be they scaled by τ or not. That is why our model, with τ, is equivalent to FvCB without τ. However, our model allows us to back-substitute the steady-state solution x_{nadp} into the first line of Eq. 30 and then use the definition of Eq. 21 and 22:

$$
\begin{aligned}
A_{nadp} = g_i\,(C_i - x_{nadp}) &= v_{carb} + \alpha \cdot v_{ox} \\
&= \tau \cdot v_{carb_max}\,\frac{x_{nadp}}{x_{nadp} + k_3} + \alpha \cdot \tau \cdot v_{ox_max}\,\frac{k_2}{x_{nadp} + k_3} \\
&= \tau \cdot v_{carb_max}\,\frac{x_{nadp} + \alpha \cdot \Lambda}{x_{nadp} + k_3}
\end{aligned}
\tag{32}
$$

Here is the point where the FvCB model breaks because it implies $\tau = 1$. The absurd conclusion would be that there is only a steady-state where $x_{nadp} = x_{rbc}$ and $A_{nadp} = A_{rbc}$.

The value of τ can be calculated with Eq. 22

$$
\begin{aligned}
\tau &= \frac{g_i\,(C_i - x_{nadp})}{v_{carb_max}\,\frac{x_{nadp}+\alpha\,\Lambda}{x_{nadp}+k_3}} \\
&= \frac{0.5\,J\,\frac{x_{nadp}+\alpha\,\Lambda}{\gamma_0\,x_{nadp}+\gamma\,\Lambda}}{v_{carb_max}\,\frac{x_{nadp}+\alpha\,\Lambda}{x_{nadp}+k_3}} \\
&= \frac{0.5\,J \cdot (x_{nadp} + k_3)}{v_{carb_max} \cdot (\gamma_0\,x_{nadp} + \gamma\,\Lambda)}
\end{aligned}
\tag{33}
$$

The amount of down-regulation of Rubisco that τ represents depends on γ but not on α. Since the carbon-neutral pathway and native photorespiration have the same γ, the amount of down-regulation represented in τ is the same. However, their NADPH-limited assimilation rates A_{nadp} are not the same because of the higher carbon efficiency of the carbon-neutral pathway.

ATP limitation

ATP limitation works in exactly the same way as NADPH limitation with two differences: β replaces γ and the number of ATP molecules per electron is 0.75 (we assume that per electron, 3 protons are pumped and that 4 protons are required to make one ATP), not 0.5. Thus

$$
A_{atp} = g_i\,(C_i - x_{atp}) = 0.75\,J\,\frac{x_{atp} + \alpha\,\Lambda}{\beta_0\,x_{atp} + \beta\,\Lambda}
\tag{34}
$$

The solution to the quadratic is given in Eq. 25 with the coefficients

$$
\begin{aligned}
a &= \beta_0 \\
b &= \frac{0.75\,J}{g_i} + \beta\,\Lambda - \beta_0\,C_i \\
c &= \alpha\,\frac{0.75\,J\,\Lambda}{g_i} - C_i\,\beta\,\Lambda
\end{aligned}
\tag{35}
$$

The value of τ can be calculated with Eq. 22

$$\begin{aligned}\tau &= \frac{g_i\,(C_i - x_{atp})}{v_{carb_max}\,\frac{x_{atp}+\alpha\,\Lambda}{x_{atp}+k_3}} \\ &= \frac{0.75\,J\cdot(x_{atp}+k_3)}{v_{carb_max}\cdot(\beta_0\,x_{atp}+\beta\,\Lambda)}\end{aligned} \tag{36}$$

Limitation by a Calvin Cycle enzyme

The FvCB model considers only a subset of the limitations of supply-demand model, namely limitation by Rubisco or light at physiological C_i. Since this model is ubiquitously applied we define the restricted supply-demanded model fixation rate explicitly as

$$A^* = \min(A_{rbc},\, A_{nadp}, A_{atp}) \tag{37}$$

As this definition shows, the FvCB model assumes that none of the Calvin Cycle enzymes other than Rubisco or light are limiting carbon assimilation. This may not always be the case. For example, it has been shown that the activity of sedoheptulose-bisphosphatase (SBPase) can be rate limiting [5]. SBPase, like all other Calvin Cycle enzymes, also participates in native photorespiration because the molecules of PGA that are the outcome of native photorespiration also need to be converted into RuBP. In general, the flux of a given Calvin Cycle enzyme v_{cbb} can be expressed as

$$v_{cbb} = \eta_{ox}\,v_{ox} + \eta_{carb}\,v_{carb} \tag{38}$$

The values of η_{ox} and η_{carb} depend entirely on the stoichiometry of the Calvin Cycle and photorespiration or the synthetic photorespiratory shunt replacing it. They can be read off Table 4. For many enzymes $\eta_{ox} = \eta_{carb}$.

Parametrisation of a Calvin Cycle enzyme

We parametrize v_{cbb} with reversible Michaelis-Menten kinetics adopting the decomposition of [6]. Here the definitions are given for a uni-uni reaction but they can be easily applied to more complex reaction schemes.

$$v = v^+ \cdot \frac{S/K_s}{1 + S/K_s + P/K_p} \cdot (1 - \frac{P/S}{K'_{eq}}) \tag{39}$$

The three factors in this decomposition are the forward maximal rate , the fractional saturation and the thermodynamic driving force of the reaction. The maximal rate is defined by $v^+ = k^+_{cat} \cdot E$, where E is the total concentration of enzyme and k^+_{cat} is the maximal forward rate per unit of enzyme. S and P are the concentrations of substrate and product respectively, and K_s and K_p are their respective Michaelis constants. K'_{eq} is the ratio of P to S at equilibrium.

We now combine all three terms of Eq. 39 into a single parameter φ that combines saturation and driving force and expresses the maximal rate in units of the maximal carboxylase rate of Rubisco:

	η_{carb}	η_{ox}				
Pathway	Calvin cycle	Native PR[a]	TrCoA	Ru1P	Ar5P	Eu	Xu
Phosphoglycerate kinase	2	1.5	2	1			
GAP DH	2	1.5	2	1			
Sink	1/3	−1/6	1/3	0			
TPI	2/3			1	0	1	1
Aldolase	2/3			0	0	1	1
FBPase	1/3			0	0	0	1
Transketolase	2/3			0	0	1	0
SBPase	1/3			0	0	1	0
Isomerase	1/3			0	0	1	0
Epimerase	2/3			0	0	0	1
Phosphoribulokinase	1			0	1	1	1

Table 4: Flux distribtion of the Calvin cycle and photorespiratory bypasses. The table gives the stoichiometric coefficent η for each enzyme. The table gives the stoichiometric coefficent η for each enzyme, that has to be multiplied with the respective base rates, v_{carb} and v_{ox}. The total flux through an enzyme is the superposition of the fluxes originating from RuBP oxygenase and carboxylase (Eq. 38). Note that all pathways that channel G2P back to PGA have the same coefficients below the sink. The reason for this is that the entire flux goes via the regenerative part of the Calvin cycle. The carbon neutral shunts do not proceed in this manner, and there are differences within that group. The Ru1P and Ar5P shunts effectively bypass the majority of enzymes of the regenerative phase of the pentose phosphate cycle (i.e. their flux deriving from photorespiration is zero). Aldolase and transketolase catalyse two reactions each so their coefficient is the sum of the two reactions. Furthermore it is assumed that the Calvin cycle aldolase does not catalyse the aldolase reactions of the Ru1P and Ar5P shunts, but that transketolase does catalyse the transketolase reaction of the Eu shunt. [a] The values of η are the same for native photorespiration and the TSA shunt.

$$\varphi = \frac{v}{v_{carb_max}} = \frac{v^+ \cdot \frac{S/K_s}{1+S/K_s+P/K_p} \cdot (1 - \frac{P/S}{K'_{eq}})}{v_{carb_max}} \tag{40}$$

φ can be interpreted as the maximal rate scaled by the saturation of the enzyme and the thermodynamic driving force. expressed as a fraction of v_{carb_max}

Thus,

$$v_{cbb} = \varphi_{cbb} \cdot v_{carb_max} \tag{41}$$

The advantage of this approach is that all the unknowns (in particular substrate concentrations) are condensed into a single scaling factor φ_{cbb}. Whenever the total activity of a Calvin Cycle enzyme is known (e.g. from the literature), it can be used to remove the effect of v^+.

Generic solution

The Calvin Cycle enzyme limited carbon-fixation rate can be found by expressing v_{ox} in terms of v_{cbb} and substituting this result into a diffusion equation that is also expressed in terms of v_{ox} (of course we

could have used v_{carb} instead of v_{ox} in both cases). We are using the transformation $v_{carb} = v_{ox} \cdot x/\Lambda$.

$$\begin{aligned} v_{cbb} = \eta_{ox}\, v_{ox} + \eta_{carb}\, v_{carb} = v_{ox}\,(\eta_{ox} + \eta_{carb} \cdot x_{cbb}/\Lambda) \\ \Leftrightarrow\ v_{ox} = \frac{v_{cbb}}{\eta_{ox} + \eta_{carb} \cdot x_{cbb}/\Lambda} \end{aligned} \tag{42}$$

In the rearranged diffusion equation A is expressed in terms of v_{ox}:

$$A_{cbb} = g_i\,(C_i - x_{cbb}) = v_{carb} + \alpha\, v_{ox} = v_{ox} \cdot (x_{cbb}/\Lambda + \alpha) \tag{43}$$

Combining these two equation yields

$$A_{cbb} = g_i\,(C_i - x_{cbb}) = v_{cbb}\,\frac{x_{cbb}/\Lambda + \alpha}{\eta_{ox} + \eta_{carb} \cdot x_{cbb}/\Lambda} = v_{cbb}\,\frac{x_{cbb} + \alpha \cdot \Lambda}{\eta_{carb} \cdot\ x_{cbb} + \eta_{ox} \cdot \Lambda} \tag{44}$$

which is a quadratic in x_{cbb}. The only positive root is defined by the coefficients

$$\begin{aligned} a &= \eta_{carb} \\ b &= v_{cbb}/g_i + \eta_{ox} \cdot \Lambda - \eta_{carb} \cdot C_i \\ c &= \Lambda \cdot (\alpha \cdot v_{cbb}/g_i - \eta_{ox} \cdot C_i) \end{aligned} \tag{45}$$

Native photorespiration

On examination of Table 4, one observes that η_{ox} is equal to η_{carb} for a large subset of Calvin Cycle enzymes in native photorespiration and for all Calvin Cycle enzymes in the tartronyl-CoA pathway (but not in the carbon-neutral shunts). The subset of enzymes in native photorespiration consists of all enzymes beyond GAP (where the sink is situated) and includes all enzymes that are known to be poorly expressed, i.e. SBPase, FBPase, transketolase and aldolase [7, 8]. With this in mind, we can afford to ignore the general case $\eta_{ox} \neq \eta_{carb}$. Instead, for these two pathways we set $\eta_{ox} = \eta_{carb}$:

$$A_{cbb} = g_i\,(C_i - x_{cbb}) = v_{cbb}\,\frac{x_{cbb} + \alpha \cdot \Lambda}{\eta_{carb} \cdot\ x_{cbb} + \eta_{carb} \cdot \Lambda} = \frac{v_{cbb}}{\eta_{carb}} \cdot \frac{x_{cbb} + \alpha \cdot \Lambda}{x_{cbb} + \Lambda} \tag{46}$$

If $\alpha = -0.5$ as in native photorespiration, this is again a quadratic and the solution is given in Eq. 45.

Tatronyl-CoA shunt

However when $\alpha = 1$, the second fraction of Eq. 46 is equal to one and the solution is

$$A_{cbb} = v_{cbb}/\eta_{carb} \tag{47}$$

Therefore, in the tartronyl-CoA pathway, the rate of carbon fixation when a Calvin Cycle enzymes is limiting takes on a very simple form (Eq. 47) that is not dependent on C_i, i.e. it is always the same no matter what the CO_2 concentrations inside and outside the chloroplast are. In contrast, the rate of fixation of native photorespiration (Eq. 44) depends on the CO_2 concentrations x_{cbb} and C_i. The fixation rate A is proportional to the fraction $\frac{\Lambda+\alpha\, x_{cbb}}{\Lambda+x_{cbb}} < 1$ for any positive value of x_{cbb} and $\alpha = -0.5$. Thus, the carbon-fixing shunt always achieves a higher fixation rate A than native photorespiration at a fixed but limiting rate v_{cbb}.

The Ru1P/Ar5P carbon-neutral shunts

Here, Table 4 tells us that η_{ox} is not equal to η_{carb} for all enzymes. However, these carbon-neutral shunts redirects all flux resulting from photorespiration away from the regenerative part of the pentose phosphate pathway; η_{ox} is zero for all Calvin Cycle enzymes situated between the triose phosphates and Ru5P/RuBP. For these enzymes that, as stated earlier, are the ones most likely to be limiting in vivo, we arrive at a much simpler version of Eq. 44: if we substitute $\alpha = 0$ (carbon-neutral shunts) and $\eta_{ox} = 0$, we arrive at

$$A_{cbb} = g_i\,(C_i - x_{cbb}) = v_{cbb}\,\frac{x_{cbb} + 0\cdot\Lambda}{\eta_{carb}\cdot x_{cbb} + 0\cdot\Lambda} = v_{cbb}/\eta_{carb} \tag{48}$$

This is the same result we obtained for the carbon-fixing shunt. By the same token, the limitation by a Calvin Cycle enzyme in these carbon-neutral shunts is also independent of CO_2 concentrations and always higher than native photorespiration at a fixed and limiting value of v_{cbb}.

The Erythrulose/Xylulose carbon-neutral shunts

These pathways have have several things in common: flux through aldolase and one of the bisphosphatases (FBPase and SBPase). This is a case for the generic equation (Eq. 45).

Limitation by a photorespiration shunt enzyme

The argument for a photorespiration shunt enzyme is analogous to the previous section.

First we observe that v_{pr}, the flux a photorespiration enzyme has to carry, is equal to v_{ox} in all synthetic shunts.

In the tartronyl-CoA pathway, photorespiratory enzymes operate between phosphoglycolate and phosphoglycerate. Since the pathway is linear, it is easy to see that the flux at each step has to be equal to v_{ox}. In contrast, in the arabinose-5P pathway, 2PG is first converted to glycolaldehyde (GA) in a linear chain of reactions. The flux has to be equal to v_{ox}. In the next step, GA is stoichiometrically combined with GAP (which is produced by Calvin Cycle enzymes); the rate again has to be equal to v_{ox} if GA is to be produced and consumed at the same rate (i.e. at steady state). The amount of GAP that is produced from every RuBP oxygenase reaction is also equal to v_{ox}. The remainder of the pathway is linear, and has to be equal to v_{ox} (Table 4). Therefore the rate of every enzyme of the arabinose-5P pathway has to be equal to v_{ox}. The argument would be analogous for the other carbon-neutral shunts described in the main text.

Native photorespiration is more complicated: it is linear in character, however, the flux stoichiometry changes from 1 to 0.5 times the flux of v_{ox} at the decarboxylation step that is carried out by the glycine cleavage complex. Therefore we define

$$v_{pr} = \epsilon \cdot v_{ox} \tag{49}$$

where ϵ is always equal to one for all synthetic shunts, also equal to one for all enzymes in native photorespiration leading up to the decarboxylation reaction and 0.5 thereafter (and also the transaminases and the ammonia refixation reactions). To be conservative, i.e. to assume the conditions that are most favourable to native photorespiration and the glycerate shunt, we can simply use $\epsilon = 0.5$, so requirements for a photorespiration enzyme are only half that for an enzyme in a synthetic shunt.

Next we parametrise this enzyme with reversible Michaelis-Menten kinetics and simplify by introducing the scaling factor φ_{pr} in the same manner as we did for the Calvin Cycle enzyme (Eq.41).

$$v_{pr} = \varphi_{pr} \cdot v_{carb_max} \tag{50}$$

We combine the rearranged diffusion equation (Eq. 43) with results from the previous two equations:

$$\begin{aligned} A_{pr} = g_i\,(C_i - x_{pr}) &= v_{ox}\,(x_{pr}/\Lambda + \alpha) \\ &= v_{pr}/\epsilon \cdot (x_{pr}/\Lambda + \alpha) \\ &= \varphi_{pr} \cdot v_{carb_max} \cdot (x_{pr}/\Lambda + \alpha)/\epsilon \end{aligned} \tag{51}$$

Solving for x_{pr} yields

$$x_{pr} = \frac{C_i - \alpha \frac{\varphi_{pr} \cdot v_{carb_max}}{\epsilon\, g_i}}{1 + \frac{\varphi_{pr} \cdot v_{carb_max}}{\epsilon\, g_i\, \Lambda}} \tag{52}$$

Limitation by a photorespiratory shunt enzyme that is a carboxylase

We now investigate the more difficult case where the photorespiration shunt enzyme is a carboxylase, and hence its rate will depend on x directly. The argument is very similar to a non-carboxylating enzyme therefore we can use Eq. 49 without modification except that we call the rate of the carboxylating enzyme v_{cbx} and not v_{pr}. The reaction it catalysis is

$$S + CO_2 \rightarrow P \tag{53}$$

The difference appears in the parametrisation of the kinetic rate v_{cbx}:

$$v_{cbx} = v_{cbx_max}\,\frac{x}{x + K_{cbx}} \cdot \frac{S}{S + K_M} = \varphi_{cbx}\, v_{carb_max}\,\frac{x}{x + K_{cbx}} \tag{54}$$

As in the previous section, φ_{pr} is a scaling factor expressed in units of v_{carb_max} that include the saturation with all substrates other than CO_2, in this case the acceptor molecule of the carboxylation reaction. v_{cbx_max} is the maximal rate. K_{cbx} is the apparent Michaelis constant for CO_2. When the substrate of the carboxylase is HCO_3^-, we calculate K_{cbx} based on the assumption that CO_2 is in equilibrium with HCO_3^- due to the activity of carbonic anhydrase in the chloroplast. The carboxylation enzyme in the tartronyl-CoA pathway is indeed HCO_3^--dependent. The K_M of a typical carboxylases for HCO_3^- is in the range of $1\,mM$ [9]. Since the equilibrium constant for the hydration of carbon dioxide is about 100 in favour of HCO_3^- we arrive at $K_{cbx} = 1\,mM/100 = 10\,\mu M$ at a pH of 8.0 and an ionic strength of 0.25 which are the conditions in the stroma of the chloroplast (see Section 2.1.4).

Finally, we substitute Eq. 54 into the rearranged diffusion equation (Eq. 43). We also set $\alpha = 1$ since this only applies to the carbon-fixing shunt.

$$\begin{aligned} A_{cbx} = g_i\,(C_i - x) &= v_{ox}\,(x/\Lambda + 1) \\ &= v_{cbx}\;(x/\Lambda + 1) \\ &= \varphi_{cbx}\,v_{carb_max}\,(x/\Lambda + 1)\,\frac{x}{x + K_{cbx}} \end{aligned} \tag{55}$$

The equation is again quadratic in x and the generic solution is given in Eq. 25 with the coefficients

$$\begin{aligned} a &= 1 + \frac{\varphi_{cbx}\,v_{carb_max}}{g_i\,\Lambda} \\ b &= \alpha\,\frac{\varphi_{cbx}\,v_{carb_max}}{g_i} + K_{cbx} - C_i \\ c &= -C_i\,K_{cbx} \end{aligned} \tag{56}$$

2.1.4 Physicochemical parameters

Henri's law

The solubility of a gas like CO_2 or O_2 in water is given by Henri's law [10]

$$c_{aq} = p_{gas} \cdot k_H \tag{57}$$

where c_{aq} is the molar concentration of the gas in solution, p_{gas} is the partial pressure of the gas and k_H is Henri's constant. For CO_2, $k_H = 0.034\ M\ (atm)^{-1}$ at $25\,^\circ\,C$ (298.15 K). The partial pressure of CO_2 in the intercellular airspace when the stomate are open is about 220 ppm [11], therefore the equivalent concentration of CO_2 in water at $25\,^\circ\,C$ is 7.5 μM. The temperature dependence of k_H can be expressed in the form of the van't Hoff equation [10]:

$$k_H(T) = k_H(T_{ref}) \cdot e^{C\cdot(\frac{1}{T} - \frac{1}{T_{ref}})} \tag{58}$$

The constant $C = 2400\,K$ for CO_2; T is absolute temperature and T_{ref} is the reference temperature, 298.15 K. For example, at $30\,^\circ\,C$, k_H is 12% lower than at $25\,^\circ\,C$.

For O_2, Henri's constant is $k_H = 0.0013\,M\ (atm)^{-1}$ and the temperature dependency is $C = 1700\,K$.

The equilibrium constant and steady state concentration of HCO_3^-

It is assumed that the concentration of aqueous CO_2 (representing both the species of $CO_2(aq)$ and H_2CO_3) is in equilibrium with the gaseous phase (see Henri's law). Furthermore, dissolved CO_2 is assumed to be in equilibrium with HCO_3^- by the action of carbonic anhydrase. Then the concentration of HCO_3^- is given by

$$HCO_3^- = K_{eq} \cdot CO_2 \tag{59}$$

where K_{eq} is the equilibrium constant of the combined hydration and dissociation reaction

$$CO_2 + H_2O \rightleftharpoons HCO_3^- + H^+ \tag{60}$$

K_{eq} is often expressed on the log scale as a pK_a,

$$K_{eq} = 10^{-pK_a} \Leftrightarrow pK_a = -log_{10}(K_{eq}) \tag{61}$$

so that the well-known Henderson-Hasselbach [12] equation can be applied to find the concentration of HCO_3^- when pH, the pK_a and the concentration of CO_2 are known.

$$pH = pK_a + log_{10}\frac{HCO_3^-}{CO_2} \Leftrightarrow\ HCO_3^- = CO_2 \cdot 10^{pH-pK_a} \tag{62}$$

Calculating the pK_a

The pK_a for Eq. 60 is often over- or underestimated because it is either measured in pure water or seawater, while the chloroplast has a salinity and ionic strength between pure water and saltwater.

Empirical formulae for this pK_a have been developed, depending on (absolute) temperature T and salinity S. According to Roy et al.[13] the $ln(K_{eq})$ (not the decadic logarithm) is calculated with:

$$ln(K_{eq}) = a_1 + \frac{a_2}{T} + a_3\,ln(T) + (b_1 + \frac{b_2}{T})\,S^{0.5} + c\,S + d\,S^{1.5} \tag{63}$$

With the parameters

$$\begin{aligned} a_1 &= 2.83655 \\ a_2 &= -2307.1266 \\ a_3 &= -1.5529413 \\ b_1 &= -0.20760841 \\ b_2 &= -4.0484 \\ c &= 0.08468345 \\ d &= -.00654208 \end{aligned} \tag{64}$$

The pK_a on the more common negative decadic logarithm scale is

$$pK_a = log_{10}(exp(ln(K_{eq}))) \tag{65}$$

Salinity and ionic strength

The above formulae have been developed for and applied to seawater where salinity is well-defined. The term has some correspondence to the ionic strength (I) of intracellular fluids, however, since both salinity and ionic strength depend on the composition of the 'salts' in solution, again an empirical conversion has to be used [14]:

$$I = \frac{19.92\,S}{1 + 1.005\,S} \;\Leftrightarrow\; S = \frac{I}{19.92 + 1.005\,I} \tag{66}$$

Calculation of K_{eq} in the chloroplast

We assume that the pH in the chloroplast is 8.0 [15] and that ionic strength is 0.25 [16]. The latter corresponds to a salinity of 0.14 (roughly 40% of the salinity of seawater, Eq. 61). Applying Eq. 64, we obtain a pK_a of 5.98. Since there are close to two pH units between the pK_a and the pH, the equilibrium contant is $K_{eq} = 10^2 = 100$. This is the value we use in our calculations.

Mathematical model of the light reactions

The light reactions are modelled by the empirical equation [17]

$$J = \frac{I_2 + J_{max} - \sqrt{(I_2 + J_{max})^2 - 4\,\theta\,J_{max}\,I_2}}{2\,\theta} \tag{67}$$

where

$$I_2 = \frac{a\,(1-f)}{2} \cdot I \tag{68}$$

I_2 is the amount of light photosystem II absorbs; $a = 0.85$ is the absorptance of leaves, $f = 0.15$ is a correction for spectral quality. The denominator of two is a consequence of each photosystem absorbing half of the light. I is irradiance and $\theta = 0.7$ is a curvature factor (values taken from [3]. J_{max} is the maximal electron flux.

Light limitation

When light is limiting, the model is either ATP or NADPH limited (Eq. 34 and Eq. 30 respectively). However, at steady state the correct amounts of both ATP and NADPH that are dictated by the stoichiometry of the Calvin Cycle and photorespiration need to be produced. We take the approach that we assume that the amount of ATP that is produced is limiting, and that the excess of NADPH that would produced by linear electron flow at the required electron flux for ATP production is by some mechanism corrected to the necessary stoichiometric amount. It has been shown that in this scenario, the maximal capacity of the electron chain can be extrapolated by calculating $J_{max} = 2.3 \cdot v_{carb_max}$ [18]. Conversely, we could have used the approach that assumes that NADPH is limiting, in which case the ratio of J_{max} to v_{carb_max} would be close to 2.0. Both approaches give very similar results.

Carbon dioxide transfer conductances

We only use a single conductance because our synthetic pathways and the glycerate shunt are contained in a single compartment, the chloroplast. Including more conductances into the model would only affect native photorespiration, and in a negative manner. By using a single conductance we are being conservative in the sense that native photorespiration is described as being 100% efficient in recycling photorespiratory CO_2 from mitochondria to chloroplasts, which is unrealistic. As von Caemmerer and colleagues have shown, several conductances/resistances can be modelled, resulting in a worse performance of native photorespiration with respect to the glycerate shunt [3].

2.2 Results

In this Section, we will put the methodology that we established in the previous one into practice. To this end we need to put some actual numbers on the kinetics constants of Rubisco's rate equation and carbon dioxide diffusion that are representative of C3 photosynthesis.

2.2.1 Parameters and variables

The parameters listed in Table 5 are mostly identical to the ones von Caemmerer and colleagues have used in numerous publications [19, 17]. All partial pressures have been converted to molar concentrations. We note that the maximal carboxylation rate is close to four times the maximal oxygenation rate. The units of the rates are $\mu mole\ m^{-2}\ s^{-1}$, and the value of v_{carb_max} =80 $\mu mole\ m^{-2}\ s^{-1}$ corresponds to a k_{cat} of 3.5 s^{-1} and 2.3 mM Rubisco. The effective Michaelis constant for both the carboxylase and oxygenase reactions, k_3, is 18.7 μM. The value of Λ, which is related to the carbon-compensation point [19] of native photorespiration ($\Gamma_* = 0.5\,\Lambda$) is 2.6 μM.

Light irradiance and intercellular airspace CO_2 concentration are the key environmental variables that

determine the carbon fixation rate A. We use a light range between 200 and 1500 $\mu E\, m^{-2}\, s^{-1}$ (from 'low' to 'high' light). The latter is considered 'saturating', in the sense that A levels off well before that point. However, light irradiance can be as high as 2000 on a clear day in central Europe [20]. The intercellular airspace CO_2 concentration C_i is the concentration of CO_2 inside the leaf but outside the photosynthetic cells. Since diffusion of CO_2 from the atmosphere into the leaf is correlated to diffusion of water vapour out of the leaf, there are experimental methods to estimate C_i [11]. Atmospheric CO_2 (partial pressure of 400 ppm) corresponds to a concentration of $13.6 \mu M$ (Henri's law) and the C_i is considerably lower than that. Recently, careful measurements of C_i gave a value of about 220 ppm (7.5 μM) when the stomata were open and about 65 ppm (2.2 μM) when the stomata were closed [11]. Therefore, to cover the full range of physiologically possible values, we use a 'high' C_i value of 8 μM and a 'low' C_i value of C_i =2 μM.

The CO_2 transfer conductance from intercellular airspace to chloroplast (g_i) is also central to our model. We use a value of 9 $\mu mole\, m^{-2}\, s^{-1}\, \mu mole^{-1}$ which corresponds to 0.3 $\mu mole\, m^{-2}\, s^{-1}\, \mu bar^{-1}$, which is in the middle of the range that has been measured in plants [17]. A recent microscale simulation of photosynthesis also arrived at the same value of g_i [21].

2.2.2 Rubisco kinetics

The two equations that govern the carboxylase and oxygenase activity of Rubisco are given in Section 2.1.3. On plugging in the numbers from our parameter set, we obtain two curves that show the dependence of these rates on the chloroplast CO_2 concentration x (Fig. 4A). The oxygenase activity, $v_{ox}^{\dagger}$ only depends on x indirectly, as a competitive inhibitor of its substrate O_2 (which is constant). Therefore the curve falls moderately but monotonically with increasing x. In contrast, the carboxylation rate $v_{carb}^{\dagger}$ displays apparent Michaelis-Menten kinetics with respect to x. The the † superscript indicates the consumer model where x which determines ρ is a free variable. In the kinetic-stoichiometric model, x cannot be chosen freely but is fully determined by the limitations imposed on the model, e.g. CO_2 diffusion into the chloroplast. It monotonically increases with x. The apparent Michaelis constant k_3 =18.7 μM which takes into account competitive inhibition by O_2 is considerably higher than the

Name	Explanation	value	units
O	Chloroplast oxygen concentration	252	μM
x	Chloroplast CO_2 concentration	0 - 8	μM
C_i	Intercellular CO_2 concentration	2 - 8	μM
g_i	Transfer conductance	9.0	$\mu mole\, m^{-2}\, s^{-1}\, (\mu M)^{-1}$
v_{carb_max}	RuBP carboxylase V_{max}	80	$\mu mole\, m^{-2}\, s^{-1}$
k_c	Rubisco Michaelis constant for CO_2	8.6	μM
v_{ox_max}	RuBP oxygenase V_{max}	20	$\mu mole\, m^{-2}\, s^{-1}$
k_o	Rubisco Michaelis constant for O_2	215	μM
k_2	$k_2 = k_c\, O/k_o$	10.1	μM
k_3	$k_3 = k_c\, (1 + O/k_o)$	18.7	μM
Λ	$\Lambda = \frac{v_{ox_max}\, k_c}{v_{carb_max}\, k_o}\, O$	2.6	μM
I	Irradiance	200 - 1500	$\mu E\, m^{-2}\, s^{-1}$
v_{ox_max}/v_{carb_max}	Ratio of maximal velocities	0.255	
k_{cat}	Catalytic constant	3.5	s^{-1}

Table 5: Kinetic constants of Rubisco and CO_2 diffusion. The values are taken from [19, 17] and were converted from partial pressures in bar to molar concentrations. g_i is the transfer conductance of CO_2 from the intercellular airspace to the chloroplast.

upper limit of x ($x \leq C_i \leq 8\ \mu M$). Importantly, Rubisco is always sub-saturated with respect to CO_2, which limits its rate to less than 30 % of its nominal v_{max} (which is measured at saturating CO_2).

The net carbon fixation rate $A^\dagger$ is a linear combination of the rates $v^\dagger_{carb}$ and $v^\dagger_{ox}$. The contribution of $v^\dagger_{carb}$, i.e. the Calvin cycle, is always positive with respect to $A^\dagger$ (i.e. carbon-fixing) no matter which photorespiration pathway it is paired with. The contribution of $v^\dagger_{ox}$ can be negative (i.e. releasing CO_2, as in native photorespiration), neutral or positive. This is encapsulated in the parameter α of each pathway (Table 3). Fig. 4B graphically depicts the net fixation rates of the four photorespiration pathways when each is paired with the Calvin cycle. Each combination of photorespiration pathway with the Calvin cycle is a function of x, but since there is no interaction between the Calvin cycle and the photorespiration pathway or shunt, one can simply view the resulting carbon fixation rate $A^\dagger$ as a superposition of the two base rates $v^\dagger_{carb}$ and $v^\dagger_{ox}$: The carbon-fixing pathway paired with the Calvin cycle is the sum of the $v^\dagger_{carb}$ and $v^\dagger_{ox}$ and native photorespiration or glycerate shunt paired with the Calvin cycle correspond to $v^\dagger_{carb} - 0.5\, v^\dagger_{ox}$. A carbon-neutral shunt paired with the Calvin cycle is identical to $v^\dagger_{carb}$ since there is neither a positive nor a negative contribution from $v^\dagger_{ox}$.

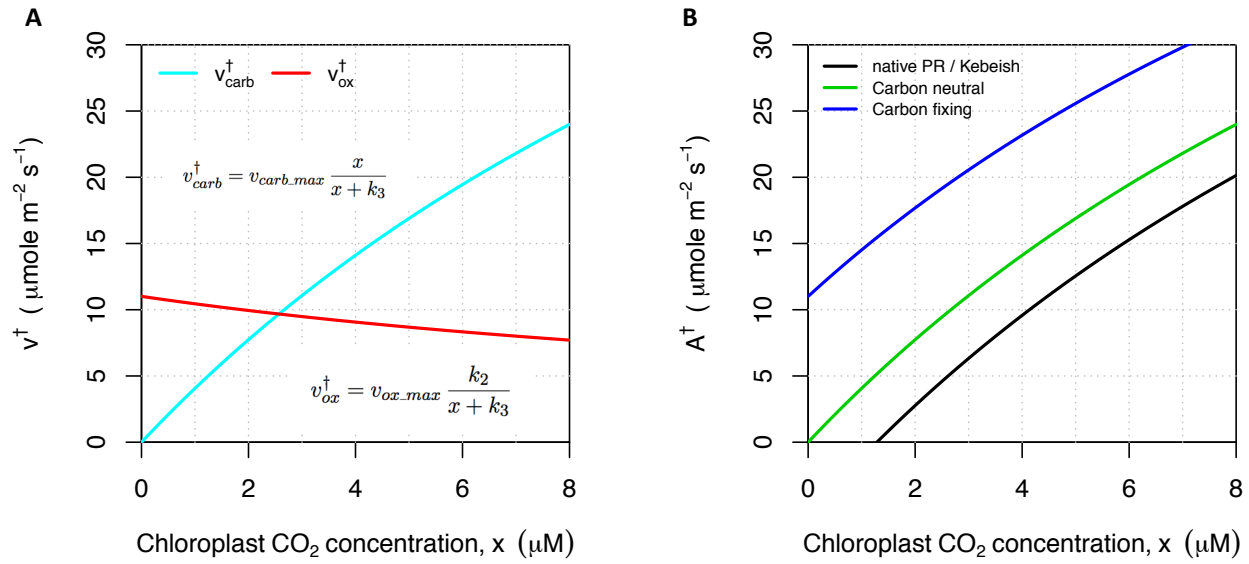


Figure 4: Rubisco kinetics and net carbon fixation rates in the consumer model. A: The rates of RuBP carboxylase ($v^\dagger_{carb}$, cyan) and RuBP oxygenase ($v^\dagger_{ox}$, red) are plotted as a function of chloroplast CO_2 concentration. The two rate equations are shown in the plot. B: Net carbon fixation rates of photosynthesis, i.e. the Calvin cycle paired with a photorespiration pathway as a function of chloroplast CO_2 concentration. The blue and green lines are the net fixation rates of the Calvin cycle paired with a carbon-fixing or a carbon-neutral shunts respectively (it applies equally to all four carbon-neutral shunts). Native photorespiration and Calvin cycle is shown as black line (the glycerate shunt gives identical results). The underlying assumption of the consumer model (Section 2.1.2) is that the rates are not limited by the supply of CO_2, ATP or NADPH. The kinetic parameters for these plots can be found in Table 5.

2.2.3 Limitation by light

The variable x is of central importance, as the curves in Fig 4 clearly show. The ratio of activities of either of our synthetic shunts to native photorespiration, i.e. the activity of a synthetic photorespiration shunt paired with the Calvin cycle expressed divided by the the activity of native photorespiration

paired with the Calvin cycle [1] at the same x, is much bigger at low C_i than at high x. Unfortunately, x cannot be controlled in an experiment or even directly measured and has to be inferred, i.e. calculated from a model. The variables that can be manipulated are light irradiance and C_i. Therefore it is common practice to measure the response curves of the net carbon fixation rate with respect to light and C_i [17].

The restricted kinetic-stoichiometric model, which is equivalent to the FvCB model [17] calculates the carbon fixation rate A^* using the assumption that only light and Rubisco are limiting. We will also use this assumption for the present time, but we will later introduce additional factors that can limit A (see Section 2.2.4). The light response curve of the restricted kinetic-stoichiometric model (Fig. 5) has two readily distinguishable phases, first rising monotonically with increasing irradiance and then reaching a plateau. The two phases correspond to light limitation (here via ATP) and limitation by Rubisco. The amount of Rubisco and the available CO_2 that diffuses into the cell define the upper limit A_{rbc} above which it does not matter if additional energy in the form of ATP and NADPH becomes available.

The light response curve has a different appearance at low $C_i = 2\mu M$ (Fig. 5B) compared to high $C_i = 8\mu M$ (Fig. 5A): The curves reach the plateau much earlier. In other words, Rubisco becomes limiting much earlier and this is because at low C_i the contribution of v_{ox} is much more important than at high C_i. The effect is two-fold: first, for pathways with negative α (native photorespiration and glycerate shunt), v_{ox} is subtracted from v_{carb}. Second, in pathways where α is non-negative, the much lower absolute rate of v_{carb} leads to the largest decrease in the fixation rate. For example, in the carbon-fixing shunt, the two rates, v_{carb} and v_{ox} are added 1:1. One may naively expect that in these circumstances the loss in v_{carb} might be compensated by the increase in v_{ox}. This is not the case: the maximal rate of v_{carb} is four times higher than v_{ox}, and also the fall in its activity is much sharper at low C_i than the corresponding rise in v_{ox}.

The differences between the different photorespiration pathways is immediately visible in their light response curves. The carbon-neutral shunts peaks later in the light curve than native photorespiration and glycerate shunt. The carbon-fixing shunt is just about Rubisco-limited at 'saturating light' (I =1500$\mu E\ m^{-2}\ s^{-1}$). The reason is not that the synthetic shunts use light more efficiently (which they do), but that their limitation by Rubisco/CO_2 only kicks in at a higher level (because $A_{rbc}^{fix} > A_{rbc}^{nat} > A_{rbc}^{nat}$, where the superscript indicate the carbon-fixing shunt, carbon-neutral shunts and native photorespiration respectively). The higher A_{rbc}, the less it is limiting, and the curve is dominated by ATP-limitation. Thus our first conclusion is that our synthetic pathways should lead to higher net fixation rates (for the present time ignoring limitation by enzymatic activities other than Rubisco). This result does not depend on Rubisco kinetic parameters but is largely a consequence of the non-negativity of α. The second conclusion is that the advantage of the synthetic pathways should be greater the lower C_i falls.

2.2.4 Limitation by enzyme activities other than Rubisco

The FvCB steady-state model assumes that light and Rubisco/CO_2 are the only factors that limit the net carbon assimilation rate. In our mathematical framework, we can in addition treat three more limitations (see Section 2.1.3).

[1] Since we are only concerned with situations where a photorespiration pathway operates alongside the Calvin cycle, and never without it, we will hereafter no longer explicitly write 'paired with the Calvin cycle' whenever we mention 'the activity' or 'the fixation rate' of a photorespiration pathway.

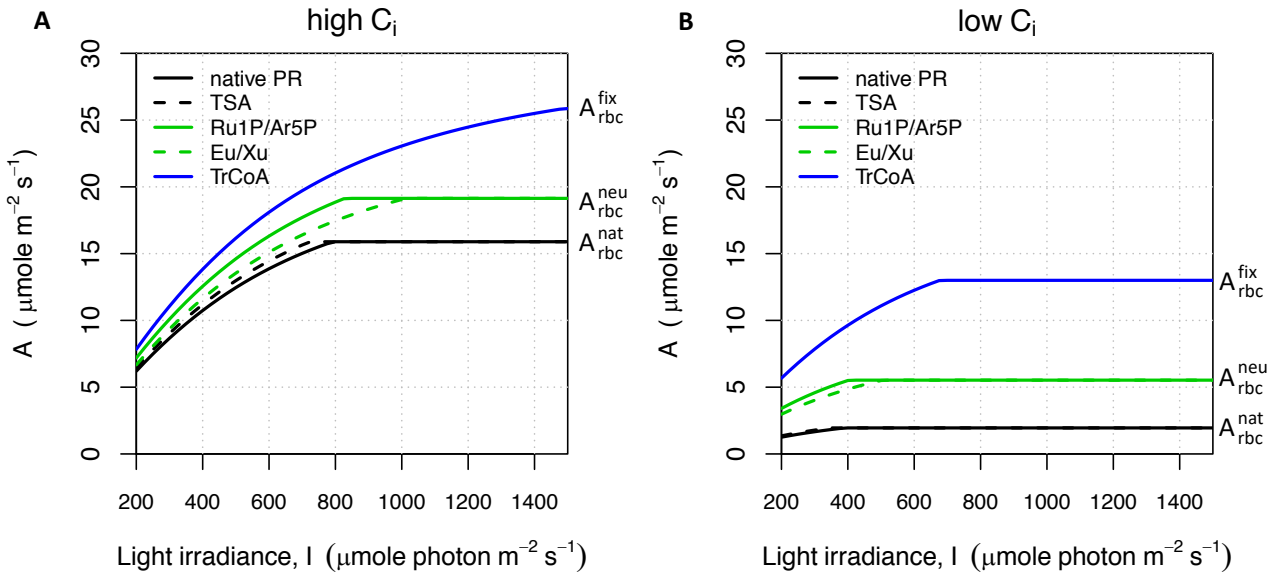


Figure 5: Light response curves of the restricted kinetic-stoichiometric model. A: Light response curves at high intercellular CO_2 concentration (C_i =8 μM). B: Light response curves at low intercellular CO_2 concentration (C_i =2 μM). Net carbon fixation rates of photosynthesis, i.e. the Calvin cycle paired with a photorespiration pathway, are plotted against light irradiance. In the restricted kinetic-stoichiometric model, the net fixation rate A^* is the minimum of the Rubisco-limited and the light-limited rate (here: ATP limited). The five pathways (native photorespiration, glycerate shunt, Ru1P/Ar5P shunt, Eu/Xu shunt, carbon-fixing shunt) are coloured according to the legend. The parameters for these plots can be found in Table 5. The values of the Rubisco-limited rate of the carbon-fixing shunt (A_{rbc}^{fix}), carbon-neutral shunts (A_{rbc}^{neu}) and native photorespiration (A_{rbc}^{nat}) are indicated.

2.2.5 Limitation by a Calvin cycle enzyme

It is known that Calvin cycle enzymes other than Rubisco can limit photosynthesis. The best known example is sedoheptulose bisphosphatase (SBPase) [5]. From our theoretical analysis we know that this limitation would be more severe for native photorespiration than for the synthetic shunts. In native photorespiration, SBPase activity must carry one third of the flux generated by v_{carb} and one third of the flux generated by v_{ox}. This is the same as in the carbon-fixing shunt. However, the consumer model tells us that native photorespiration fixes only 2.5 carbons in the same time that the carbon-fixing shunt fixes 4 (i.e. the carbon-fixing shunt is 60% more carbon efficient at high C_i and saturating light). Therefore, with a fixed amount of Calvin cycle enzyme activity, a higher carbon fixation rate is achieved with the carbon-fixing shunt.

In contrast, the Ru1P/ Ar5P carbon-neutral shunts have a different mechanism of reducing the load of a potentially limiting Calvin cycle enzymes such as SBPase: the flux originating from photorespiration is completely redirected past all Calvin cycle enzymes that lie sequentially behind GAP (Fig. 2). Careful analysis (Section 2.1.3) shows that the SBPase-limited rate would be same in the carbon-fixing and these carbon-neutral shunts and that this Calvin cycle-limited rate (A_{cbb}) is independent of the C_i. This analysis applies equally to FBPase, aldolase and transketolase (for all of these $\eta_{carb} = \eta_{ox}$ in native photorespiration and the carbon-fixing shunt and $\eta_{ox} = 0$ in the Ru1P/Ar5P carbon-neutral shunts, see Table 4).

To illustrate the effect of a limiting Calvin cycle enzyme, we define the scaling factor φ_{cbb} (Section 2.1.3)

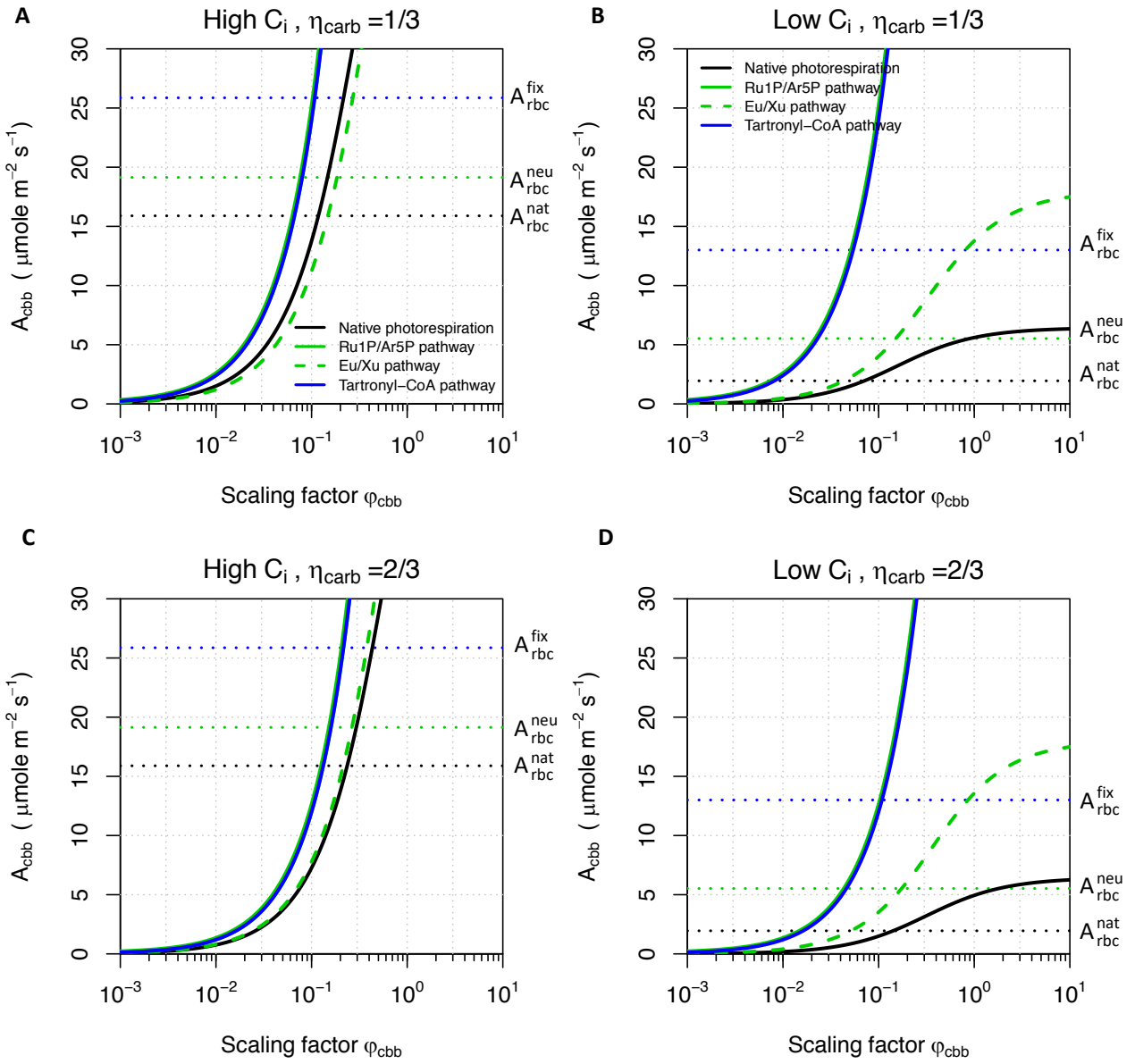


Figure 6: Limitation by a Calvin cycle enzyme. A and C: Carbon fixation rates assuming only a Calvin cycle enzyme activity is limiting (A_{cbb}), at high intercellular CO_2 concentrations (C_i =8 μM). B and D: A_{cbb} at low intercellular CO_2 concentrations (C_i =2 μM). The scaling factor φ_{cbb} expresses the activity of the Calvin cycle enzyme as the fraction of the maximal activity of RuBP carboxylase ($v_{cbb} = \varphi_{cbb} \cdot v_{carb_max}$). The curve shown in panels A and B belongs to an enzyme that carries one third of the Calvin cycle flux ($\eta_{carb} = 1/3$), such as SBPase and FBPase. The curve shown in panels C and D belongs to an enzyme that carries two thirds of the Calvin cycle flux ($\eta_{carb} = 2/3$), such as aldolase and transketolase. Horizontal dotted lines represent the Rubisco-limited rate of the carbon-fixing shunt (A^{fix}_{rbc}, blue), carbon-neutral shunts (A^{neu}_{rbc}, green) and native photorespiration (A^{nat}_{rbc}, black, identical to glycerate shunt). The blue and solid green line are identical and are shifted slightly to the right to make them both visible.

that expresses the activity of a Calvin cycle enzyme in units of v_{carb_max}, the maximal carboxylation rate of Rubisco [2]. In Fig. 6, we plot the fixation rate A_{cbb} against φ_{cbb} for an enzyme like SBPase

[2] φ_{cbb} absorbs the v_{max} of an enzyme and all factors concerning the saturation with substrates and products and the thermodynamics driving force of the reaction [6]. Thus, a value of $\varphi_{cbb} = 0.1$ does not describe a single state, it could

($\eta_{carb} = 1/3$) and aldolase ($\eta_{carb} = 1/3$ - aldolase catalyses two reactions). We compare this to the pathway-dependent Rubisco-limited rate A_{rbc} (horizontal dotted lines). At the intersection of the A_{cbb} with A_{rbc} of the same pathway (e.g. the black solid line with the black dotted line), Rubisco becomes limiting, and at values of φ_{cbb} lower than this point, the Calvin cycle enzyme is limiting. We call the value of φ_{cbb} at the intersection point the threshold φ_{cbb}^{TH}. About 1/10 of the maximal activity of Rubisco is required for a Calvin cycle enzyme to be non-limiting in native photorespiration. At high C_i, also the carbon-fixing and carbon-neutral pathways require about 1/10 of v_{carb_max} to be at least as high as their respective A_{rbc}. In contrast, both synthetic pathways only require a much smaller fraction of v_{carb_max} at low C_i.

Although $\varphi_{cbb} \approx 1/10$ seems to be sufficient in all cases to support a pathway's Rubisco-limited rate, it is important to stress that the synthetic pathways are capable of supporting a higher absolute rate of carbon fixation than native photorespiration with the same φ_{cbb}. With a given amount of enzyme, the synthetic pathways achieve much higher rates of carbon fixation, especially at low C_i. In other words, our synthetic pathways are more parsimonious.

Activity	Full name	EC	Latzko	Peterkofsky	Normalised range
Rubisco	rubisco (CO_2)	4.1.1.39	0.9	150	1
PGAK	phosphoglycerate kinase	2.7.2.3	130	4500	30-140
GAPDH	GAP dehydrogenase	1.2.1.13	15	260	2-17
TPI	triose phosphate isomerase	5.3.1.1	110	1100	7-120
ALD	aldolase (F6P)	4.1.2.13	5.3	300	2-6
FBPase	fructose bisphosphatase	3.1.3.11	2.4	39	0.3-3
SBPase	sedoheptulose bisphosphatase	3.1.3.37	0.2	8	0.05-0.2
TK	transketolase	2.2.1.1	10	300	2-11
ISO	phosphopentose isomerase	5.3.1.6	16	510	3-18
EPI	phosphopentose epimerase	5.1.3.1	8.4	1600	9-11
R5PK	phosphoribulokinase	2.7.1.19	16	600	4-18

Table 6: Calvin cycle enzyme total activities. The datasets of Latzko [7] and Peterkofsky [8] were rounded to two significant digits. The activity of GAPDH is the geometric mean of two values in the original tables that were obtained with different co-factors. The normalised ranges (last column) were obtained by scaling each set separately with the activity of Rubisco. SBP aldolase activity was not measured. The units of Latzko and Peterkofsky are $\mu mole\,(\text{mg protein})^{-1}\,h^{-1}$ and $\mu mole\,(\text{mg chlorophyll})^{-1}\,h^{-1}$ respectively, while the combined activity is unitless.

In order to compare our theoretical results with measured activities of Calvin cycle enzymes in relation to Rubisco, we present two datasets from spinach in Table 6. Clearly the lowest v_{max} is the activity of SBPase (between 0.05 and 0.2 $\cdot v_{carb_max}$). Overall the results in Table 6 indicate that SBPase activity in spinach that is most likely to be limiting (in agreement with e.g. [5]). The average activity calculated from the two datasets is very close to $\varphi_{cbb} = 0.1$, the threshold value of native photorespiration that we calculated from our model. The activity of SBPase will in practice be lower than the total activity shown in Table 6 because the latter are measured at full saturation at maximal substrate saturation and thermodynamic driving force. Thus, there is very good agreement between the model and experimental evidence. In contrast, both of our synthetic shunts achieve higher rates of carbon fixation because their A_{cbb} curve is shifted to lower values of φ_{cbb}, in other words they can do more with less. In summary, native photorespiration (and the glycerate shunt) are considerably more vulnerable to the limiting activity of a Calvin cycle enzyme such as SBPase.

equally mean that the enzyme is saturated with substrate and the v_{max} is 1/10 of v_{carb_max}, or that the enzyme is only 10% saturated and the v_{max} is equal to v_{carb_max}. Maximal rates such v_{carb_max} are defined as $v_{max} = k_{cat}\,E_{tot}$, where E_{tot} is the total concentration (abundance) of enzyme.

2.2.6 Limitation by a photorespiratory enzyme

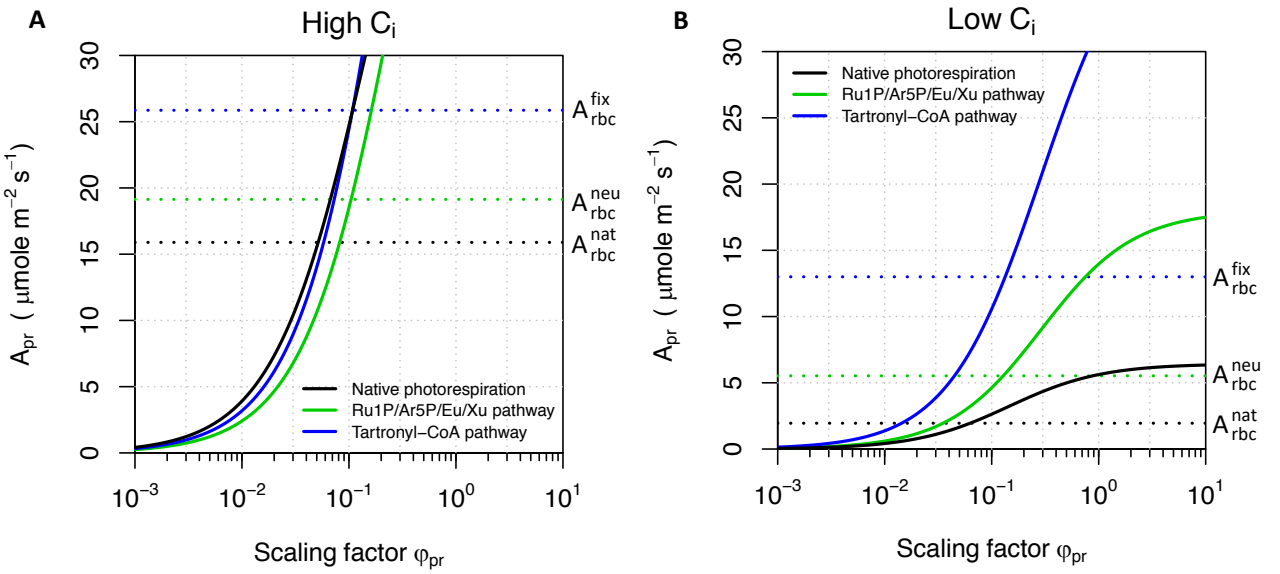


Figure 7: Limitation by a photorespiration enzyme. A: photorespiration (shunt) enzyme-limited carbon assimilation rates (A_{pr}) at high intercellular CO_2 concentrations (C_i =8 μM). B: photorespiration (shunt) enzyme-limited carbon assimilation rates at low intercellular CO_2 concentrations (C_i =2 μM). The scaling factor φ_{cbb} expresses the activity of the shunt enzyme as the fraction of the maximal activity of RuBP carboxylase ($v_{cbb} = \varphi_{cbb} \cdot v_{carb_max}$). The native photorespiration curve shown belongs to an enzyme that carries one half of photorespiration flux ($\epsilon = 1/2$), whereas in the two synthetic pathways the enzyme is assumed to carry all of v_{ox}. The glycerate shunt is identical to native photorespiration. Horizontal dotted lines represent the Rubisco-limited rate of the carbon-fixing shunt (A^{fix}_{rbc}, blue), carbon-neutral shunts (A^{neu}_{rbc}, green) and native photorespiration (A^{nat}_{rbc}, black, identical to glycerate shunt).

The enzymes of an synthetic pathway need to be expressed at a sufficient level to achieve the improvements calculated in Section 2.2.3. Equally, in native photorespiration, also the non-Calvin cycle enzymes need to be expressed in sufficient amounts to achieve a Rubisco or light-limited fixation rate. In the following we will calculate this level of activity relative to Rubisco's v_{carb_max}.

The application of this principle to the synthetic pathways is straight-forward since they consist of two linear branches that carry equal flux (since RuBP oxygenase creates equal amounts of PGA and G2P, 2-phosphoglycolate) and are eventually combined in a 1:1 condensation reaction. Thus all enzymatic steps carry the same flux, v_{ox}. In contrast, the description of native photorespiration is not as straightforward because the flux decreases from v_{ox} to $0.5\,v_{ox}$ in the middle of the G2P branch at the point of glycine condensation. We solve this problem by taking a conservative approach and assuming that in native photorespiration and the glycerate shunt, the limiting enzyme needs to only support $0.5\,v_{ox}$.

Theoretical consideration show that, at low C_i, the fixation rate A_{pr} of synthetic pathways is always higher than native photorespiration (and glycerate shunt). This is illustrated in Fig. 7B. However, at high C_i (Fig. 7A) and low values of φ_{pr} native photorespiration performs almost identically to the carbon-fixing shunt and slightly better than the carbon-neutral shunts. We again compare the value of φ^{TH}_{pr} at which the photorespiration-limited rate equals the Rubisco-limited activity: At high C_i this value for all pathways is between 1/10 and 1/20. At low C_i, the main difference is that the two synthetic pathways reach the level of A_{rbc} of native photorespiration much earlier, at $\varphi^{TH}_{pr} = 1/28$ and $\varphi^{TH}_{pr} = 1/69$ for carbon-neutral and carbon-fixing pathways. In summary, the synthetic pathways are

less likely to be limited by a PR enzyme at low C_i which is the more relevant condition. At high C_i all pathways are relatively similar in performance.

2.2.7 Limitation by a photorespiration carboxylase

The carbon-fixing pathway contains an enzyme that is a carboxylase. Unlike all other enzymes except Rubisco, this enzyme depends directly on chloroplast CO_2 concentration. For this reason, we separate the saturation with CO_2 from the saturation with all other metabolites (Eq. 54) which is described by φ_{cbx}. The effective Michaelis constant for CO_2 is expected to lie close to $10\,\mu M$ (see Section 2.1.4, equivalent to a K_M of 1mM for bicarbonate). However, the flux in photorespiration shunt depends on RuBP oxygenase. Therefore, the situation may become critical at low values of x where v_{ox} is moderately increased (disinhibited), but the rate of the carboxylase is severely reduced.

This is indeed what we see in Figure 8B: at low C_i the scaling factor φ_{cbx} needs to be 2.6 (i.e. the rate needs to be 2.6 times Rubisco's maximal carboxylation rate) to reach the Rubisco-limited rate of the carbon-fixing shunt. However, it is not necessary to reach to theoretical maximum of A to achieve an improvement. To reach the carbon-neutral's theoretical maximum only one third of v_{carb_max} is required, and to compete with A_{rbc} of native photorespiration, only one tenth. The values are considerably higher than for a non-carboxylase shunt enzyme (Fig 7B), where the three values were 1/9, 1/17 and 1/19. Thus, to achieve the same level of net carbon fixation as the carbon-neutral pathway, the carboxylase of the carbon-fixing pathway needs to have six times more activity than any non-carboxylase enzyme.

At high C_i (Figure 8A), the requirements for the rate of the carboxylase are also considerable: 1/3 of v_{carb_max} to match its own A_{rbc}, 1/5 of v_{carb_max} to match the A_{rbc} of the carbon-fixing pathway and 1/7 of v_{max} to match the A_{rbc} of native photorespiration. These values are much closer to the requirements of a non-carboxylase enzyme in either of the synthetic shunts. What is required to reap the full benefits of the carbon-fixing shunt is about 3 times the activity of SBPase.

3 Kinetic models of photorespiration shunts

3.1 Introduction

The purpose of kinetic modeling of photorespiration shunts is to gain understanding how the kinetic parameters of every enzyme influence both one another and the concentration of the metabolic intermediates. In particular, we want to find the kinetic parameters that would allow the pathway to carry the full photorespiratory flux while keeping all metabolic intermediates within a biologically plausible range.

For this purpose, every reaction of a photorespiration shunt is parametrized with reversible Michaelis-Menten kinetics (Section 3.3); Michaelis constants are taken from the literature (Section 3.4); missing values were substituted with database averages (Section 3.4.2), equilibrium constants are estimated with eQuilibrator (Section 3.4.3) and Calvin cycle metabolite and co-factor concentrations are taken from the literature (see Section 3.4.5). The rate equations are solved one-by-one (Section 3.5.2) and the solutions of a large number of simulations in which all parameters are randomly sampled (Section 3.4.6) lead to distributions of all metabolite concentrations. The fluxes are calculated with the kinetic-stoichiometric photosynthesis model (Section 3.4.9).

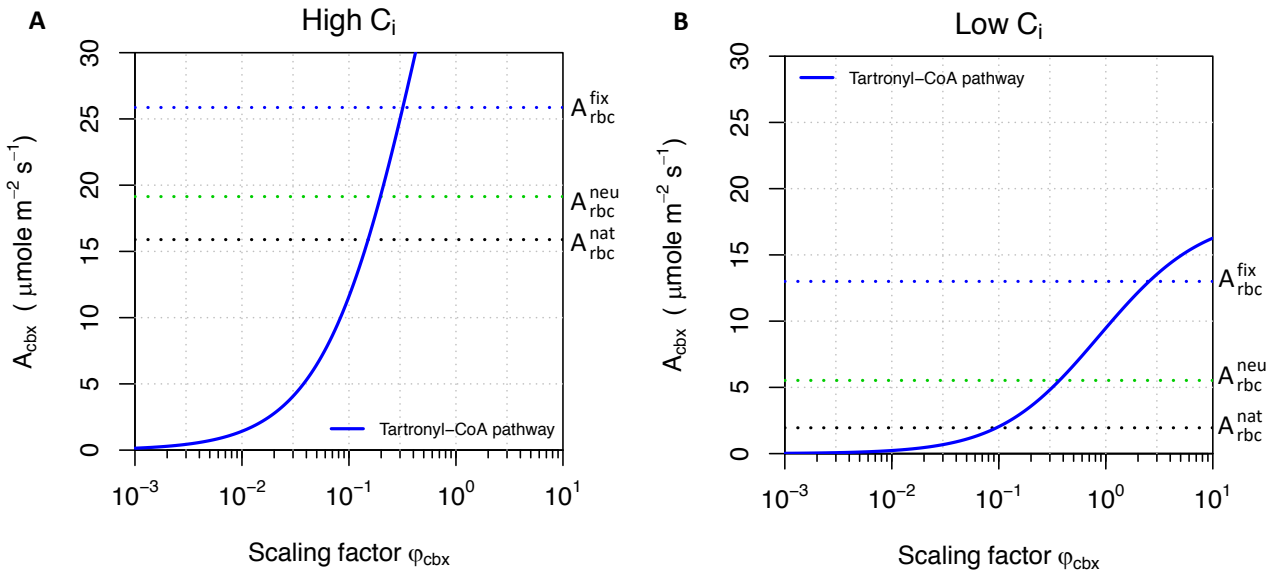


Figure 8: Limitation by a photorespiration carboxylase. A: photorespiration-carboxylase-limited carbon assimilation rates (A_{cbx}) at high intercellular CO_2 concentrations (C_i =8 μM). B: photorespiration-carboxylase-limited carbon assimilation rates at low intercellular CO_2 concentrations (C_i =2 μM). This enzyme only exists in the carbon-fixing pathway. It is assumed to have an apparent Michaelis constant for CO_2 of 10 μM (which corresponds to a K_M of 1 mM for HCO_3^-, see Section 2.1.3). The scaling factor φ_{cbx} expresses the activity of the carboxylase as the fraction of the maximal activity of RuBP carboxylase ($v_{cbx} = \varphi_{cbx} \cdot v_{carb_max}$). Horizontal dotted lines represent the Rubisco-limited rate of the carbon-fixing shunt (A_{rbc}^{fix}, blue), carbon-neutral shunts (A_{rbc}^{neu}, green) and native photorespiration (A_{rbc}^{nat}, black, identical to glycerate shunt).

Ultimately we identify optimal kinetic parameter pairs - the lowest k_{cat} at a given K_M - for each enzyme that are able to support a given flux through a photorespiration shunt at substrate and product concentrations within a biologically meaningful range. We call this a Pareto analysis (Section 3.5).

3.2 Generic form of a random order rate reaction with one substrate, one products and a variable number of co-factors

3.2.1 Abbreviations

$$
\begin{aligned}
= &\text{ Concentration of metabolite X} \\
K_{\mathrm{X}} = &\text{ Michaelis constant of metabolite X} \\
K_{\mathrm{X}}^{*} = &\text{ Apparent Michaelis constant of metabolite X} \\
V_{max} = &\text{ Maximal rate} \\
K'_{eq} = &\text{ Equilibrium constant (ratio of products over substrates at equilibrium)} \\
\kappa = &\text{ Co-factor adjusted equilibrium constant (dimensionless)} \\
\omega = &\text{ Co-factor saturation (dimensionless)} \\
v = &\text{ Flux (forward minus reverse rate) of a reaction. Assumed to be positive.}
\end{aligned}
\tag{69}
$$

The unit of all concentrations and Michaelis constants is mM. The unit if flux is mM/s.

3.2.2 Linear reactions

All concentrations of co-factors and Calvin cycle intermediates (see Table 8) are assumed to be constant, which for every reaction leaves a single concentration variable on either side of the reaction. We call these reactions linear because they give rise to a linear relation between the only remaining substrate and the only remaining product.

3.2.3 Generic example of a linear reaction

The example here is a random order bi-bi reaction where X and Y are a co-factor pair such as ATP and ADP.

$$\mathrm{A} + \mathrm{X} \rightleftharpoons \mathrm{B} + \mathrm{Y} \tag{70}$$

3.2.4 Canonical form of the equation

We want to define a canonical form of the rate equation that makes finding analytical solutions straightforward. This can be achieved in the following manner:

$$\begin{aligned} v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{A}] - [\mathrm{B}]/\kappa}{[\mathrm{A}] + K_\mathrm{A}^*} \\ \omega &= \frac{[\mathrm{X}]}{[\mathrm{X}] + K_\mathrm{X} \cdot (1 + [\mathrm{Y}]/K_\mathrm{Y})} \\ \kappa &= K'_{eq} \cdot \frac{[\mathrm{X}]}{[\mathrm{Y}]} \\ K_\mathrm{A}^* &= K_\mathrm{A} \cdot (1 + [B]/K_\mathrm{B}) \end{aligned} \tag{71}$$

The constants κ and ω are dimensionless and the apparent Michaelis constant for the substrate, K_A^* is in the same units as all Michaelis constants.

3.2.5 Linear relation between the concentrations of substrate and product

Given the concentration of product ([B]) one can straight-forwardly calculate the concentration of substrate ([A])

$$[\mathrm{A}] = \frac{(\frac{K_\mathrm{A}}{K_\mathrm{B}} + \frac{V_{max} \cdot \omega}{v \cdot \kappa}) \cdot [\mathrm{B}] + K_\mathrm{A}}{\frac{V_{max} \cdot \omega}{v} - 1} = \frac{K_\mathrm{A}^* + \frac{V_{max} \cdot \omega}{v \cdot \kappa} \cdot [\mathrm{B}]}{\frac{V_{max} \cdot \omega}{v} - 1} \tag{72}$$

This solution is always available if the rate equation can be expressed in the canonical form. We will show in the following that this is possible for all reactions in our pathways, under the above-mentioned assumptions (constant co-factor and Calvin cycle intermediate concentrations and known flux v).

3.2.6 Minimal required activity

The denominator of Eq. 72 implies that, to support a positive flux v, a minimum activity $\tilde{V}_{max}$ is required:

$$\begin{aligned} \frac{V_{max} \cdot \omega}{v} - 1 &> 0 \\ V_{max} &\geq \tilde{V}_{max} \\ \tilde{V}_{max} &= v/\omega \end{aligned} \tag{73}$$

If $\omega = 1$ the trivial result $\tilde{V}_{max} = v$ is obtained.

3.2.7 Thermodynamic limitations

Even if the reaction is kinetically feasible, i.e. when $V_{max} \geq v/\omega$, it can be thermodynamically infeasible. This is because we require the flux to be positive and thus

$$\begin{aligned} -[\mathrm{B}]/\kappa &> 0 \\ [\mathrm{B}]/\kappa &< [\mathrm{A}] \end{aligned} \tag{74}$$

This can become a problem if we set a - biological - upper limit on [A], e.g. an upper limit on [glycolaldehyde] of 1 mM because it would otherwise become toxic. If such a limit, $[\mathrm{A}]_{\mathrm{max}}$ exists, it effectively restricts the concentration of [B], too:

$$\begin{aligned} /\kappa &< [\mathrm{A}]_{\mathrm{max}} \\ [\mathrm{B}]_{\mathrm{max}} &= [\mathrm{A}]_{\mathrm{max}} \cdot \kappa \end{aligned} \tag{75}$$

The limit on [A] is a biological one, which necessarily leads to a thermodynamic limit on [B].

3.3 Rate laws of all photorespiration shunt reactions

In this section, we formulate the rate laws in the canonical form (Eq 71).

3.3.1 Compound abbreviations

$$
\begin{aligned}
\text{Ac} &= \text{acetate} \\
\text{AcCoa} &= \text{acetyl CoA} \\
\text{G} &= \text{glycolate} \\
\text{G2P} &= \text{glycolate 2-phosphate} \\
\text{GlCoA} &= \text{glycolyl-CoA} \\
\text{GA} &= \text{glycolaldehyde} \\
\text{GAP} &= \text{glyceraldehyde 3-phosphate} \\
\text{DHAP} &= \text{dihydroxyacetone phosphate} \\
\text{PGA} &= \text{glycerate 3-phosphate} \\
\text{TrCoA} &= \text{tartronyl-CoA} \\
\text{Eu} &= \text{erythrulose} \\
\text{Eu4P} &= \text{erythrulose 4-phosphate} \\
\text{Ar5P} &= \text{arabinose 5-phosphate} \\
\text{Ru1P} &= \text{ribulose 1-phosphate} \\
\text{RuBP} &= \text{ribulose 1,5-bisphosphate} \\
\text{Xu} &= \text{xylulose} \\
\text{Xu5P} &= \text{xylulose 5-phosphate} \\
\text{Ru5P} &= \text{ribulose 5-phosphate} \\
\text{R5P} &= \text{ribose 5-phosphate} \\
\text{F6P} &= \text{fructose 6-phosphate} \\
\text{S7P} &= \text{sedoheptulose 7-phosphate} \\
\text{P}_i &= \text{Inorganic phosphate} \\
\text{PP}_i &= \text{Inorganic pyrophosphate}
\end{aligned}
\tag{76}
$$

3.3.2 Glycolate 2-phosphatase

$$
\text{G2P} \rightleftharpoons \text{G} + \text{P}_i \tag{77}
$$

Rate law in canonical form

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{G2P}] - [\text{G}]/\kappa}{[\text{G2P}] + K^*_{\text{G2P}}} \\
\omega &= 1 \\
\kappa &= K'_{eq}/[\text{P}_i] \\
K^*_{\text{G2P}} &= K_{\text{G2P}} \cdot (1 + [\text{G}]/K_{\text{G}}) \cdot (1 + [\text{P}_i]/K_{\text{P}_i})
\end{aligned}
\tag{78}
$$

3.3.3 Glycolyl-CoA synthetase

Here we are assuming two sites on the enzyme, one for ATP, AMP and PP_i and the other for glycolate, glycolyl-CoA (GlCoA) and CoA.

$$\mathrm{G} + \mathrm{ATP} + \mathrm{CoA} \rightleftharpoons \mathrm{GlCoA} + \mathrm{AMP} + \mathrm{PP}_i \tag{79}$$

Rate law in canonical form

$$\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{G}] - [\mathrm{GlCoA}]/\kappa}{[\mathrm{G}] + K^*_{\mathrm{G}}} \\
\omega &= \frac{[\mathrm{ATP}]}{[\mathrm{ATP}] + [\mathrm{AMP}] + K_{\mathrm{ATP}} + [\mathrm{PP}_i] \cdot (1 + [\mathrm{AMP}]/K_{\mathrm{ATP}})} \cdot \frac{[\mathrm{CoA}]}{[\mathrm{CoA}] + K_{\mathrm{CoA}}} \\
\kappa &= \frac{K'_{eq} \cdot [\mathrm{CoA}] \cdot [\mathrm{ATP}]}{[\mathrm{AMP}] \cdot [\mathrm{PP}_i]} \\
K^*_{\mathrm{G}} &= K_{\mathrm{G}} \cdot (1 - (1 - \omega) \cdot [\mathrm{GlCoA}]/K_{\mathrm{CoA}})
\end{aligned} \tag{80}$$

3.3.4 Glycolyl-CoA transferase

$$\mathrm{AcCoA} + \mathrm{G} \rightleftharpoons \mathrm{GlCoA} + \mathrm{Ac} \tag{81}$$

This is a ping-pong mechanism.

Rate law in canonical form

$$\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{G}] - [\mathrm{GlCoA}]/\kappa}{K^*_{\mathrm{G}} + [\mathrm{G}]} \\
D &= [\mathrm{AcCoA}] + K_{\mathrm{CoA}} + [\mathrm{GlCoA}] \\
\omega &= [\mathrm{AcCoA}]/D \\
\kappa &= K'_{eq} \cdot [\mathrm{AcCoA}]/[\mathrm{Ac}] \\
K^*_{\mathrm{G}} &= K_{\mathrm{G}} \cdot \left(1 + [\mathrm{Ac}]/K_{\mathrm{Ac}} - K_{\mathrm{CoA}}/D\right)
\end{aligned} \tag{82}$$

3.3.5 Glycolaldehyde dehydrogenase, CoA-acylating (Glycolyl-CoA reductase)

$$\mathrm{GlCoA} + \mathrm{NADPH} \rightleftharpoons \mathrm{GA} + \mathrm{NADP} + \mathrm{CoA} \tag{83}$$

Rate law in canonical form

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{GlCoA}] - [\text{GA}]/\kappa}{[\text{GlCoA}] + K^*_{\text{GlCoA}}} \\
\omega &= \frac{[\text{NADPH}]}{[\text{NADPH}] + K_{\text{NADPH}} + [\text{NADP}]} \\
\kappa &= K'_{eq} \cdot \frac{[\text{NADPH}]}{[\text{NADP}] \cdot [\text{CoA}]} \\
K^*_{\text{GlCoA}} &= K_{\text{GlCoA}} \cdot (1 + [\text{GA}]/K_{\text{GA}}) \cdot (1 + [\text{CoA}]/K_{\text{CoA}})
\end{aligned}
\tag{84}
$$

3.3.6 Ar5P aldolase

$$\text{GA} + \text{GAP} \rightleftharpoons \text{Ar5P} \tag{85}$$

In this reaction, GA is the donor and GAP is the acceptor.

Rate law in canonical form

We are assuming an ordered reaction mechanism.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{GA}] - [\text{Ar5P}]/\kappa}{K^*_{\text{GA}} + [\text{GA}]} \\
\omega &= \frac{[\text{GAP}]}{[\text{GAP}] + K_{\text{GAP}}} \\
\kappa &= K'_{eq} \cdot [\text{GAP}] \\
K^*_{\text{GA}} &= K_{\text{GA}} \cdot \frac{1 + [\text{Ar5P}]/K_{\text{Ar5P}}}{1 + [\text{GAP}]/K_{\text{GAP}}}
\end{aligned}
\tag{86}
$$

3.3.7 Ru1P aldolase

$$\text{DHAP} + \text{GA} \rightleftharpoons \text{Ru1P} \tag{87}$$

In this reaction, DHAP is the donor and GA is the acceptor.

Rate law in canonical form

Again we are assuming an ordered reaction mechanism.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{GA}] - [\mathrm{Ru1P}]/\kappa}{K^*_{\mathrm{GA}} + [\mathrm{GA}]} \\
\omega &= 1 \\
\kappa &= K'_{eq} \cdot [\mathrm{DHAP}] \\
K^*_{\mathrm{GA}} &= K_{\mathrm{GA}} \cdot \frac{1 + [\mathrm{Ru1P}]/K_{\mathrm{Ru1P}} + [\mathrm{DHAP}]/K_{\mathrm{DHAP}}}{[\mathrm{DHAP}]/K_{\mathrm{DHAP}}}
\end{aligned}
\tag{88}
$$

3.3.8 Transketolase

$$
\mathrm{S7P} + \mathrm{GA} \rightleftharpoons \mathrm{R5P} + \mathrm{Eu} \tag{89}
$$

Rate law in canonical form

This is a ping-pong mechanism.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{GA}] - [\mathrm{Eu}]/\kappa}{K^*_{\mathrm{GA}} + [\mathrm{GA}]} \\
D &= [\mathrm{S7P}] + K_{\mathrm{S7P}} \cdot (1 + [\mathrm{Eu}]/K_{\mathrm{Eu}}) \\
\omega &= [\mathrm{S7P}]/D \\
\kappa &= K'_{eq} \cdot [\mathrm{S7P}]/[\mathrm{R5P}] \\
K^*_{\mathrm{GA}} &= K_{\mathrm{GA}} \cdot \left(1 + [\mathrm{R5P}]/K_{\mathrm{R5P}} - K_{\mathrm{S7P}}/D\right)
\end{aligned}
\tag{90}
$$

3.3.9 Transaldolase

$$
\mathrm{F6P} + \mathrm{GA} \rightleftharpoons \mathrm{GAP} + \mathrm{Xu} \tag{91}
$$

Rate law in canonical form

This is a ping-pong mechanism.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{GA}] - [\mathrm{Xu}]/\kappa}{K^*_{\mathrm{GA}} + [\mathrm{GA}]} \\
D &= [\mathrm{F6P}] + K_{\mathrm{F6P}} \cdot (1 + [\mathrm{Xu}]/K_{\mathrm{Xu}}) \\
\omega &= [\mathrm{F6P}]/D \\
\kappa &= K'_{eq} \cdot [\mathrm{F6P}]/[\mathrm{GAP}] \\
K^*_{\mathrm{GA}} &= K_{\mathrm{GA}} \cdot \left(1 + [\mathrm{GAP}]/K_{\mathrm{GAP}} - K_{\mathrm{F6P}}/D\right)
\end{aligned}
\tag{92}
$$

3.3.10 Erythrulose 4-kinase

$$\mathrm{Eu} + \mathrm{ATP} \rightleftharpoons \mathrm{Eu4P} + \mathrm{ADP} \tag{93}$$

Rate law in canonical form

$$\begin{aligned} v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{Eu}] - [\mathrm{Eu4P}]/\kappa}{[\mathrm{Eu}] + K^*_{\mathrm{Eu}}} \\ \omega &= \frac{[\mathrm{ATP}]}{[\mathrm{ATP}] + K_{\mathrm{ATP}} + [\mathrm{ADP}]} \\ \kappa &= K'_{eq} \cdot \frac{[\mathrm{ATP}]}{[\mathrm{ADP}]} \\ K^*_{\mathrm{Eu}} &= K_{\mathrm{Eu}} \cdot (1 + [\mathrm{Eu4P}]/K_{\mathrm{Eu4P}}) \end{aligned} \tag{94}$$

3.3.11 Xylulose 5-kinase

$$\mathrm{Xu} + \mathrm{ATP} \rightleftharpoons \mathrm{Xu5P} + \mathrm{ADP} \tag{95}$$

Rate law in canonical form

$$\begin{aligned} v &= V_{max} \cdot \omega \cdot \frac{[\mathrm{Xu}] - [\mathrm{Xu5P}]/\kappa}{[\mathrm{Xu}] + K^*_{\mathrm{Xu}}} \\ \omega &= \frac{[\mathrm{ATP}]}{[\mathrm{ATP}] + K_{\mathrm{ATP}} + [\mathrm{ADP}]} \\ \kappa &= K'_{eq} \cdot \frac{[\mathrm{ATP}]}{[\mathrm{ADP}]} \\ K^*_{\mathrm{Xu}} &= K_{\mathrm{Xu}} \cdot (1 + [\mathrm{Xu5P}]/K_{\mathrm{Xu5P}}) \end{aligned} \tag{96}$$

3.3.12 Ribulose-1-phosphate 5-kinase

$$\mathrm{Ru1P} + \mathrm{ATP} \rightleftharpoons \mathrm{RuBP} + \mathrm{ADP} \tag{97}$$

Rate law in canonical form

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{Ru1P}] - [\text{RuBP}]/\kappa}{[\text{Ru1P}] + K^*_{\text{Ru1P}}} \\
\omega &= \frac{[\text{ATP}]}{[\text{ATP}] + K_{\text{ATP}} + [\text{ADP}]} \\
\kappa &= K'_{eq} \cdot \frac{[\text{ATP}]}{[\text{ADP}]} \\
K^*_{\text{Ru1P}} &= K_{\text{Ru1P}} \cdot (1 + [\text{RuBP}]/K_{\text{RuBP}})
\end{aligned}
\tag{98}
$$

3.3.13 Glycolyl-CoA carboxylase

$$
\text{GlCoA} + \text{CO}_2 + \text{ATP} \rightleftharpoons \text{TrCoA} + \text{ADP} + \text{P}_i \tag{99}
$$

Rate law in canonical form

We assume independent binding sites for CO_2, ATP/ADP/P_i, and GlCoA/TrCoA.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{GlCoA}] - [\text{TrCoA}]/\kappa}{[\text{GlCoA}] + K^*_{\text{GlCoA}}} \\
\omega &= \frac{[\text{ATP}]}{[\text{ATP}] + [\text{ADP}] + K_{\text{ATP}} \cdot (1 + [\text{P}_i]/K_{\text{P}_i})} \cdot \frac{[\text{CO}_2]}{[\text{CO}_2] + K_{\text{CO}_2}} \\
\kappa &= K'_{eq} \cdot \frac{[\text{ATP}] \cdot [\text{CO}_2]}{[\text{ADP}] \cdot [\text{P}_i]} \\
K^*_{\text{GlCoA}} &= K_{\text{CoA}} + [\text{TrCoA}]
\end{aligned}
\tag{100}
$$

3.3.14 Tartronyl-CoA reductase

$$
\text{TrCoA} + 2\ \text{NADPH} \rightleftharpoons \text{glycerate} + 2\ \text{NADP} + \text{CoA} \tag{101}
$$

Rate law in canonical form

The two-step reduction is modeled as a single reaction assuming that the intermediate, tartronate semialdehyde, does not leave the active site.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{TrCoA}] - [\text{glycerate}]/\kappa}{[\text{TrCoA}] + K^*_{\text{TrCoA}}} \\
\omega &= \frac{[\text{NADPH}]}{[\text{NADPH}] + K_{\text{NADPH}} + [\text{NADP}]} \\
\kappa &= K'_{eq} \cdot \frac{[\text{NADPH}]^2}{[\text{NADP}]^2 \cdot [\text{CoA}]} \\
K^*_{\text{TrCoA}} &= K_{\text{CoA}} \cdot (1 + [\text{glycerate}]/K_{\text{glycerate}} + [\text{CoA}]/K_{\text{CoA}})
\end{aligned}
\tag{102}
$$

3.3.15 Glycerate 3-kinase

$$
\text{glycerate} + \text{ATP} \rightleftharpoons \text{PGA} + \text{ADP} \tag{103}
$$

Rate law in canonical form

We include an inhibition term for RuBP.

$$
\begin{aligned}
v &= V_{max} \cdot \omega \cdot \frac{[\text{glycerate}] - [\text{PGA}]/\kappa}{[\text{glycerate}] + K^*_{\text{glycerate}}} \\
\omega &= \frac{[\text{ATP}]}{[\text{ATP}] + K_{\text{ATP}} + [\text{ADP}]} \\
\kappa &= K'_{eq} \cdot \frac{[\text{ATP}]}{[\text{ADP}]} \\
K^*_{\text{glycerate}} &= K_{\text{glycerate}} \cdot (1 + [\text{PGA}]/K_{\text{PGA}} + [\text{RuBP}]/K_{\text{RuBP}})
\end{aligned}
\tag{104}
$$

3.4 Parameters

3.4.1 Imputing unknown parameters

3.4.2 Kinetic parameters

Values for kinetic parameters were either taken directly from the literature, or from a database (BRENDA [22] and/or [23]).

Wherever possible, K_M values from the literature were taken for the actual enzyme, reaction and compound. When several values were available they were averaged (median). In most cases, since the enzymes of the synthetic shunts perform reactions that do not exist in nature, values of similar enzymes, compounds and reactions were averaged (median). For CoA-compounds, a database analysis showed that most values are in a similar range (median K_M of 30 μM). Therefore one generic value (named 'class average' hereafter) was used for all CoA-compounds including free CoA. For adenosine

nucleotides, the same approach was used: a class average was used for ATP, ADP and AMP. Similarly, a class average K_M value for NADPH and NADP was used.

All reactions are modelled as reversible; since K_M values for products (i.e. the substrate in the reverse direction) are often not available at all, default values of 1 mM were used.

3.4.3 Thermodynamic parameters

All $\Delta G'^m$ values and equilibrium constants were calculated with eQuilibrator [24], at pH 8.0 and ionic strength of 0.25. Equilibrium constants for reactions that are not available directly in eQuilibrator were calculated as the average of several proxy reactions. The proxy reactions for Ru1P kinase were ribulose kinase, fructose 1-phosphate 6-kinase and fructose 6-phosphate 1-kinase. Proxy reactions for glycolyl-CoA synthetase were reactions with acetate, propionate, hydroxypropionate, butyrate and acetoacetate. Proxy reactions for acetyl-CoA:glycolate CoA transferase were reactions with acetyl-CoA, propionyl-CoA and hydroxypropionyl-CoA. The proxy reactions for glycolyl-CoA carboxylase were acetyl-CoA to malonyl-CoA and propionyl-CoA to methylmalonyl-CoA. The proxy reactions for tartronyl-CoA reductase were succinyl-CoA, malonyl-CoA and methylmalonyl-CoA reduction to the corresponding hydroxyacid. The proxy reactions for glycolyl-CoA reductase were reverse aldehyde dehydrogenase (CoA-acylating) reactions with propionyl-CoA, butyryl-CoA and hydroxypropionyl-CoA. The proxy reaction for Ru1P aldolase was fuculose 1-phosphate aldolase. When applicable, the arithmetic mean of the $\Delta G'$ values was calculated, which is equivalent to the geometric mean of the equilibrium constants.

3.4.4 Kinetic and thermodynamic parameters of the simulation

	Pathway	Reaction	Parameter	Value	Unit	Method
1		Generic	K_{ATP}	0.2	mM	Average[1]
2			K_{CoA}	0.03	mM	Average[2]
3			K_{NADPH}	0.03	mM	Average[3]
4	Xu	Xylulose kinase	K_{Xu5P}	1	mM	Default
5			K'_{eq}	8e4		eQuilibrator
6	Xu	Transaldolase	K'_{eq}	1		eQuilibrator
7			K_{F6P}	1.2	mM	[25]
8			K_{Xu}	1	mM	Default
9			K_{GAP}	0.04	mM	[25]
10	Eu	Transketolase	K'_{eq}	0.4		eQuilibrator
11			K_{Eu}	1	mM	Default
12			K_{S7P}	2	mM	[26]
13			K_{R5P}	0.5	mM	[26]
14	Eu	Eu kinase	K'_{eq}	800		eQuilibrator
15			K_{Eu4P}	1	mM	Default
16	Eu	Erythrose isomeras	K'_{eq}	6		eQuilibrator
17	Eu	Erythrulose isomerase	K'_{eq}	1		Assumed
18	TcoA	Reductase	K'_{eq}	76		eQuilibrator
19			$K_{glycerate}$	1	mM	Default
20	TcoA	Carboxylase	K_{HCO_3}	1	mM	Average
21			K'_{eq}	67		eQuilibrator
22	TcoA	Glycerate kinase	K_{PGA}	1.5	mM	[27]
23			K_{RuBP}	2.5	mM	[27]
24			K'_{eq}	800		eQuilibrator
25	Ru1P	Ru1P kinase	K'_{eq}	5.4e4		eQuilibrator
26			K_{RuBP}	5	mM	Assumed
27	Ru1P	Aldolase	K'_{eq}	3	$(mM)^{-1}$	eQuilibrator
28			K_{DHAP}	0.4	mM	Average
29			K_{Ru1P}	0.1	mM	Assumed
30	Ar5P	Aldolase	K'_{eq}	0.5	$(mM)^{-1}$	eQuilibrator
31			K_{GAP}	0.8	mM	[28]
32			K_{Ar5P}	0.1	mM	Assumed
33	Ar5P	Ar5P isomerase	K'_{eq}	0.35		eQuilibrator
34	Carbon neutral	Reductase	K'_{eq}	3	mM	eQuilibrator
35			K_{GA}	1	mM	Default
36	All	Phosphatase	K'_{eq}	1e8		eQuilibrator
37			K_G	1	mM	Default
38	All	CoA synthetase	K'_{eq}	4		eQuilibrator
39	All	CoA transferase	K'_{eq}	0.06		eQuilibrator
40			K_{Ac}	same as K_G		Assumed

Table 7: Parameters. Averages were calculated with the database from [23], with some more recent additions from BRENDA [22]. Mutant enzymes were excluded. Wherever the value was taken from a single publication, this is indicated. The default value for product K_M's is 1 mM. The default for a K_M of a product is 1 mM. [1] Class average for ATP, ADP and AMP. [2] Class average for CoA and all acyl-CoA compounds. [3] Class average for NADPH and NADP. Some K_M's, e.g. of the reverse reaction of a kinase, have never been measured, therefore the values were 'assumed'.

3.4.5 Steady-state concentrations of co-factors and Calvin cycle intermediates

Compound	Concentration (mM)
P_i	2.000
PP_i	0.300
AMP	0.500
ADP	1.000
ATP	2.000
PGA	5.000
RuBP	5.000
NADPH	0.500
NADP	0.500
CoA	0.005
AcCoA	0.050
acetate	0.010
Xu5P	0.100
R5P	0.100
Ru5P	0.100
S7P	1.000
DHAP	0.500
GAP	0.050
E4P	0.100
F6P	1.000

Table 8: The values shown are rounded and averaged values from [29, 30, 31].

3.4.6 Sampling

To reflect the uncertainty in the parameters, we draw parameter values from a log-normal distribution centered on the literature value and a with log-standard deviation of 2 (i.e. 70% of all values are within a factor of 2 of the log-mean). All parameters and fixed concentrations that enter the calculations are treated in this manner.

3.4.7 Lower and upper limits on pathway metabolites

The default lower limit is 1 μM for all compounds. The default upper limit is 10 mM. For reasons of toxicity/known inhibition of Calvin cycle enzymes, the upper limits for [GA] and [G2P] were set to 1 mM and 0.1 mM respectively [32, 33, 34, 35]. The upper limit of all CoA-species was set to 0.05 mM because of the relatively low abundance of these compounds in the stroma [36]. The upper limit for glycerate was also set to 0.1 mM for similar reasons [37]. For a number of compounds, an effective thermodynamic upper/lower limit needs to be considered too.

Ru1P pathway: upper limit on [Ru1P]

Since the upper limit on [GA] is 1 mM, there is an effective thermodynamic upper limit on [Ru1P]. According to Eq. 75

$$
\begin{aligned}
_{\max} &= [\mathrm{GA}]_{\max} \cdot \kappa \\
[\mathrm{Ru1P}]_{\max} &= [\mathrm{GA}]_{\max} \cdot K'_{eq}[\mathrm{DHAP}] \\
&= 1\,mM \cdot 3\ (mM)^{-1} \cdot 0.5 = 1.5\ mM
\end{aligned}
\tag{105}
$$

To allow for some degrees of freedom in the kinetic parameters, we set the limit to 1 mM.

Ar5P pathway: upper and lower limit on [Ar5P]

The upper limit for Ar5P is derived in the same manner as for Ru1P:

$$
\begin{aligned}
_{\max} &= [\mathrm{GA}]_{\max} \cdot \kappa \\
[\mathrm{Ar5P}]_{\max} &= [\mathrm{GA}]_{\max} \cdot K'_{eq} \cdot [\mathrm{GAP}] \\
&= 1\,mM \cdot 0.5\ (mM)^{-1} \cdot 0.05 = 0.025\ mM
\end{aligned}
\tag{106}
$$

This upper limit is in fact very low. In addition, there is a lower limit on [Ar5P] because of the isomerisation to Ru5P that follows the aldolase reaction. The concentration of Ru5P is below 0.1 mM, perhaps as low as 0.02 mM. The equilibrium constant is close to 1; eQuilibrator calculates 0.35.

$$
\begin{aligned}
_{\min} &= [\mathrm{Ru5P}]_{\max}/K'_{eq} \\
[\mathrm{Ar5P}]_{\min} &= 0.02/0.35 \approx 0.067mM
\end{aligned}
\tag{107}
$$

Thus the lower limit for Ar5P is higher than the upper limit. Even if we changed both equilibrium constants and [Ru5P] to more favorable values, we would end up with an extremely narrow range for [Ar5P]. Thus, to stay in this range, the kinetics of the enzyme would have to be spot on, a situation which is unlikely to be achievable in vivo. However, if it were possible to substantially increase [GAP], the Ar5P pathway would become thermodynamically feasible.

Eu pathway: upper limit on [Eu]

$$
\begin{aligned}
_{\max} &= [\mathrm{GA}]_{\max} \cdot \kappa \\
[\mathrm{Eu}]_{\max} &= [\mathrm{GA}]_{\max} \cdot K'_{eq} \cdot [\mathrm{S7P}]/[\mathrm{R5P}] \quad = 1\,mM \cdot 0.4 \cdot 1\ mM/0.1\ mM = 4\ mM
\end{aligned}
\tag{108}
$$

Due to the ratio of [S7P] to [Ru5P] (or equally, [F6P] to [E4P]) this upper limit is thermodynamically unproblematic. Since millimolar concentrations of erythrulose may not be tolerated by the chloroplast, we decided to set the upper limit to 0.1 mM. The more restrictive upper limit has two consequences: the kinetic parameters of transketolase need to be 'better' (e.g. higher k_{cat}), and, conversely, those of erythrulose kinase can be 'worse'.

Xu pathway: upper limit on [Xu]

$$
\begin{aligned}
_{\max} &= [\mathrm{GA}]_{\max} \cdot \kappa \\
[\mathrm{Xu}]_{\max} &= [\mathrm{GA}]_{\max} \cdot K'_{eq} \cdot [\mathrm{F6P}]/[\mathrm{GAP}] \quad = 1\,mM \cdot 1 \cdot 1\ mM/0.05mM = 20\ mM
\end{aligned}
\tag{109}
$$

The thermodynamic driving force is to a large part derived from the high ratio of [F6P] to [E4P]. Again, for biological reasons we decided to set the upper limit to 0.1 mM.

3.4.8 Summary

	lower bound	upper bound
glycolate	0.001	10.000
glycolate 2-phosphate	0.0001	0.100
glycolaldehyde	0.001	1.000
ribulose 1-phosphate	0.001	1.000
erythrulose	0.001	0.100
xylulose	0.001	0.100
glycerate	0.001	0.100
glycolyl-CoA	0.001	0.050
tartronyl-CoA	0.001	0.050

Table 9: Upper and lower limits in millimolar units.

3.4.9 Calculating fluxes

For every pathway, two types of fluxes are calculated at high and low C_i: the Rubisco-limited flux of the pathway, and the flux through the pathway that results in the same carbon-fixation rate as native photorespiration under the same circumstances. The former serves as an upper limit as it corresponds to the maximal carbon fixation rate of the pathway ('maximal gain'), and the latter as a lower limit in the sense that above this flux the pathway has a higher carbon fixation rate than native photorespiration ('minimal gain'). In each case it is assumed that the photorespiration shunt completely replaces native photorespiration.

	Native	Carbon neutral	Carbon positive
High C_i, maximal gain	0.83	0.84	0.86
High C_i, minimal gain	0.83	0.66	0.46
Low C_i, maximal gain	1.00	1.02	1.07
Low C_i, minimal gain	1.00	0.28	0.12

Table 10: Photorespiration fluxes assuming Rubisco limitation in units of mM/s. All carbon neutral pathways have the same flux. Maximal and minimal gain are explained in the text.

3.5 Pareto analysis

By Pareto analysis we mean that, for every enzyme in a pathway, we try to find the lowest k_{cat} over a range of K_M values at a given flux under biological constraints on metabolic concentrations.

3.5.1 Assumptions

1. We assume the flux through the shunt is known from the photosynthesis model. Flux is calculated at either at high or low C_i (8 and 2 μM respectively), and for maximal or minimal gain (Section 3.4.9).

2. We assume that co-factor and Calvin cycle metabolite and concentrations will not be shifted by a synthetic photorespiration bypass. They are treated as fixed concentrations (Section 3.4.5).

3. For convenience we set the abundance of every enzyme (the total concentration of enzyme, $[E_0]$) to 1 mM. Results for a different value of $[E_0]$ can be simply obtained by adjusting the value of k_{cat} since what matters is the maximal activity $V_{max} = [E_0]k_{cat}$.

4. We assume that there are upper and lower limits for the concentration of each metabolite (Section 3.4.7).

5. Isomerase reactions are not explicitly modeled. We assume that all isomerases are at equilibrium.

3.5.2 Solving the rate equations

Given the rate laws, and fixed co-factor and Calvin cycle intermediate concentrations, it is possible to formulate an analytical solution for each reaction step (see Section 3.2.5). Thus, one by one, we obtain solutions for the concentrations of all metabolites in the pathway:

1. A parameter set is drawn with log-normal sampling (see Section 3.4.6)

2. The rate equation of the last reaction of the pathway (for which the concentration of product, a Calvin cycle intermediate, is known) is solved, leading to a solution for the concentration of substrate.

3. This substrate is the product of the previous reaction. Each reaction in turn is solved, using the solution of the previous reaction.

3.5.3 Metabolite distributions

The procedure described in the previous section can be carried out a very large number of times, leading to distributions of metabolite concentrations. We define a parameter combination as feasible if the 20^{th} and 80^{th} percentile of the distribution of every metabolite stay within the bounds of the constraints that have been defined (Section 3.4.7).

3.5.4 Carrying out the Pareto analysis

Ranges for the k_{cat} and K_M for the substrate of each reaction are defined on a logarithmic grid. K_M's are varied between 0.01 mM and 100 mM in 161 increments. k_{cat}'s are varied between 1 s^{-1} and 1000 s^{-1} in 129 increments.

The following algorithm finds the minimal k_{cat} (if any) at a given K_M:

For each value of k_{cat} on the grid, starting with lowest value that can support the flux (Section 3.2.6), do the following:

1. Calculate metabolite distributions from 1e6 simulations with randomly drawn parameters.

2. If the parameter set is feasible stop. If not try the next-higher k_{cat}.

The Pareto pairs over the K_M range constitute the Pareto front shown in the main article. The results depend on the flux through the pathway, therefore the procedure is repeated for every flux value.

3.5.5 Results for all photorespiratory shunts

For each pathway the Pareto analysis of each enzyme is shown at high and low C_i and at maximal and minimal gain.

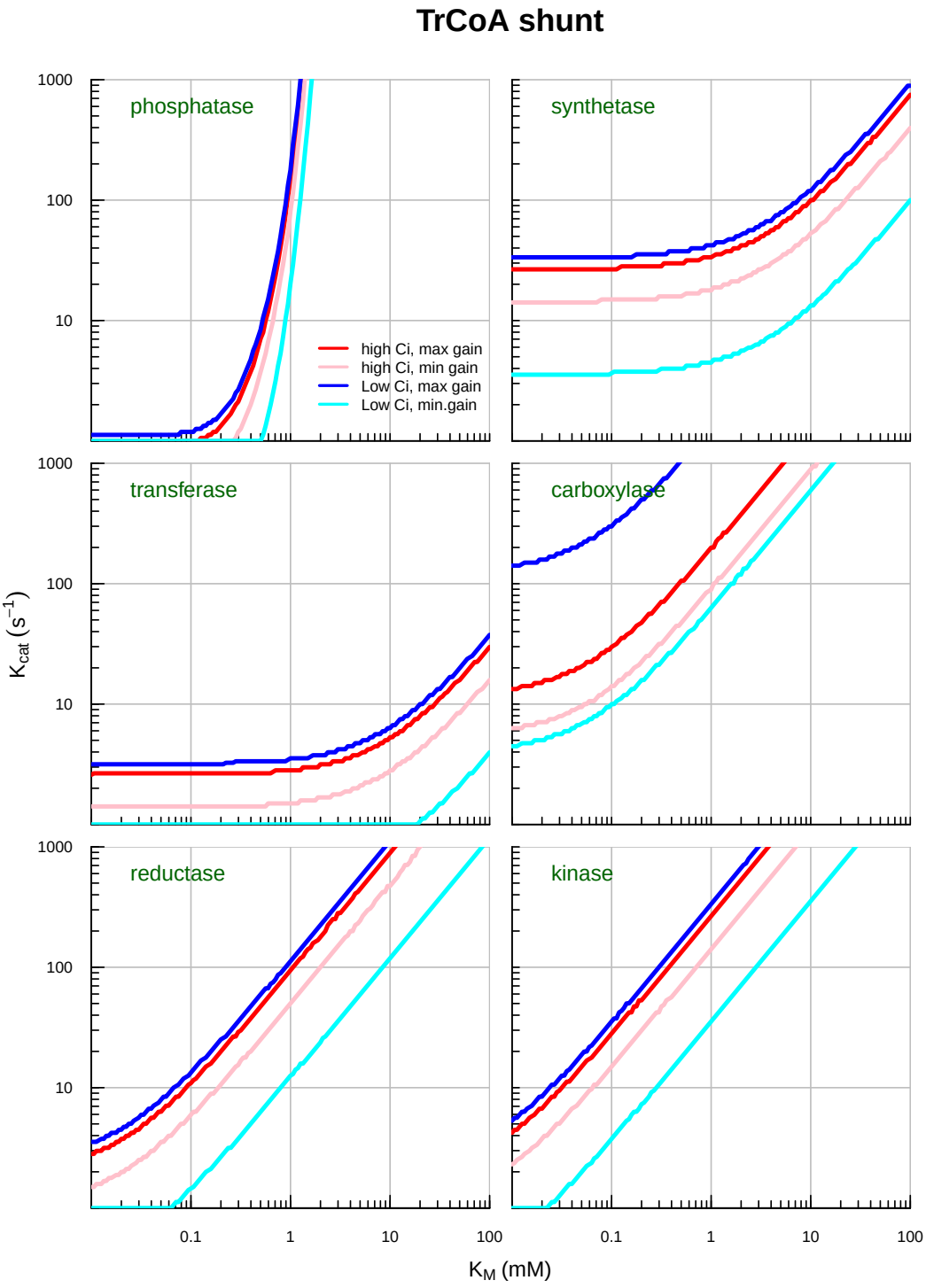


Figure 9: Pareto analysis of the Tartronyl-CoA shunt. Any point on a line represents the minimal k_{cat} at the given K_M that would support the flux through a photorespiration shunt. The fluxes were calculated with the photosynthesis model assuming either high or low intercellular CO_2 concentrations (C_i of 8 and 2 μM respectively). We show both the maximal gain curve (where the maximal possible carbon fixation rate of a shunt is achieved) and the minimal gain curve (where the flux through the shunt matches the carbon fixation rate of native photorespiration).

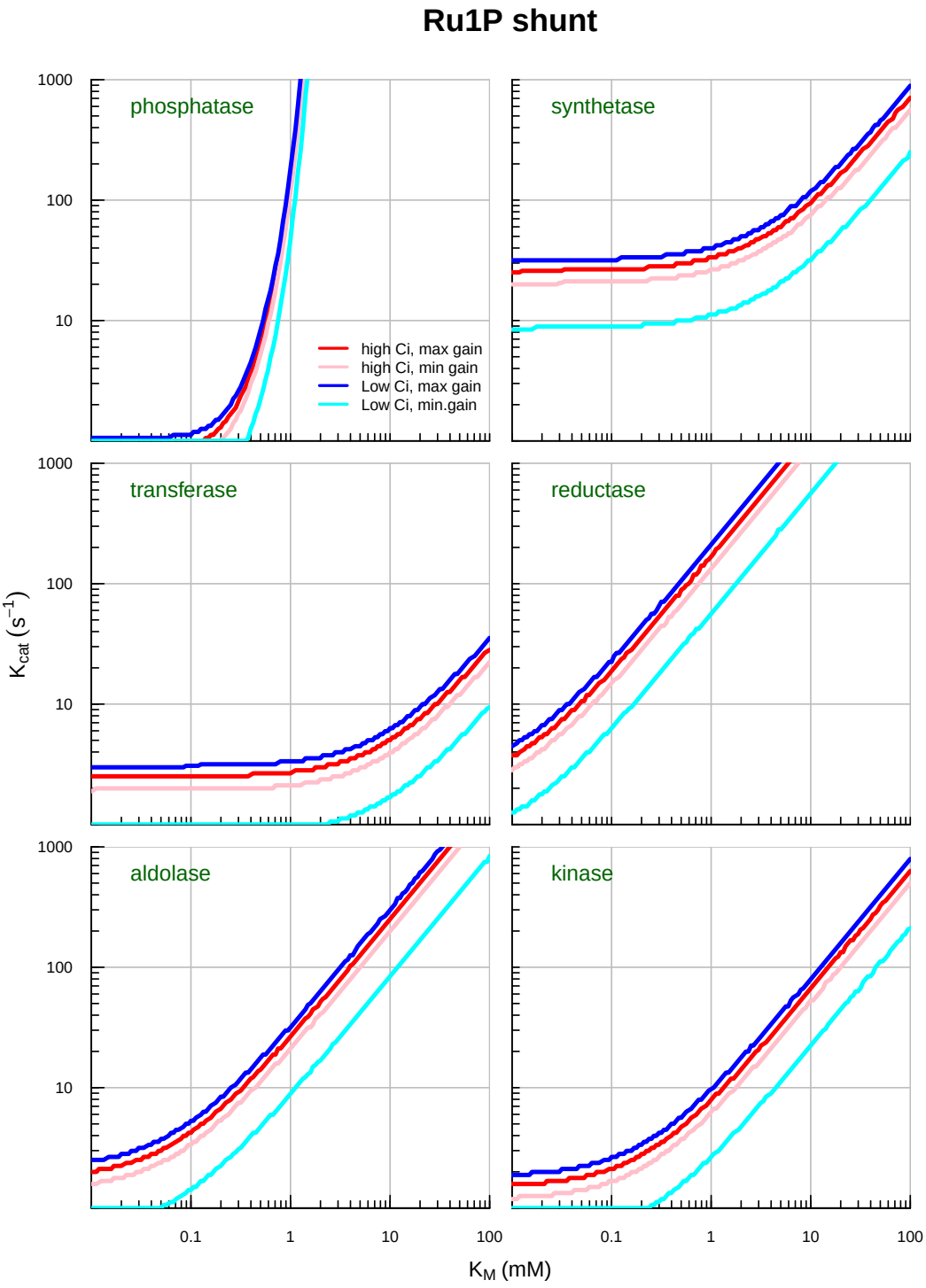


Figure 10: Pareto analysis of the Ru1P shunt. Any point on a line represents the minimal k_{cat} at the given K_M that would support the flux through a photorespiration shunt. The fluxes were calculated with the photosynthesis model assuming either high or low intercellular CO_2 concentrations (C_i of 8 and 2 μM respectively). We show both the maximal gain curve (where the maximal possible carbon fixation rate of a shunt is achieved) and the minimal gain curve (where the flux through the shunt matches the carbon fixation rate of native photorespiration).

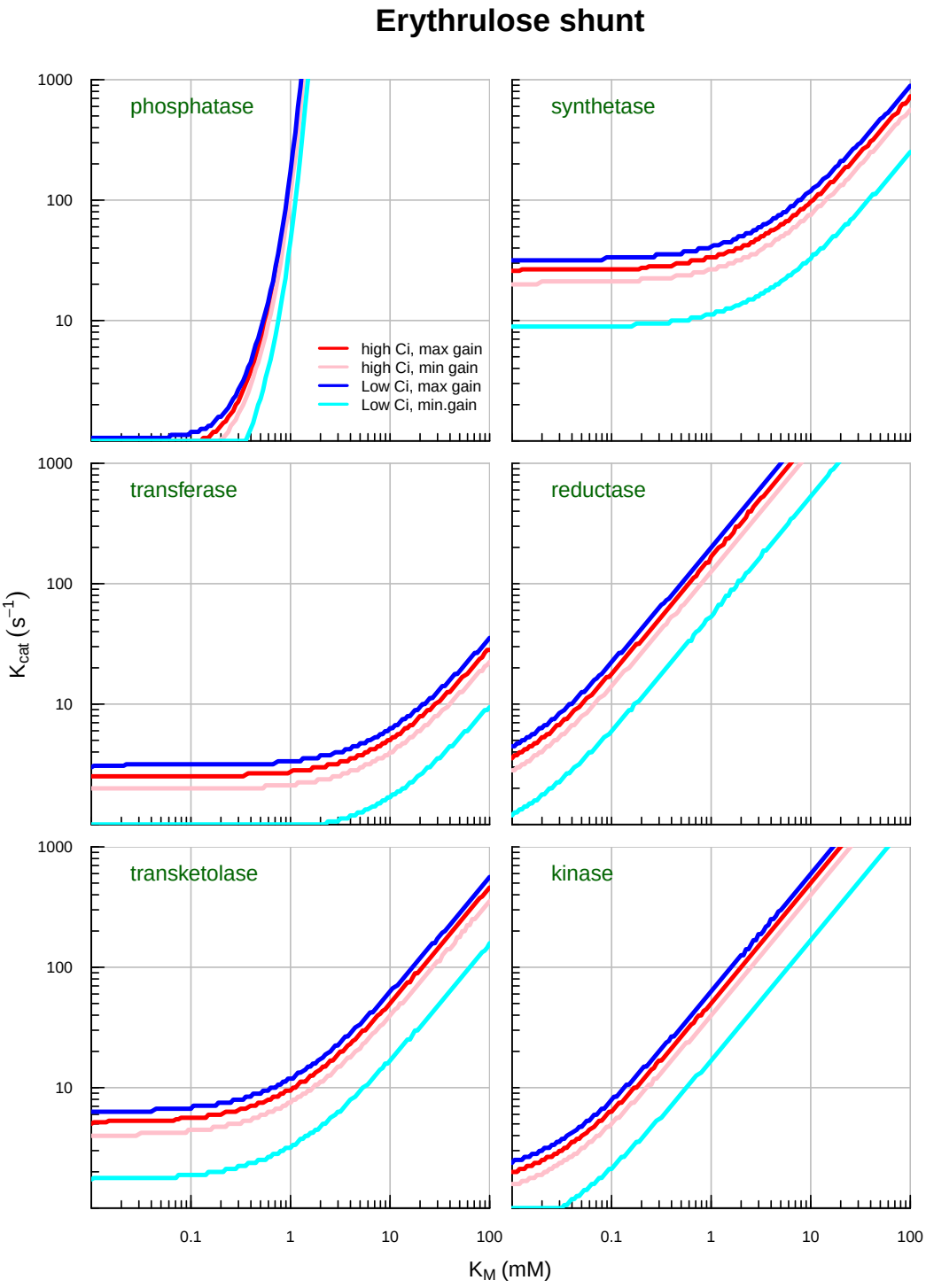


Figure 11: Pareto analysis of the Erythrulose shunt. Any point on a line represents the minimal k_{cat} at the given K_M that would support the flux through a photorespiration shunt. The fluxes were calculated with the photosynthesis model assuming either high or low intercellular CO_2 concentrations (C_i of 8 and 2 μM respectively). We show both the maximal gain curve (where the maximal possible carbon fixation rate of a shunt is achieved) and the minimal gain curve (where the flux through the shunt matches the carbon fixation rate of native photorespiration).

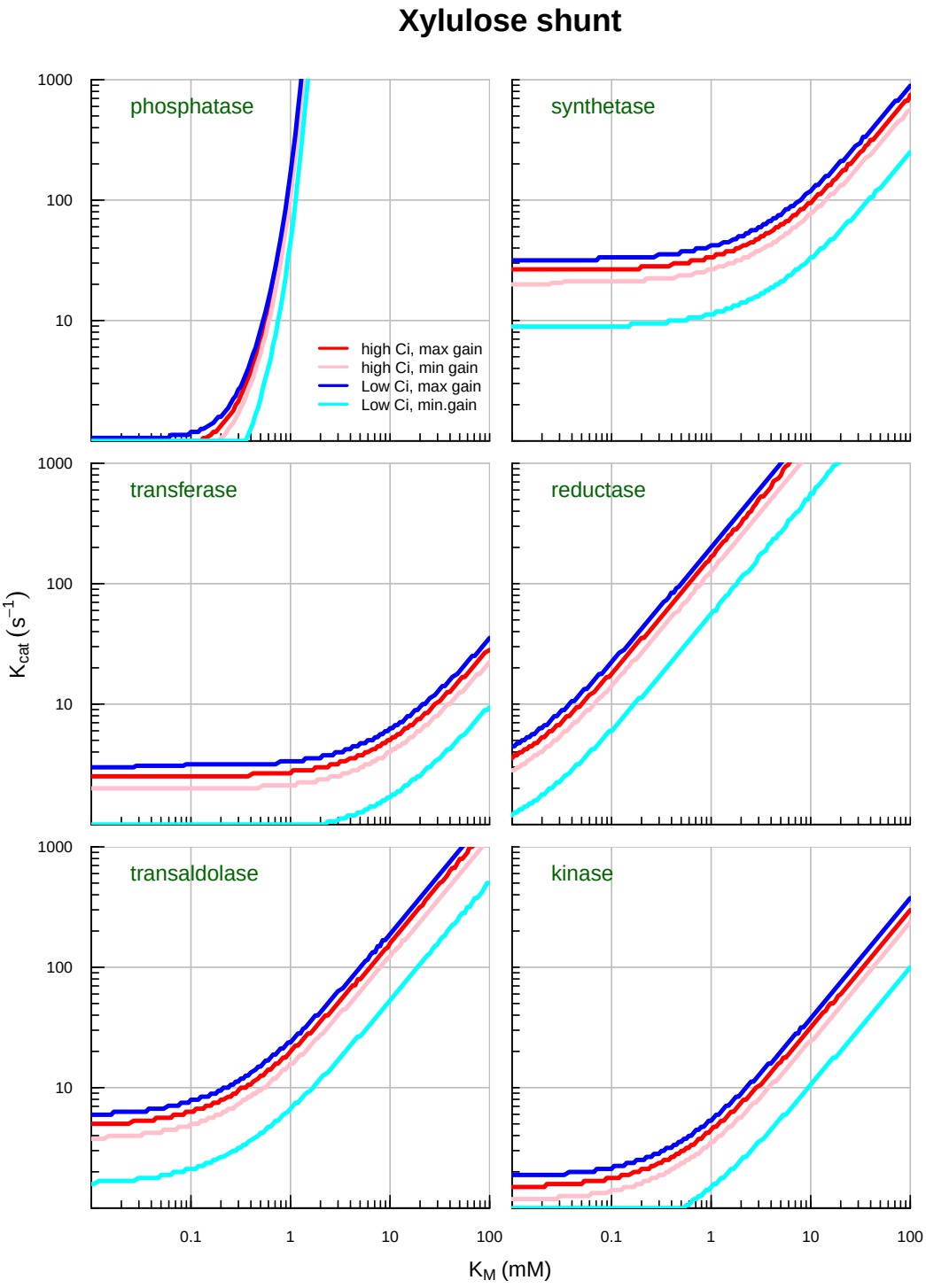
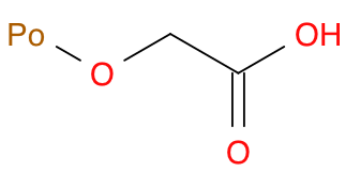


Figure 12: Pareto analysis of the Xylulose shunt. Any point on a line represents the minimal k_{cat} at the given K_M that would support the flux through a photorespiration shunt. The fluxes were calculated with the photosynthesis model assuming either high or low intercellular CO_2 concentrations (C_i of 8 and 2 μM respectively). We show both the maximal gain curve (where the maximal possible carbon fixation rate of a shunt is achieved) and the minimal gain curve (where the flux through the shunt matches the carbon fixation rate of native photorespiration).

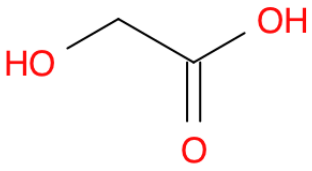
4 pathSeekR pathway architectures

4.1 Glycolyl-CoA pathways

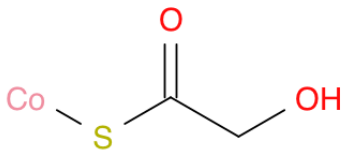
Pathway 1



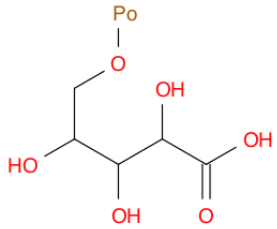
1: glycolate 2-phosphate



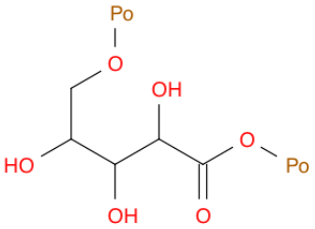
2: glycolate



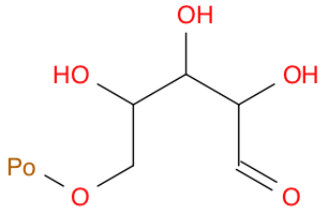
3: glycolyl-CoA



4:



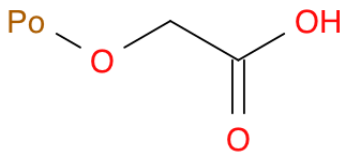
5:



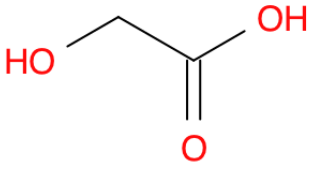
6: C5-aldose 5-phosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(=O)(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	GAP
Co-product	

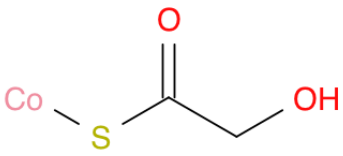
Pathway 2



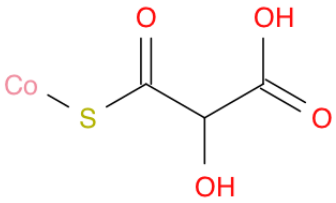
1: glycolate 2-phosphate



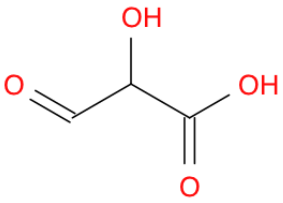
2: glycolate



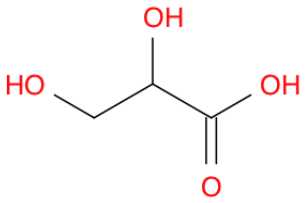
3: glycolyl-CoA



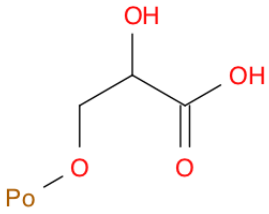
4: tartronyl-CoA



5: tartronic semialdehyde



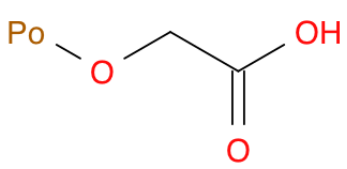
6: glycerate



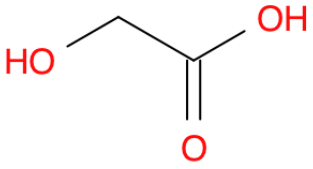
7: glycerate 3-phosphate

Number of reactions	6
Pathway superclass	2:3
Key C2 intermediate	glycolyl-CoA
Key C3 intermediate	tartronyl-CoA
Calvin cycle sink	glycerate 3-phosphate
Key reaction	Thioester activated carboxylation
Co-reactant	
Co-product	

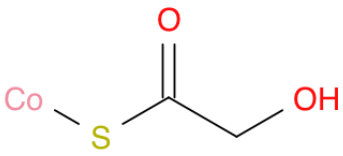
Pathway 3



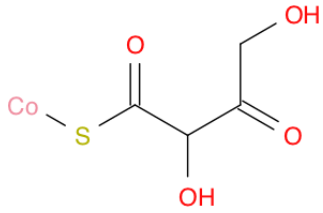
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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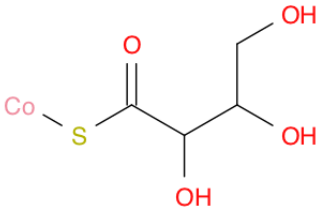

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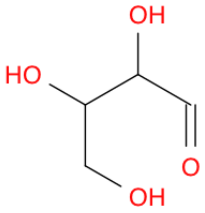
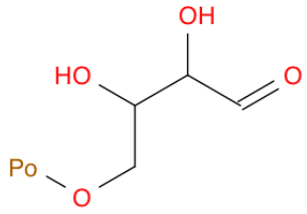


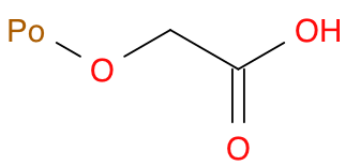
6: C4-aldose



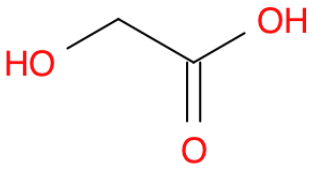
7: C4-aldose 4-phosphate

Number of reactions	6
Pathway superclass	2:4
Key C2 intermediate	glycolyl-CoA
Key C4 intermediate	C(=O)(S[CoA])C(O)C(=O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Thioester activated acyl-CoA C-transferase (acceptor and donor)
Co-reactant	glycolyl-CoA
Co-product	

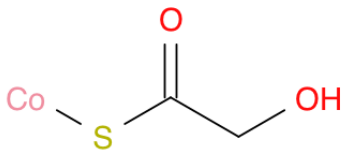
Pathway 4



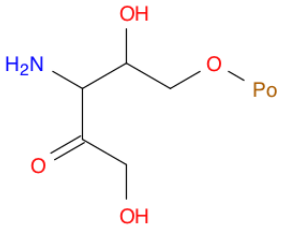
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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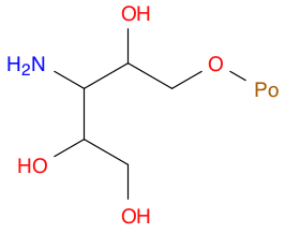

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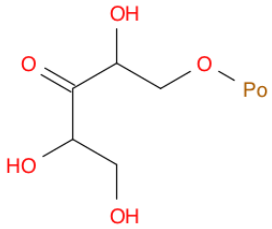

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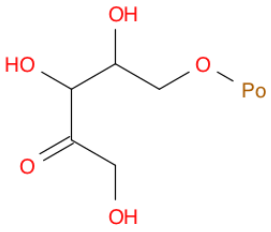
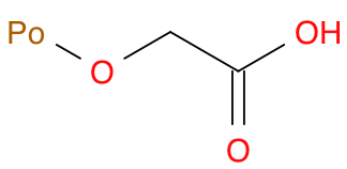


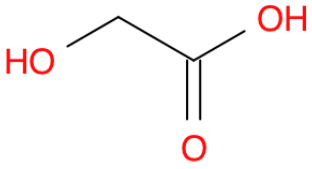
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(O[PO3])C(O)C(N)C(=O)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

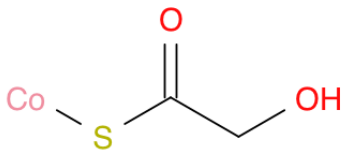
Pathway 5



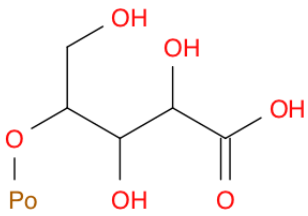
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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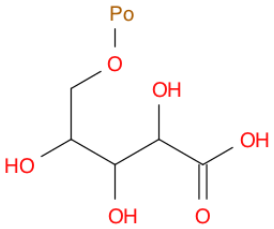

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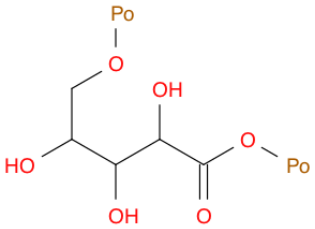

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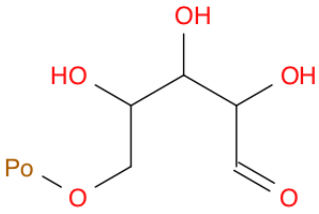
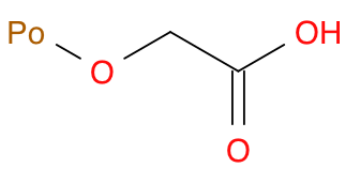


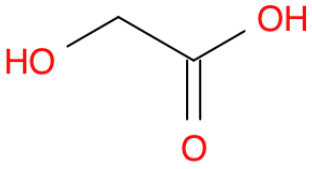
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(=O)(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

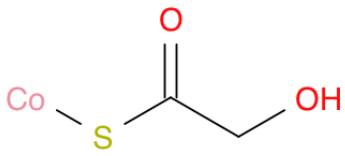
Pathway 6



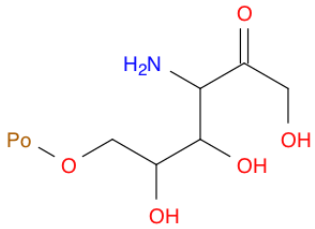
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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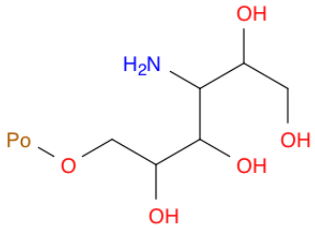

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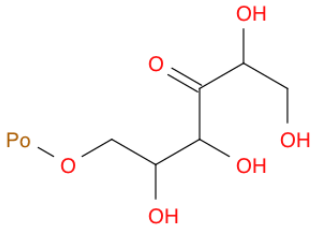

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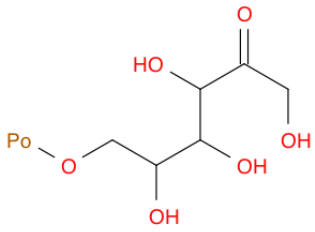
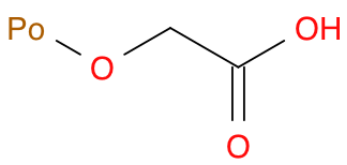


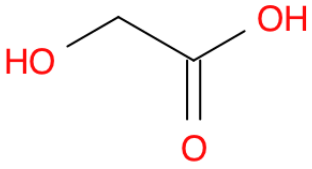
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(O[PO3])C(O)C(O)C(N)C(=O)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

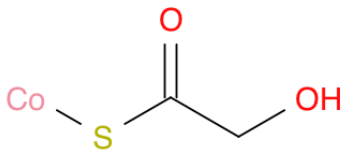
Pathway 7



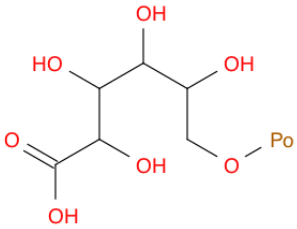
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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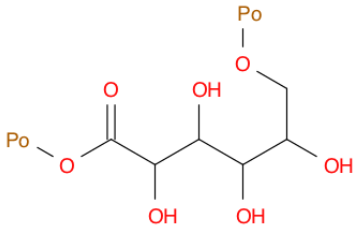

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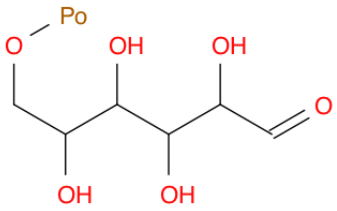
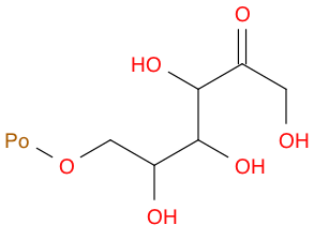


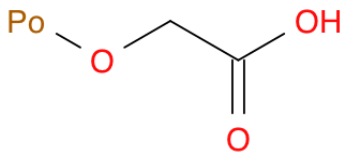
6: C6-aldose 6-phosphate



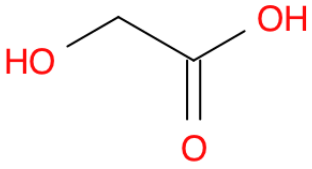
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

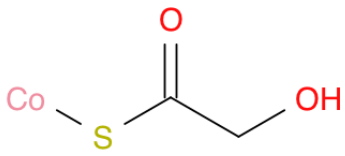
Pathway 8



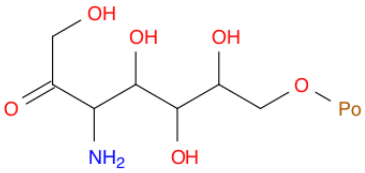
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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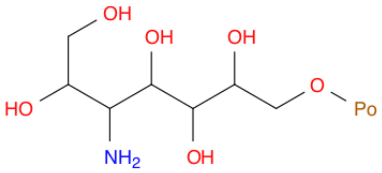

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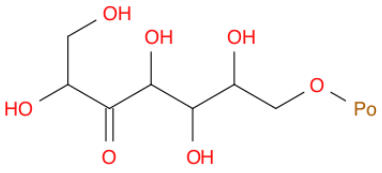

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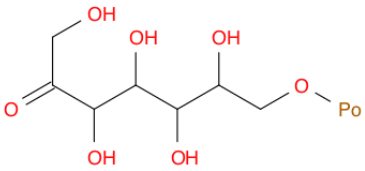
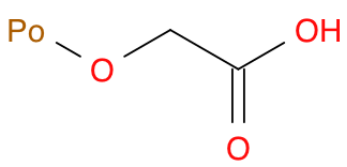


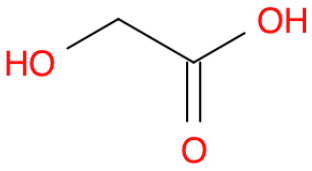
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(=O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

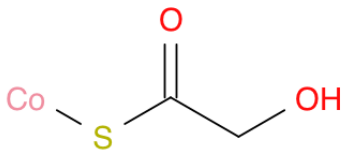
Pathway 9



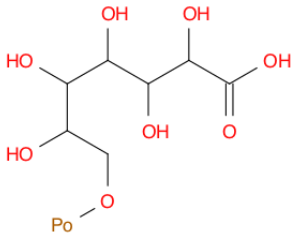
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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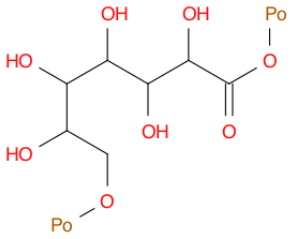

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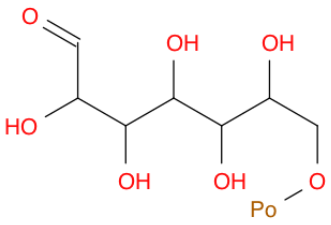
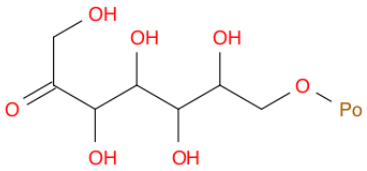


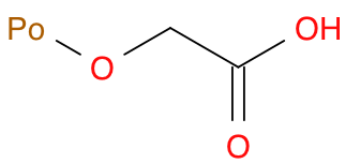
6: C7-aldose 7-phosphate



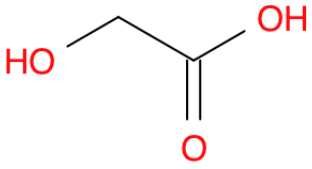
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

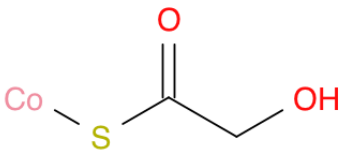
Pathway 10



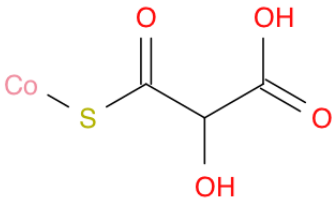
1: glycolate 2-phosphate



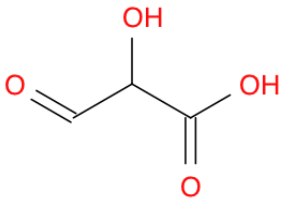
2: glycolate



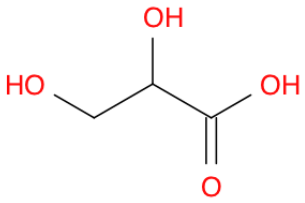
3: glycolyl-CoA



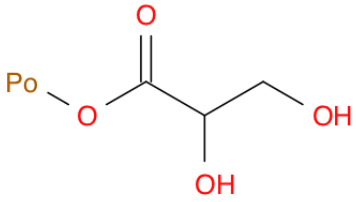
4: tartronyl-CoA



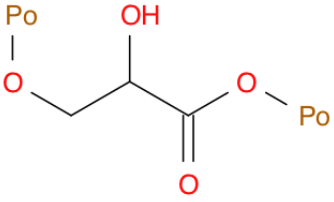
5: tartronic semialdehyde



6: glycerate



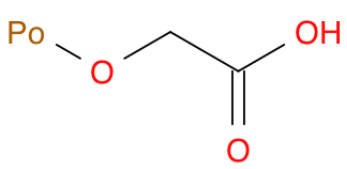
7: glyceroyl phosphate



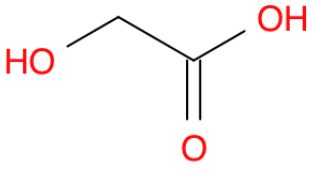
8: glycerate 1,3-bisphosphate

Number of reactions	7
Pathway superclass	2:3
Key C2 intermediate	glycolyl-CoA
Key C3 intermediate	tartronyl-CoA
Calvin cycle sink	glycerate 1,3-bisphosphate
Key reaction	Thioester activated carboxylation
Co-reactant	
Co-product	

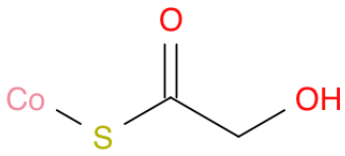
Pathway 11



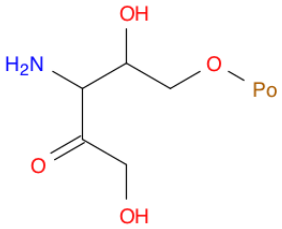
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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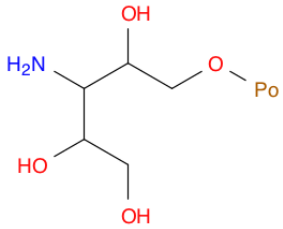

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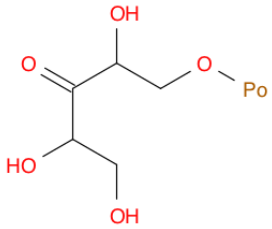

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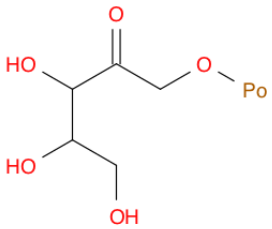
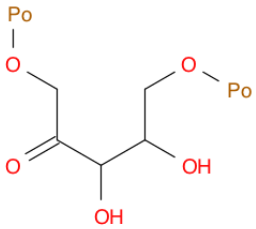


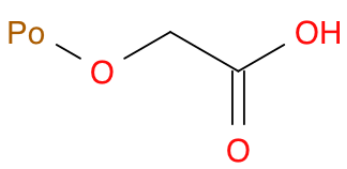
7: C5-ketose 1-phosphate



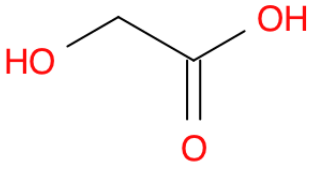
8: C5-ketose 1,5-bisphosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(O[PO3])C(O)C(N)C(=O)C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

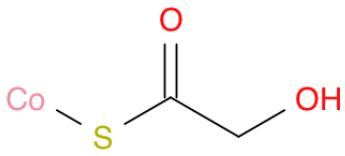
Pathway 12



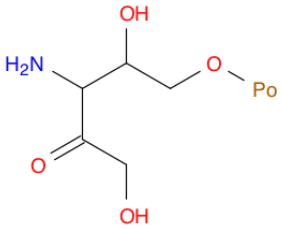
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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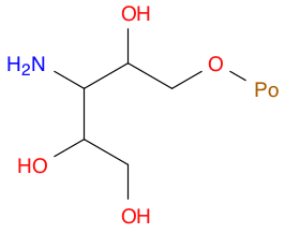

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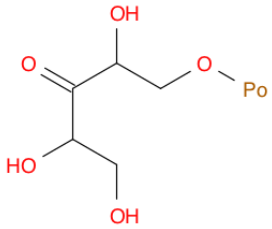

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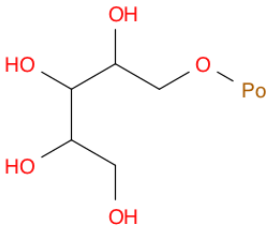

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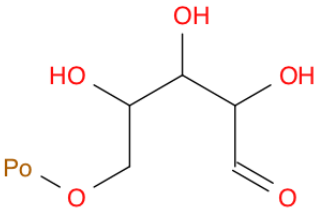
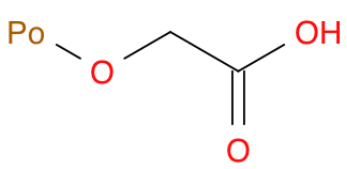


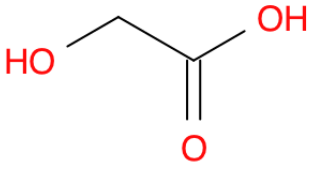
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(O[PO3])C(O)C(N)C(=O)C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

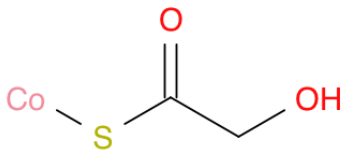
Pathway 13



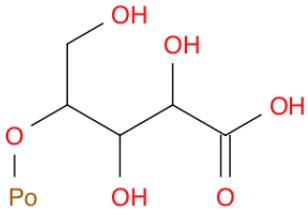
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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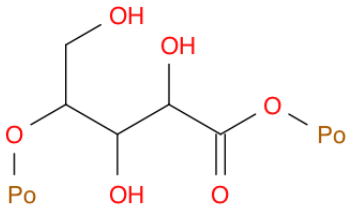

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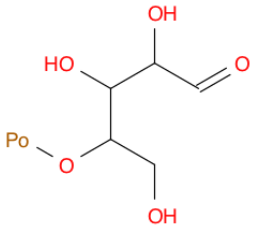

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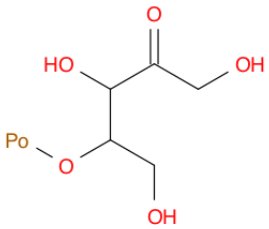

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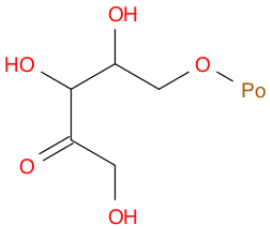
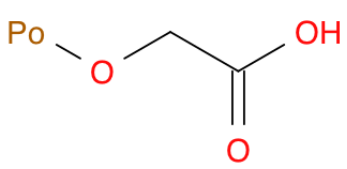


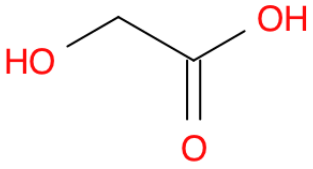
8: C5-ketose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(=O)(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

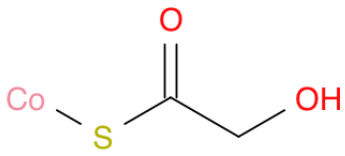
Pathway 14



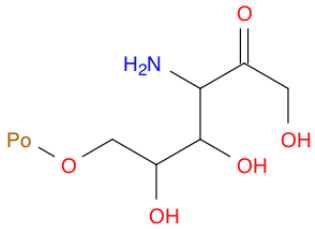
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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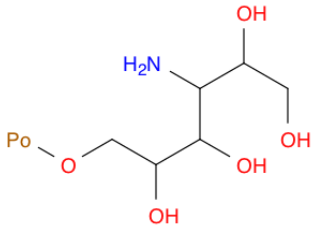

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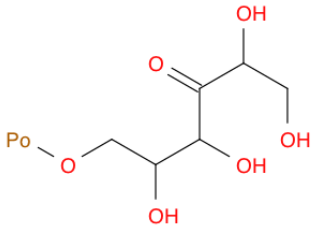

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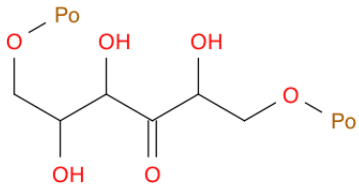

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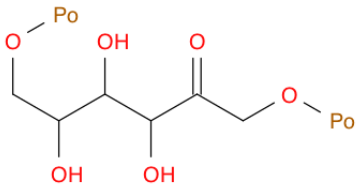
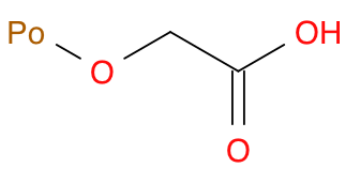


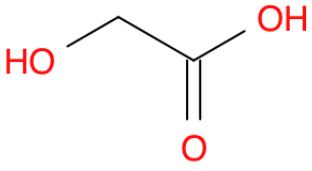
8: C6-ketose 1,6-bisphosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(O[PO3])C(O)C(O)C(N)C(=O)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

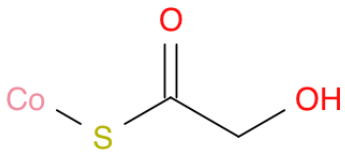
Pathway 15



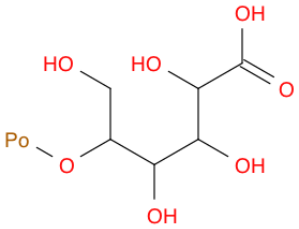
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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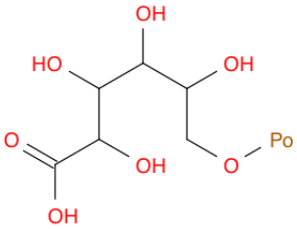

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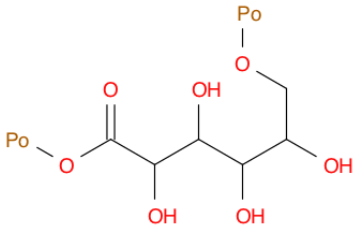

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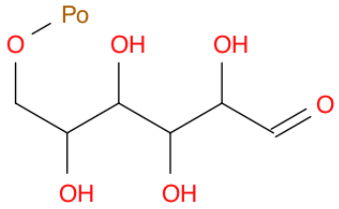
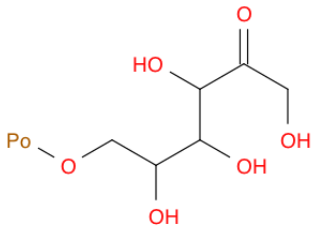


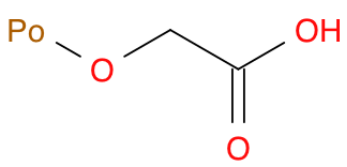
7: C6-aldose 6-phosphate



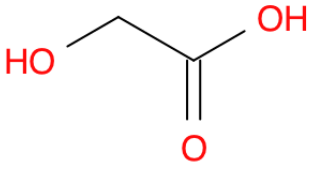
8: C6-ketose 6-phosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

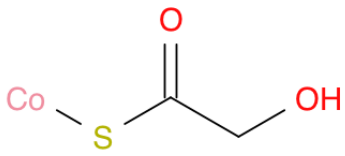
Pathway 16



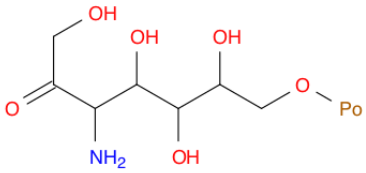
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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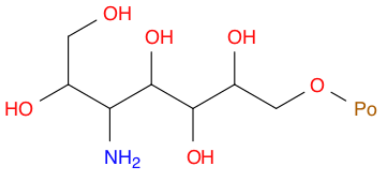

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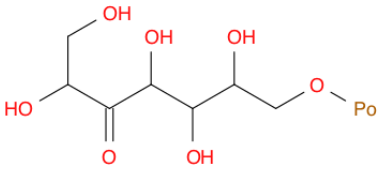

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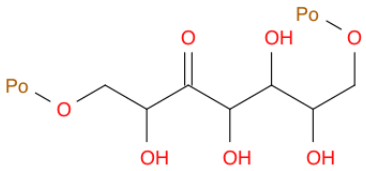

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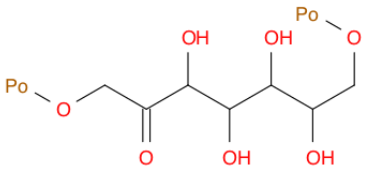
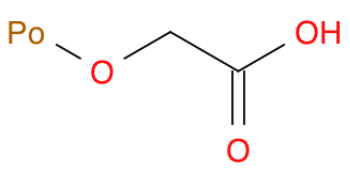


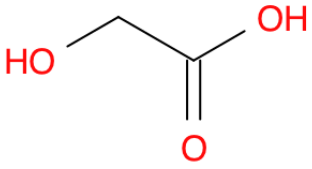
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(=O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Amine activated acyl-CoA C-transferase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

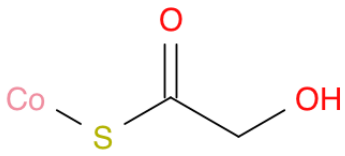
Pathway 17



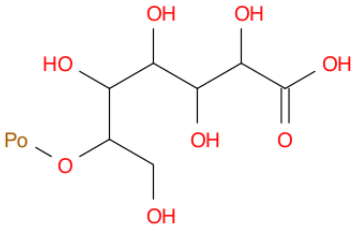
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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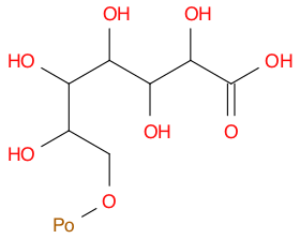

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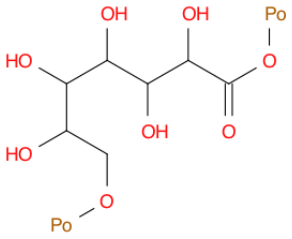

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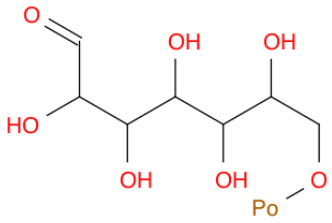
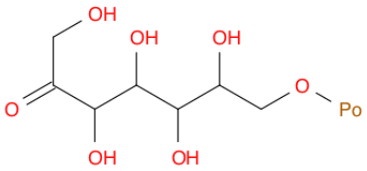


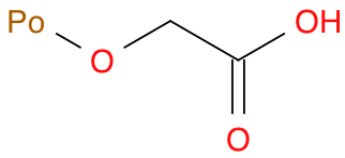
7: C7-aldose 7-phosphate



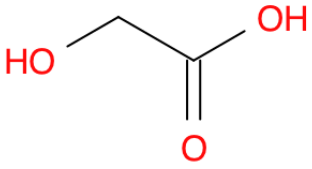
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

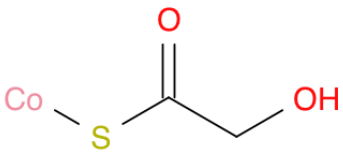
Pathway 18



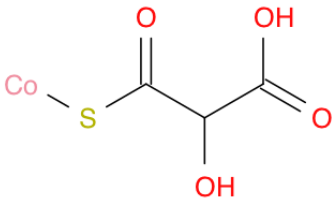
1: glycolate 2-phosphate



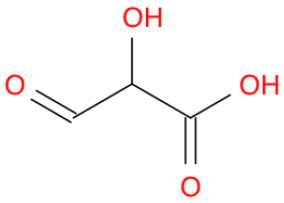
2: glycolate



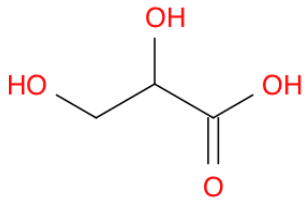
3: glycolyl-CoA



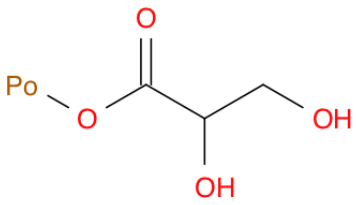
4: tartronyl-CoA



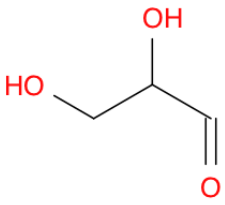
5: tartronic semialdehyde



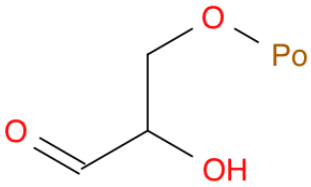
6: glycerate



7: glyceroyl phosphate



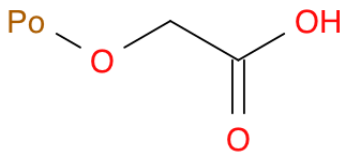
8: glyceraldehyde



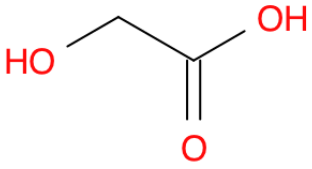
9: GAP

Number of reactions	8
Pathway superclass	2:3
Key C2 intermediate	glycolyl-CoA
Key C3 intermediate	tartronyl-CoA
Calvin cycle sink	GAP
Key reaction	Thioester activated carboxylation
Co-reactant	
Co-product	

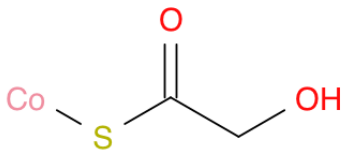
Pathway 19



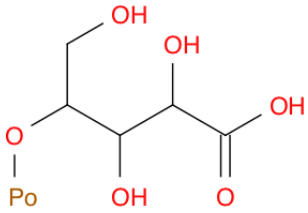
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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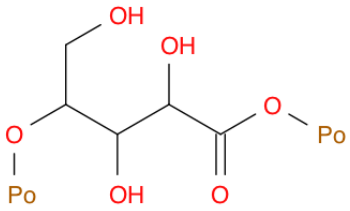

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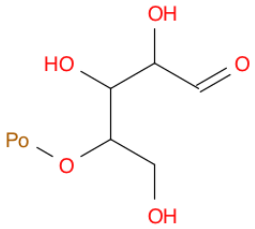

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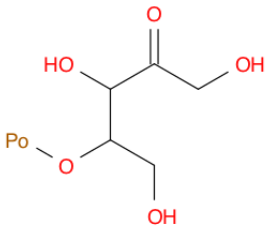

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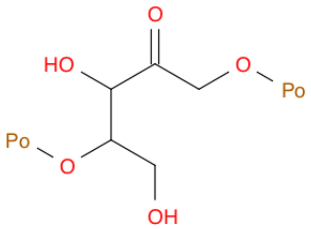

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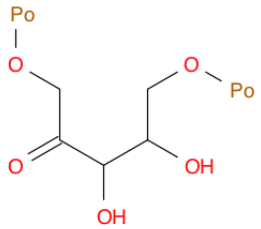
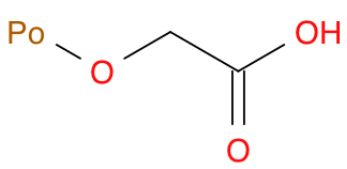


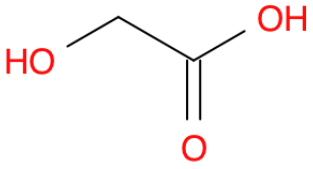
9: C5-ketose 1,5-bisphosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glycolyl-CoA
Key C5 intermediate	C(=O)(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

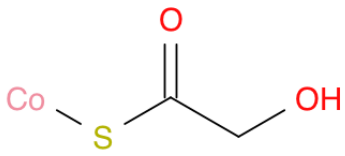
Pathway 20



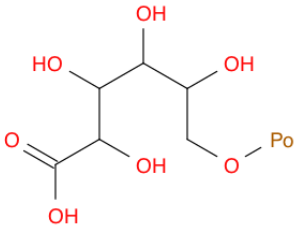
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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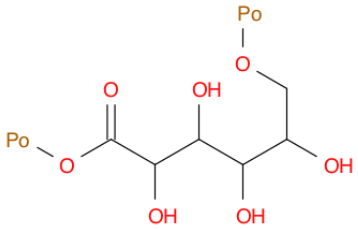

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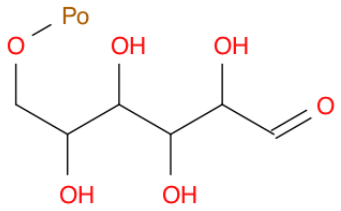
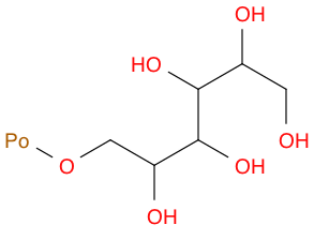


6: C6-aldose 6-phosphate



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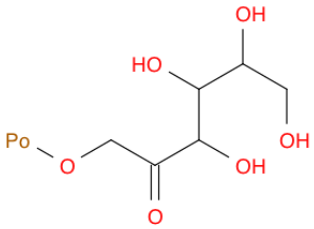
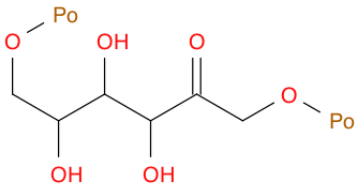


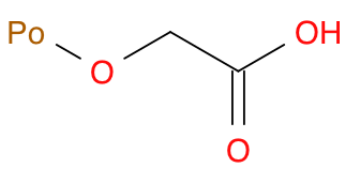
8: C6-ketose 1-phosphate



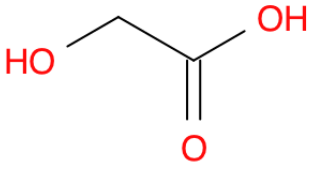
9: C6-ketose 1,6-bisphosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

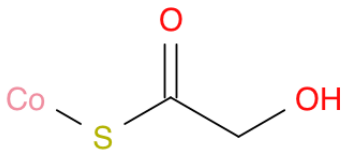
Pathway 21



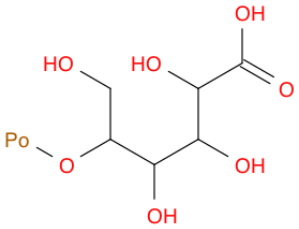
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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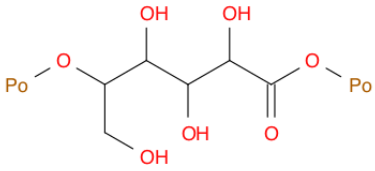

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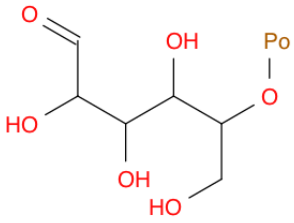

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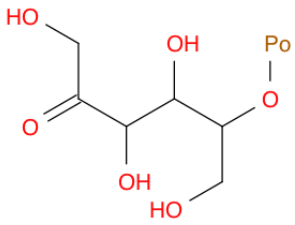

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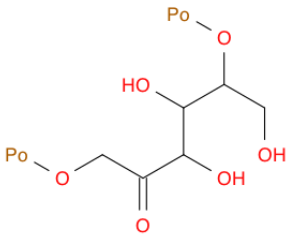

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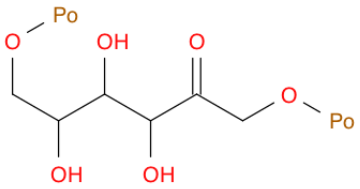
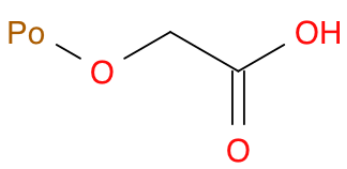


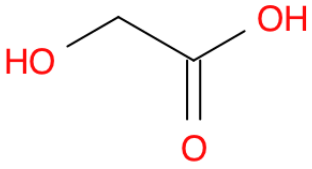
9: C6-ketose 1,6-bisphosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glycolyl-CoA
Key C6 intermediate	C(=O)(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

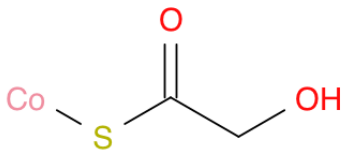
Pathway 22



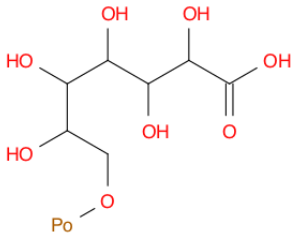
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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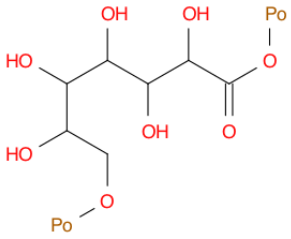

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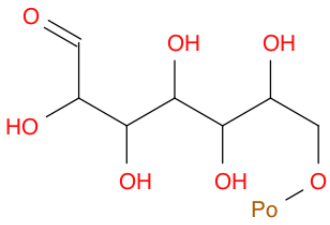
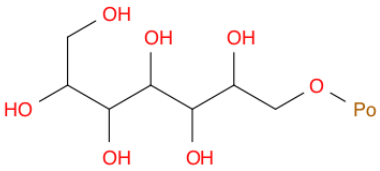


6: C7-aldose 7-phosphate



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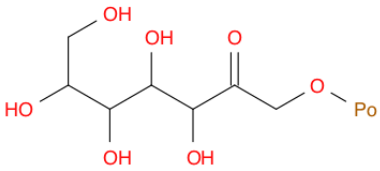
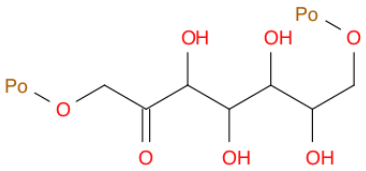


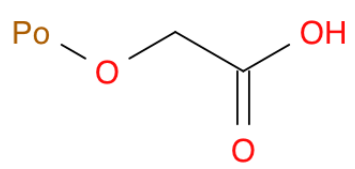
8: C7-ketose 1-phosphate



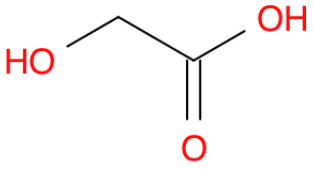
9: C7-ketose 1,7-bisphosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

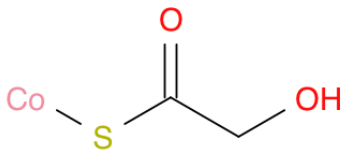
Pathway 23



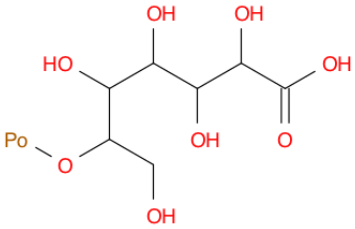
1: glycolate 2-phosphate



2: glycolate



3: glycolyl-CoA



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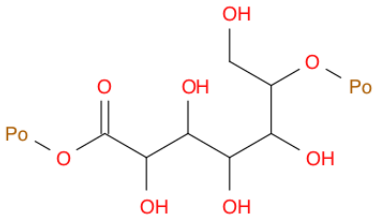

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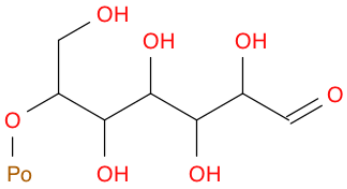

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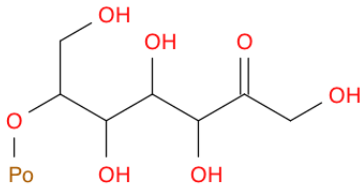

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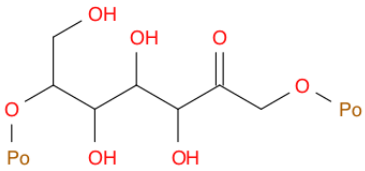

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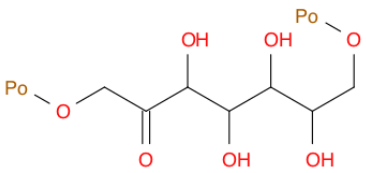
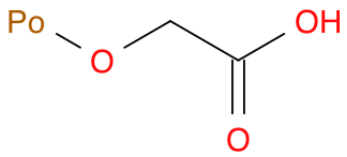


9: C7-ketose 1,7-bisphosphate

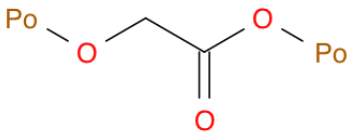
Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glycolyl-CoA
Key C7 intermediate	C(=O)(O)C(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Thioester aldolase with thioester hydrolysis (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

4.2 Glycolaldehyde 2-phosphate pathways

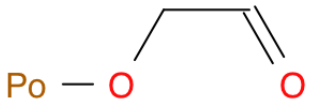
Pathway 24



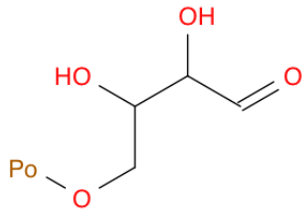
1: glycolate 2-phosphate



2: glycolate bisphosphate



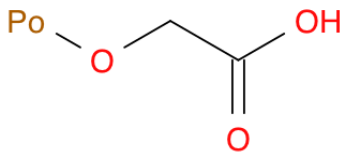
3: glycolaldehyde 2-phosphate



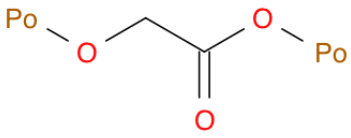
4: C4-aldose 4-phosphate

Number of reactions	3
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C4 intermediate	C4-aldose 4-phosphate
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	glycolaldehyde
Co-product	

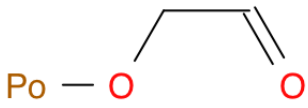
Pathway 25



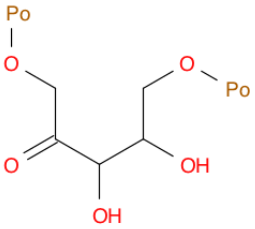
1: glycolate 2-phosphate



2: glycolate bisphosphate



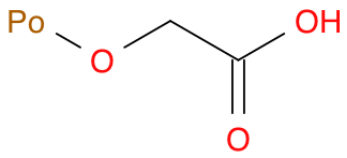
3: glycolaldehyde 2-phosphate



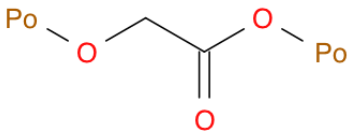
4: C5-ketose 1,5-bisphosphate

Number of reactions	3
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C5-ketose 1,5-bisphosphate
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Carbonyl aldolase (acceptor) or Ketolase (donor)
Co-reactant	GAP or DHAP
Co-product	

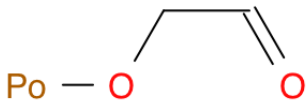
Pathway 26



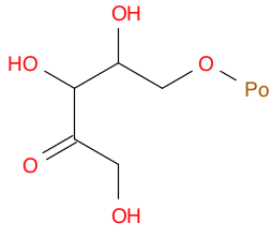
1: glycolate 2-phosphate



2: glycolate bisphosphate



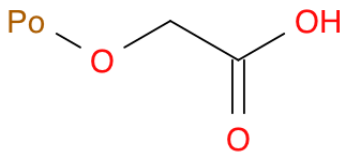
3: glycolaldehyde 2-phosphate



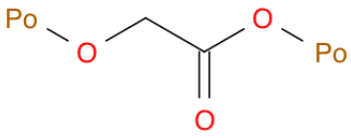
4: C5-ketose 5-phosphate

Number of reactions	3
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C5-ketose 5-phosphate
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

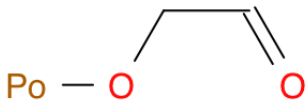
Pathway 27



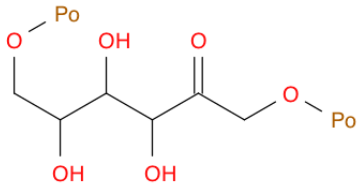
1: glycolate 2-phosphate



2: glycolate bisphosphate



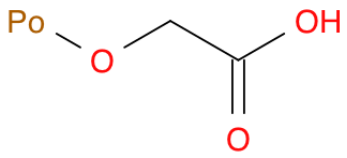
3: glycolaldehyde 2-phosphate



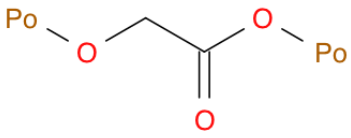
4: C6-ketose 1,6-bisphosphate

Number of reactions	3
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C6 intermediate	C6-ketose 1,6-bisphosphate
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

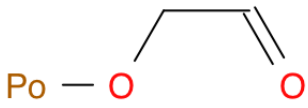
Pathway 28



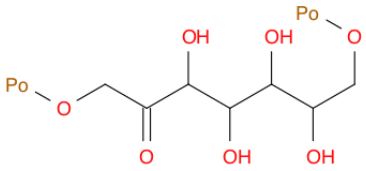
1: glycolate 2-phosphate



2: glycolate bisphosphate



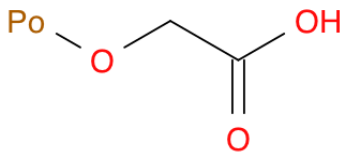
3: glycolaldehyde 2-phosphate



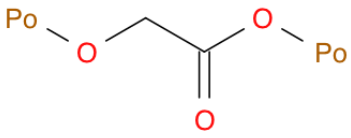
4: C7-ketose 1,7-bisphosphate

Number of reactions	3
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C7-ketose 1,7-bisphosphate
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

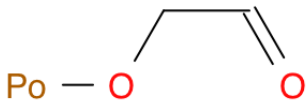
Pathway 29



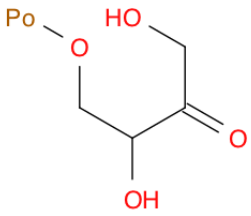
1: glycolate 2-phosphate



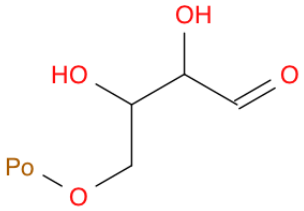
2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



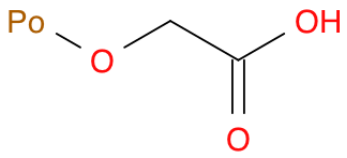
4: C4-ketose 4-phosphate



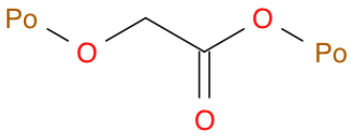
5: C4-aldose 4-phosphate

Number of reactions	4
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C4 intermediate	C4-ketose 4-phosphate
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Ketolase (acceptor) or Transketolase (acceptor)
Co-reactant	glycolaldehyde
Co-product	

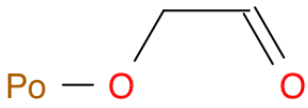
Pathway 30



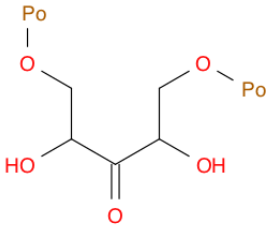
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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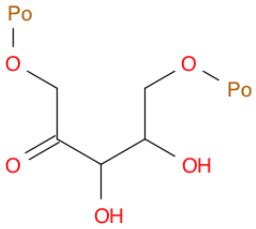
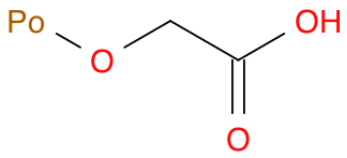


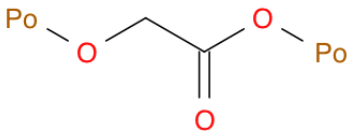
5: C5-ketose 1,5-bisphosphate

Number of reactions	4
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

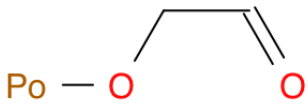
Pathway 31



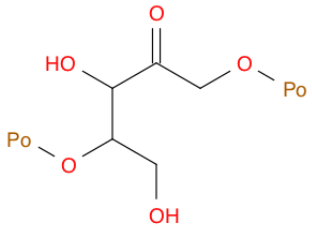
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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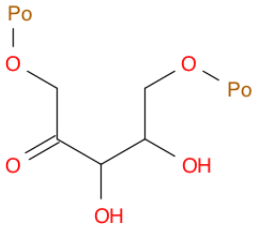
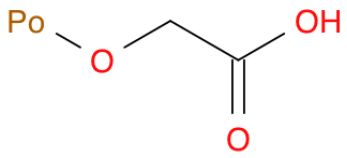


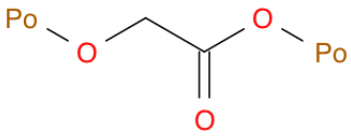
5: C5-ketose 1,5-bisphosphate

Number of reactions	4
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C(O[PO3])C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

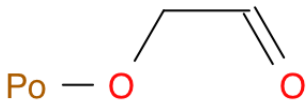
Pathway 32



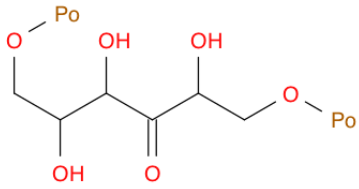
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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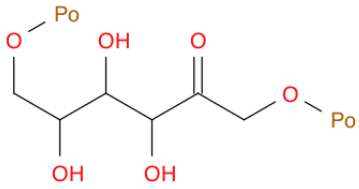
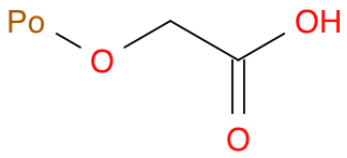


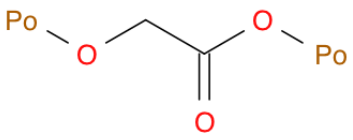
5: C6-ketose 1,6-bisphosphate

Number of reactions	4
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C6 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Carbonyl aldolase (acceptor) or Ketolase (acceptor)
Co-reactant	C4-ketose 4-phosphate or C4-aldose 4-phosphate
Co-product	

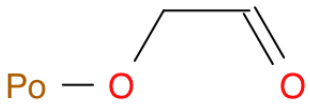
Pathway 33



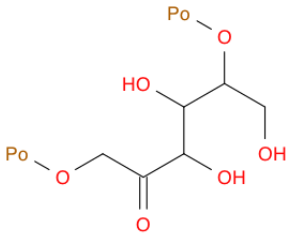
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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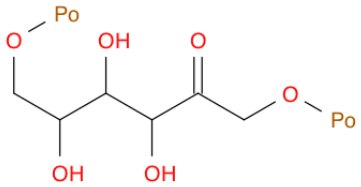
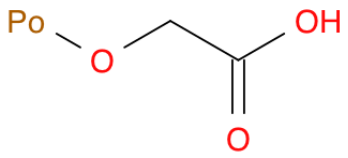


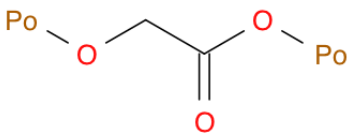
5: C6-ketose 1,6-bisphosphate

Number of reactions	4
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C6 intermediate	C(O[PO3])C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

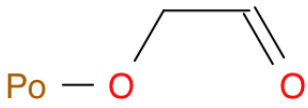
Pathway 34



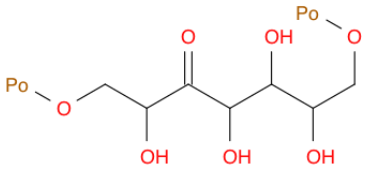
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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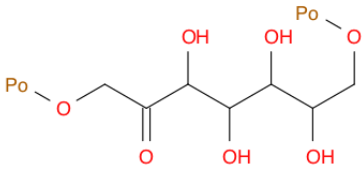
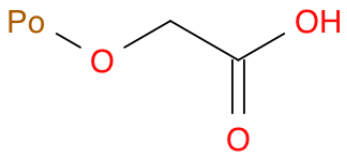


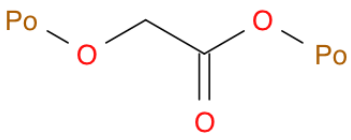
5: C7-ketose 1,7-bisphosphate

Number of reactions	4
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

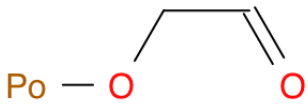
Pathway 35



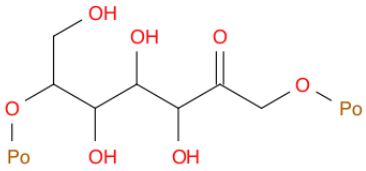
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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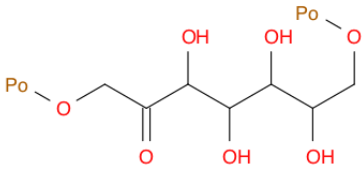
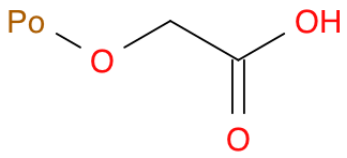


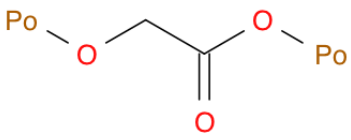
5: C7-ketose 1,7-bisphosphate

Number of reactions	4
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

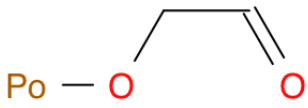
Pathway 36



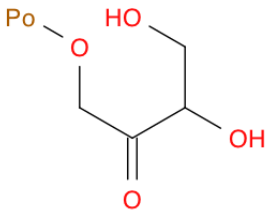
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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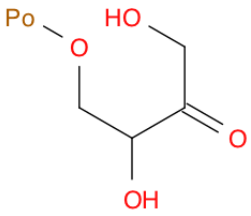
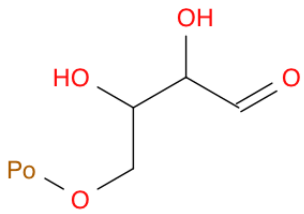


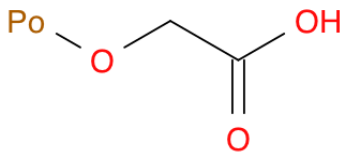
5: C4-ketose 4-phosphate



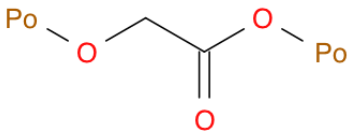
6: C4-aldose 4-phosphate

Number of reactions	5
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C4 intermediate	C(O[PO3])C(=O)C(O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Ketolase (donor)
Co-reactant	glycolaldehyde
Co-product	

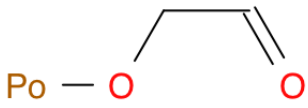
Pathway 37



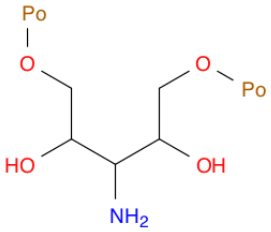
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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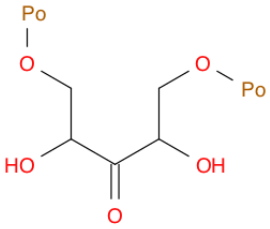

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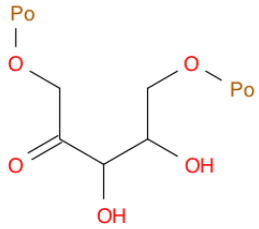
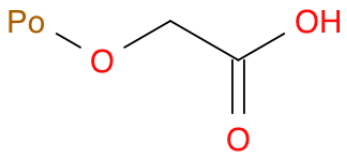


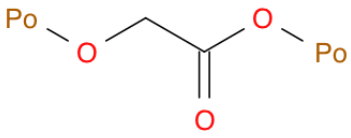
6: C5-ketose 1,5-bisphosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C(O[PO3])C(O)C(N)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

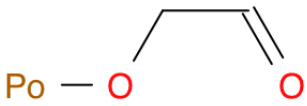
Pathway 38



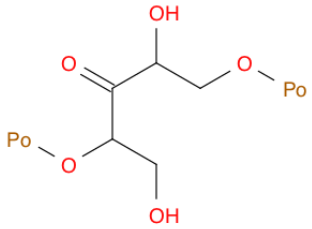
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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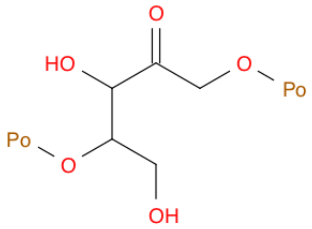

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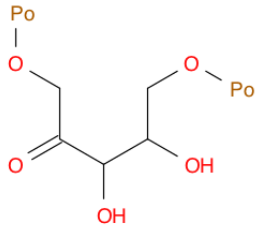
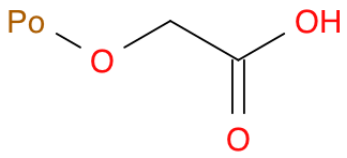


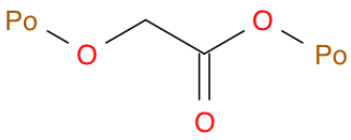
6: C5-ketose 1,5-bisphosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C5 intermediate	C(O[PO3])C(O)C(=O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

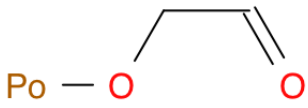
Pathway 39



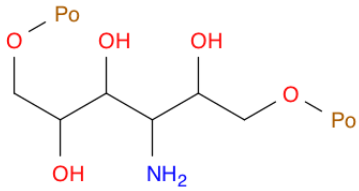
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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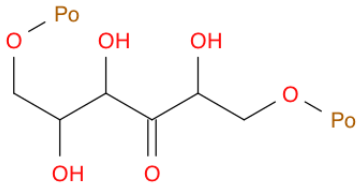
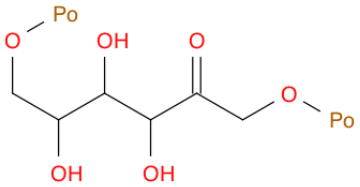


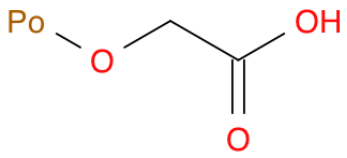
5:



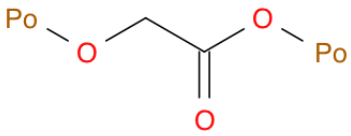
6: C6-ketose 1,6-bisphosphate

Number of reactions	5
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C6 intermediate	C(O[PO3])C(O)C(N)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

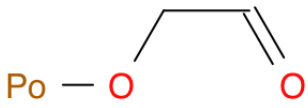
Pathway 40



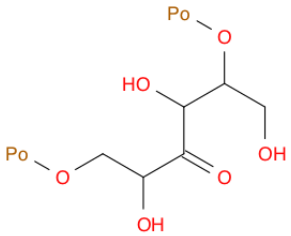
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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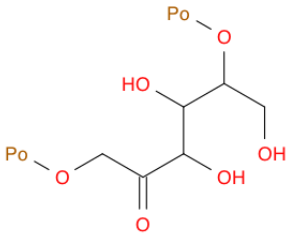

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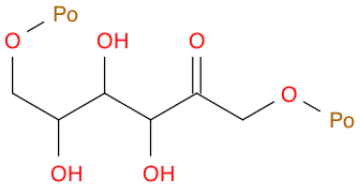
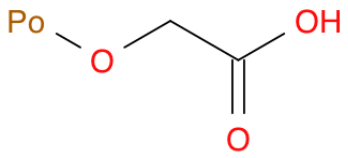


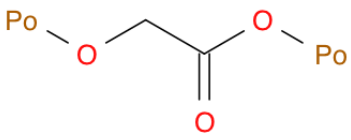
6: C6-ketose 1,6-bisphosphate

Number of reactions	5
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C6 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

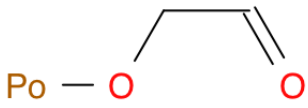
Pathway 41



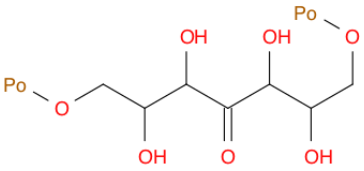
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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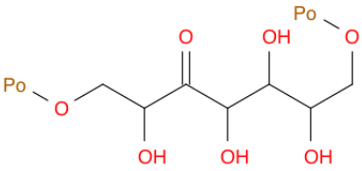

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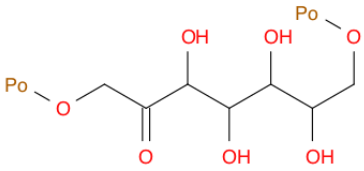
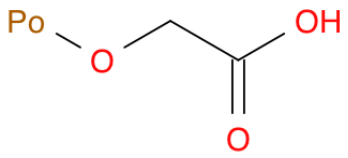


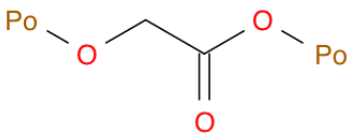
6: C7-ketose 1,7-bisphosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C5-ketose 5-phosphate
Co-product	

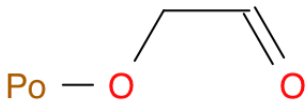
Pathway 42



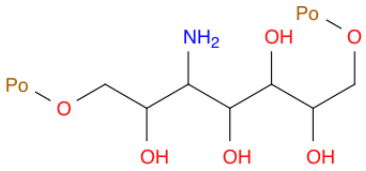
1: glycolate 2-phosphate



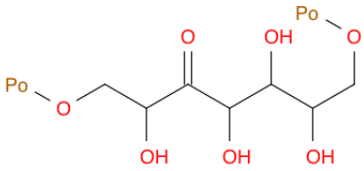
2: glycolate bisphosphate



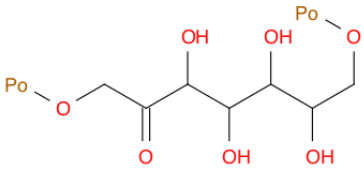
3: glycolaldehyde 2-phosphate



4:



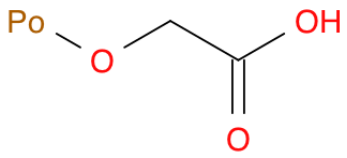
5:



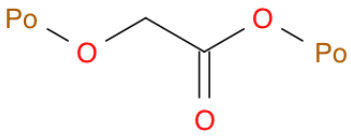
6: C7-ketose 1,7-bisphosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(O)C(N)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

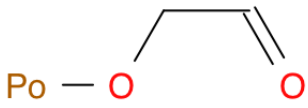
Pathway 43



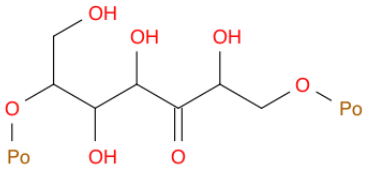
1: glycolate 2-phosphate



2: glycolate bisphosphate



3: glycolaldehyde 2-phosphate



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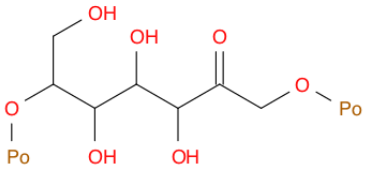

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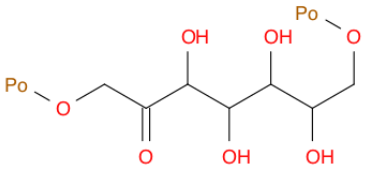
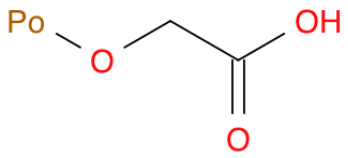


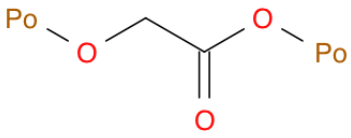
6: C7-ketose 1,7-bisphosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

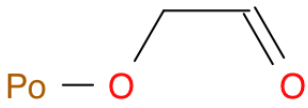
Pathway 44



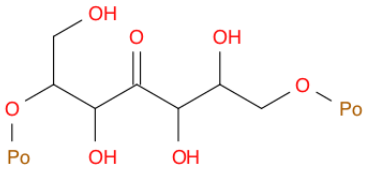
1: glycolate 2-phosphate



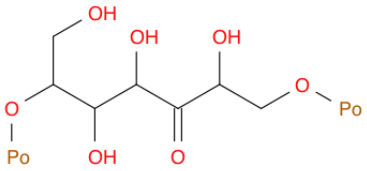
2: glycolate bisphosphate



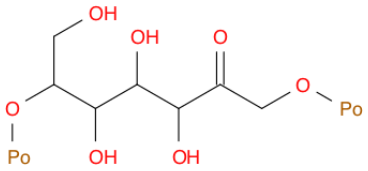
3: glycolaldehyde 2-phosphate



4:



5:



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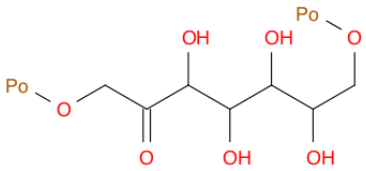
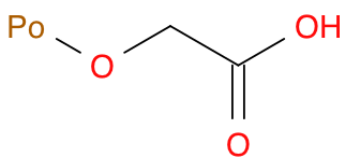


7: C7-ketose 1,7-bisphosphate

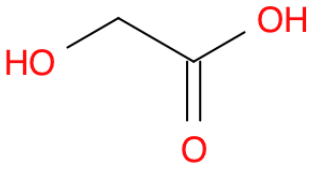
Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde 2-phosphate
Key C7 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C(O)C(=O)C(O)C(O[PO3])C(O)
Co-product	

4.3 Glycolaldehyde pathways

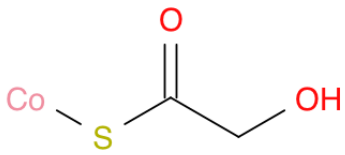
Pathway 45



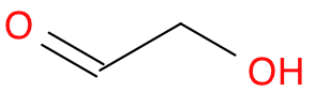
1: glycolate 2-phosphate



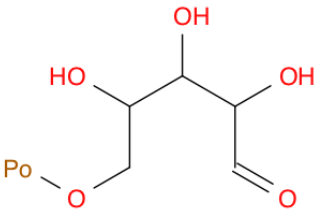
2: glycolate



3: glycolyl-CoA



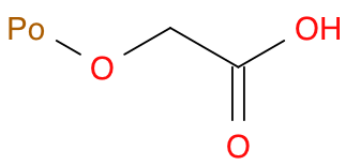
4: glycolaldehyde



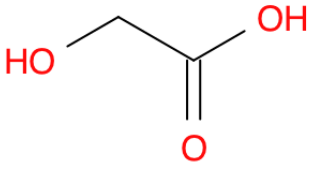
5: C5-aldose 5-phosphate

Number of reactions	4
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-aldose 5-phosphate
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	GAP
Co-product	

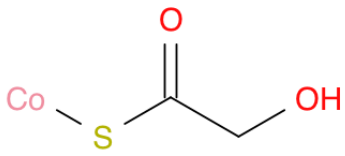
Pathway 46



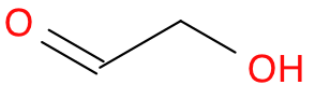
1: glycolate 2-phosphate



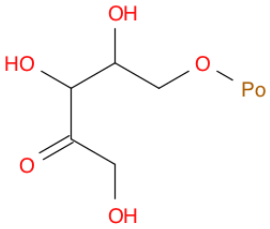
2: glycolate



3: glycolyl-CoA



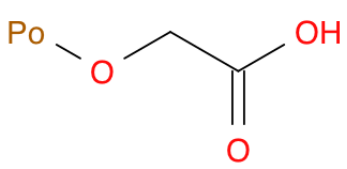
4: glycolaldehyde



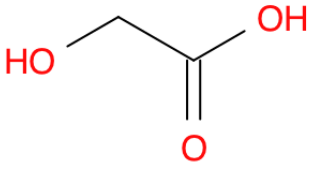
5: C5-ketose 5-phosphate

Number of reactions	4
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose 5-phosphate
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	GAP
Co-product	

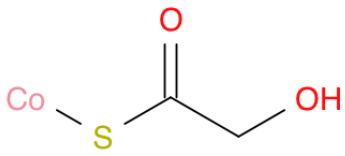
Pathway 47



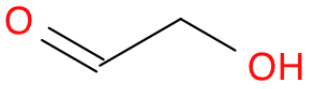
1: glycolate 2-phosphate



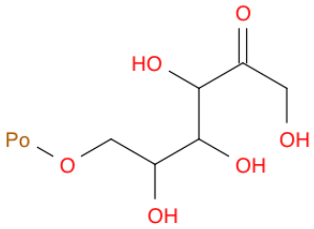
2: glycolate



3: glycolyl-CoA



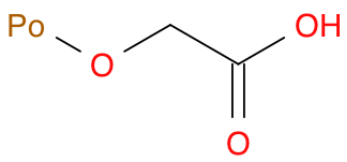
4: glycolaldehyde



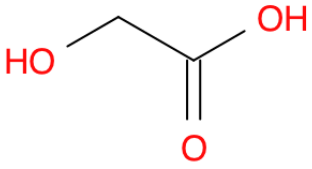
5: C6-ketose 6-phosphate

Number of reactions	4
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C6-ketose 6-phosphate
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

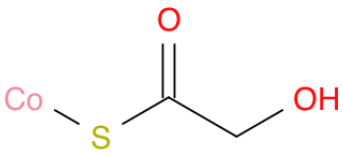
Pathway 48



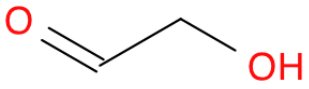
1: glycolate 2-phosphate



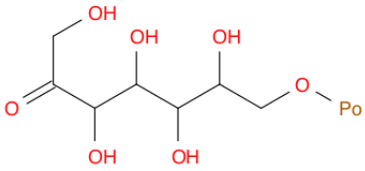
2: glycolate



3: glycolyl-CoA



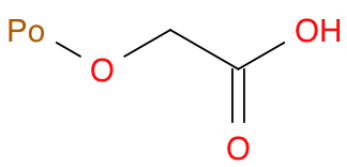
4: glycolaldehyde



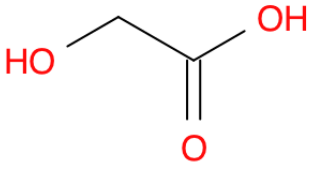
5: C7-ketose 7-phosphate

Number of reactions	4
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C7-ketose 7-phosphate
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

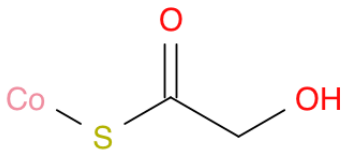
Pathway 49



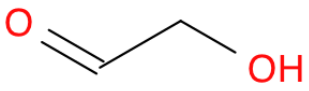
1: glycolate 2-phosphate



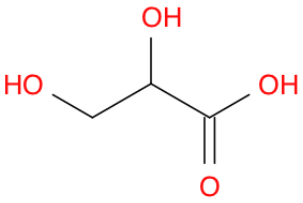
2: glycolate



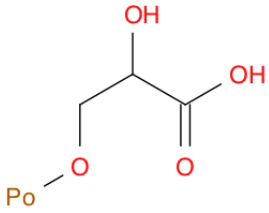
3: glycolyl-CoA



4: glycolaldehyde



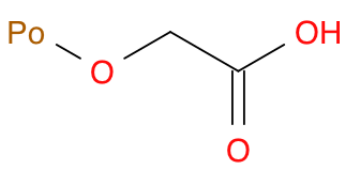
5: glycerate



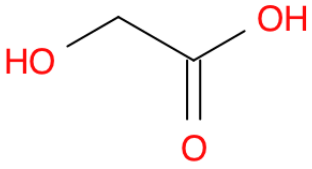
6: glycerate 3-phosphate

Number of reactions	5
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	glycerate
Calvin cycle sink	glycerate 3-phosphate
Key reaction	Reductive carbonyl α-carboxylation
Co-reactant	
Co-product	

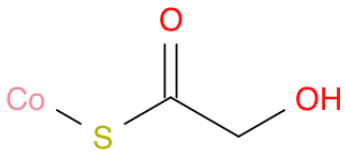
Pathway 50



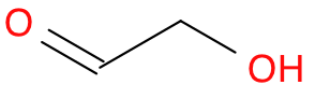
1: glycolate 2-phosphate



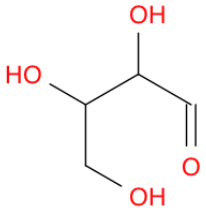
2: glycolate



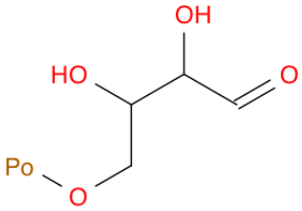
3: glycolyl-CoA



4: glycolaldehyde



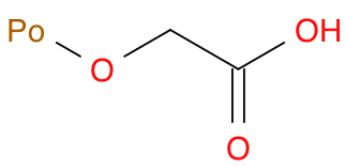
5: C4-aldose



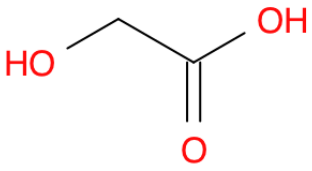
6: C4-aldose 4-phosphate

Number of reactions	5
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde
Key C4 intermediate	C4-aldose
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Carbonyl aldolase (acceptor and donor)
Co-reactant	glycolaldehyde
Co-product	

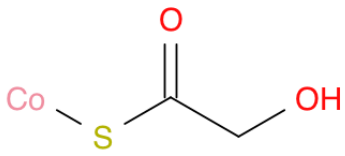
Pathway 51



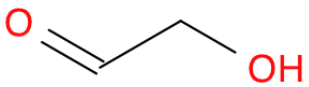
1: glycolate 2-phosphate



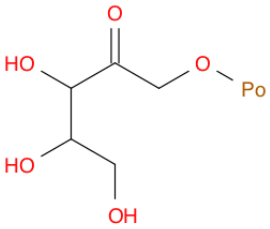
2: glycolate



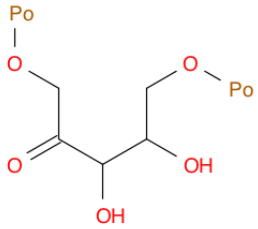
3: glycolyl-CoA



4: glycolaldehyde



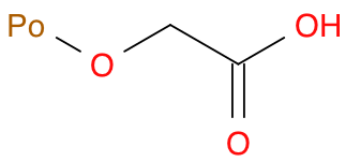
5: C5-ketose 1-phosphate



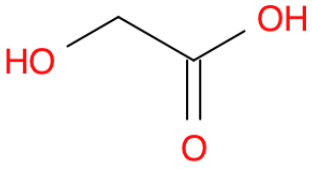
6: C5-ketose 1,5-bisphosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose 1-phosphate
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	DHAP
Co-product	

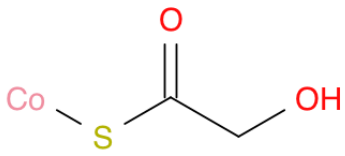
Pathway 52



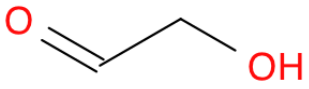
1: glycolate 2-phosphate



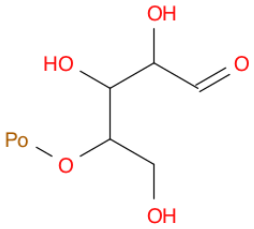
2: glycolate



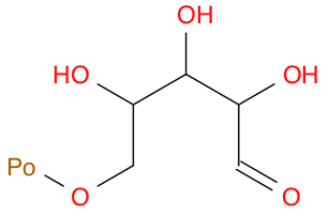
3: glycolyl-CoA



4: glycolaldehyde



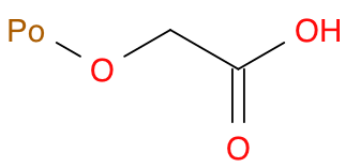
5:



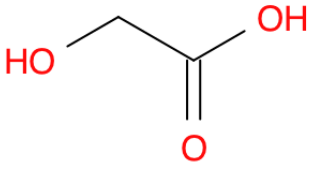
6: C5-aldose 5-phosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

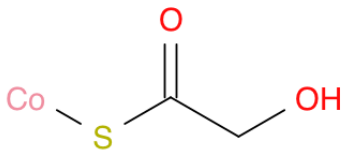
Pathway 53



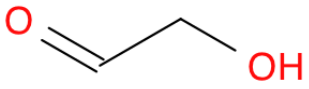
1: glycolate 2-phosphate



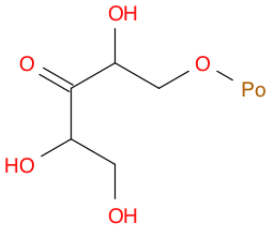
2: glycolate



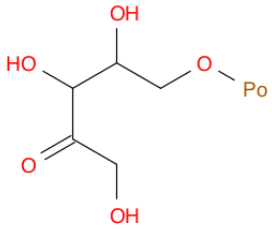
3: glycolyl-CoA



4: glycolaldehyde



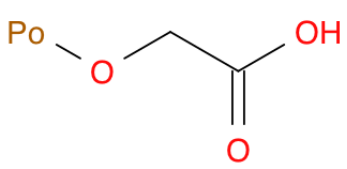
5:



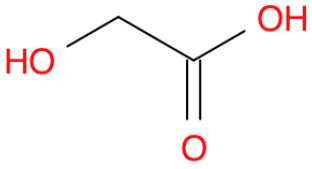
6: C5-ketose 5-phosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

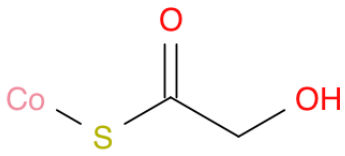
Pathway 54



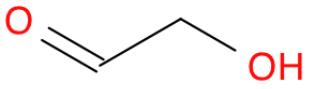
1: glycolate 2-phosphate



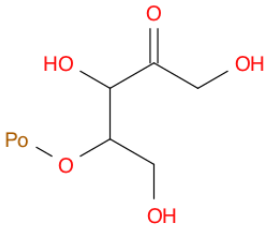
2: glycolate



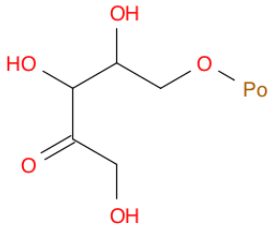
3: glycolyl-CoA



4: glycolaldehyde



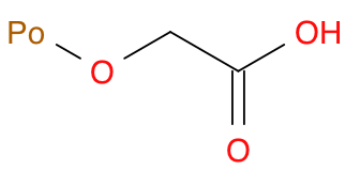
5:



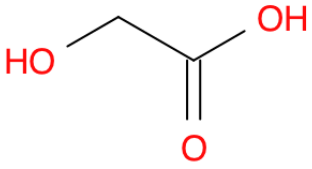
6: C5-ketose 5-phosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

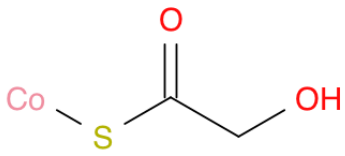
Pathway 55



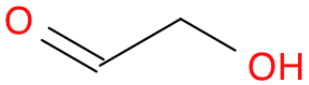
1: glycolate 2-phosphate



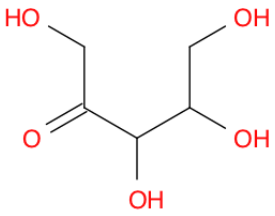
2: glycolate



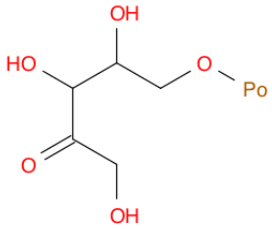
3: glycolyl-CoA



4: glycolaldehyde



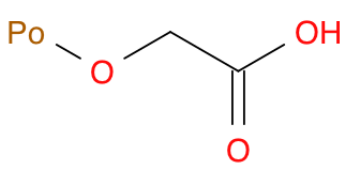
5: C5-ketose



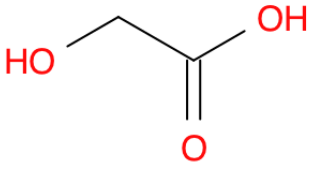
6: C5-ketose 5-phosphate

Number of reactions	5
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

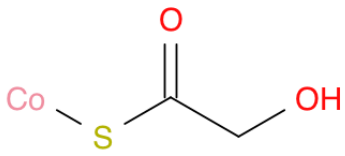
Pathway 56



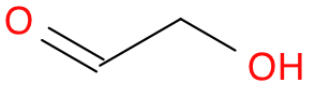
1: glycolate 2-phosphate



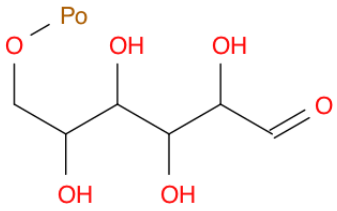
2: glycolate



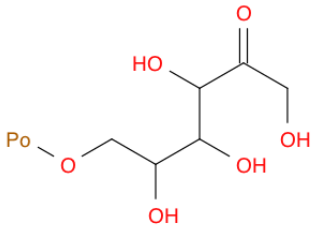
3: glycolyl-CoA



4: glycolaldehyde



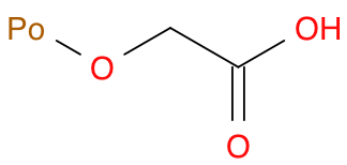
5: C6-aldose 6-phosphate



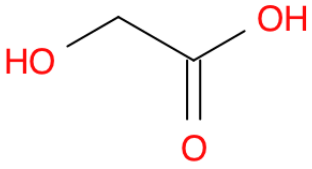
6: C6-ketose 6-phosphate

Number of reactions	5
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C6-aldose 6-phosphate
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

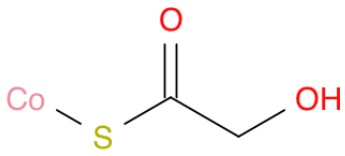
Pathway 57



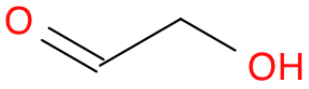
1: glycolate 2-phosphate



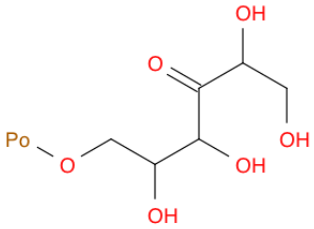
2: glycolate



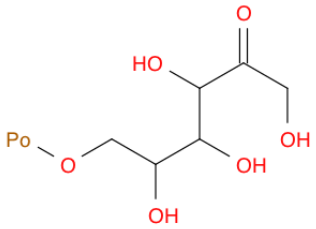
3: glycolyl-CoA



4: glycolaldehyde



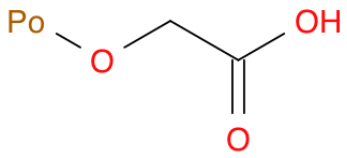
5:



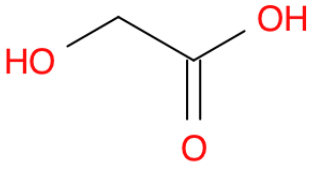
6: C6-ketose 6-phosphate

Number of reactions	5
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

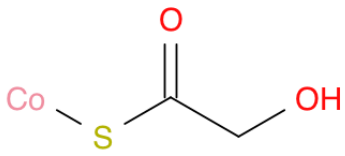
Pathway 58



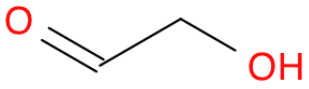
1: glycolate 2-phosphate



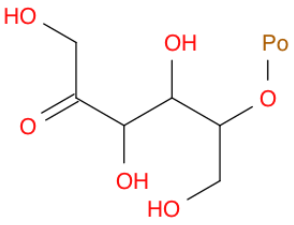
2: glycolate



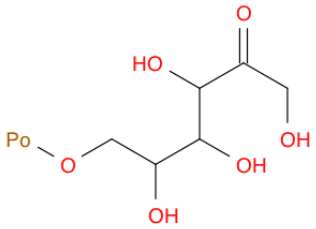
3: glycolyl-CoA



4: glycolaldehyde



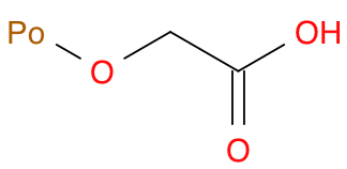
5:



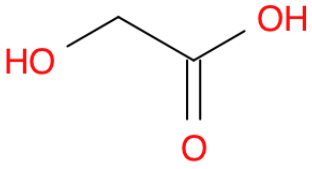
6: C6-ketose 6-phosphate

Number of reactions	5
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O)C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

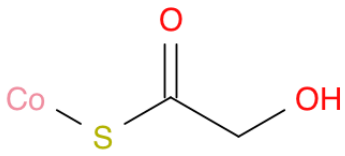
Pathway 59



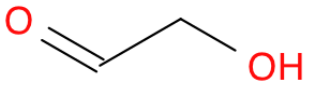
1: glycolate 2-phosphate



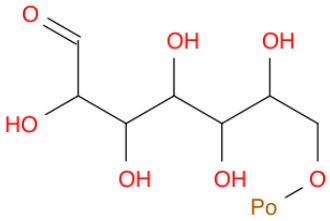
2: glycolate



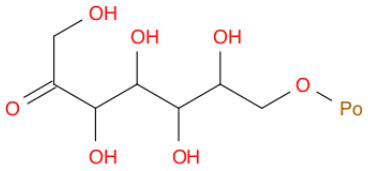
3: glycolyl-CoA



4: glycolaldehyde



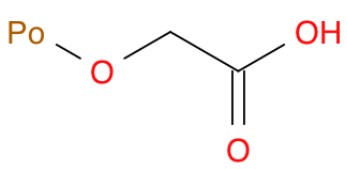
5: C7-aldose 7-phosphate



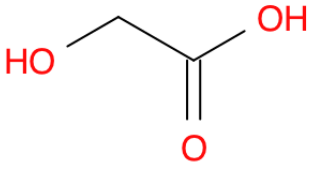
6: C7-ketose 7-phosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C7-aldose 7-phosphate
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

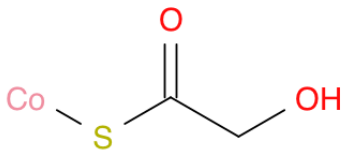
Pathway 60



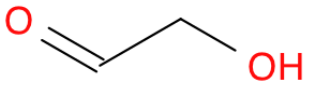
1: glycolate 2-phosphate



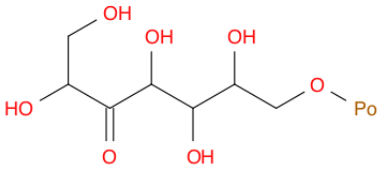
2: glycolate



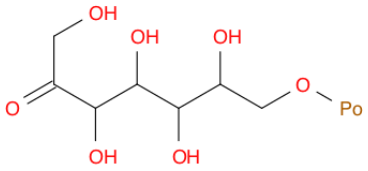
3: glycolyl-CoA



4: glycolaldehyde



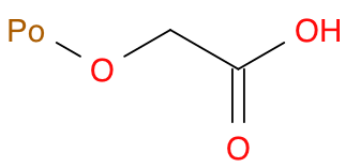
5:



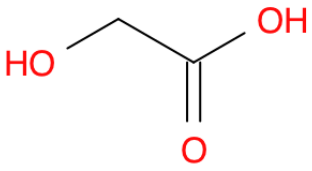
6: C7-ketose 7-phosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

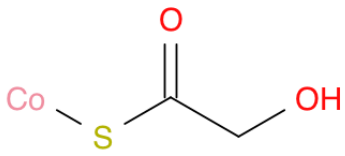
Pathway 61



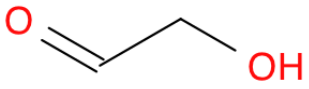
1: glycolate 2-phosphate



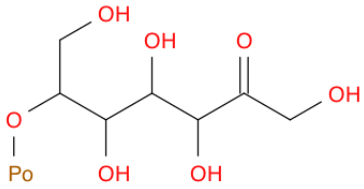
2: glycolate



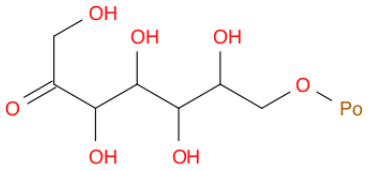
3: glycolyl-CoA



4: glycolaldehyde



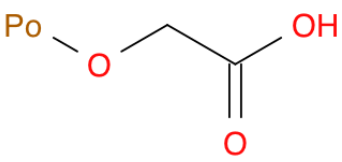
5:



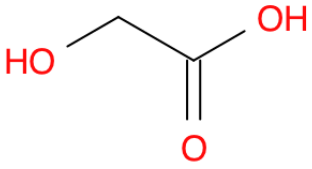
6: C7-ketose 7-phosphate

Number of reactions	5
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

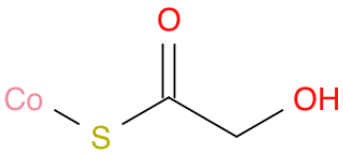
Pathway 62



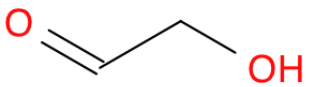
1: glycolate 2-phosphate



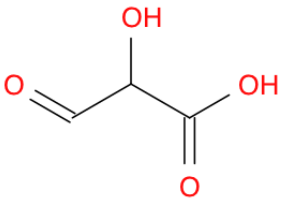
2: glycolate



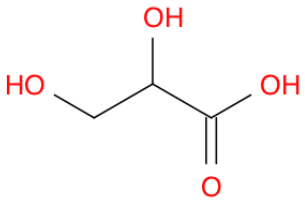
3: glycolyl-CoA



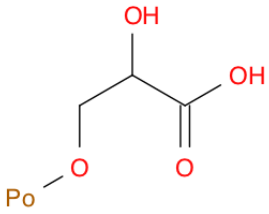
4: glycolaldehyde



5: tartronic semialdehyde



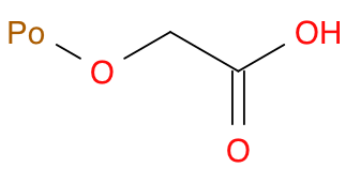
6: glycerate



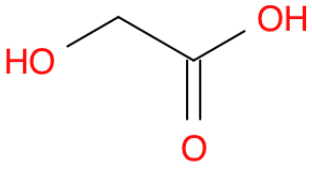
7: glycerate 3-phosphate

Number of reactions	6
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	tartronic semialdehyde
Calvin cycle sink	glycerate 3-phosphate
Key reaction	Carbonyl α-carboxylation
Co-reactant	
Co-product	

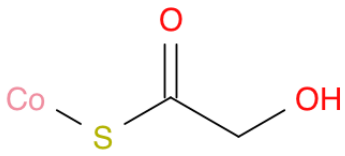
Pathway 63



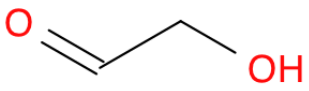
1: glycolate 2-phosphate



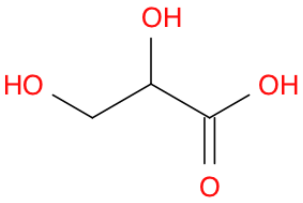
2: glycolate



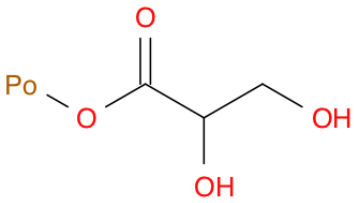
3: glycolyl-CoA



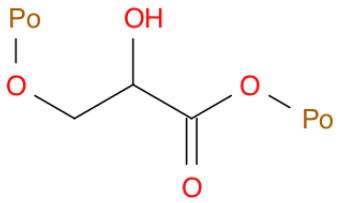
4: glycolaldehyde



5: glycerate



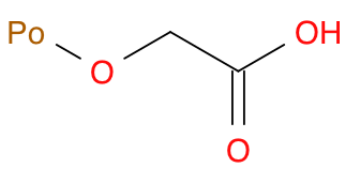
6: glyceroyl phosphate



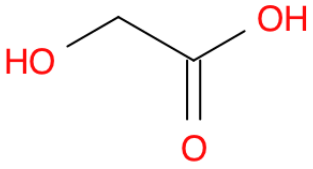
7: glycerate 1,3-bisphosphate

Number of reactions	6
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	glycerate
Calvin cycle sink	glycerate 1,3-bisphosphate
Key reaction	Reductive carbonyl α-carboxylation
Co-reactant	
Co-product	

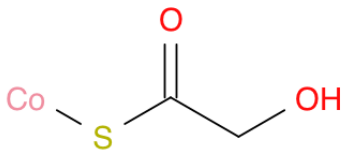
Pathway 64



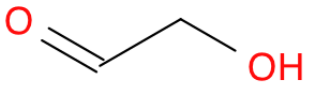
1: glycolate 2-phosphate



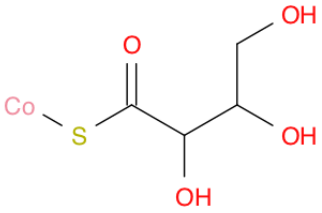
2: glycolate



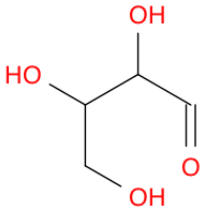
3: glycolyl-CoA



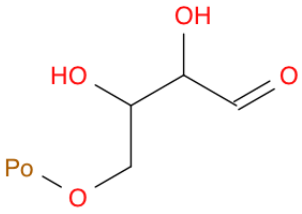
4: glycolaldehyde



5:



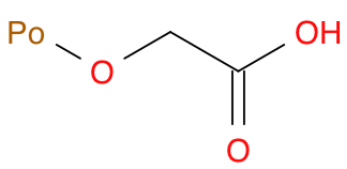
6: C4-aldose



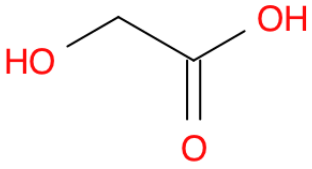
7: C4-aldose 4-phosphate

Number of reactions	6
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde
Key C4 intermediate	C(=O)(S[CoA])C(O)C(O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Thioester aldolase (acceptor)
Co-reactant	glycolyl-CoA
Co-product	

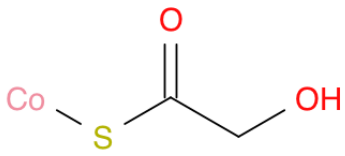
Pathway 65



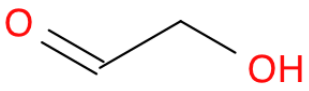
1: glycolate 2-phosphate



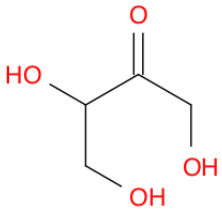
2: glycolate



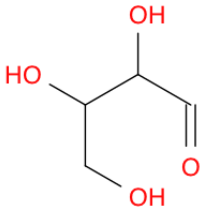
3: glycolyl-CoA



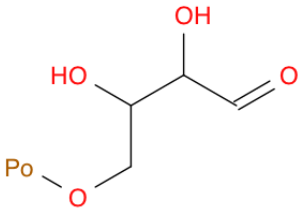
4: glycolaldehyde



5: C4-ketose



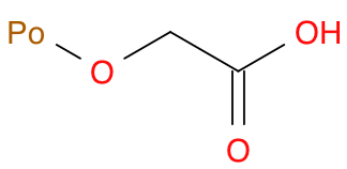
6: C4-aldose



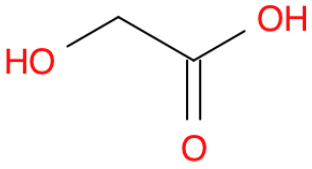
7: C4-aldose 4-phosphate

Number of reactions	6
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde
Key C4 intermediate	C4-ketose
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Ketolase (acceptor and donor) or Transketolase (acceptor)
Co-reactant	glycolaldehyde
Co-product	

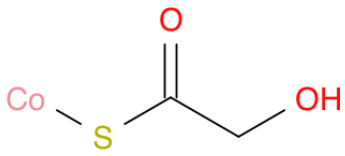
Pathway 66



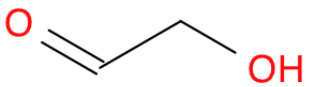
1: glycolate 2-phosphate



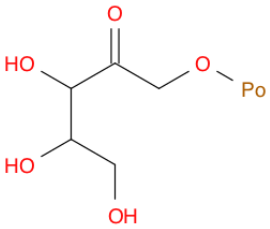
2: glycolate



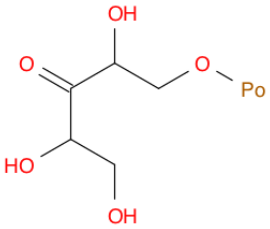
3: glycolyl-CoA



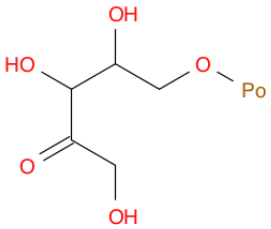
4: glycolaldehyde



5: C5-ketose 1-phosphate



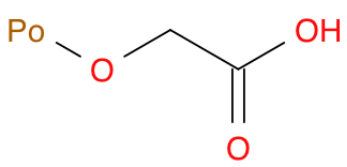
6:



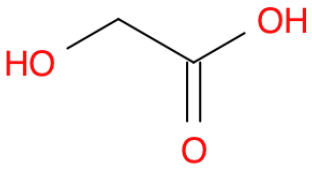
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose 1-phosphate
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	DHAP
Co-product	

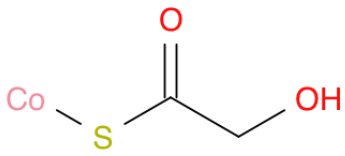
Pathway 67



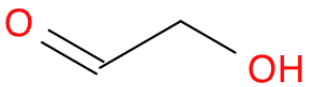
1: glycolate 2-phosphate



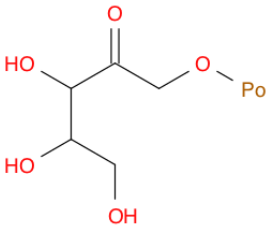
2: glycolate



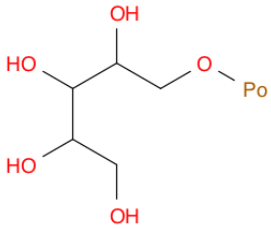
3: glycolyl-CoA



4: glycolaldehyde



5: C5-ketose 1-phosphate



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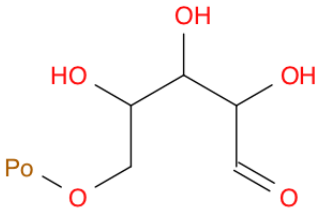
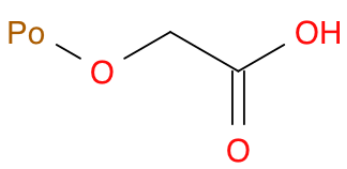


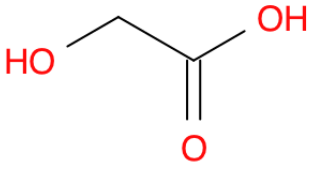
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose 1-phosphate
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	DHAP
Co-product	

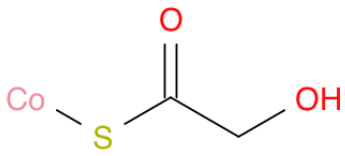
Pathway 68



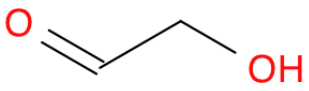
1: glycolate 2-phosphate



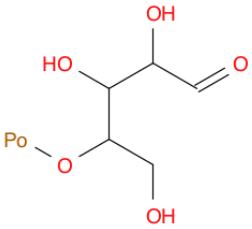
2: glycolate



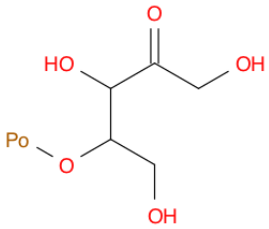
3: glycolyl-CoA



4: glycolaldehyde



5:



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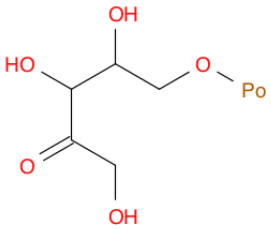
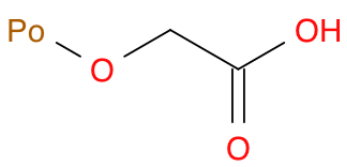


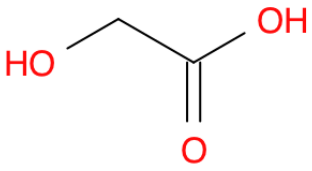
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

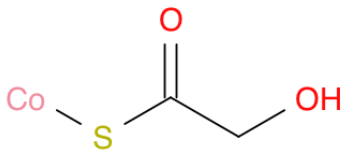
Pathway 69



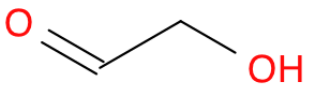
1: glycolate 2-phosphate



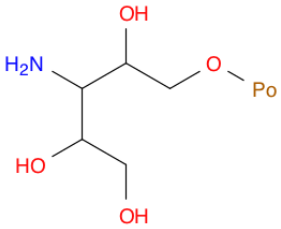
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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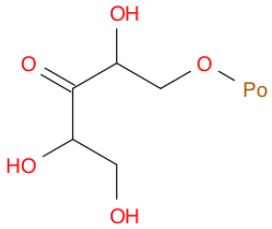

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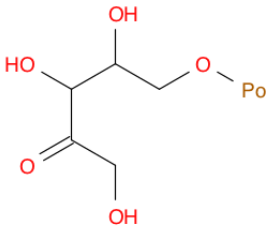
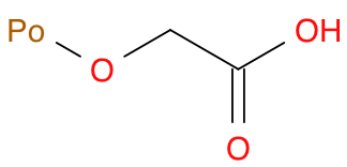


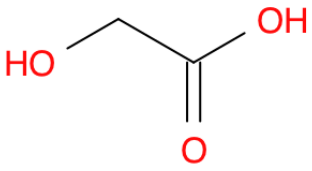
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O[PO3])C(O)C(N)C(O)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

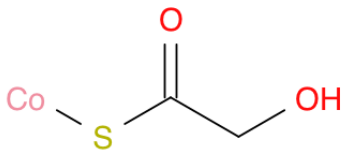
Pathway 70



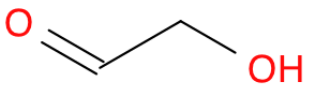
1: glycolate 2-phosphate



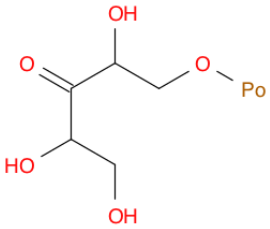
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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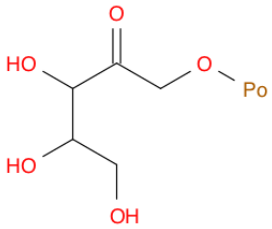
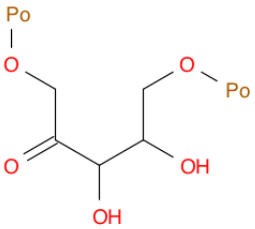


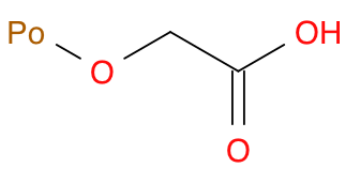
6: C5-ketose 1-phosphate



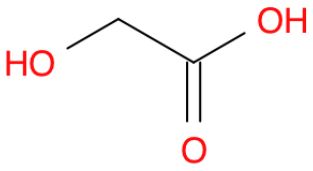
7: C5-ketose 1,5-bisphosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

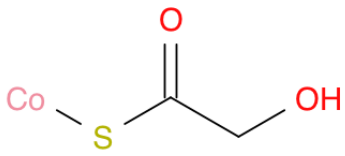
Pathway 71



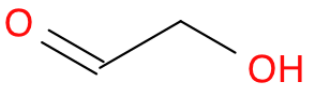
1: glycolate 2-phosphate



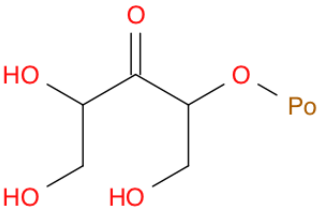
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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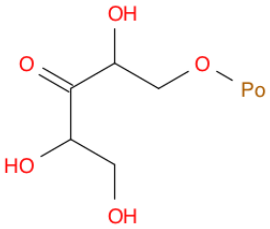

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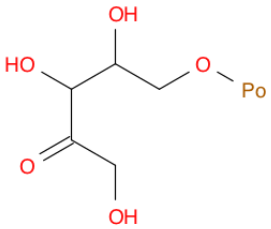
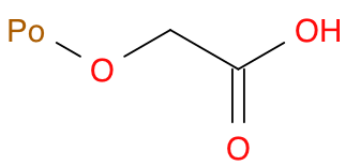


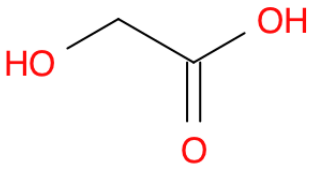
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(O[PO3])C(=O)C(O)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

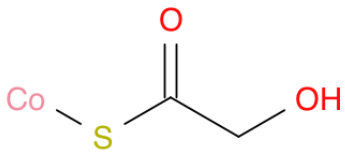
Pathway 72



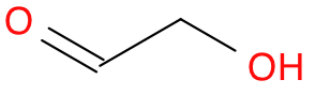
1: glycolate 2-phosphate



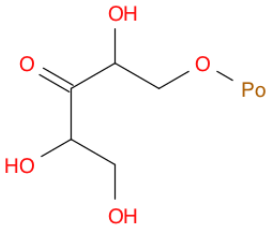
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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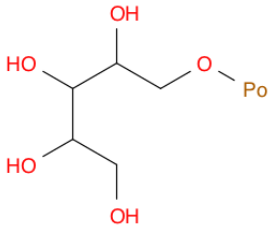

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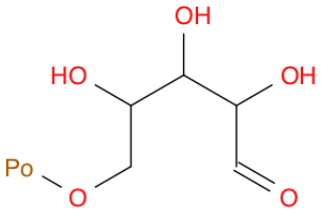
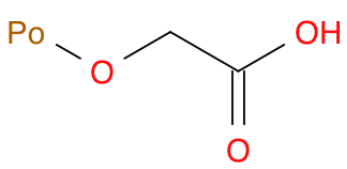


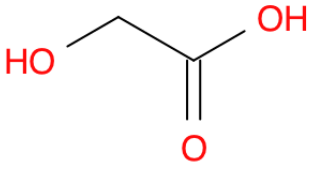
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

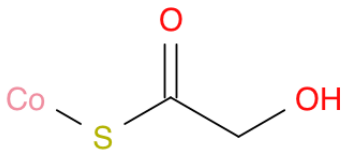
Pathway 73



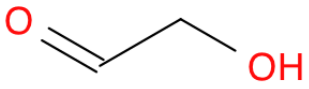
1: glycolate 2-phosphate



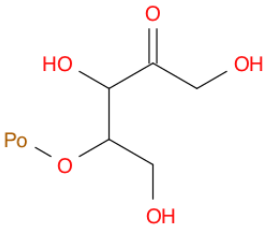
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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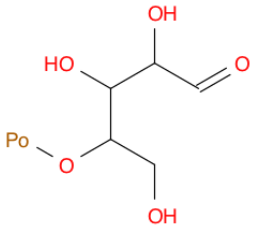

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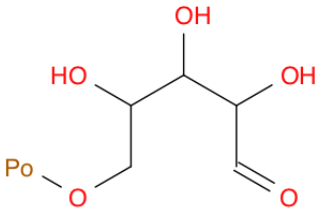
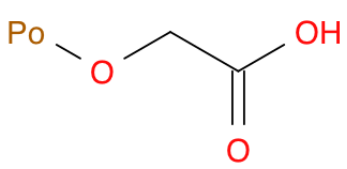


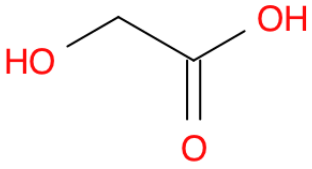
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

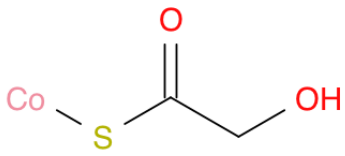
Pathway 74



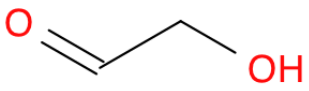
1: glycolate 2-phosphate



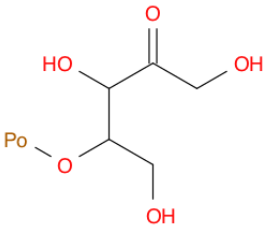
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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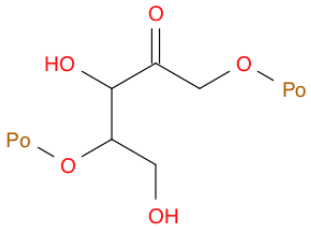

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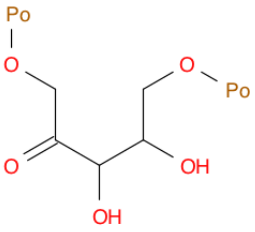
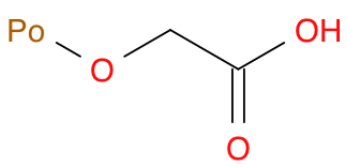


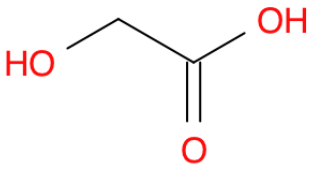
7: C5-ketose 1,5-bisphosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

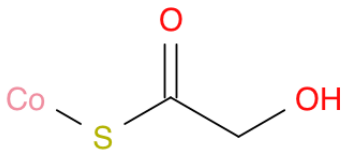
Pathway 75



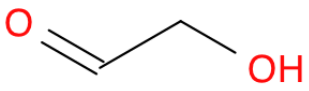
1: glycolate 2-phosphate



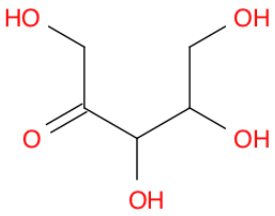
2: glycolate



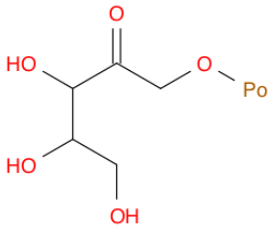
3: glycolyl-CoA



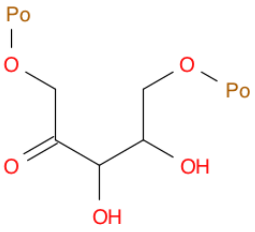
4: glycolaldehyde



5: C5-ketose



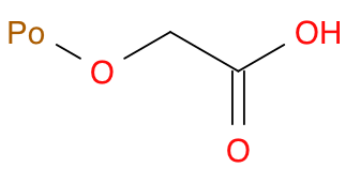
6: C5-ketose 1-phosphate



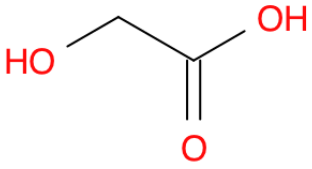
7: C5-ketose 1,5-bisphosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

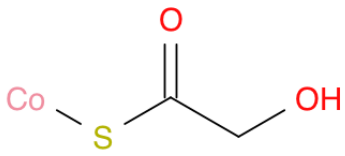
Pathway 76



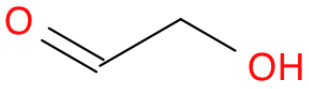
1: glycolate 2-phosphate



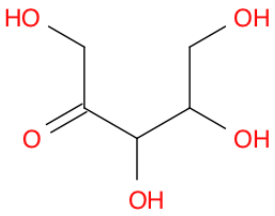
2: glycolate



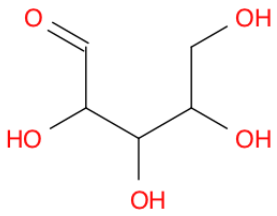
3: glycolyl-CoA



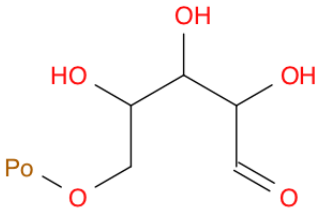
4: glycolaldehyde



5: C5-ketose



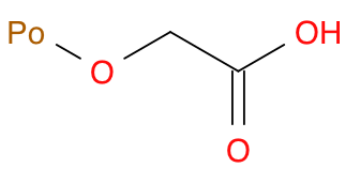
6: C5-aldose



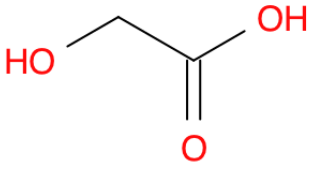
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C5-ketose
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

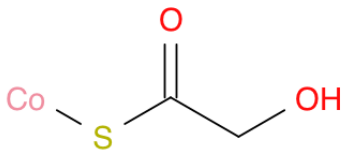
Pathway 77



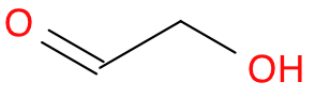
1: glycolate 2-phosphate



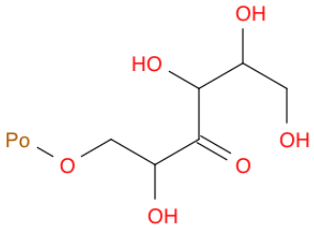
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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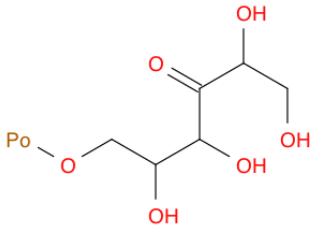

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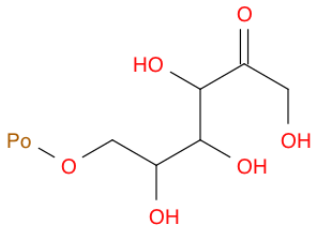
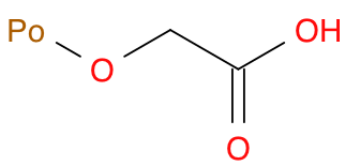


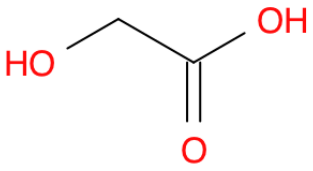
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C4-ketose 4-phosphate
Co-product	

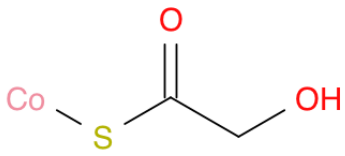
Pathway 78



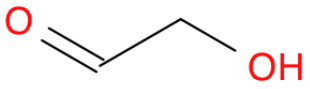
1: glycolate 2-phosphate



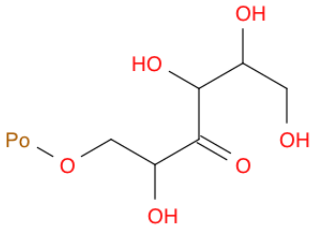
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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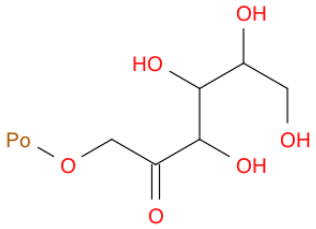
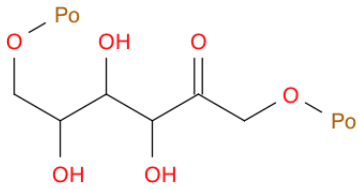


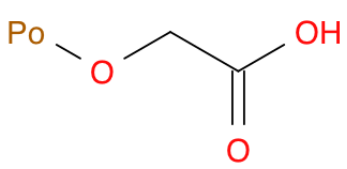
6: C6-ketose 1-phosphate



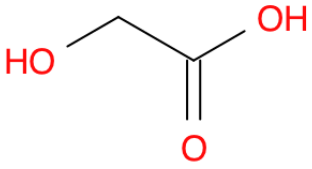
7: C6-ketose 1,6-bisphosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C4-ketose 4-phosphate
Co-product	

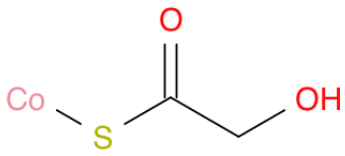
Pathway 79



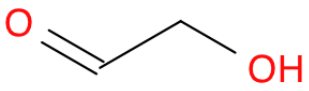
1: glycolate 2-phosphate



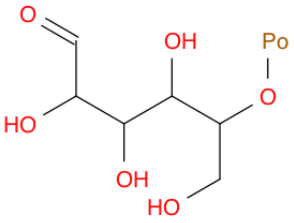
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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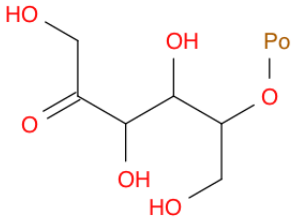

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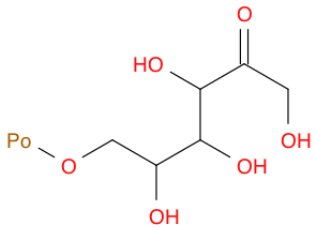
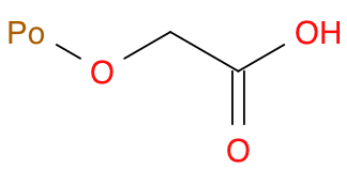


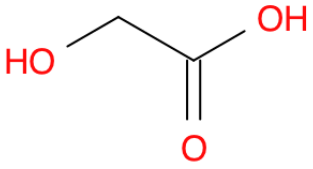
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

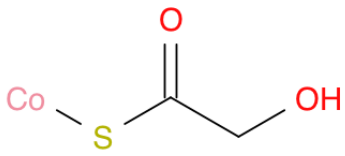
Pathway 80



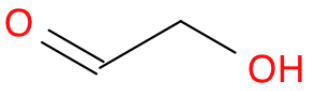
1: glycolate 2-phosphate



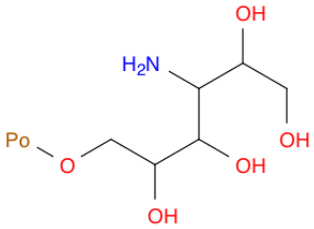
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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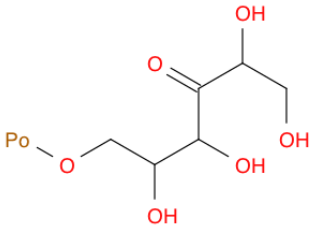

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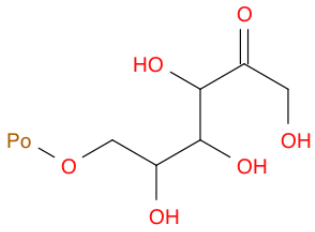
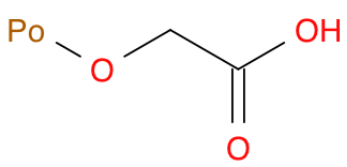


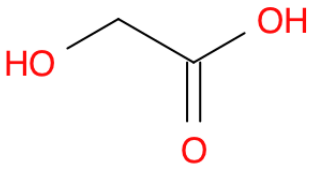
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(O)C(N)C(O)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

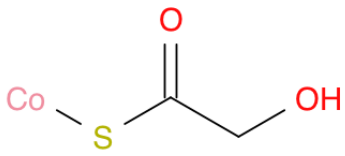
Pathway 81



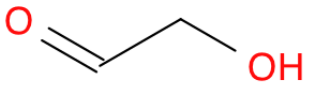
1: glycolate 2-phosphate



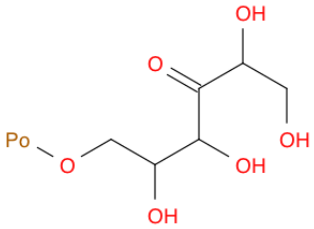
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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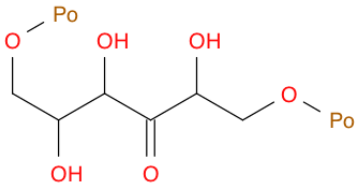

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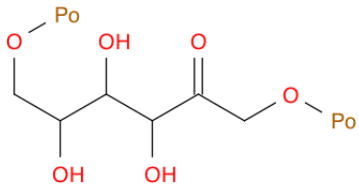
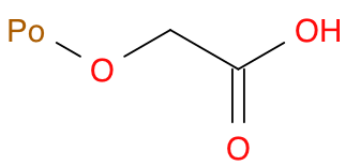


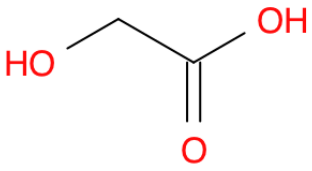
7: C6-ketose 1,6-bisphosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

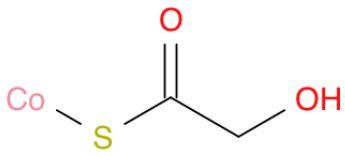
Pathway 82



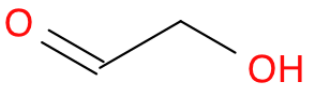
1: glycolate 2-phosphate



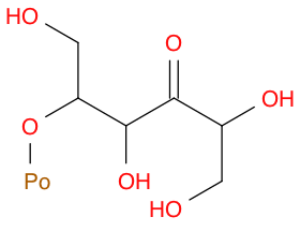
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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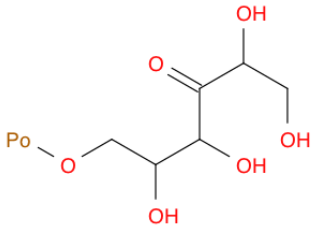

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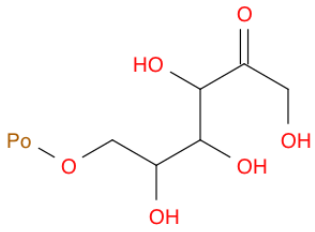
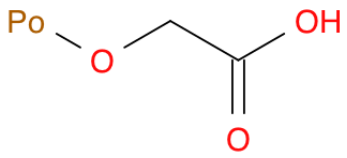


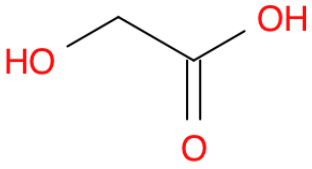
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O)C(O[PO3])C(O)C(=O)C(O)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

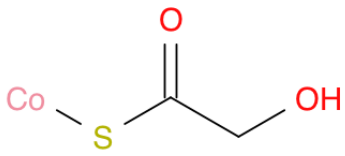
Pathway 83



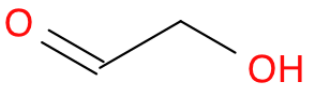
1: glycolate 2-phosphate



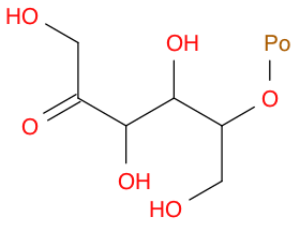
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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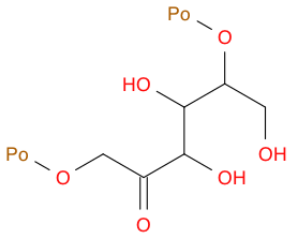

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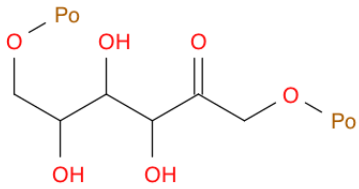
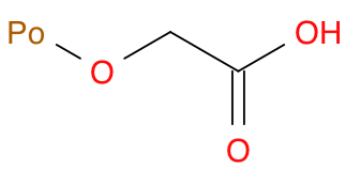


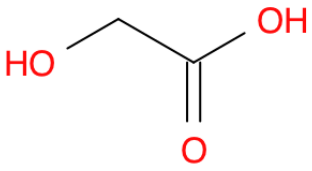
7: C6-ketose 1,6-bisphosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O)C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

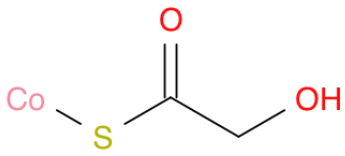
Pathway 84



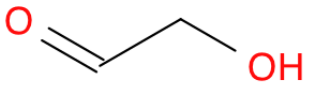
1: glycolate 2-phosphate



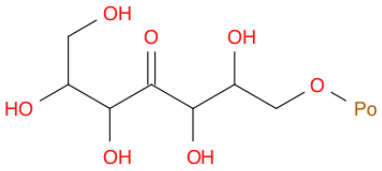
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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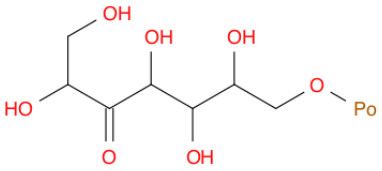

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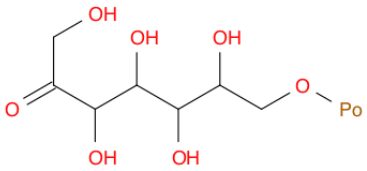
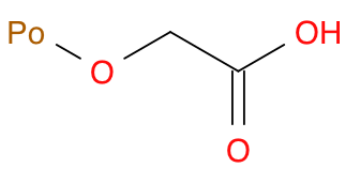


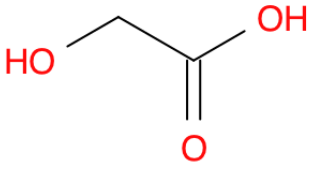
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C5-ketose 5-phosphate
Co-product	

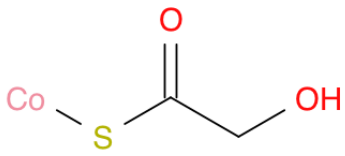
Pathway 85



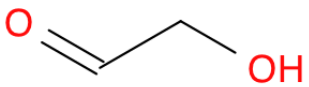
1: glycolate 2-phosphate



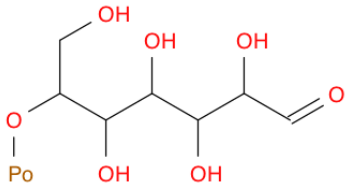
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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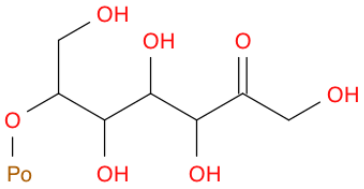

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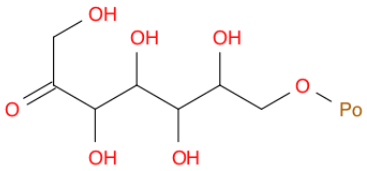
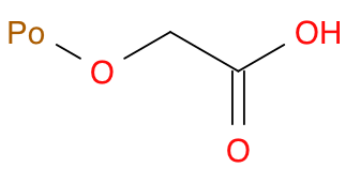


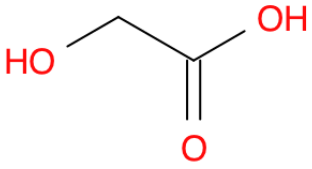
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(=O)C(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

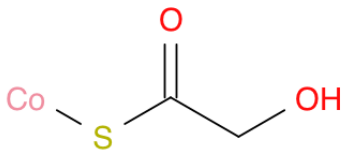
Pathway 86



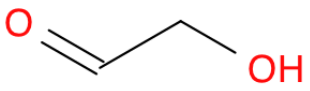
1: glycolate 2-phosphate



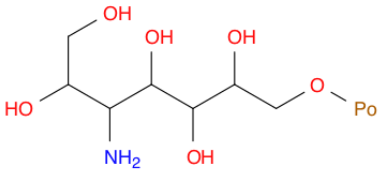
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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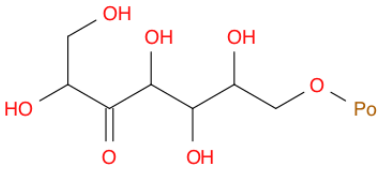

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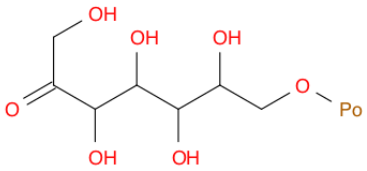
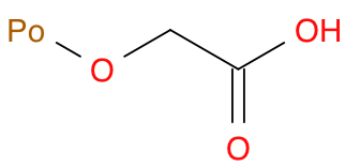


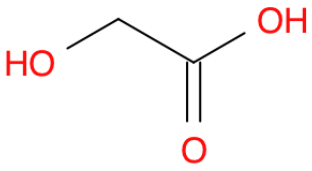
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

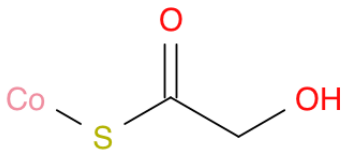
Pathway 87



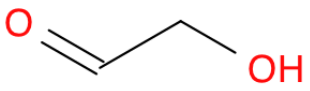
1: glycolate 2-phosphate



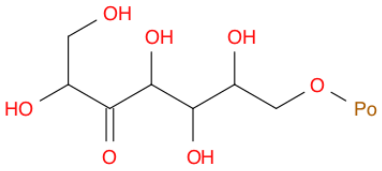
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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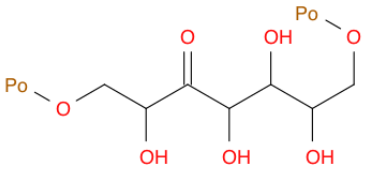

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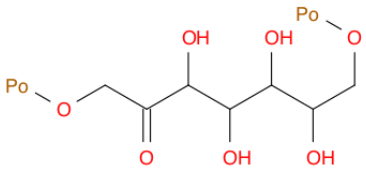
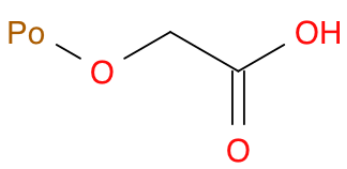


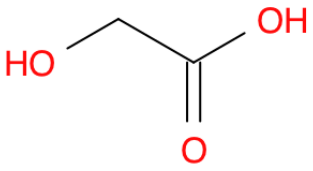
7: C7-ketose 1,7-bisphosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

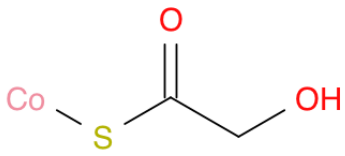
Pathway 88



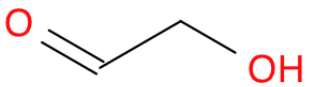
1: glycolate 2-phosphate



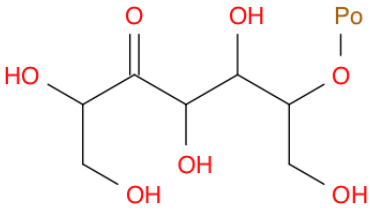
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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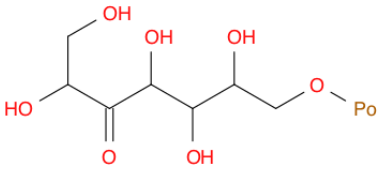

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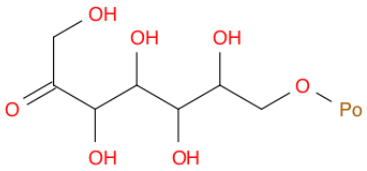
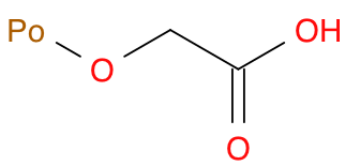


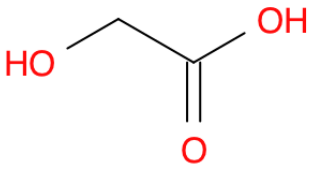
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(O[PO3])C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

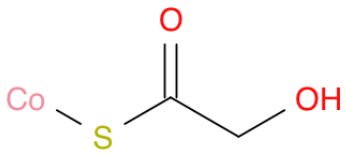
Pathway 89



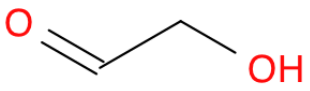
1: glycolate 2-phosphate



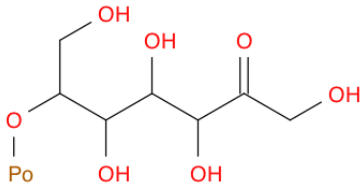
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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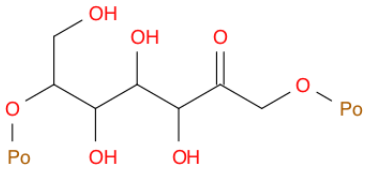

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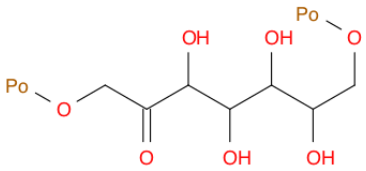
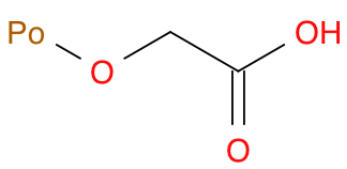


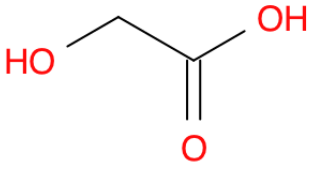
7: C7-ketose 1,7-bisphosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

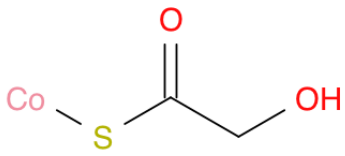
Pathway 90



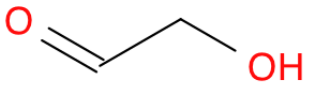
1: glycolate 2-phosphate



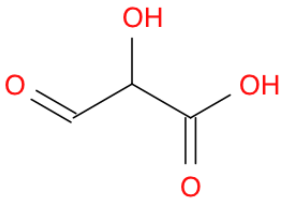
2: glycolate



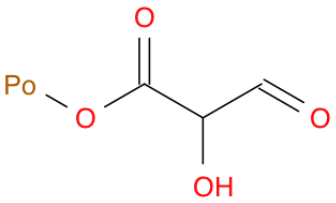
3: glycolyl-CoA



4: glycolaldehyde



5: tartronic semialdehyde



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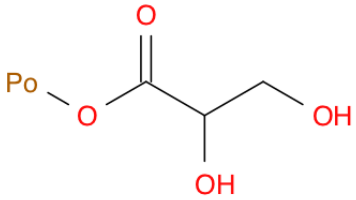
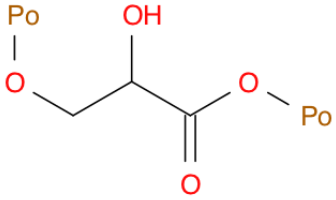


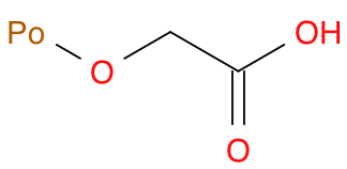
7: glyceroyl phosphate



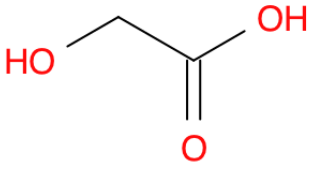
8: glycerate 1,3-bisphosphate

Number of reactions	7
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	tartronic semialdehyde
Calvin cycle sink	glycerate 1,3-bisphosphate
Key reaction	Carbonyl α-carboxylation
Co-reactant	
Co-product	

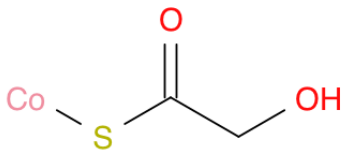
Pathway 91



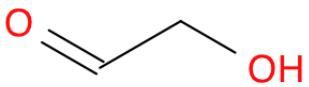
1: glycolate 2-phosphate



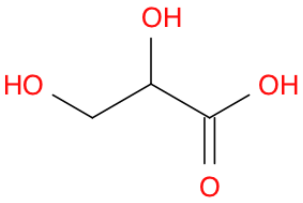
2: glycolate



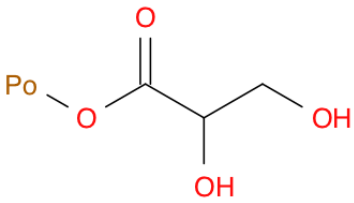
3: glycolyl-CoA



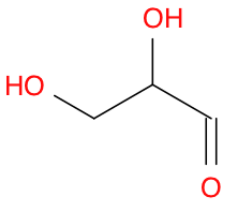
4: glycolaldehyde



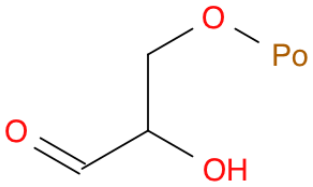
5: glycerate



6: glyceroyl phosphate



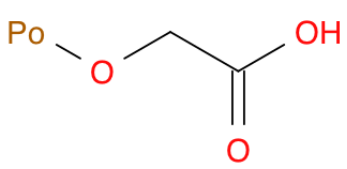
7: glyceraldehyde



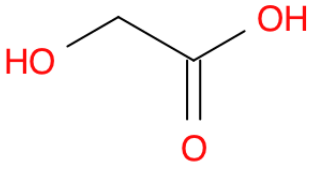
8: GAP

Number of reactions	7
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	glycerate
Calvin cycle sink	GAP
Key reaction	Reductive carbonyl α-carboxylation
Co-reactant	
Co-product	

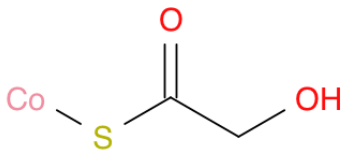
Pathway 92



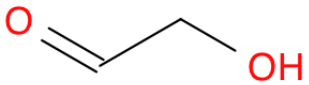
1: glycolate 2-phosphate



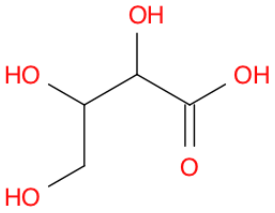
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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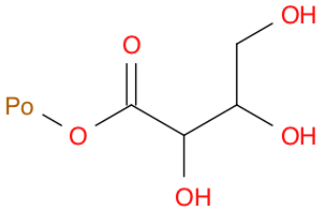

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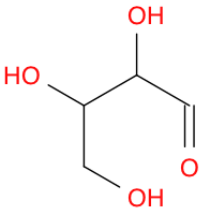
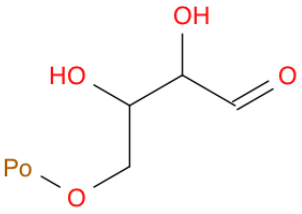


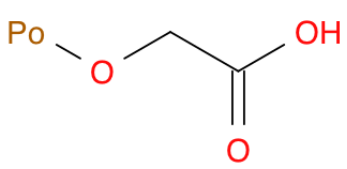
7: C4-aldose



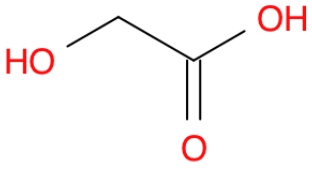
8: C4-aldose 4-phosphate

Number of reactions	7
Pathway superclass	2:4
Key C2 intermediate	glycolaldehyde
Key C4 intermediate	C(=O)(O)C(O)C(O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Thioester aldolase with thioester hydrolysis (acceptor)
Co-reactant	glycolyl-CoA
Co-product	

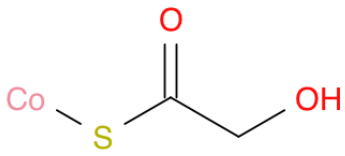
Pathway 93



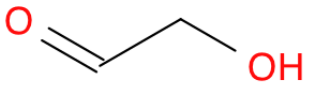
1: glycolate 2-phosphate



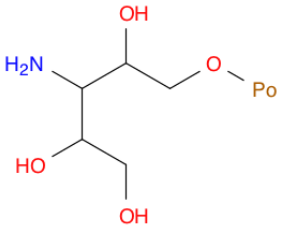
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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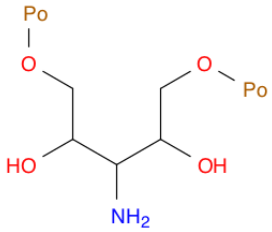

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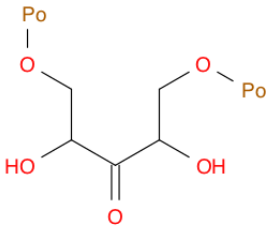

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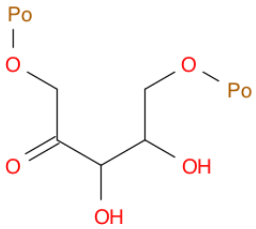
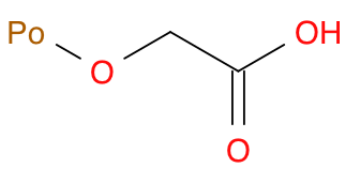


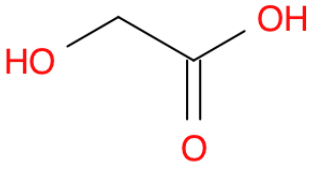
8: C5-ketose 1,5-bisphosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O[PO3])C(O)C(N)C(O)C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

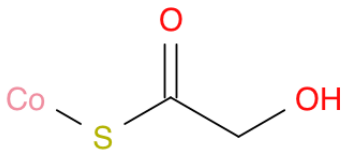
Pathway 94



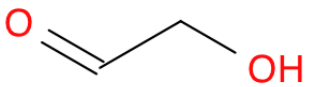
1: glycolate 2-phosphate



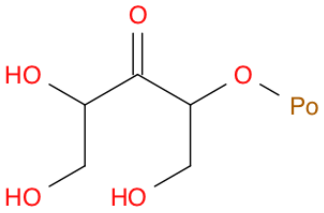
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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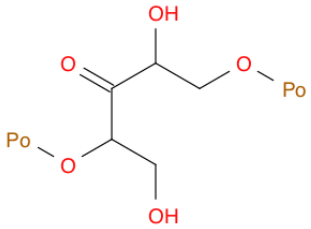

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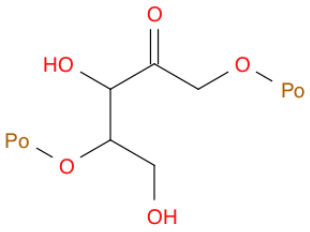

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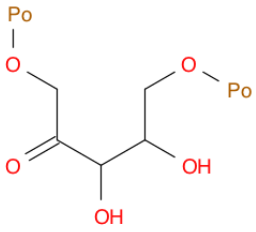
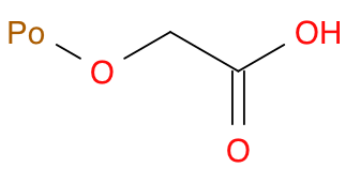


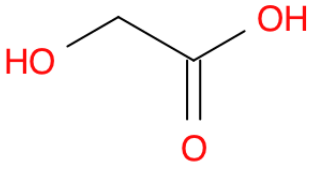
8: C5-ketose 1,5-bisphosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(O[PO3])C(=O)C(O)C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

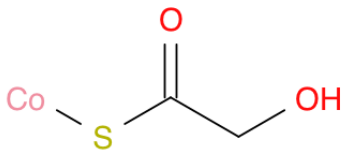
Pathway 95



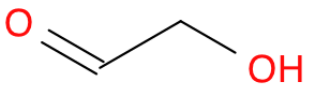
1: glycolate 2-phosphate



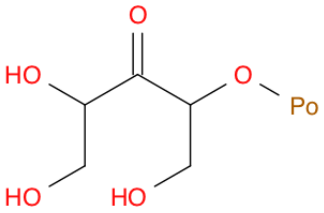
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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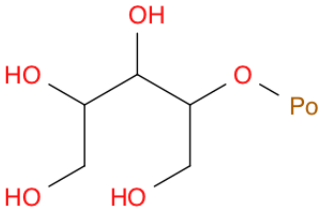

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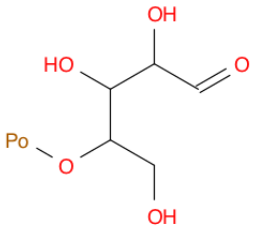

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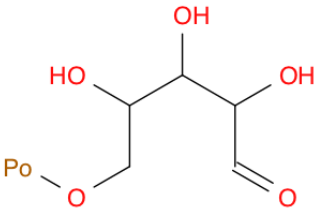
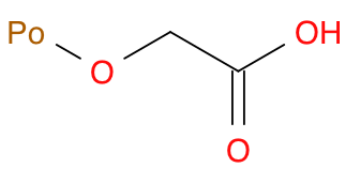


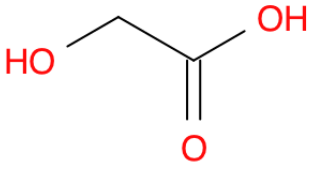
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(O)C(O[PO3])C(=O)C(O)C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

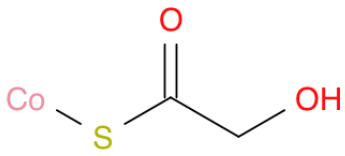
Pathway 96



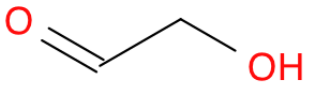
1: glycolate 2-phosphate



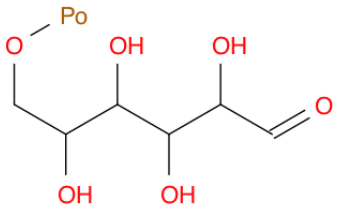
2: glycolate



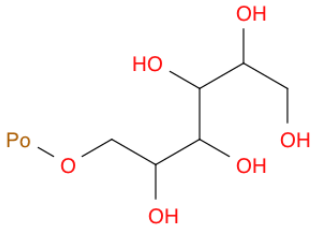
3: glycolyl-CoA



4: glycolaldehyde



5: C6-aldose 6-phosphate



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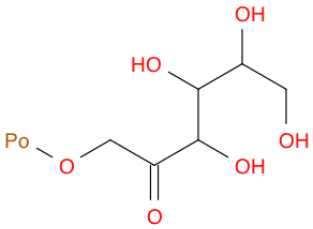
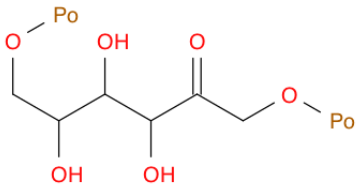


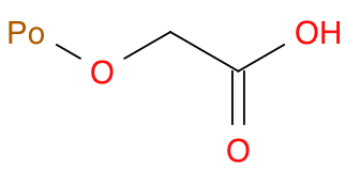
7: C6-ketose 1-phosphate



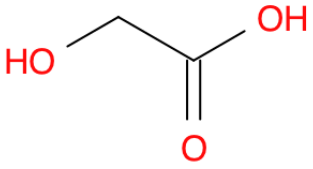
8: C6-ketose 1,6-bisphosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C6-aldose 6-phosphate
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

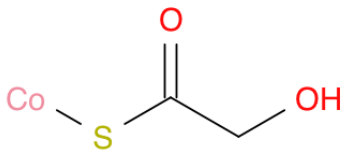
Pathway 97



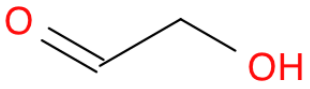
1: glycolate 2-phosphate



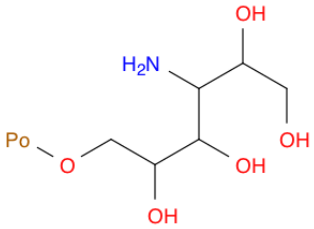
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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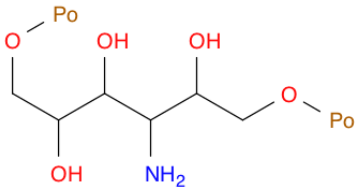

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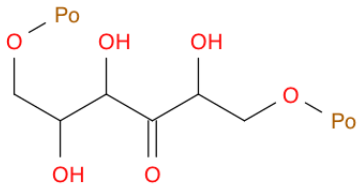

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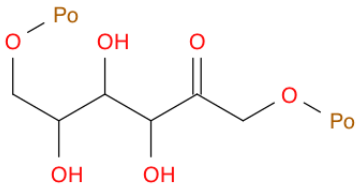
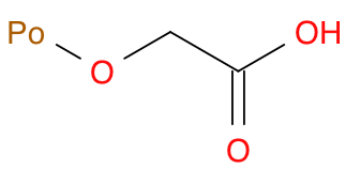


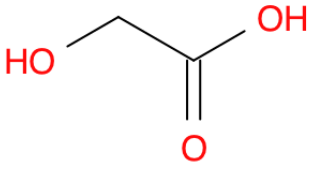
8: C6-ketose 1,6-bisphosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O[PO3])C(O)C(O)C(N)C(O)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

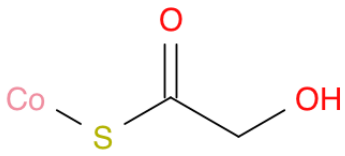
Pathway 98



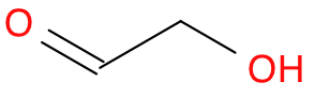
1: glycolate 2-phosphate



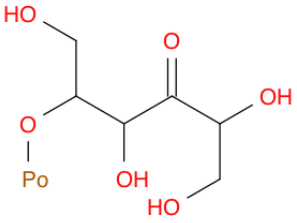
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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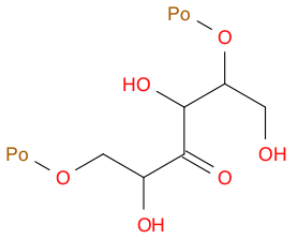

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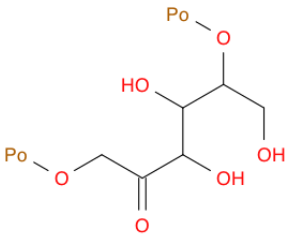

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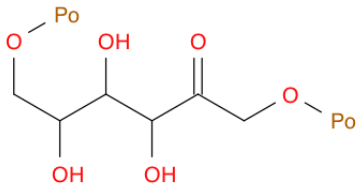
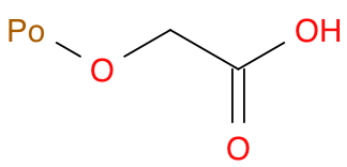


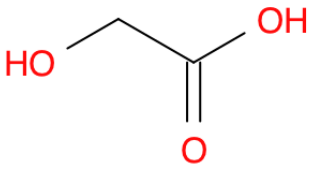
8: C6-ketose 1,6-bisphosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(O)C(O[PO3])C(O)C(=O)C(O)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

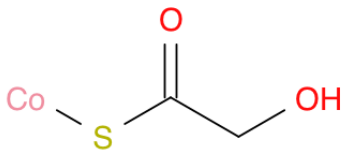
Pathway 99



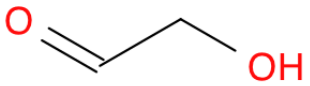
1: glycolate 2-phosphate



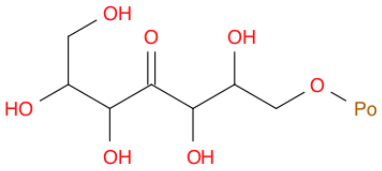
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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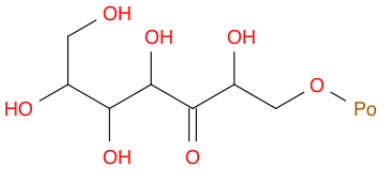

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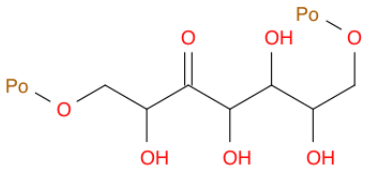

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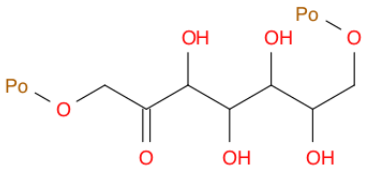
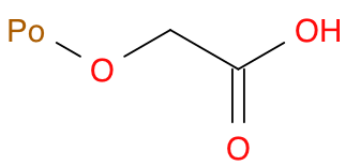


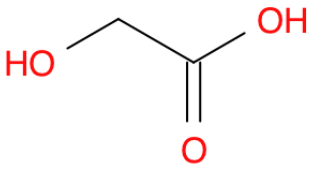
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C5-ketose 5-phosphate
Co-product	

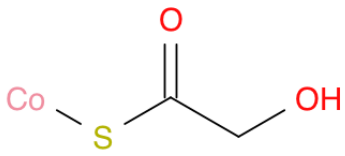
Pathway 100



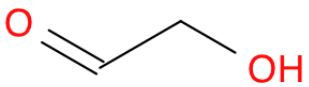
1: glycolate 2-phosphate



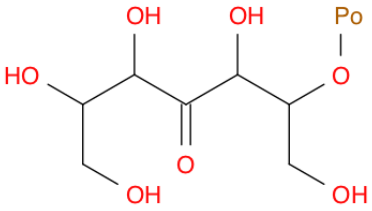
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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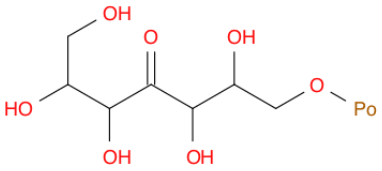

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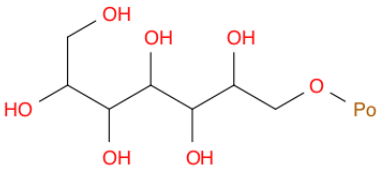

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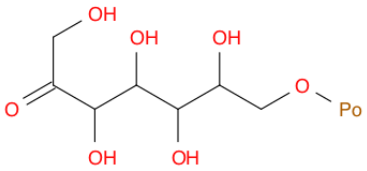
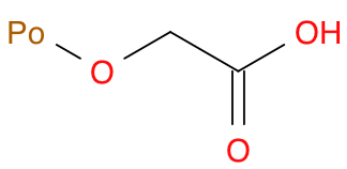


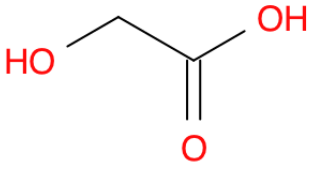
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(O[PO3])C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C(O)C(=O)C(O)C(O[PO3])C(O)
Co-product	

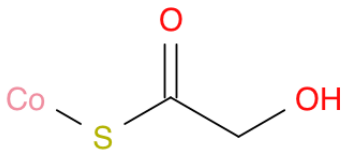
Pathway 101



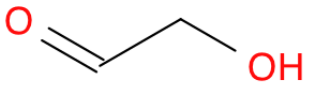
1: glycolate 2-phosphate



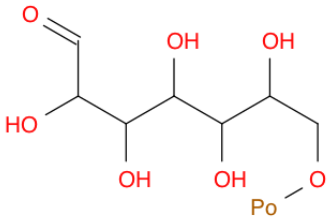
2: glycolate



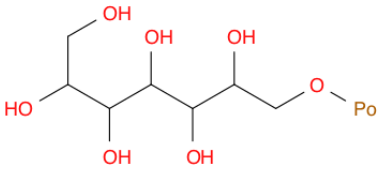
3: glycolyl-CoA



4: glycolaldehyde



5: C7-aldose 7-phosphate



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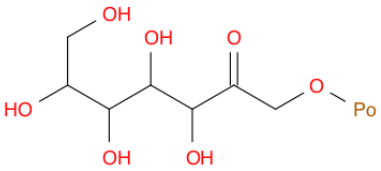
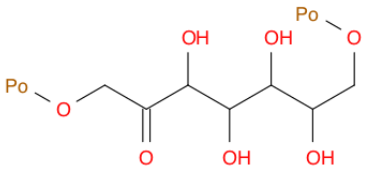


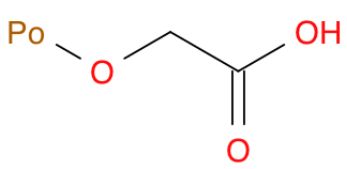
7: C7-ketose 1-phosphate



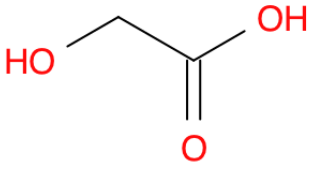
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C7-aldose 7-phosphate
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

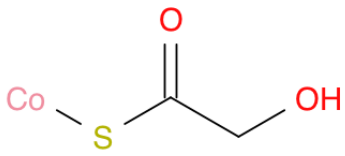
Pathway 102



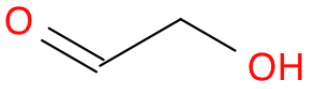
1: glycolate 2-phosphate



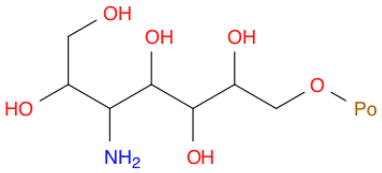
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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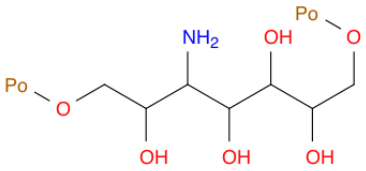

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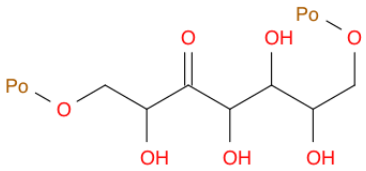

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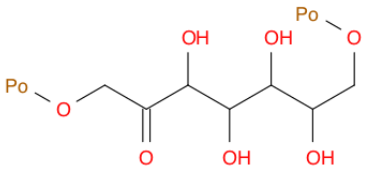
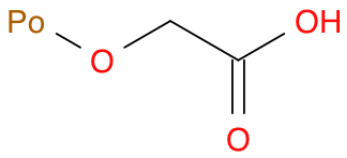


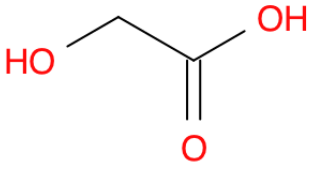
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

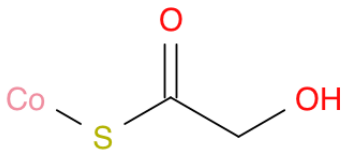
Pathway 103



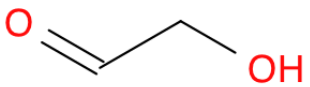
1: glycolate 2-phosphate



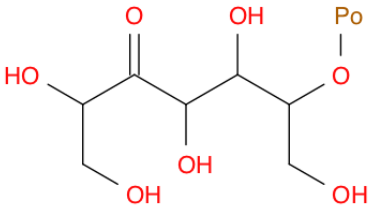
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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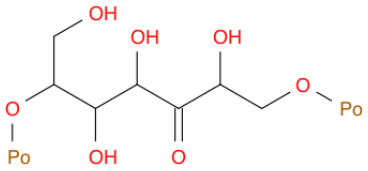

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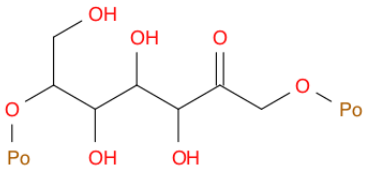

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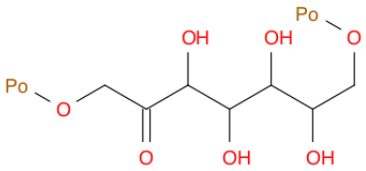
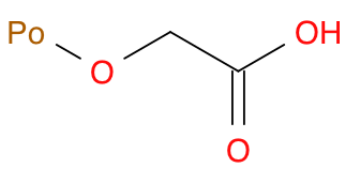


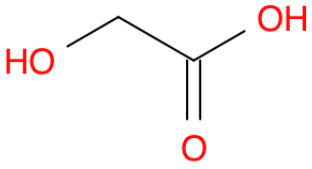
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(O[PO3])C(O)C(O)C(=O)C(O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

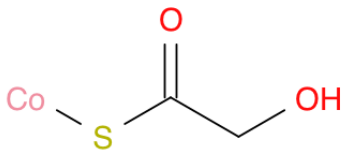
Pathway 104



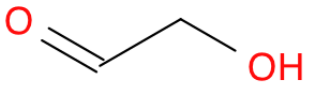
1: glycolate 2-phosphate



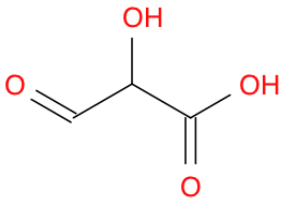
2: glycolate



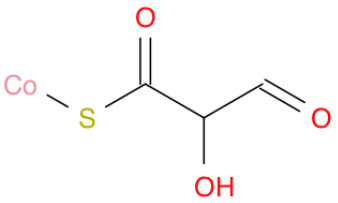
3: glycolyl-CoA



4: glycolaldehyde



5: tartronic semialdehyde



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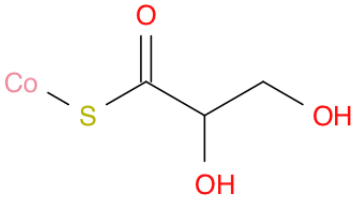
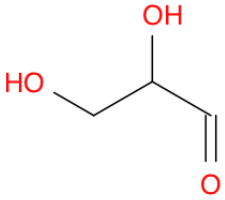
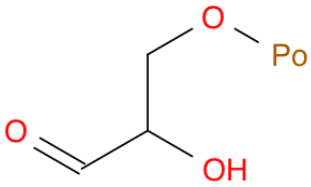


7: glyceryl-CoA



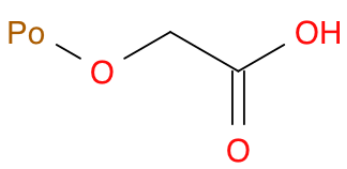
8: glyceraldehyde



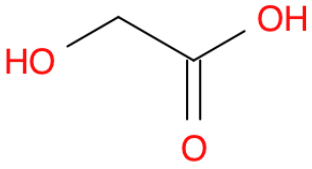
9: GAP

Number of reactions	8
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	tartronic semialdehyde
Calvin cycle sink	GAP
Key reaction	Carbonyl α-carboxylation
Co-reactant	
Co-product	

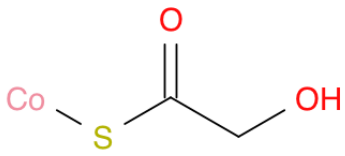
Pathway 105



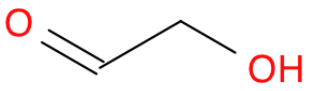
1: glycolate 2-phosphate



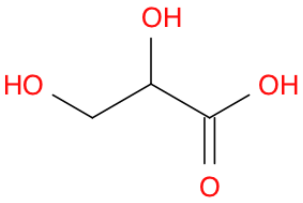
2: glycolate



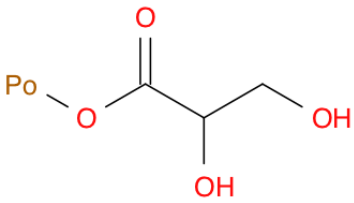
3: glycolyl-CoA



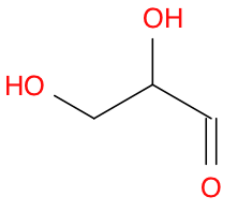
4: glycolaldehyde



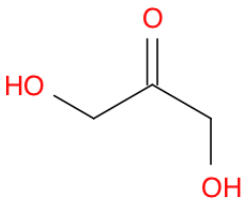
5: glycerate



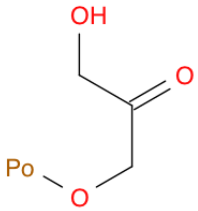
6: glyceroyl phosphate



7: glyceraldehyde



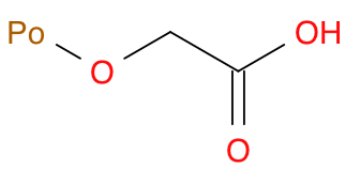
8: DHA



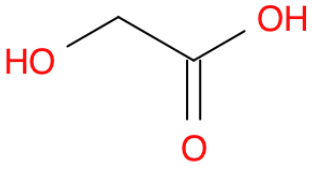
9: DHAP

Number of reactions	8
Pathway superclass	2:3
Key C2 intermediate	glycolaldehyde
Key C3 intermediate	glycerate
Calvin cycle sink	DHAP
Key reaction	Reductive carbonyl α-carboxylation
Co-reactant	
Co-product	

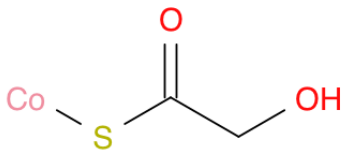
Pathway 106



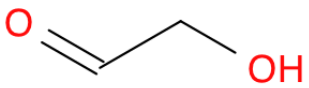
1: glycolate 2-phosphate



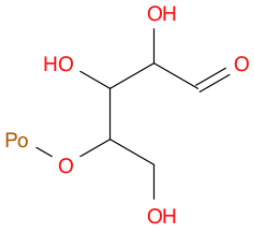
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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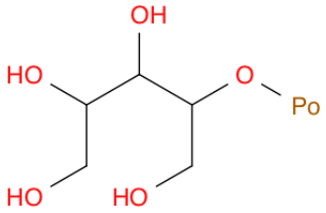
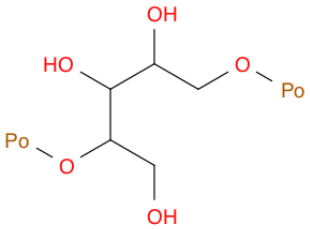


6:



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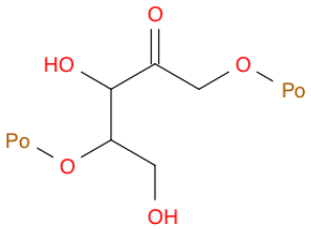
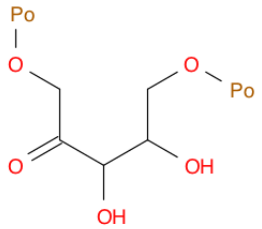


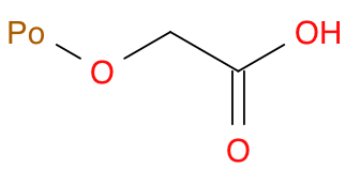
8:



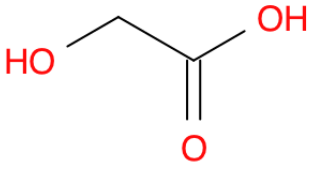
9: C5-ketose 1,5-bisphosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glycolaldehyde
Key C5 intermediate	C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

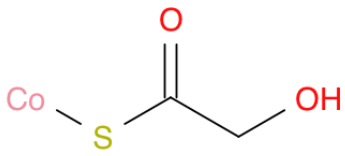
Pathway 107



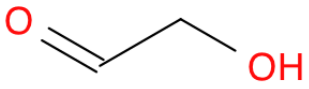
1: glycolate 2-phosphate



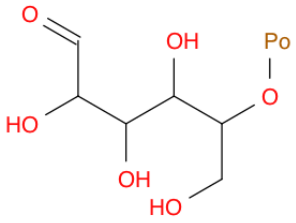
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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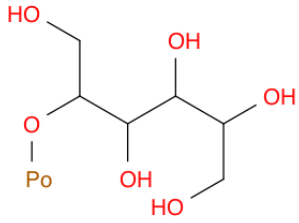

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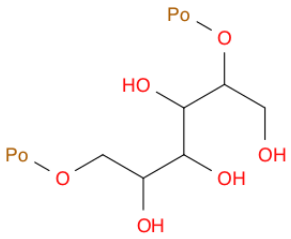

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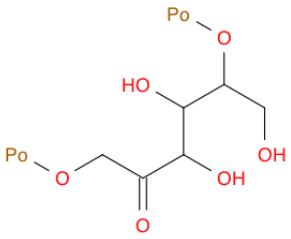

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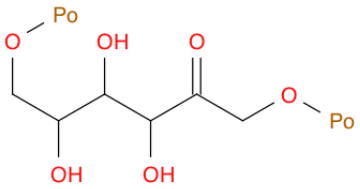
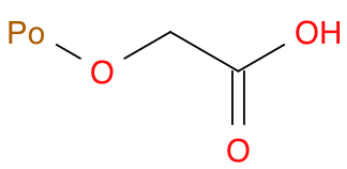


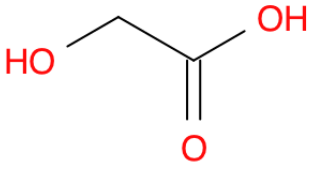
9: C6-ketose 1,6-bisphosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glycolaldehyde
Key C6 intermediate	C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

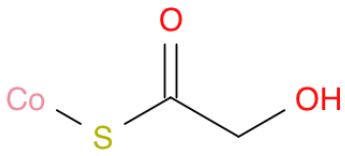
Pathway 108



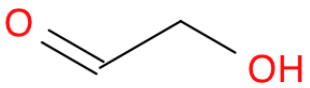
1: glycolate 2-phosphate



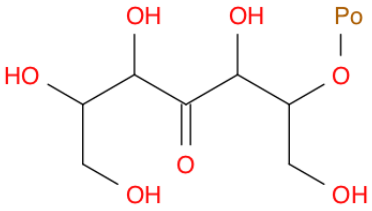
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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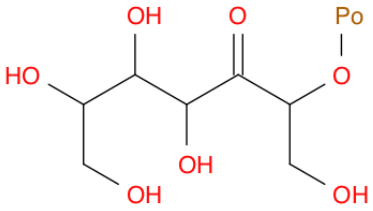

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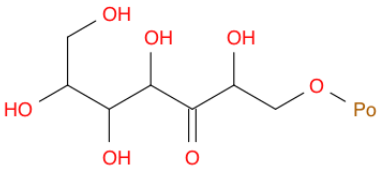

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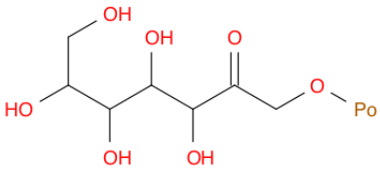
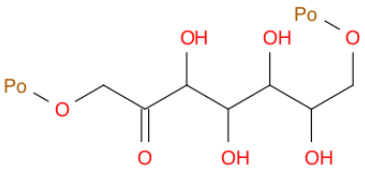


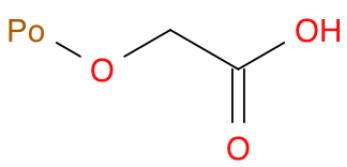
8: C7-ketose 1-phosphate



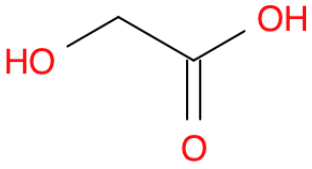
9: C7-ketose 1,7-bisphosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(O)C(O[PO3])C(O)C(=O)C(O)C(O)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C(O)C(=O)C(O)C(O[PO3])C(O)
Co-product	

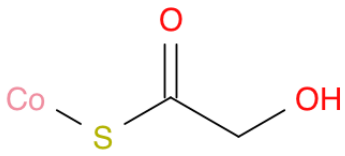
Pathway 109



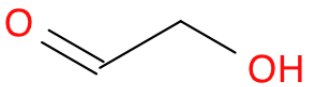
1: glycolate 2-phosphate



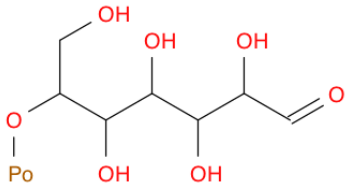
2: glycolate



3: glycolyl-CoA



4: glycolaldehyde



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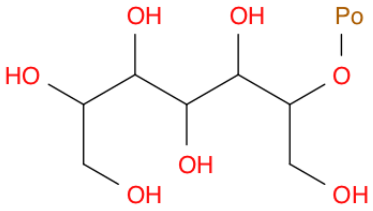

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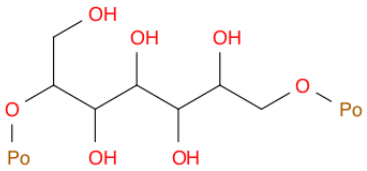
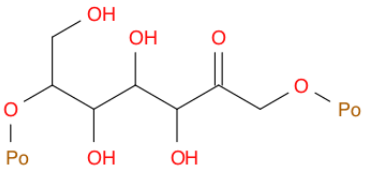
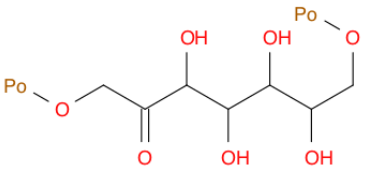


7:



8:

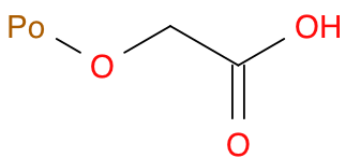


9: C7-ketose 1,7-bisphosphate

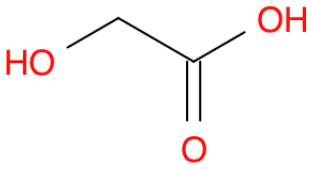
Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glycolaldehyde
Key C7 intermediate	C(=O)C(O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Carbonyl aldolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

4.4 Glyoxylate pathways

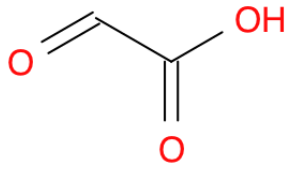
Pathway 110



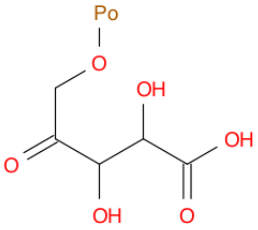
1: glycolate 2-phosphate



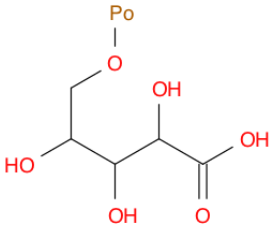
2: glycolate



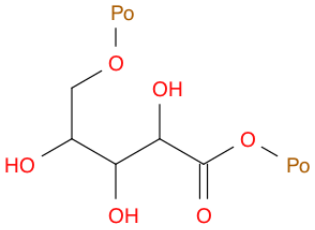
3: glyoxylate



4:



5:



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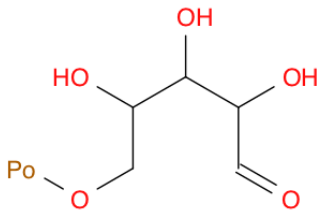
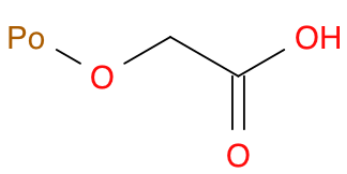


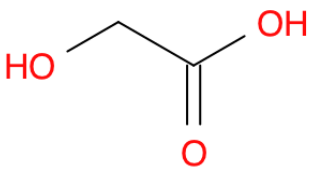
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	DHAP
Co-product	

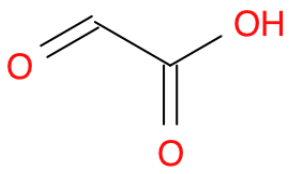
Pathway 111



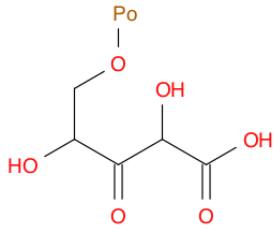
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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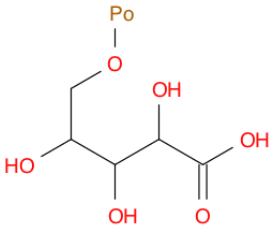

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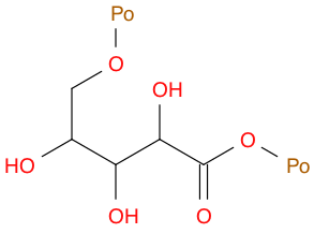

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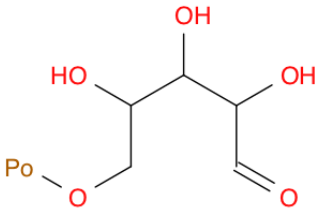
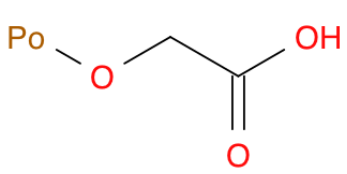


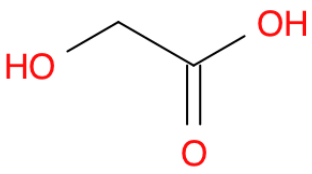
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(=O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

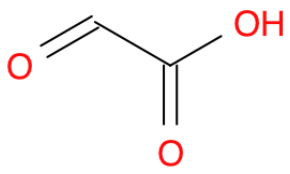
Pathway 112



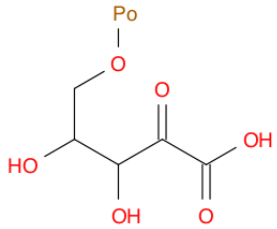
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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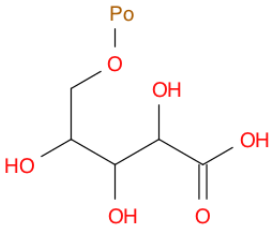

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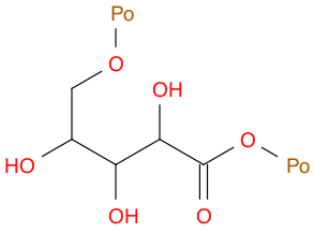

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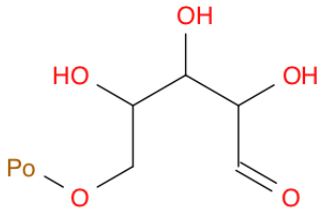
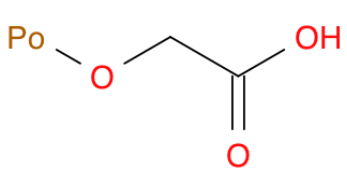


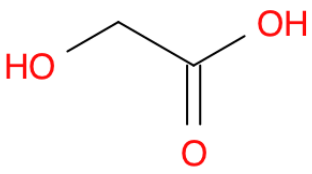
7: C5-aldose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	GAP
Co-product	

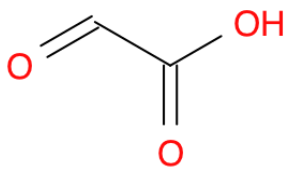
Pathway 113



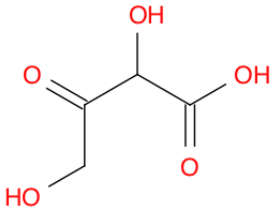
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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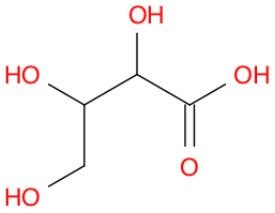

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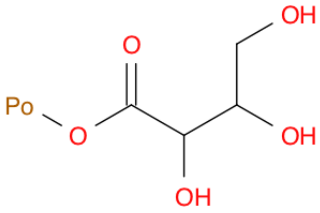

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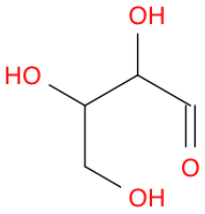
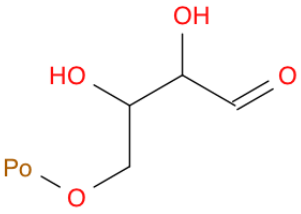


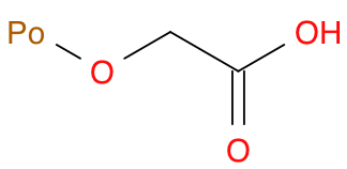
7: C4-aldose



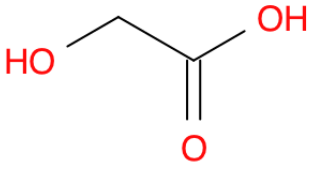
8: C4-aldose 4-phosphate

Number of reactions	7
Pathway superclass	2:4
Key C2 intermediate	glyoxylate
Key C4 intermediate	C(=O)(O)C(O)C(=O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Transketolase (acceptor)
Co-reactant	
Co-product	

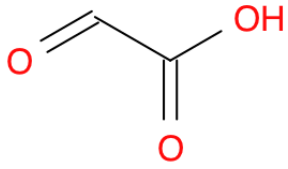
Pathway 114



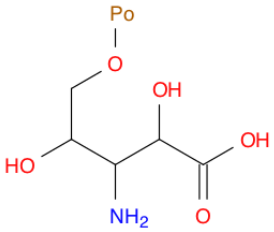
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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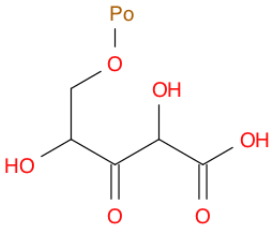

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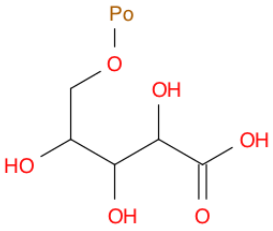

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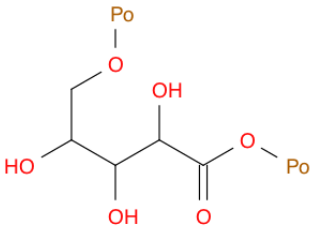

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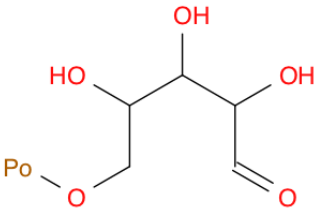
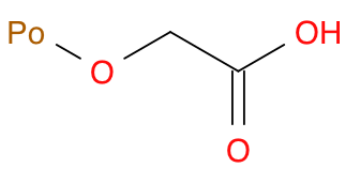


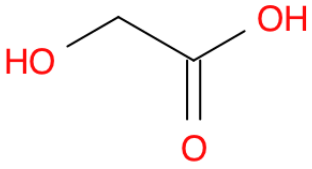
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(N)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

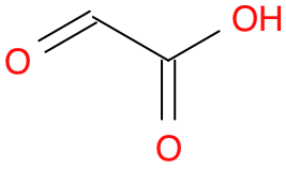
Pathway 115



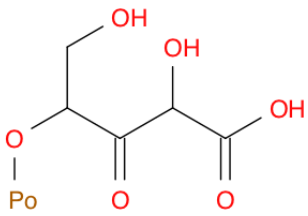
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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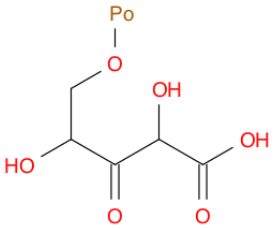

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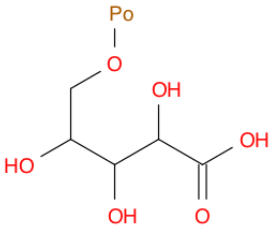

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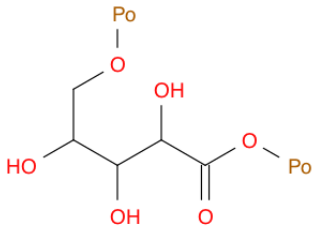

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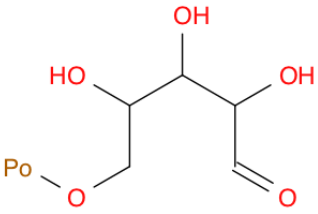
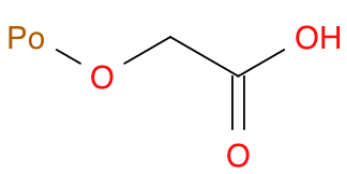


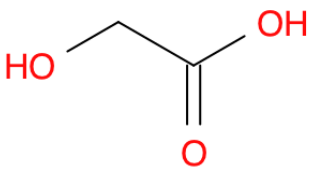
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(=O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

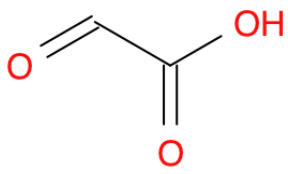
Pathway 116



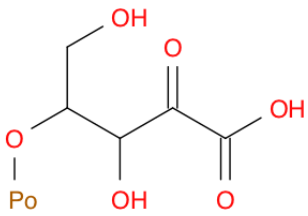
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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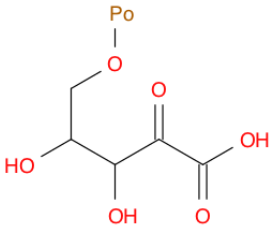

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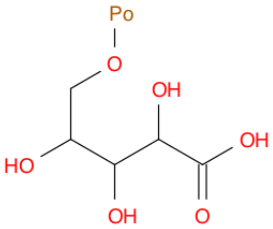

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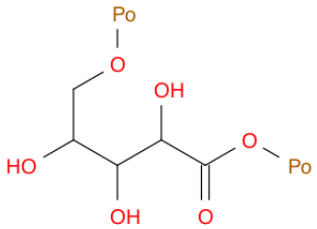

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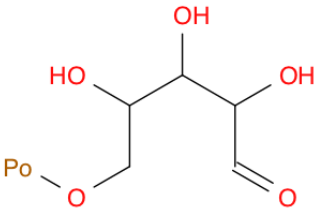
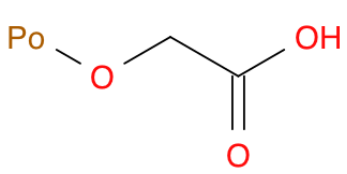


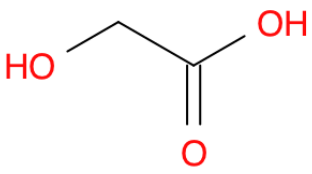
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

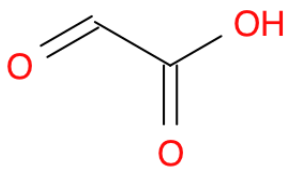
Pathway 117



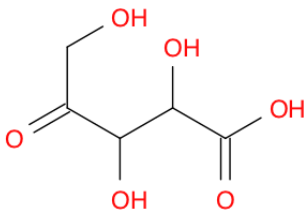
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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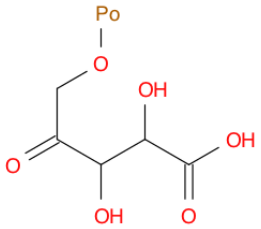

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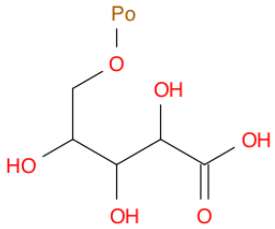

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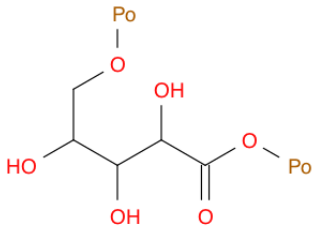

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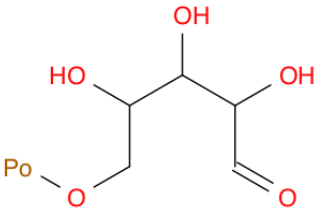
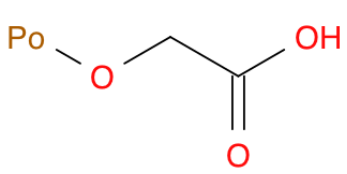


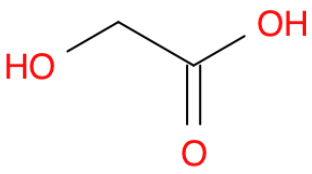
8: C5-aldose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

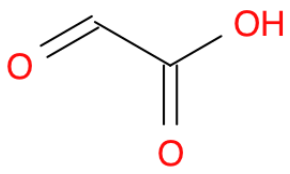
Pathway 118



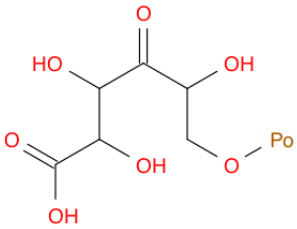
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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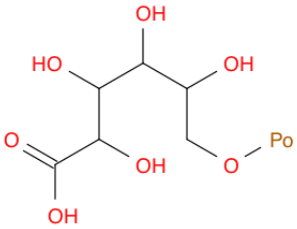

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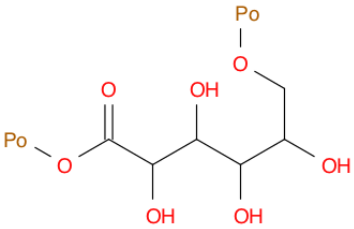

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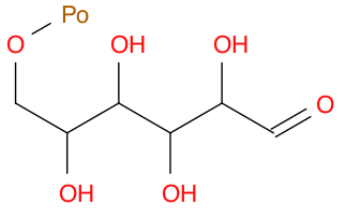
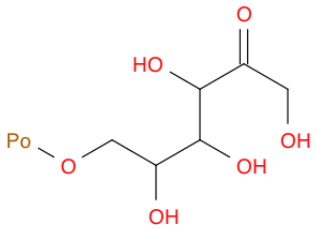


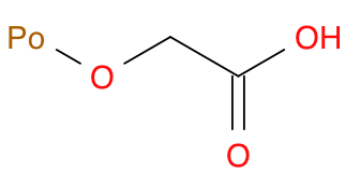
7: C6-aldose 6-phosphate



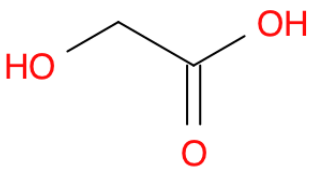
8: C6-ketose 6-phosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C4-ketose 4-phosphate
Co-product	

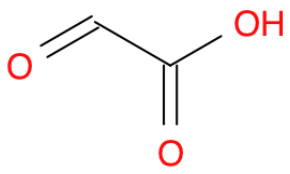
Pathway 119



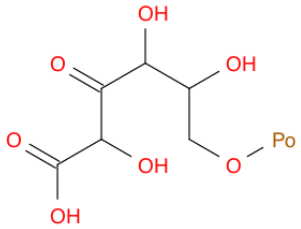
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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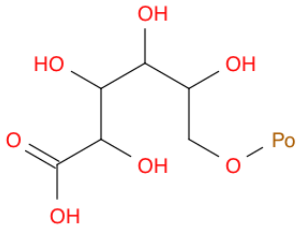

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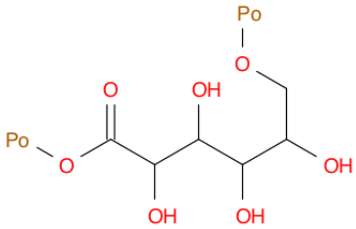

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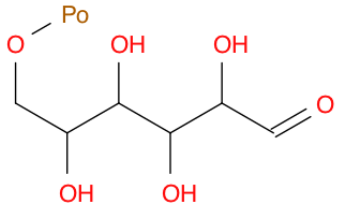
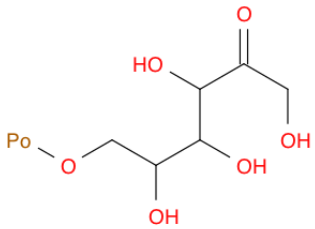


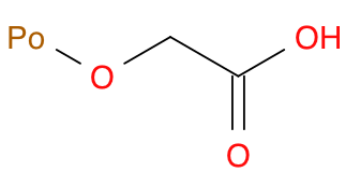
7: C6-aldose 6-phosphate



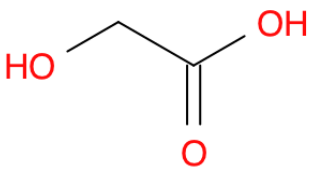
8: C6-ketose 6-phosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

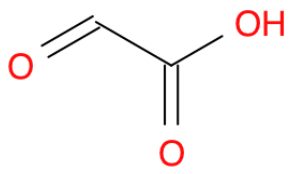
Pathway 120



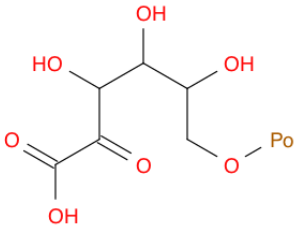
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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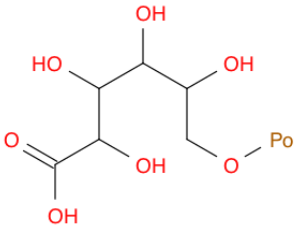

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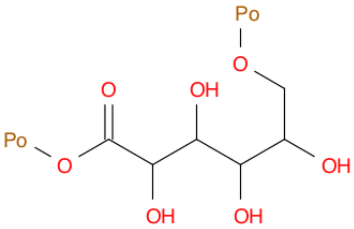

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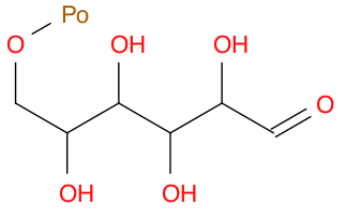
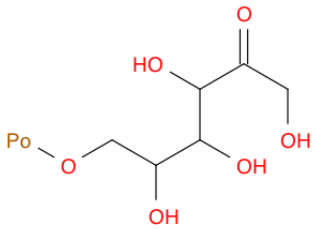


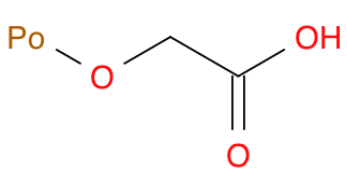
7: C6-aldose 6-phosphate



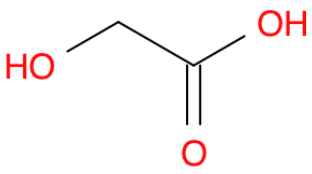
8: C6-ketose 6-phosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

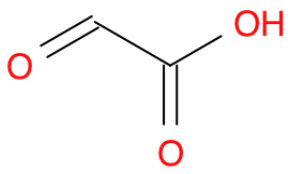
Pathway 121



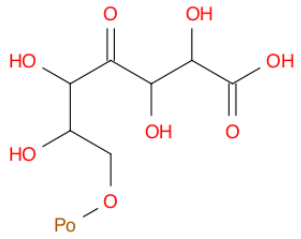
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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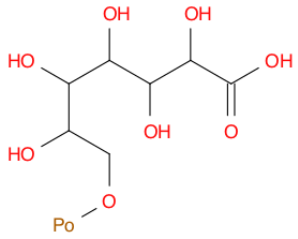

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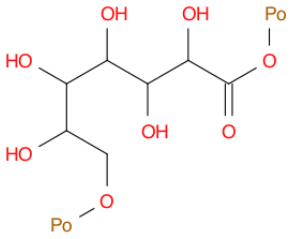

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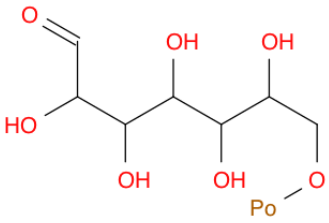
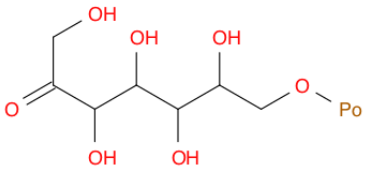


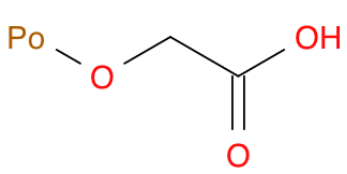
7: C7-aldose 7-phosphate



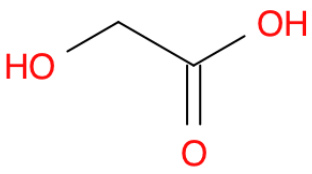
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C5-ketose 5-phosphate
Co-product	

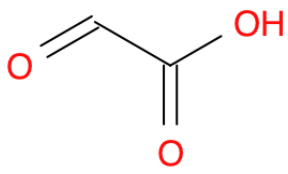
Pathway 122



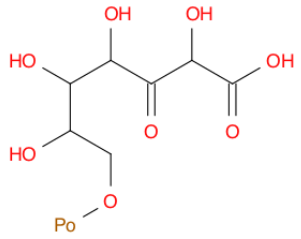
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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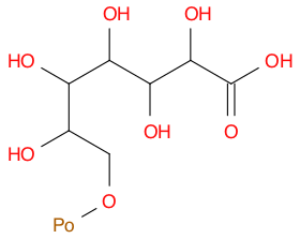

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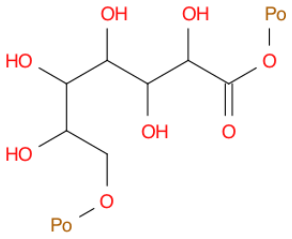

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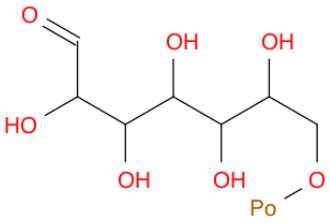
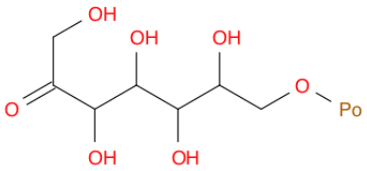


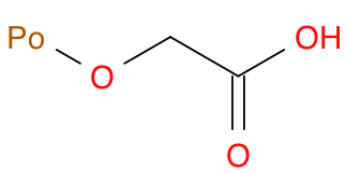
7: C7-aldose 7-phosphate



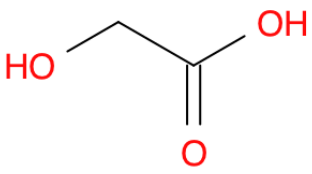
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(O)C(=O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

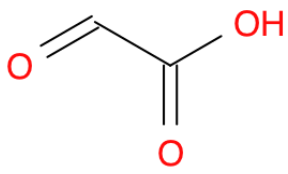
Pathway 123



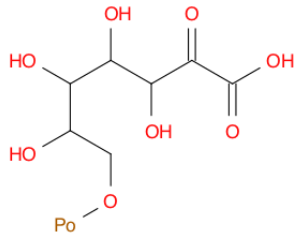
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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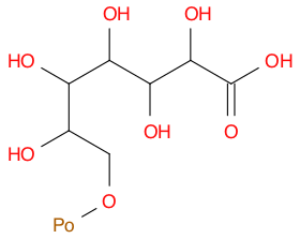

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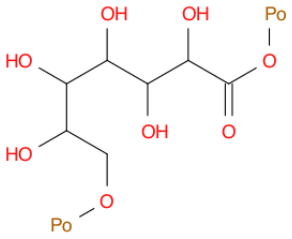

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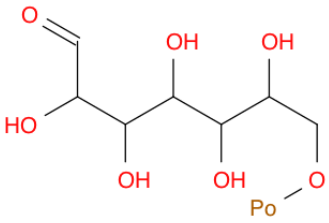
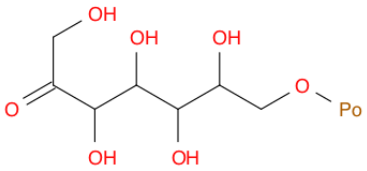


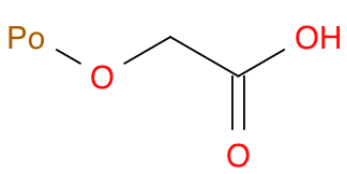
7: C7-aldose 7-phosphate



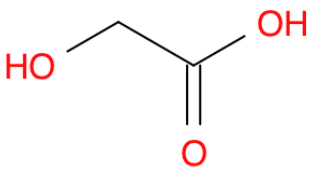
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

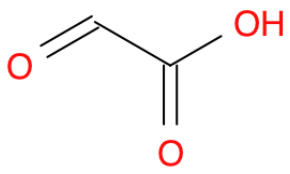
Pathway 124



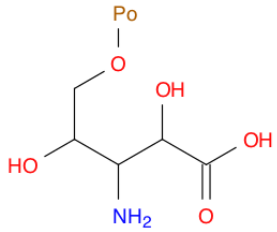
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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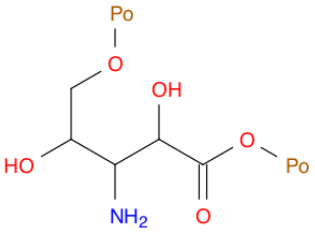

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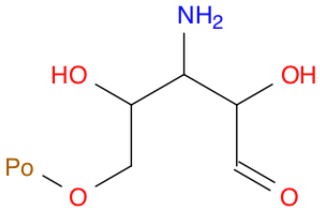

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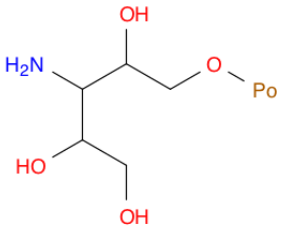

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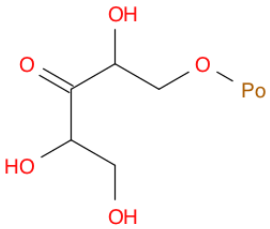

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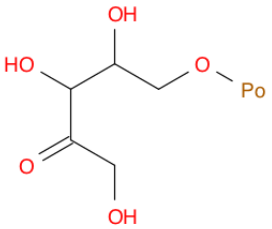
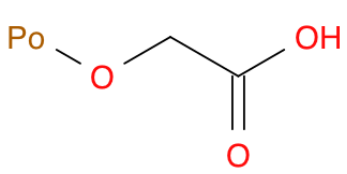


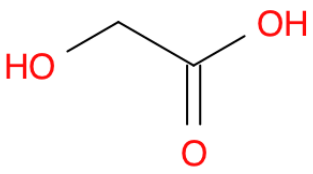
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(N)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O[PO3])
Co-product	

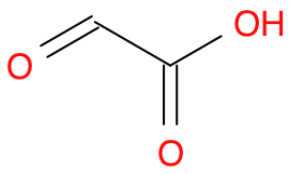
Pathway 125



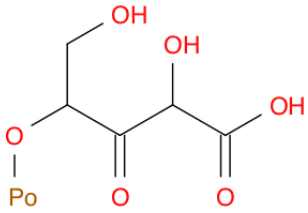
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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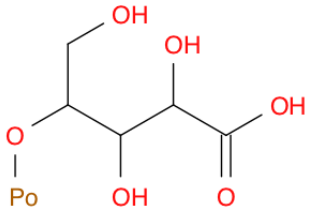

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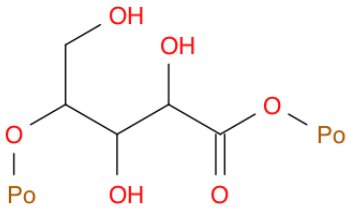

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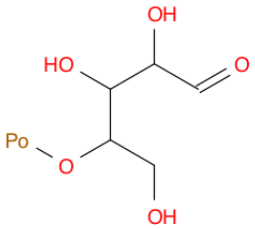

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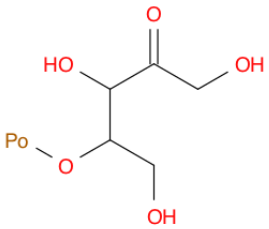

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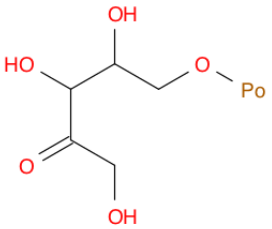
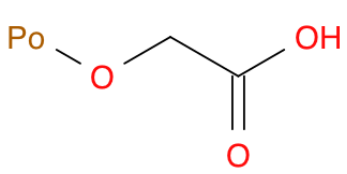


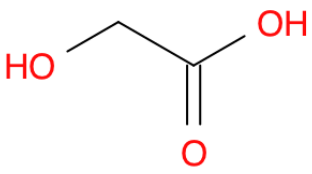
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(=O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

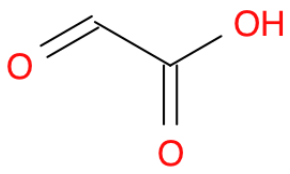
Pathway 126



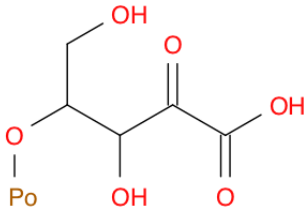
1: glycolate 2-phosphate



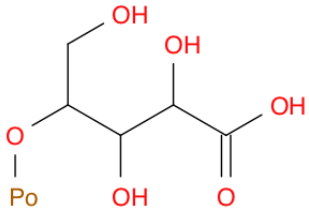
2: glycolate



3: glyoxylate



4:



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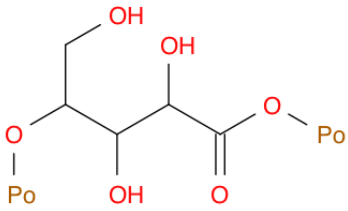

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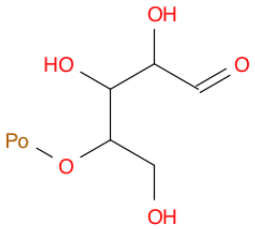

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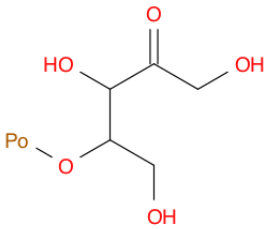

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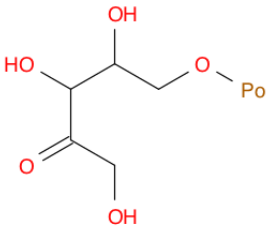
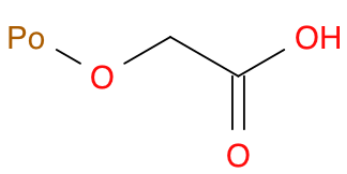


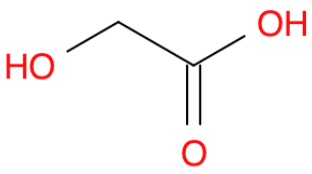
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

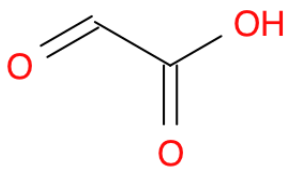
Pathway 127



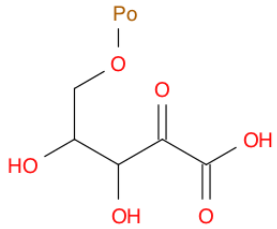
1: glycolate 2-phosphate



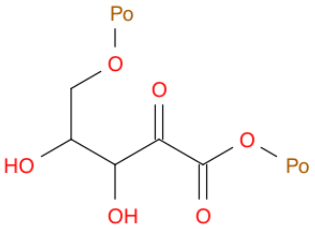
2: glycolate



3: glyoxylate



4:



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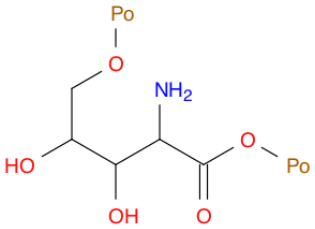

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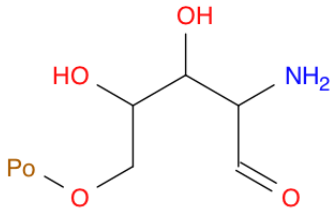

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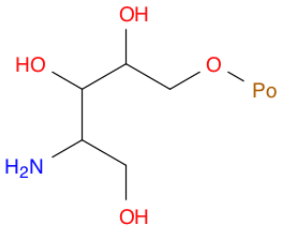

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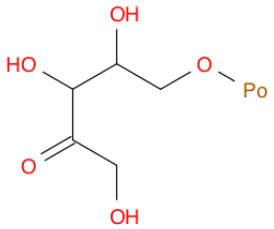
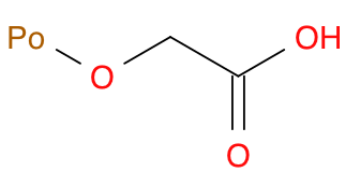


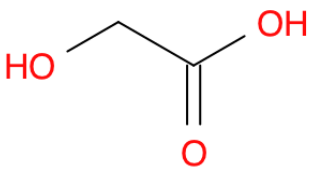
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	GAP
Co-product	

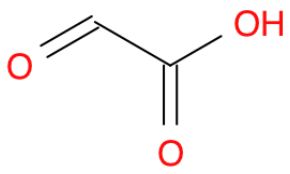
Pathway 128



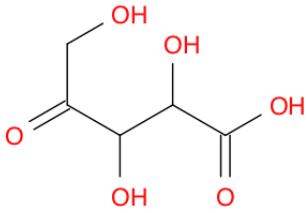
1: glycolate 2-phosphate



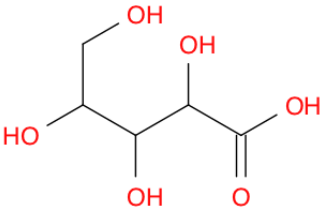
2: glycolate



3: glyoxylate



4:



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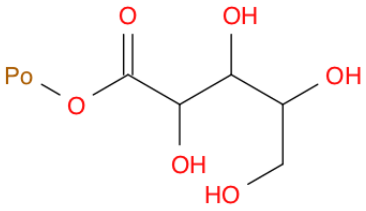

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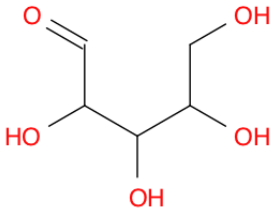
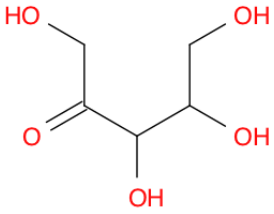
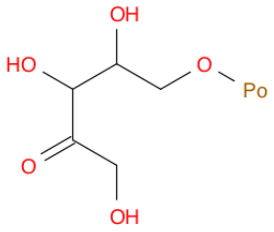


7: C5-aldose



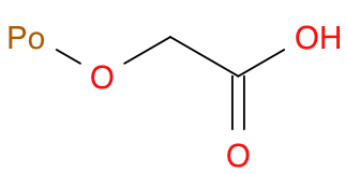
8: C5-ketose



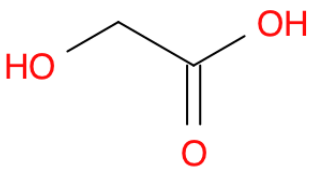
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glyoxylate
Key C5 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Transaldolase (acceptor)
Co-reactant	
Co-product	

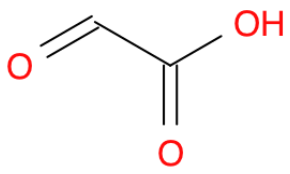
Pathway 129



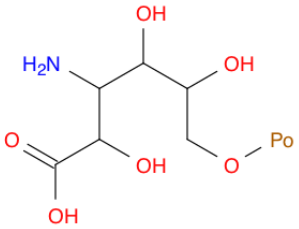
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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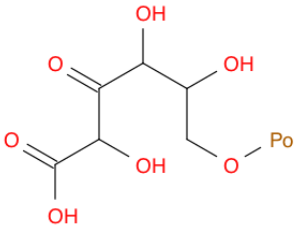

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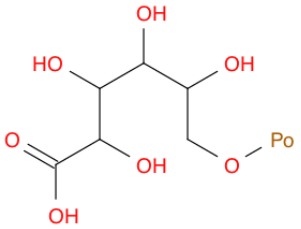

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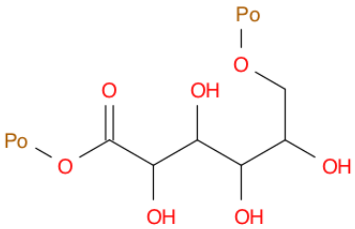

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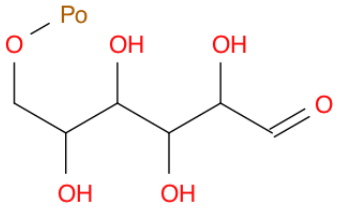
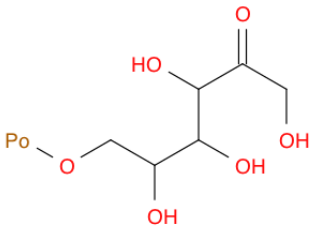


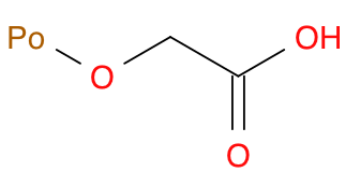
8: C6-aldose 6-phosphate



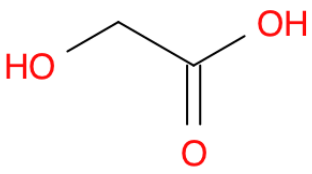
9: C6-ketose 6-phosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(O)C(N)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O[PO3])
Co-product	

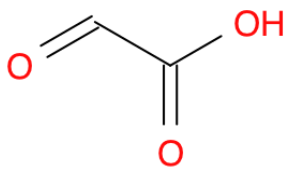
Pathway 130



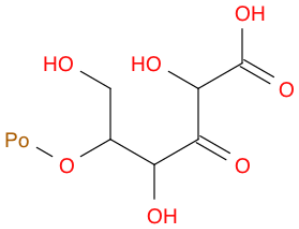
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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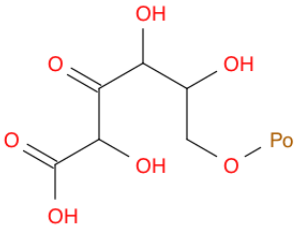

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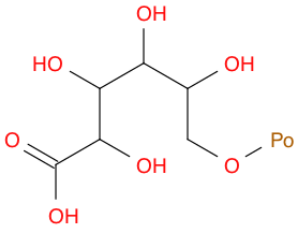

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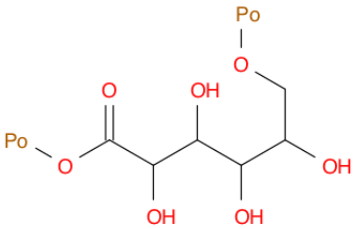

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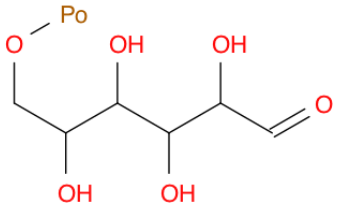
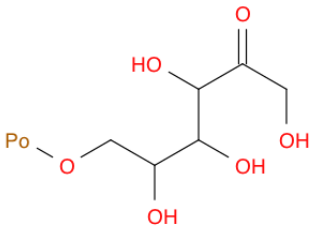


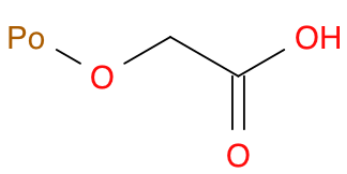
8: C6-aldose 6-phosphate



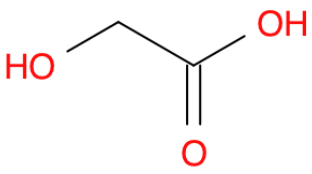
9: C6-ketose 6-phosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

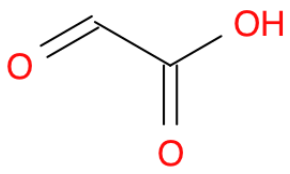
Pathway 131



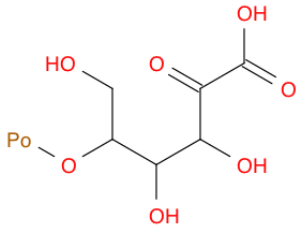
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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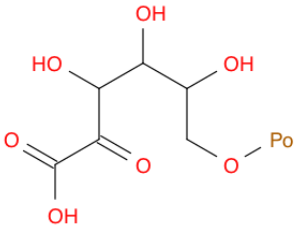

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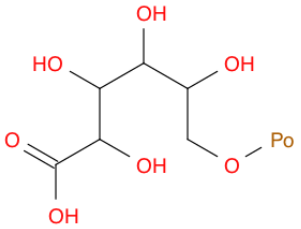

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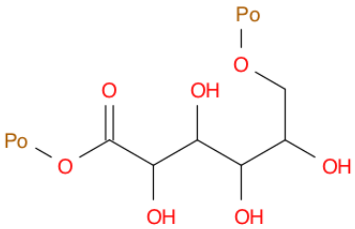

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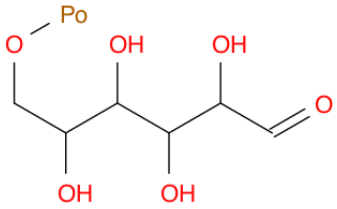
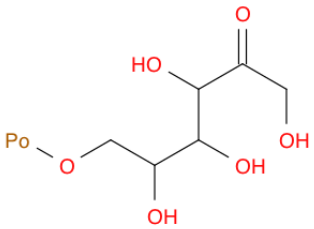


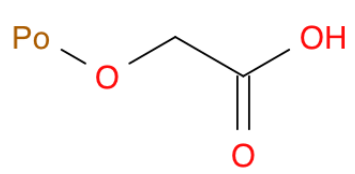
8: C6-aldose 6-phosphate



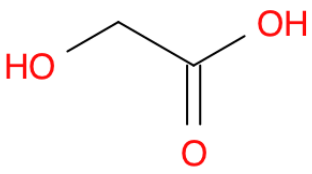
9: C6-ketose 6-phosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glyoxylate
Key C6 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

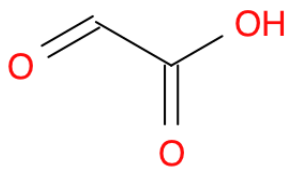
Pathway 132



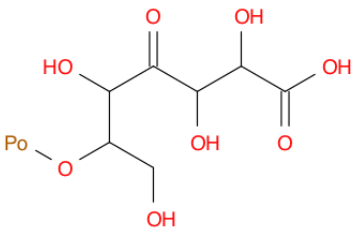
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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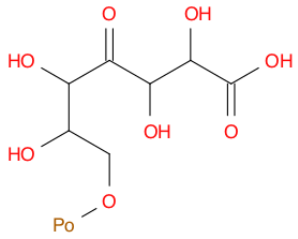

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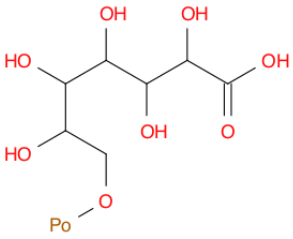

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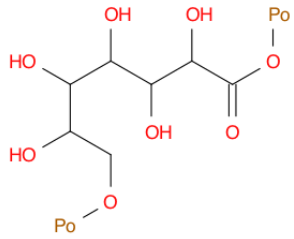

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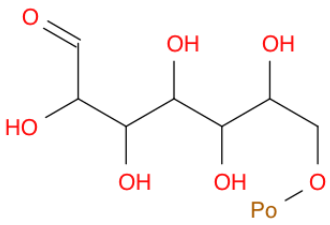
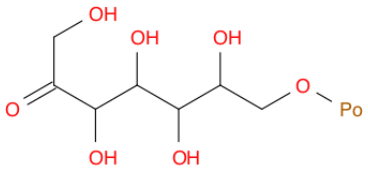


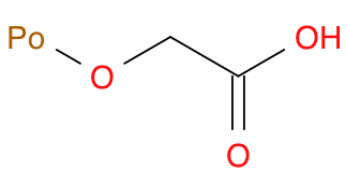
8: C7-aldose 7-phosphate



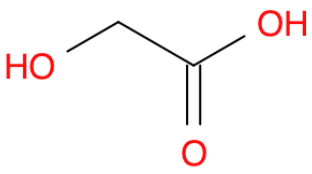
9: C7-ketose 7-phosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(O)C(O)C(=O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	C(O)C(=O)C(O)C(O[PO3])C(O)
Co-product	

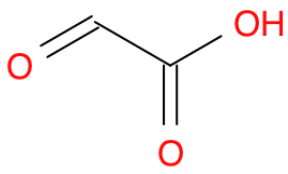
Pathway 133



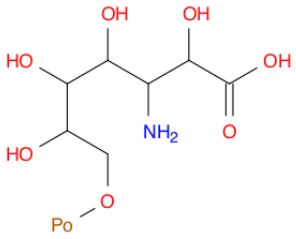
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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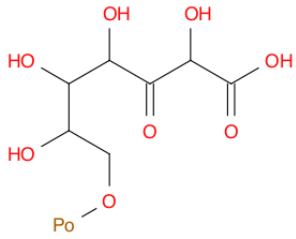

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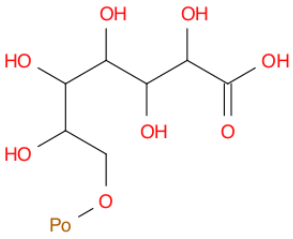

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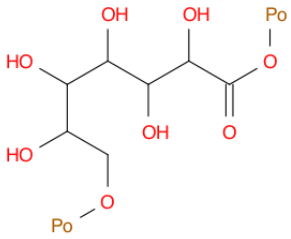

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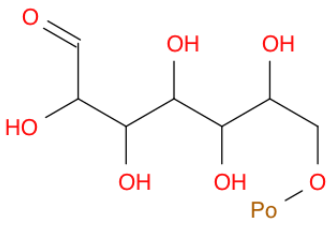
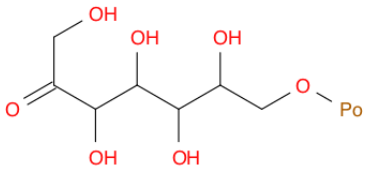


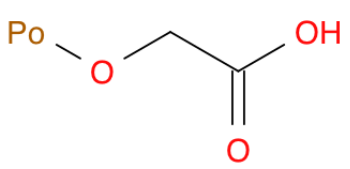
8: C7-aldose 7-phosphate



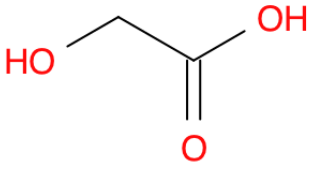
9: C7-ketose 7-phosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(O)C(N)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (acceptor)
Co-reactant	C(N)C(O)C(O)C(O)C(O[PO3])
Co-product	

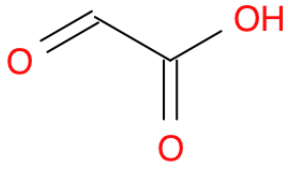
Pathway 134



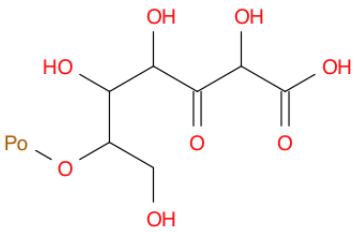
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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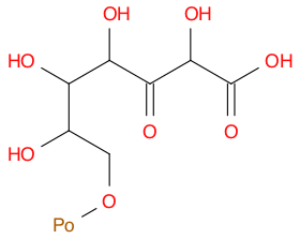

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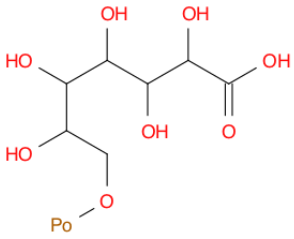

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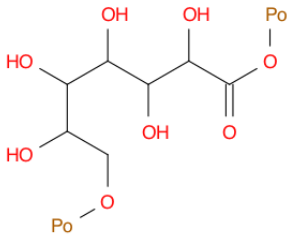

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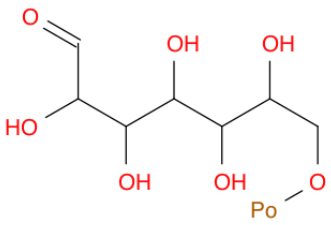
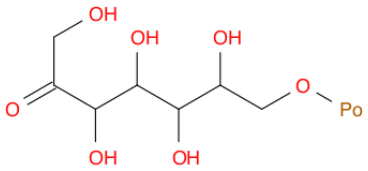


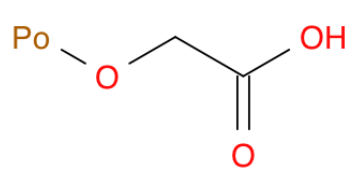
8: C7-aldose 7-phosphate



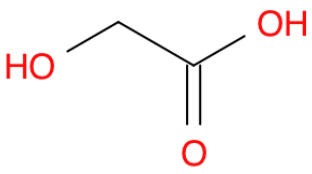
9: C7-ketose 7-phosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(O)C(=O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

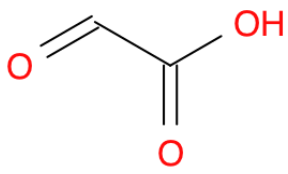
Pathway 135



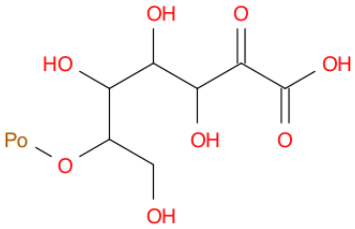
1: glycolate 2-phosphate



2: glycolate



3: glyoxylate



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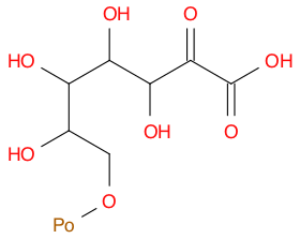

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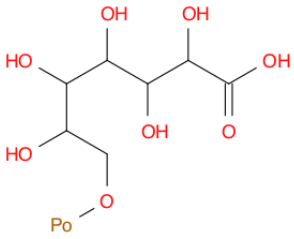

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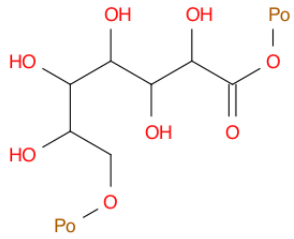

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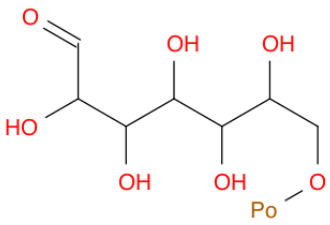
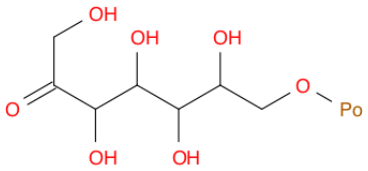


8: C7-aldose 7-phosphate

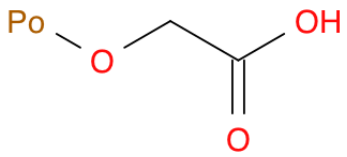


9: C7-ketose 7-phosphate

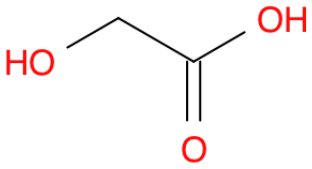
Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glyoxylate
Key C7 intermediate	C(=O)(O)C(=O)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Ketolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

4.5 Glycine pathways

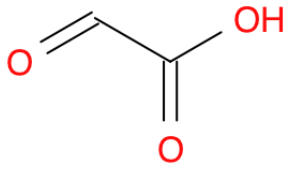
Pathway 136



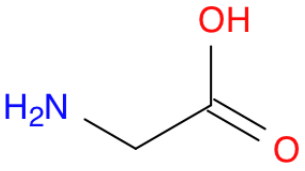
1: glycolate 2-phosphate



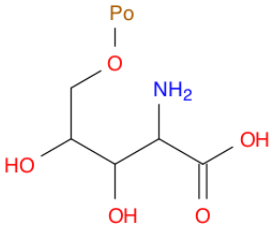
2: glycolate



3: glyoxylate



4: glycine



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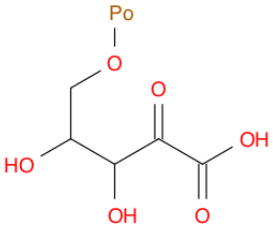

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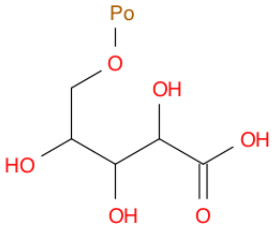
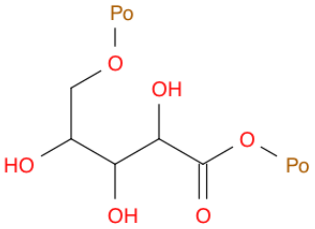
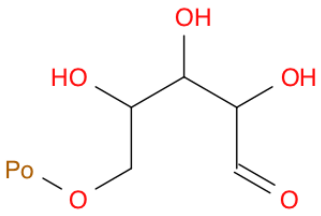


7:



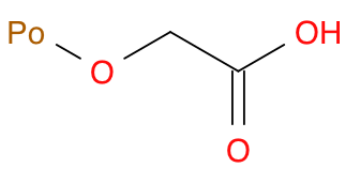
8:



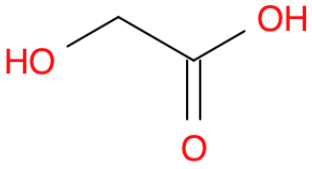
9: C5-aldose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glycine
Key C5 intermediate	C(=O)(O)C(N)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	GAP
Co-product	

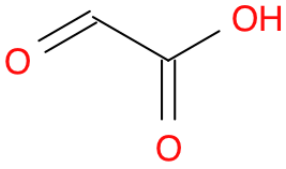
Pathway 137



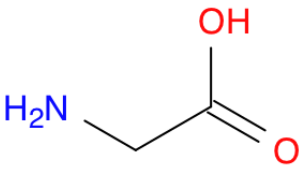
1: glycolate 2-phosphate



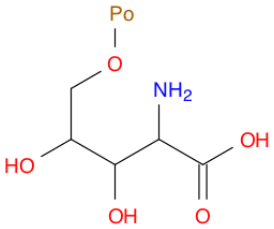
2: glycolate



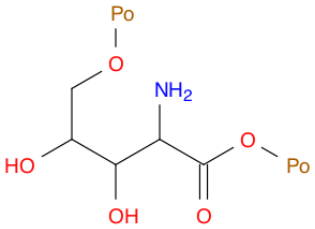
3: glyoxylate



4: glycine



5:



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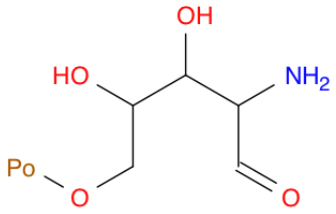

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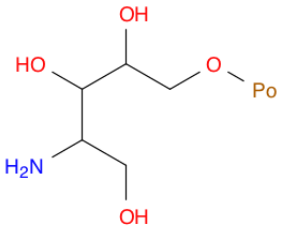

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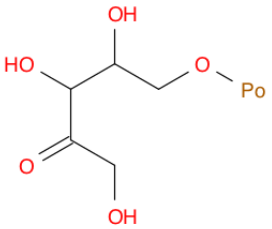
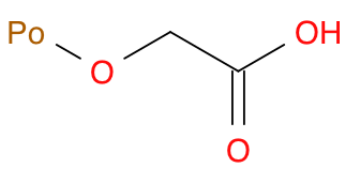


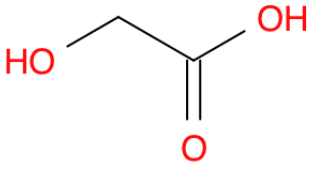
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	glycine
Key C5 intermediate	C(=O)(O)C(N)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	GAP
Co-product	

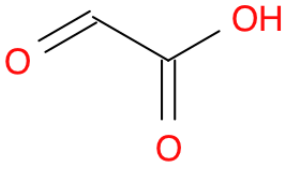
Pathway 138



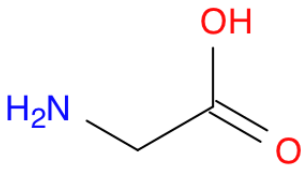
1: glycolate 2-phosphate



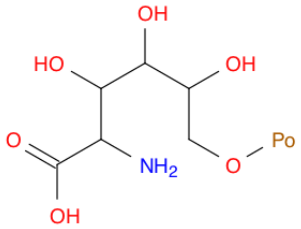
2: glycolate



3: glyoxylate



4: glycine



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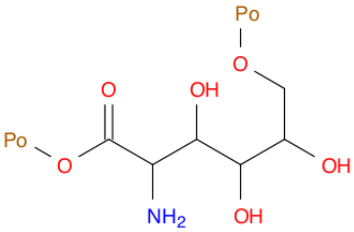

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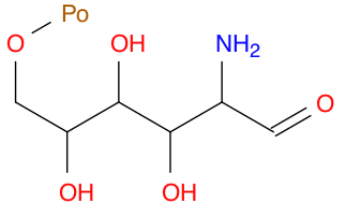

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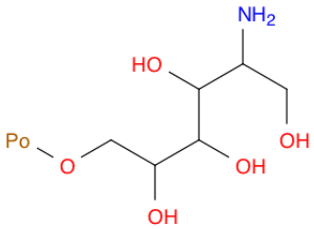

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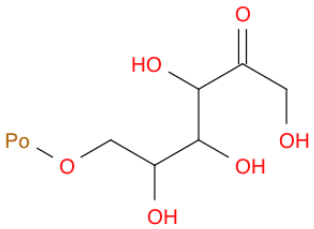
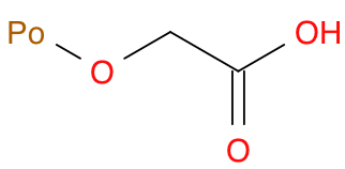


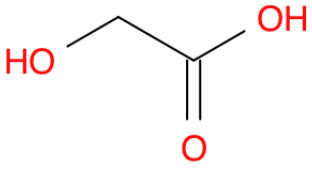
9: C6-ketose 6-phosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	glycine
Key C6 intermediate	C(=O)(O)C(N)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

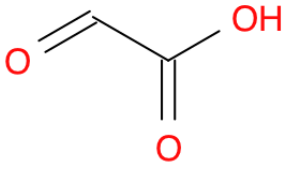
Pathway 139



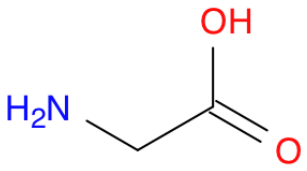
1: glycolate 2-phosphate



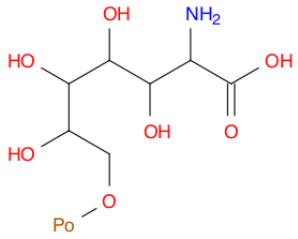
2: glycolate



3: glyoxylate



4: glycine



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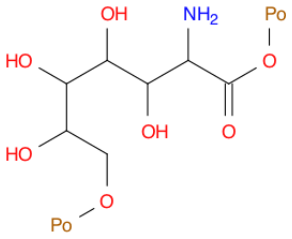

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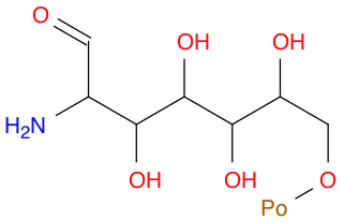

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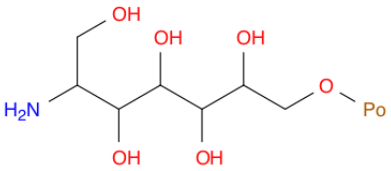
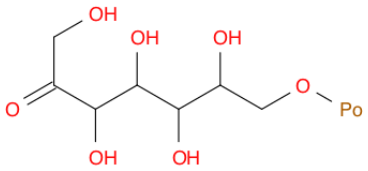


8:

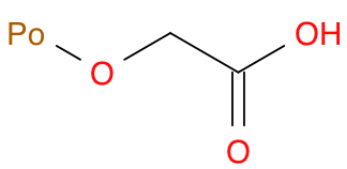


9: C7-ketose 7-phosphate

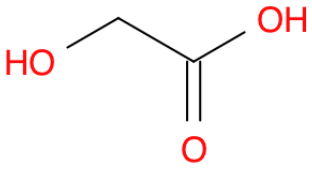
Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	glycine
Key C7 intermediate	C(=O)(O)C(N)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

4.6 Aminoethanol pathways

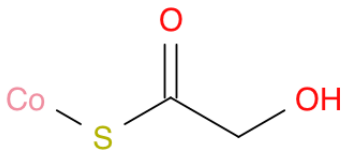
Pathway 140



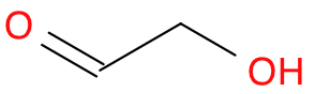
1: glycolate 2-phosphate



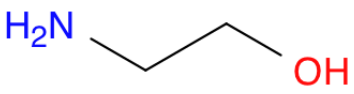
2: glycolate



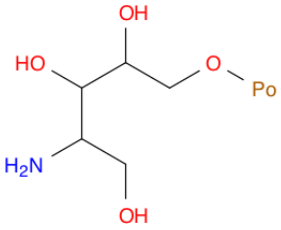
3: glycolyl-CoA



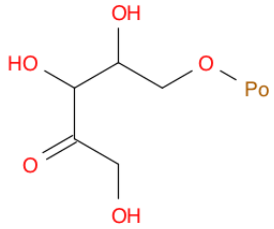
4: glycolaldehyde



5: aminoethanol



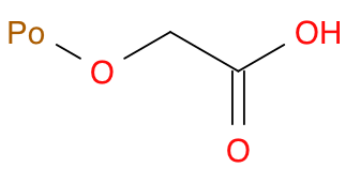
6:



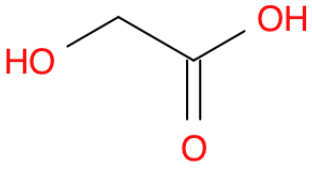
7: C5-ketose 5-phosphate

Number of reactions	6
Pathway superclass	2:5
Key C2 intermediate	aminoethanol
Key C5 intermediate	C(O[PO3])C(O)C(O)C(N)C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	GAP
Co-product	

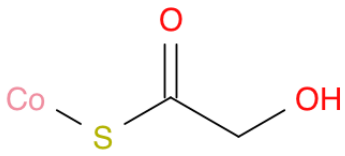
Pathway 141



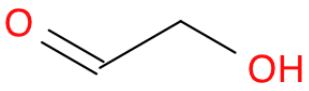
1: glycolate 2-phosphate



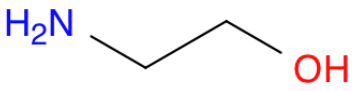
2: glycolate



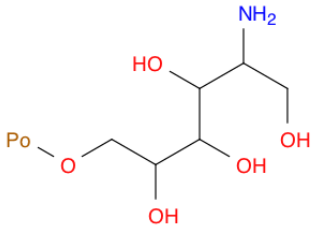
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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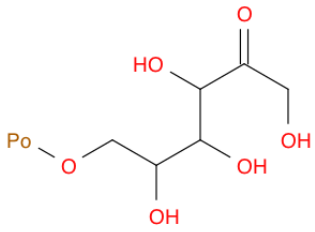
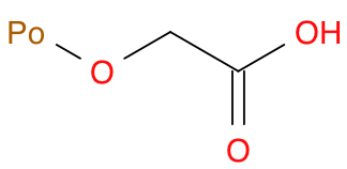


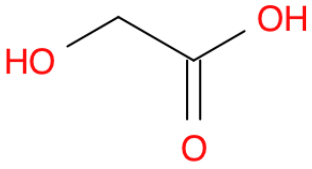
7: C6-ketose 6-phosphate

Number of reactions	6
Pathway superclass	2:6
Key C2 intermediate	aminoethanol
Key C6 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

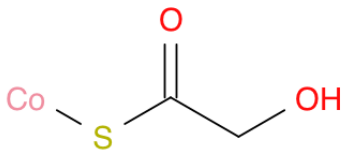
Pathway 142



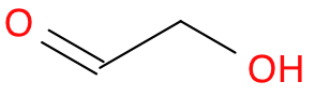
1: glycolate 2-phosphate



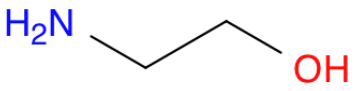
2: glycolate



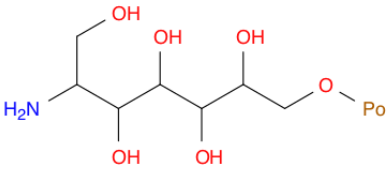
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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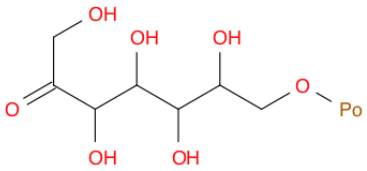
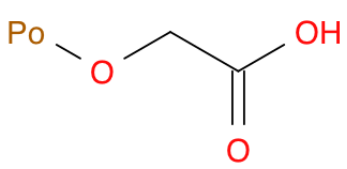


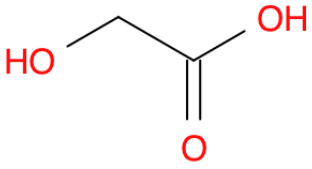
7: C7-ketose 7-phosphate

Number of reactions	6
Pathway superclass	2:7
Key C2 intermediate	aminoethanol
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(O)C(N)C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

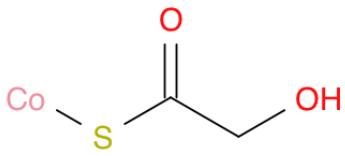
Pathway 143



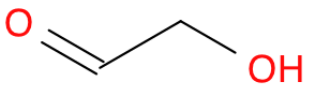
1: glycolate 2-phosphate



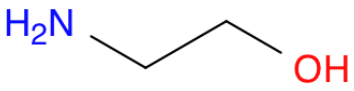
2: glycolate



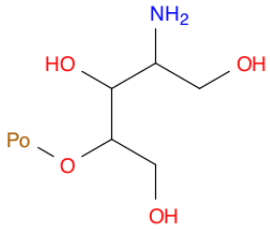
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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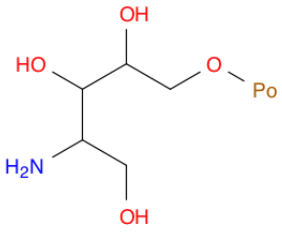

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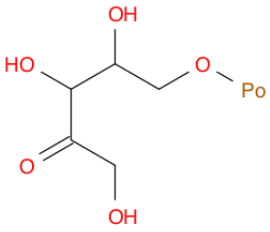
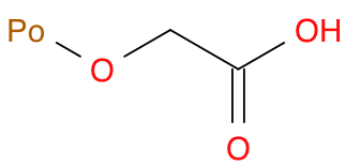


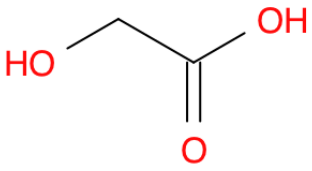
8: C5-ketose 5-phosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	aminoethanol
Key C5 intermediate	C(O)C(N)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

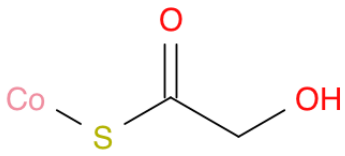
Pathway 144



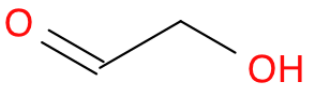
1: glycolate 2-phosphate



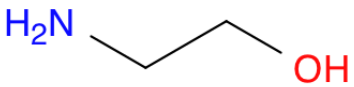
2: glycolate



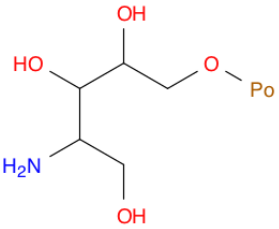
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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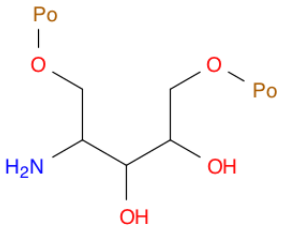

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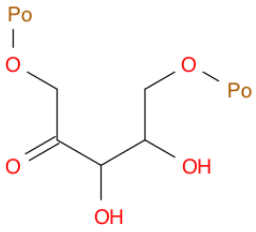
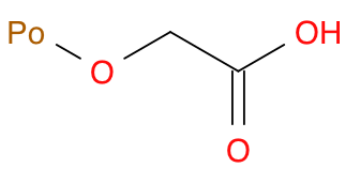


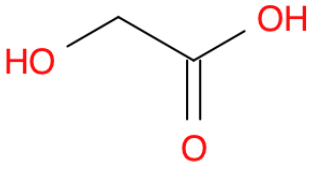
8: C5-ketose 1,5-bisphosphate

Number of reactions	7
Pathway superclass	2:5
Key C2 intermediate	aminoethanol
Key C5 intermediate	C(O[PO3])C(O)C(O)C(N)C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	GAP
Co-product	

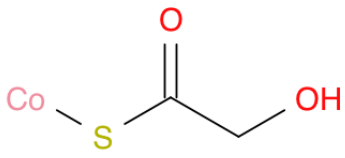
Pathway 145



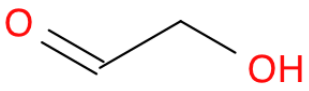
1: glycolate 2-phosphate



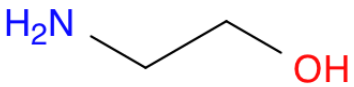
2: glycolate



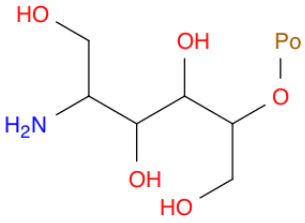
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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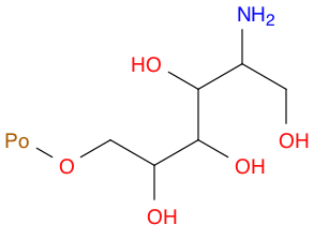

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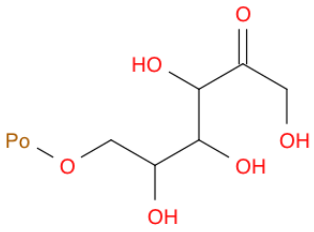
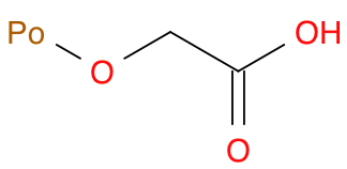


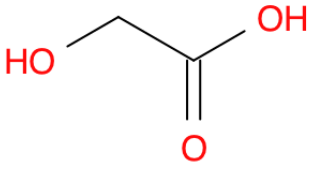
8: C6-ketose 6-phosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	aminoethanol
Key C6 intermediate	C(O)C(N)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

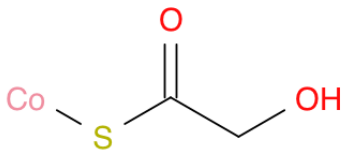
Pathway 146



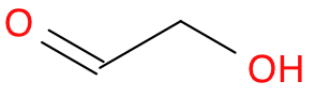
1: glycolate 2-phosphate



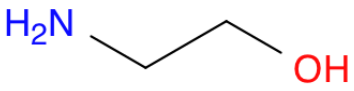
2: glycolate



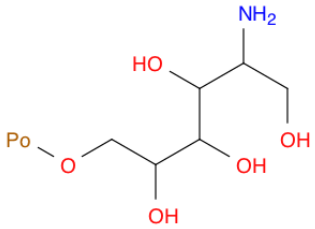
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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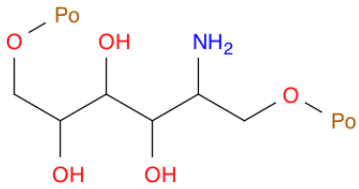

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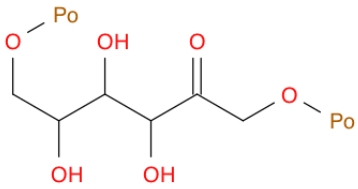
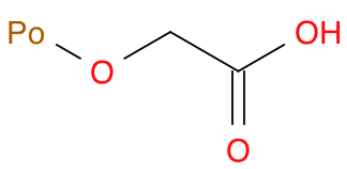


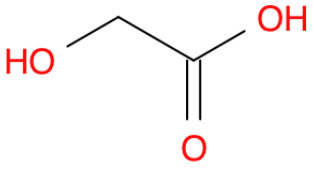
8: C6-ketose 1,6-bisphosphate

Number of reactions	7
Pathway superclass	2:6
Key C2 intermediate	aminoethanol
Key C6 intermediate	C(O[PO3])C(O)C(O)C(O)C(N)C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

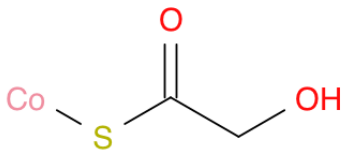
Pathway 147



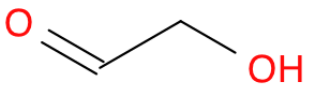
1: glycolate 2-phosphate



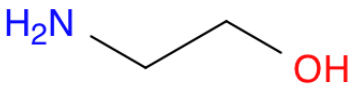
2: glycolate



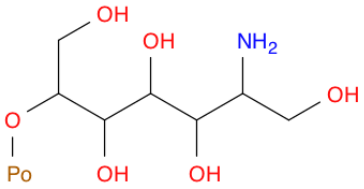
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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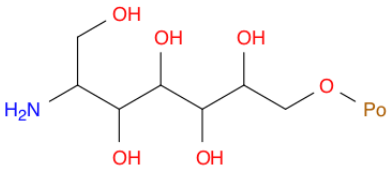
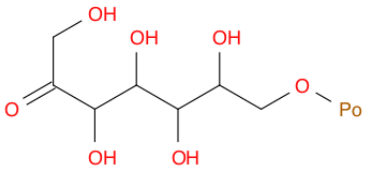


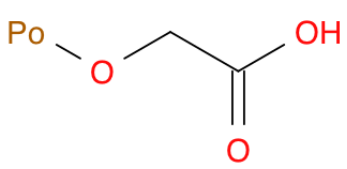
7:



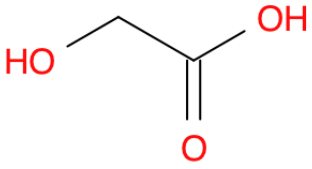
8: C7-ketose 7-phosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	aminoethanol
Key C7 intermediate	C(O)C(N)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

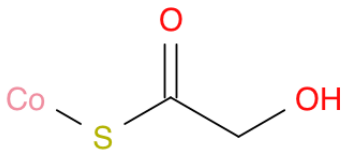
Pathway 148



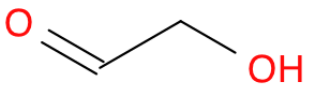
1: glycolate 2-phosphate



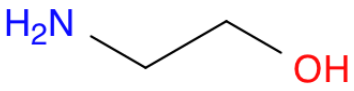
2: glycolate



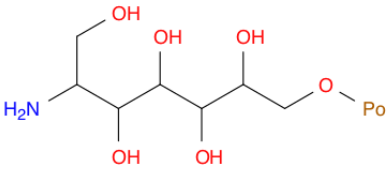
3: glycolyl-CoA



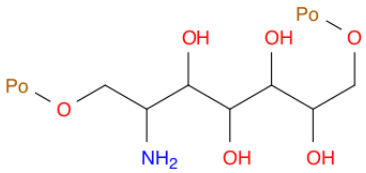
4: glycolaldehyde



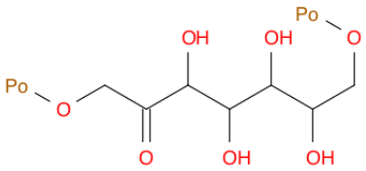
5: aminoethanol



6:



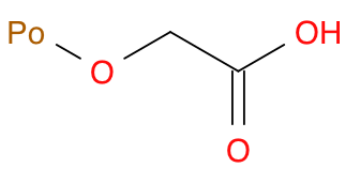
7:



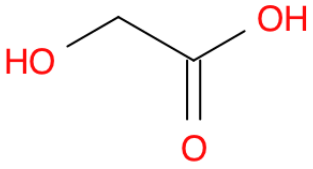
8: C7-ketose 1,7-bisphosphate

Number of reactions	7
Pathway superclass	2:7
Key C2 intermediate	aminoethanol
Key C7 intermediate	C(O[PO3])C(O)C(O)C(O)C(O)C(N)C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

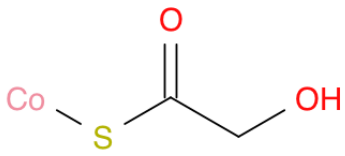
Pathway 149



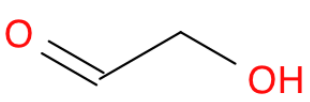
1: glycolate 2-phosphate



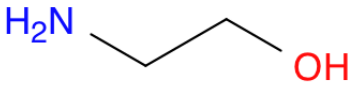
2: glycolate



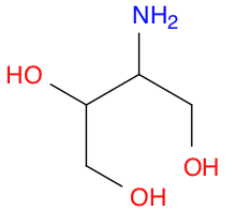
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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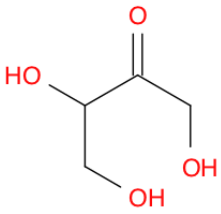
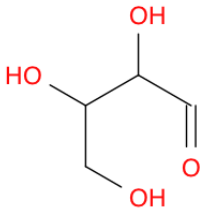
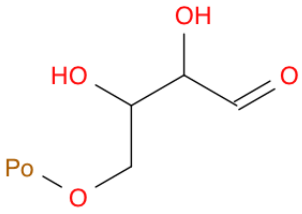


7: C4-ketose



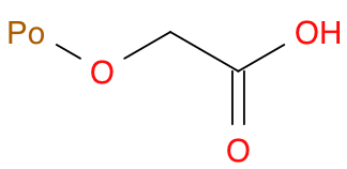
8: C4-aldose



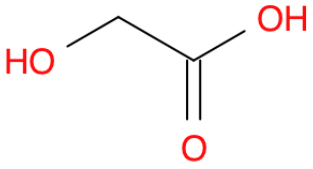
9: C4-aldose 4-phosphate

Number of reactions	8
Pathway superclass	2:4
Key C2 intermediate	aminoethanol
Key C4 intermediate	C(O)C(N)C(O)C(O)
Calvin cycle sink	C4-aldose 4-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	glycolaldehyde
Co-product	

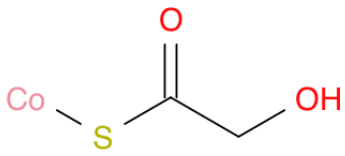
Pathway 150



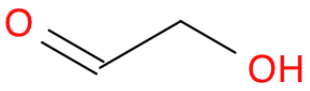
1: glycolate 2-phosphate



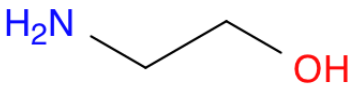
2: glycolate



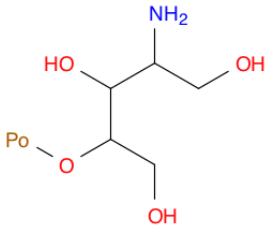
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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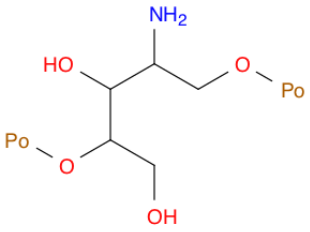

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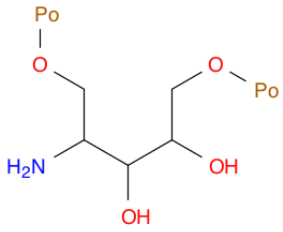

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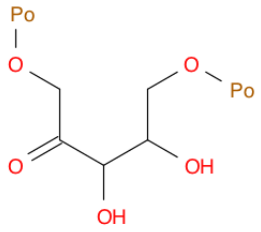
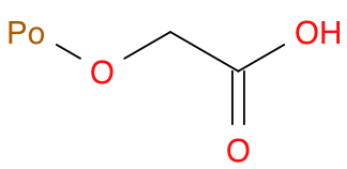


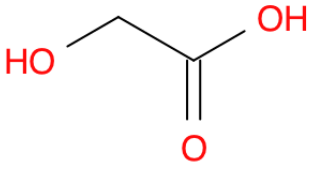
9: C5-ketose 1,5-bisphosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoethanol
Key C5 intermediate	C(O)C(N)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-ketose 1,5-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

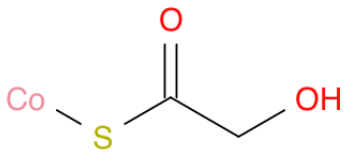
Pathway 151



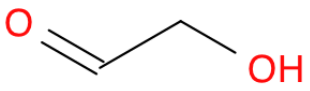
1: glycolate 2-phosphate



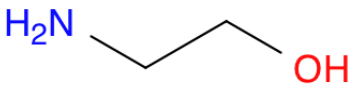
2: glycolate



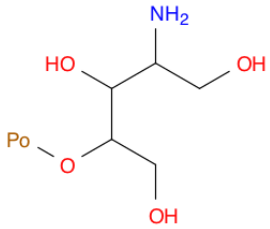
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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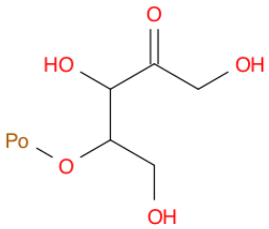

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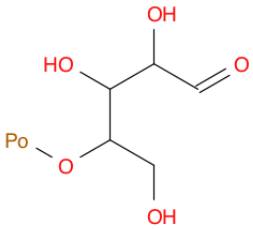

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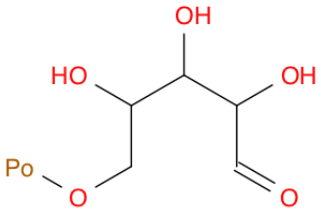
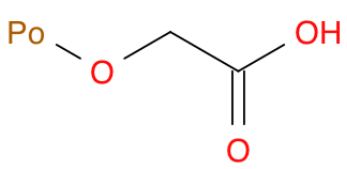


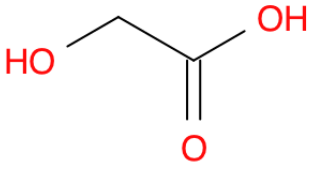
9: C5-aldose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoethanol
Key C5 intermediate	C(O)C(N)C(O)C(O[PO3])C(O)
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O[PO3])C(O)
Co-product	

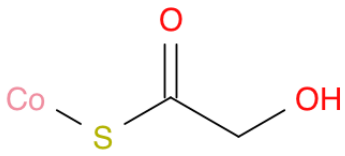
Pathway 152



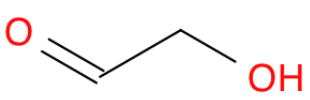
1: glycolate 2-phosphate



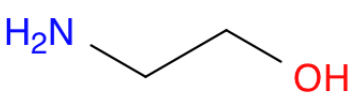
2: glycolate



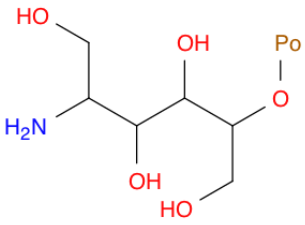
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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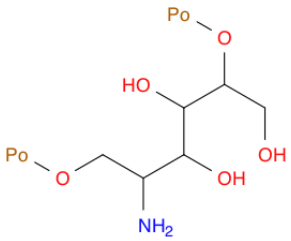
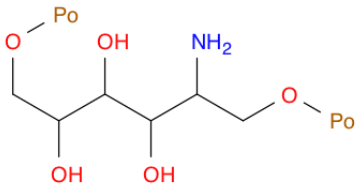


7:



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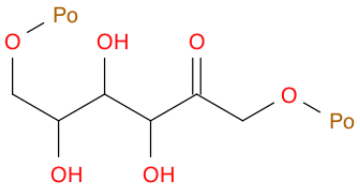
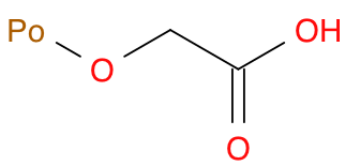


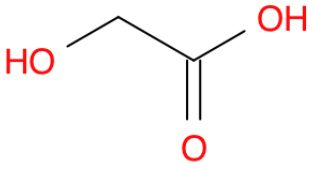
9: C6-ketose 1,6-bisphosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	aminoethanol
Key C6 intermediate	C(O)C(N)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C6-ketose 1,6-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O)C(O[PO3])C(O)
Co-product	

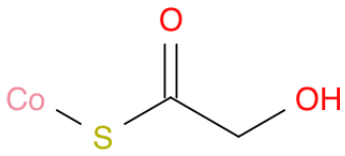
Pathway 153



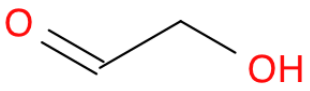
1: glycolate 2-phosphate



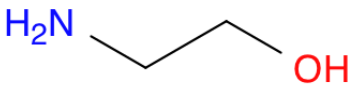
2: glycolate



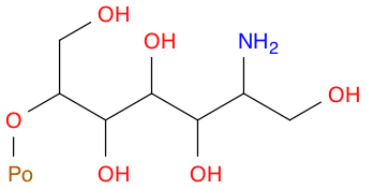
3: glycolyl-CoA



4: glycolaldehyde



5: aminoethanol



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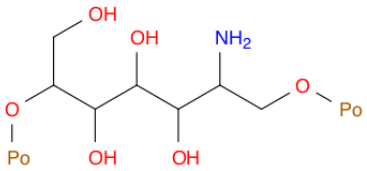

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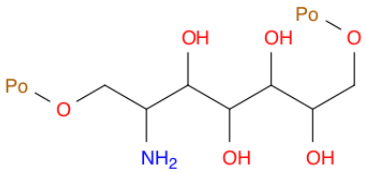

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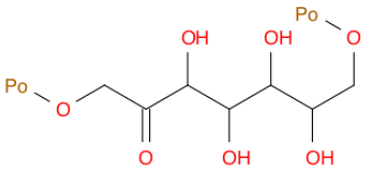
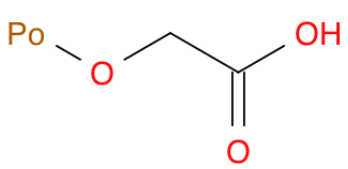


9: C7-ketose 1,7-bisphosphate

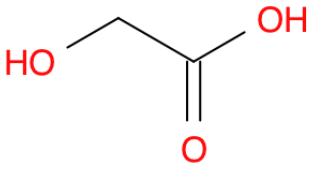
Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	aminoethanol
Key C7 intermediate	C(O)C(N)C(O)C(O)C(O)C(O[PO3])C(O)
Calvin cycle sink	C7-ketose 1,7-bisphosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C(=O)C(O)C(O)C(O[PO3])C(O)
Co-product	

4.7 Aminoacetaldehyde pathways

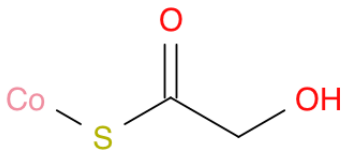
Pathway 154



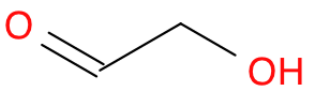
1: glycolate 2-phosphate



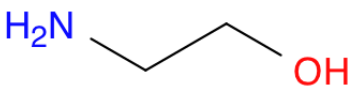
2: glycolate



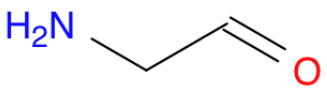
3: glycolyl-CoA



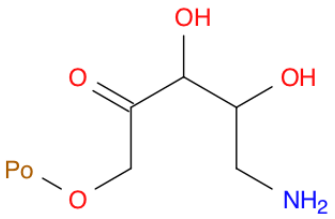
4: glycolaldehyde



5: aminoethanol



6: aminoacetaldehyde



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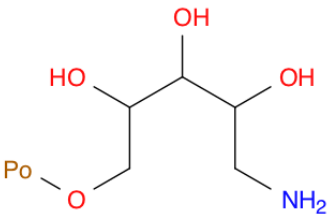

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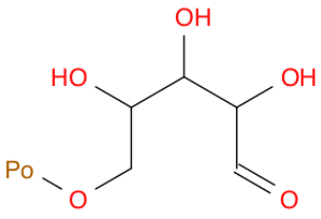
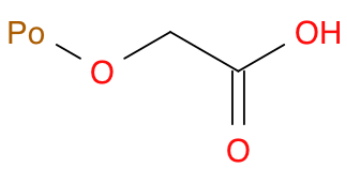


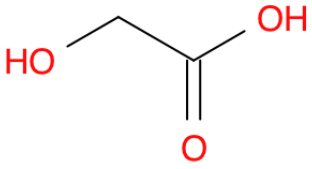
9: C5-aldose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoacetaldehyde
Key C5 intermediate	C(N)C(O)C(O)C(=O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Carbonyl aldolase (acceptor)
Co-reactant	DHAP
Co-product	

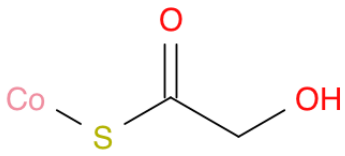
Pathway 155



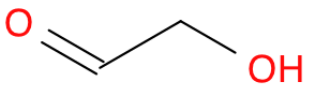
1: glycolate 2-phosphate



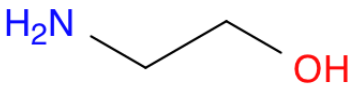
2: glycolate



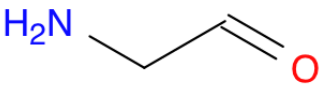
3: glycolyl-CoA



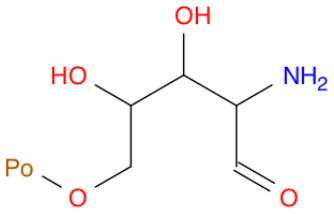
4: glycolaldehyde



5: aminoethanol



6: aminoacetaldehyde



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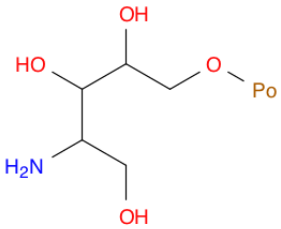

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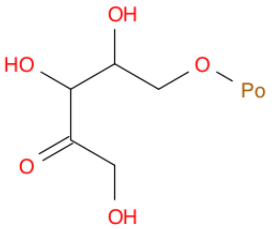
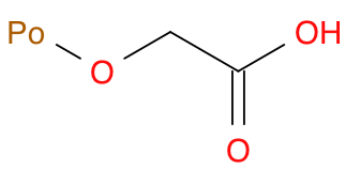


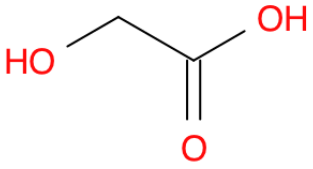
9: C5-ketose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoacetaldehyde
Key C5 intermediate	C(=O)C(N)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-ketose 5-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	GAP
Co-product	

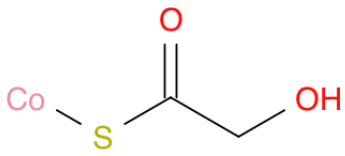
Pathway 156



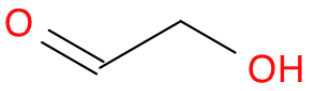
1: glycolate 2-phosphate



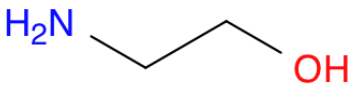
2: glycolate



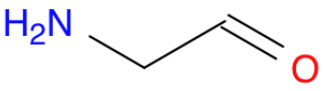
3: glycolyl-CoA



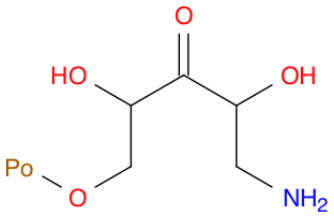
4: glycolaldehyde



5: aminoethanol



6: aminoacetaldehyde



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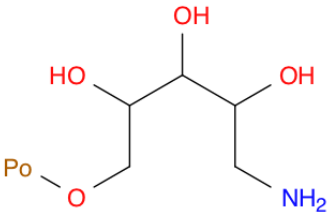

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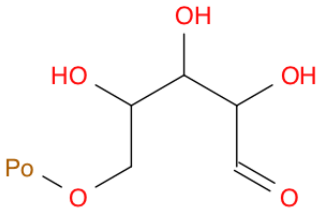
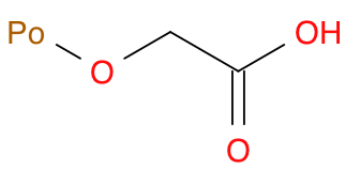


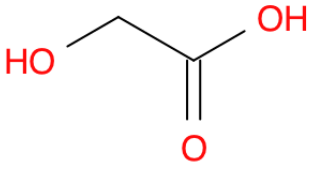
9: C5-aldose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoacetaldehyde
Key C5 intermediate	C(N)C(O)C(=O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (acceptor)
Co-reactant	GAP
Co-product	

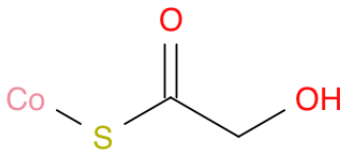
Pathway 157



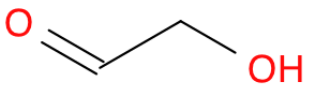
1: glycolate 2-phosphate



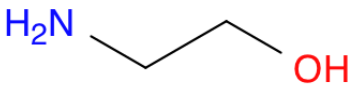
2: glycolate



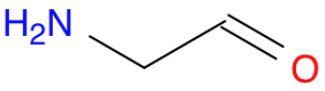
3: glycolyl-CoA



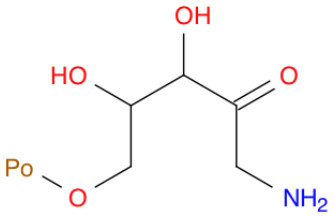
4: glycolaldehyde



5: aminoethanol



6: aminoacetaldehyde



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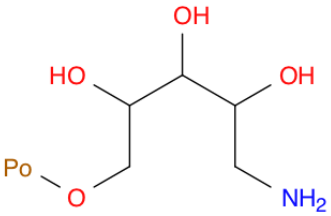

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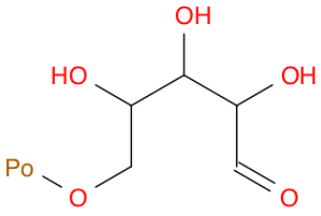
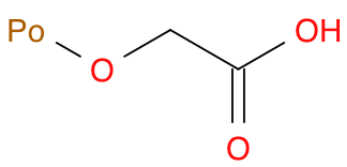


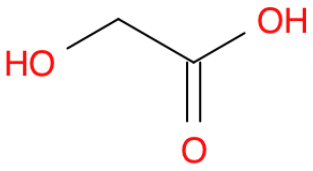
9: C5-aldose 5-phosphate

Number of reactions	8
Pathway superclass	2:5
Key C2 intermediate	aminoacetaldehyde
Key C5 intermediate	C(N)C(=O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C5-aldose 5-phosphate
Key reaction	Ketolase (donor)
Co-reactant	GAP
Co-product	

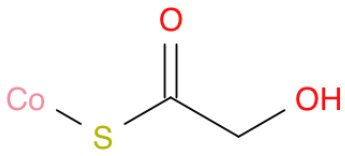
Pathway 158



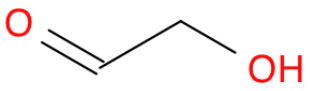
1: glycolate 2-phosphate



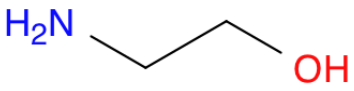
2: glycolate



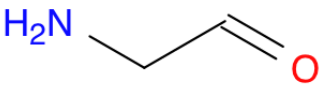
3: glycolyl-CoA



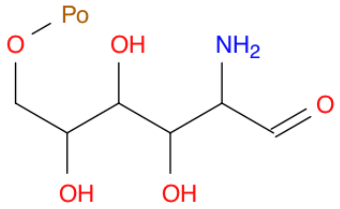
4: glycolaldehyde



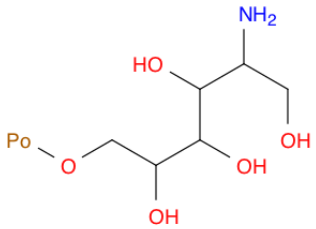
5: aminoethanol



6: aminoacetaldehyde



7:



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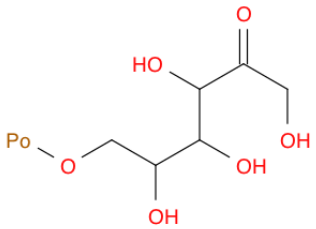
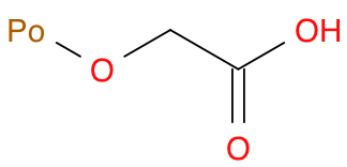


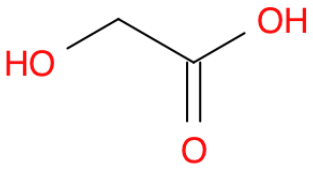
9: C6-ketose 6-phosphate

Number of reactions	8
Pathway superclass	2:6
Key C2 intermediate	aminoacetaldehyde
Key C6 intermediate	C(=O)C(N)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C6-ketose 6-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C4-aldose 4-phosphate
Co-product	

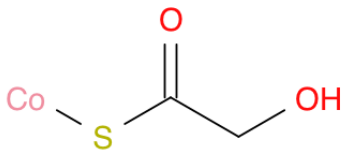
Pathway 159



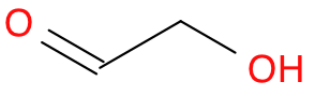
1: glycolate 2-phosphate



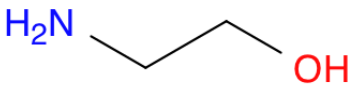
2: glycolate



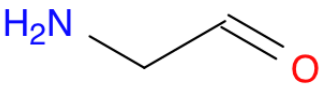
3: glycolyl-CoA



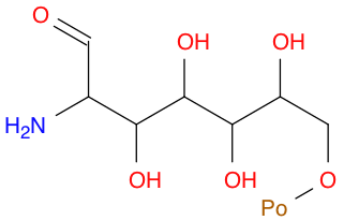
4: glycolaldehyde



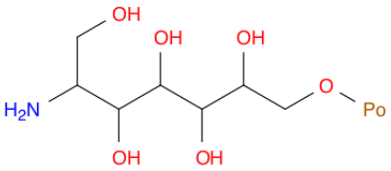
5: aminoethanol



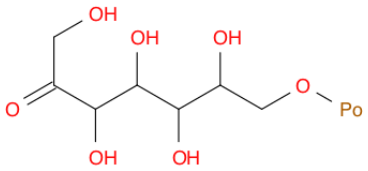
6: aminoacetaldehyde



7:



8:



9: C7-ketose 7-phosphate

Number of reactions	8
Pathway superclass	2:7
Key C2 intermediate	aminoacetaldehyde
Key C7 intermediate	C(=O)C(N)C(O)C(O)C(O)C(O)C(O[PO3])
Calvin cycle sink	C7-ketose 7-phosphate
Key reaction	Amine aldolase (donor)
Co-reactant	C5-aldose 5-phosphate
Co-product	

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