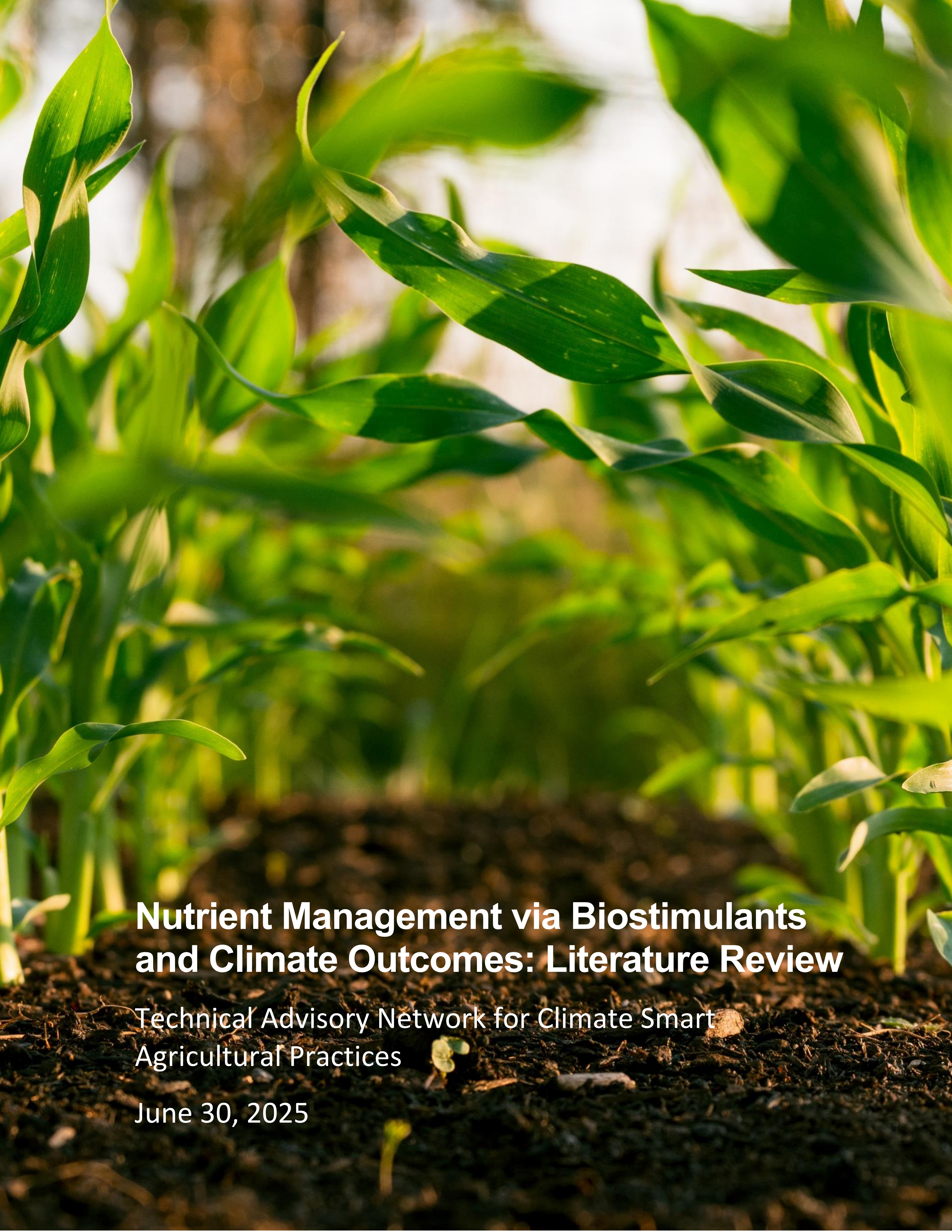

Nutrient Management via Biostimulants and Climate Outcomes: Literature Review

Technical Advisory Network for Climate Smart Agricultural Practices

June 30, 2025

ABOUT THE TECHNICAL ADVISORY NETWORK FOR CLIMATE SMART AGRICULTURAL PRACTICES

The Network emerged from a project of the Meridian Institute to better align Natural Resource Conservation Service (NRCS) Conservation Practice Standards with the best available science on GHG mitigation. In May 2024, Meridian Institute assembled six technical work groups (TWGs) to provide up-to-date scientific analysis of the impact of conservation practice adoption on GHG emissions and make technical recommendations regarding how best to achieve climate mitigation through those practices. The TWGs were comprised of academic scientists who are well regarded in their chosen field as well as scientists working within nongovernmental organizations focused on one or more of the chosen categories of interest: nutrient management, manure management, enteric methane, soil carbon, agroforestry, and grazing lands.

The technical work groups first identified agricultural practices in the six areas of interest for which evidence indicates considerable potential to reduce GHG emissions based on raw mitigation potential and adoption considerations. In August 2024 Meridian entered into a Contribution Agreement with NRCS to conduct literature reviews of the GHG emissions impacts of 39 practices, individually and in commonly practiced combinations. Meridian staff and TWG chairs and members worked with the NRCS National Discipline Leads to scope the reports and refine technical guidance regarding how best to implement practices to achieve GHG mitigation. The TWGs then conducted literature reviews, first and second-order meta-analyses, and modeling scenarios to identify the circumstances in which there is strong scientific evidence that the agricultural practices identified are likely to have net GHG benefits as well as implementation guidance for maximizing those benefits. The TWGs generated more than a dozen reports, which were delivered to NRCS in June 2025.

With the closing of Meridian Institute in mid-2025, the six technical working groups assembled by Meridian decided to establish the Network to provide a platform for sharing the work undertaken with a diversity of interested stakeholders and to support future collaboration within and/or across technical working groups. Slightly modified versions of the reports delivered to NRCS, such as this report, have been issued publicly by the Network and findings from several groups will be published in academic journals. The Network may also create briefs to summarize findings and technical recommendations for field conservationists and other interested stakeholders.

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Ruark, Matt, Monica Schauer, Justin Baker, Mike Badzmierowski, Sylvie Brouder, Michael Castellano, Heather Darby, Mark Jacobs, and Gabriel Lobet. 2025. "Nutrient Management via Biostimulants and Climate Outcomes: Literature Review and Technical Guidance." doi.org/10.5281/zenodo.15777928.

DOI: doi.org/10.5281/zenodo.15777928

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Executive Summary

Biostimulants are applied to crops to enhance performance and are gaining attention as a strategy to decrease greenhouse gas emissions and increase soil organic carbon. This report summarizes research measuring N_2O emissions and carbon sequestration on relevant biostimulants. Our findings indicate that AMF inoculation has some potential for direct N_2O reduction and low potential for indirect effects on N_2O reduction. There is no evidence that other biostimulants will lead to a reduction in N_2O emissions and can even lead to increases in N_2O emissions. Cyanobacteria are the only product that (if living) add C as well as bring in C (through CO_2) into the soil. All other products supply small amounts of C through their application, but their benefit would only occur through indirect pathways (continued increase in plant biomass C returned to the soil). Of the primary literature included in this report, only 25% of this work is conducted in field conditions, a majority of which were rice or vegetable crop fields. Unless there is substantial research to develop field-trials, and long-term field trials, it is our conclusion that these products alone will not lead to meaningful gains in C sequestration or N_2O emission reductions.

Introduction

Agricultural systems face growing pressure to enhance crop yields while simultaneously improving soil and crop resilience, decreasing greenhouse gas output, and increasing carbon sequestration. Meeting these demands requires a better understanding of the processes that govern both nutrient management and soil health, as well as the tools available for farmers for more efficient management. Loss of soil organic carbon (SOC) loss is widespread in agricultural landscapes, prompting interest in regenerative practices that aim to maintain or slow these losses to support resilient and productive soils. Agricultural greenhouse gas (GHG) emissions also remain a major concern, particularly nitrous oxide (N_2O) as it has the greatest warming potential among agricultural emissions and presents an important opportunity for mitigation. Inefficient use of nitrogen fertilizer is a huge driver in nitrogen losses from agricultural systems, largely contributing to N_2O emissions. Both nitrogen loss and carbon storage are in part determined by soil microorganisms, highlighting the significant potential for decreasing losses by altering soil microbial systems.

In recent decades, significant progress has been made in developing management strategies to improve the efficiency of crop production while reducing GHG emission and increasing SOC. One emerging strategy is the use of biostimulants applied to the crop to enhance performance and improve nutrient retention. The agricultural market is currently saturated with products and additives claiming to increase plant growth and resilience, improve yields, reduce N_2O emissions, improve soil health, sequester carbon, with key outcomes aimed at improving producer profitability. To better understand the potential impacts of these products, a synthesis of existing research is needed to clarify how these groups are categorized and what functions they serve.

The definitions of biostimulants, bioamendments, biologicals, and biofertilizers vary both in the literature and across the market, but they often refer to the same category of products. Here, we will refer to this group of products as biostimulants and utilize the definition from Du Jardin et al. (2015), “A plant biostimulant is any substance or microorganism applied to plants with the aim to enhance nutrition efficiency, abiotic stress tolerance, and/or crop quality traits, regardless of its nutrient content.". This definition of biostimulants encompasses a large range of products, each with varying functions and applications in agricultural systems.

Table 1. Inventory of technology for biostimulants explored in this work. Biostimulants are broken into groups and then include function, and mechanism for mitigating greenhouse gas emissions or carbon sequestration.

Category	Sub-category	Biostimulant	Description
Microbial	Fungi	Arbuscular mycorrhizal fungi (AMF)	Root-associated endophytic fungi.
Microbial	Bacteria	Free-living N fixers (FLNF)	Free-living diazotrophs that can fix N (may or may not be gene-regulated).
Microbial	Bacteria	Plant growth promoting bacteria (PGPR)	Soil bacteria that live in close association with plant roots (excluding cyanobacteria and free-living N fixers).
Microbial	Bacteria	Microalgae	Photosynthetic eukaryotic organisms including cyanobacteria.
Non-microbial	Plant derived	Seaweed and seaweed extracts	Macroalgae and their chemical or physical extractions, including red, brown, and green algae.
Non-microbial	Plant or animal derived	Humic and fulvic acid	Natural constituents of the soil organic matter, resulting from the decomposition of plant, animal and microbial residues, but also from the metabolic activity of microbes using these substrates.
Non-microbial	Plant or animal derived	Protein hydrolysates	Amino-acids peptide mixture obtained by chemical and enzymatic protein hydrolysis from agro-industrial by-products, from both plant sources (crop residues) and animal wastes.
Non-microbial	Animal derived	Chitosan	Co-polymers of N-acetyl-d-glucosamine and d-glucosamine from shrimp or crab shells.
Non-microbial	Mineral	Phosphite	Reduced form of phosphorus. (Considered biostimulants instead of fertilizer because no direct effect on plant nutrition)
Non-microbial	Mineral	Silicon	Biogenic silica soils, naturally occurring in earth's crust but not considered an essential element but linked to growth and development of plants.

To conduct this review, we first identified the types of biostimulants to explore, grouping by category and subcategory (Table 1); descriptions are also provided in Table 1. Using this inventory, we then created a comprehensive search strategy to investigate the effect of biostimulants on N_2O emissions and carbon sequestration. **Our search was not limited to a specific cropping system or region, as we wanted to investigate any mechanistic potential for reduction in emission or increase in sequestration**. Our search was conducted using Web of Science™ (WoS) Core Collection database. We searched using each biostimulant AND emission or sequestration AND yield (Table 2). We focused primarily on review papers, using the review paper function in WoS. This revealed 309 review papers, which was then pared down to 35 review papers to include in this report. We then used the review papers to guide us to relevant studies in the primary literature to also be included in this report. For individual biostimulants that were not well covered in a review, we conducted individual searches of the primary literature using the same search terms outlined in table 2, but only including the biostimulant of interest. We also did not want to miss any relevant work that has been conducted since the last review, so for each biostimulant we conducted searches only including years since the last included review. Of the 27 primary literature papers included in this work, 20 of these studies were conducted in greenhouse or benchtop-controlled conditions while 7 of these studies were carried out in fields, primarily paddy systems.

Table 2. Search results with outputs and number of papers included in the review. Of the 35 papers included in the review, 16 included information on biostimulant contributions to soil organic carbon, 6 to N_2O emissions, 4 covered both N_2O emissions and carbon sequestration, and 9 papers were included that gave great insight into other biostimulant properties and uses.

Search terms	Number of papers		
	Review papers from search	**Reviews included**	**Primary literature included**
(bioamendment or biostimulant or biofertilizer or "arbuscular mycorrhizal fungi" or AMF or "plant growth promo*" or PGPR or "plant growth promoting rhizobact*" or "nitrogen fix*" or "rhizob*" or "free living nitrogen fix*" or diazotroph* or symbiotic or cyanobacteria or "humic acid" or "fulvic acid" or seaweed or "seaweed extrac*" or chitosan or silicon or "protein hydrolysat*" or phosphit*) **AND** (N_2O or "nitrous oxide" or "greenhouse gas" or GHG or "carbon sequest*" or "soil c" or "SOC" or "soil organic matter" or SOM) **AND** (yield or biomass)	309	35	27

Activities with Potential Climate Benefits

(SCOPE OF ACTIVITIES CONSIDERED IN THIS REPORT)

This work will be focused on the biostimulants listed in Table 1. Biological pesticides and fumigants are not included in the scope of this review. Urease and nitrification inhibitors and controlled release products are not included here as they are being explored simultaneously as enhanced efficiency fertilizers (EEFs) by other workgroups. We are not considering biostimulants as EEFs. Biochar is also not included in this review as it is also being researched by other workgroups. Here, we will focus on the direct application of biostimulants to soil or crops, excluding their use as additives in wastewater, manure or other solid waste before reaching the field. While not included in this report, a vast body of literature exists on biostimulant additives in waste streams, warranting further exploration of those topics.

All biostimulants included in this review have demonstrated some degree of positive impact on crop yield or resilience, warranting their use and categorization as biostimulants (Abbott et al., 2018; Du Jardin, 2015; J. Li et al., 2022). While the primary focus of this report is on the effect of biostimulants on N_2O emissions and carbon sequestration, it is important to acknowledge that these products have other agronomic benefits. Biostimulants have the potential to increase plant growth, yield, and tolerance to abiotic stressors, as well as reduce need for external nutrient inputs. However, these benefits are outside the scope of this review.

The mechanism in which biostimulants can impact N_2O emissions and carbon sequestration can either be direct or indirect. A direct effect would occur when a reduction in N_2O occurs as a result of the product being applied. Indirect effects would occur when biostimulant application led to a reduction in fertilizer use or altered soil processes which then lead to N_2O reductions.

Here, we define a direct mechanism of carbon sequestration as a mechanism facilitated by microorganisms that directly provide carbon to the soil and/or aid in stabilizing soil carbon while indirect mechanisms contribute carbon to the soil by promoting plant growth and resilience and releasing organic plant exudates to the soil (Mason et al., 2023). An indirect method includes any increases in plant biomass material entering the soil, both above ground and below ground, a portion of which is subsequently converted into recalcitrant carbon pools that are resistant to decomposition. Here we acknowledge only the potential for there to be an indirect effect via biomass increase, as there are many additional factors that can influence actual C sequestration as a function of an increase in C addition to soil. We also acknowledge there have not been any continual use studies to demonstrate the potential for an indirect effect of biostimulants on C sequestration for more than two crop seasons.

State of Knowledge

ARBUSCULAR MYCORRHIZAL FUNGI

Arbuscular mycorrhizal fungi (AMF) are among the most widespread fungi in the terrestrial ecosystem and form a symbiotic relationship with plant roots. Plant roots provide photosynthates in the form of sugars or fatty acids while AMF facilitates the uptake of limited plant nutrients (Ahmed et al., 2025).

N_2O EMISSIONS

AMF transport nutrients from the soil to the roots by extending hyphal networks in the soil. A recent review by Basiru et al. (2025) found that through this symbiotic relationship, the addition of AMF into agricultural systems (via seed or soil inoculation) has the potential to influence nitrogen cycling and prevent losses both directly and indirectly. Increased abundance of AMF directly increases plant uptake of N through extraradical hyphae increasing the effective surface area of the root (Figure 1). Increased N uptake and immobilization by the plant leads to less mineral N in the soil that would be susceptible to loss as N_2O. This effect was demonstrated in several greenhouse studies where N_2O emissions were reduced via nutrient assimilation after AMF inoculation (Bender et al., 2014; Storer et al., 2018; Zhang et al., 2015).

In addition to the direct effects, AMF can also have an indirect effect on N_2O. Basiru et al. 2025 provide several examples (Figure 2) of ways in which inoculation of AMF could indirectly reduce N_2O emissions. These include synchronizing N mineralization and uptake (Leigh et al., 2009), enhancing biomass N immobilization (Bender et al., 2014; Jia et al., 2024), promoting soil aggregation (Morris et al., 2019), and altering the community structure of denitrifies (Gui et al., 2021; Zhang et al., 2022). While Basiru et al. (2025) clearly and logically explain these processes, no singular study was cited by the authors that provide direct evidence of this occurring. However, these relationships are complicated. One study attempted to evaluate both the direct effect of AMF on N_2O and the indirect of N_2O via aggregation improvement (Okiobe et al., 2019). They found that soils with denser hyphal networks (proxies for AMF populations) increased N_2O emissions, while also increasing aggregation. Through the indirect effect of increased aggregation, anaerobic zones are created which favor denitrification and increased N_2O emissions. As a result of this complexity, Basiru et al. (2025) **suggests that direct mechanisms affecting N uptake and availability in the soil may play a more significant role than indirect mechanisms related to soil aggregate functions**. However, the authors in Basiru et al. (2025) also note that evidence for there being a direct event is from research conducted in greenhouse or pot trials, as opposed to field trials.

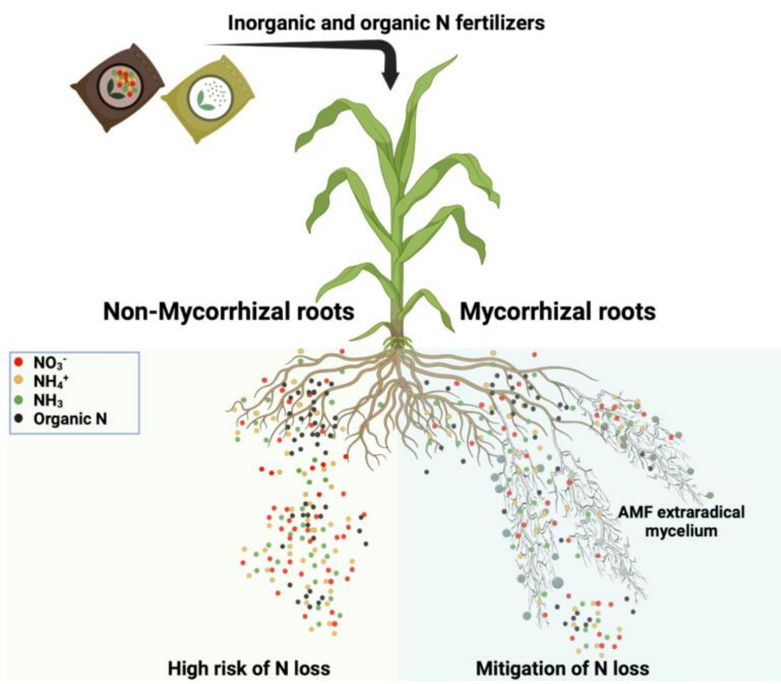

Figure 1. Direct mechanisms of N loss mitigation: AMF intercept and facilitate the uptake of inorganic N (e.g. NH_3, NH_4^+, and NO_3^-) and soluble organic N (dissolved organic N) through the extraradical mycelia after the application of chemical fertilizers or the mineralization organic N (reprinted from Basiru et al., 2025).

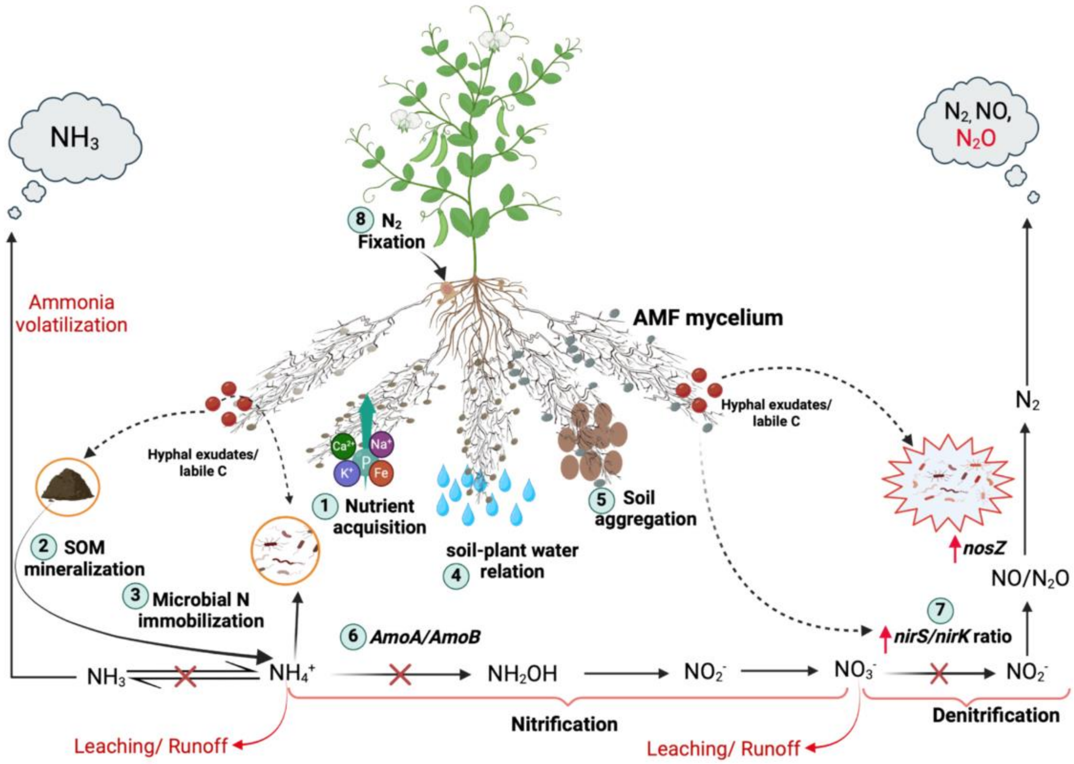

Figure 2. Indirect mechanisms by which AMF mitigate nitrogen loss. AMF can prevent or reduce nitrogen loss from plant root systems through: (1) promoting the uptake of P and other nutrients, resulting in increased N uptake and assimilation by host plants; (2) influencing SOM mineralization and facilitating the uptake of mineralized N by plants through AMF hyphae; (3) stimulating microbial respiration, resulting in N immobilization in heterotrophic bacteria; (4) promoting positive plant-soil water relations; (5) enhancing soil aggregation and structure formation; (6) altering the community structure and activities of nitrifiers and denitrifiers through changes in N availability or soil water levels; (7) affecting denitrifiers via hyphal exudation and substrate availability; and (8) promoting symbiotic N fixation, ensuring the recycling of atmospheric N2 into the system. AmoA/AmoB: Ammonium monooxygenase (archaea or bacteria). nosZ: nitrous oxide reductase gene; nirS/nirK: nitrite reductase genes (reprinted from Basiru et al., 2025).

CARBON SEQUESTRATION

AMF contributes to SOC directly through hyphal biomass and glomalin-related soil protein (GRSP) and indirectly through soil aggregate formation and enhancing plant root growth (Figure 3). Hyphal biomass alone has a high turnover rate and leads to respiration back into atmosphere upon decomposition. However, interaction between hyphal biomass and soil particles leads to formation of water stable aggregates that provide physical protection of soil C from microbial degradation (Zhu, 2003). Hyphal exudates GRSP is a more stable form of C in the soil, contributing 20 times more in SOC than microbial biomass carbon, suggested to be due to its recalcitrant nature (Schindler et al., 2007). A review by Agnihotri et al. (2022) found that the indirect effect of increased aggregation is larger contributor to SOC sequestration than the direct contribution as a C source.

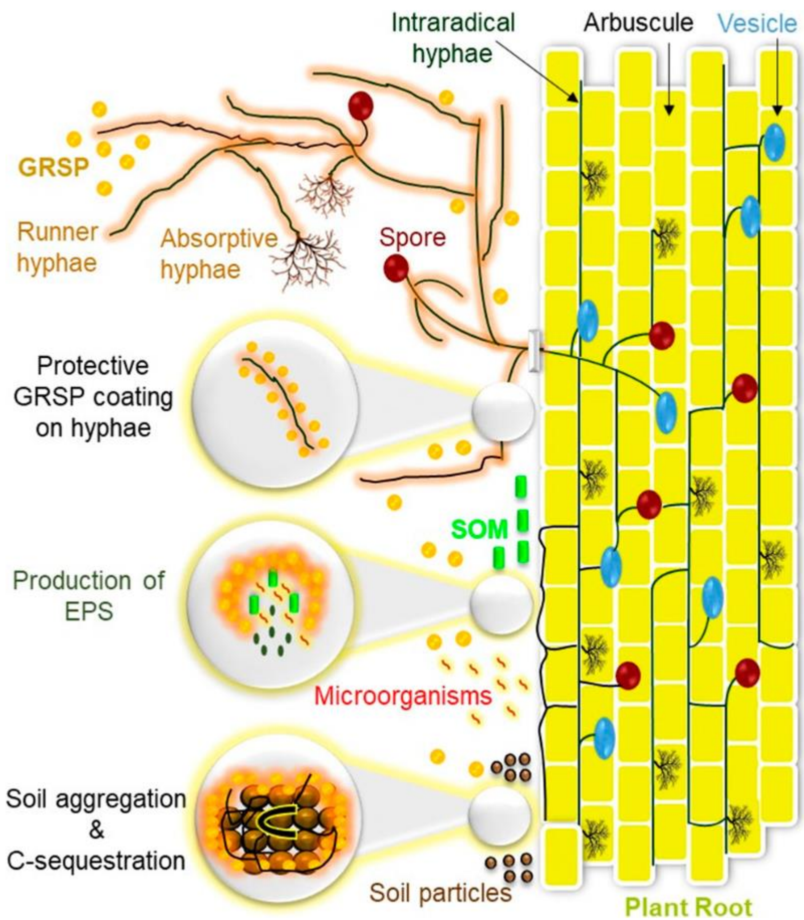

Figure 3. AMF root interaction and the mechanism for contribution to carbon sequestration (reprinted from Agnihotri et al., 2022).

The mechanism in which AMF contributes directly to carbon cycling is mainly by creating a sink demand for plant C and distributing that C from the plant to the soil. AMF contributes to soil C by adding organic residue to the soil in the form of hyphal biomass and exudates which release organic C as they decompose (Parihar et al., 2020). As obligate biotrophs, AMF receive a vast majority of their C demand from the host plant rather than mining of organic matter. By nature, AMF hyphae transport carbon away from the plant and into the soil matrix, away from the rhizosphere region with high respiration activity. This C is deposited in the soil as fungal chitin GRSP which is more stable in the soil than plant originated (Orwin et al., 2011). For AMF to contribute to a net increase in SOC, the amount of belowground rhizodeposits and residence time of these rhizodeposits resulting from the inoculant must exceed any losses from priming effect and decomposition of the inoculant itself. Given this interaction, there is a short-term priming effect but longer-term increase in soil aggregation and carbon stabilization from AMF biostimulant application (Rubin et al., 2023). This is more about how AMF plays a role in stabilizing soil carbon, rather than showing value from inoculating with AMF. While a substantial amount of carbon is retained by AMF, the length of time that carbon remains in the soil and is physically protected is crucial for prolonged carbon storage. There is no evidence that long-term inoculation of AMF increases soil C.

Current research indicates the potential for carbon sequestration by AMF, but no long-term field studies exist to validate the connection between long-term AMF inoculation with C sequestration. Research on this topic thus far has focused on AMF in greenhouse or bench top studies, and to our knowledge no work has measured the effect of an AMF product on carbon sequestration or N_2O emissions under field conditions with proper controls.

The effectiveness of AMF as a biostimulant is further undermined by the lack of regulation in commercial products, which can result in the use of non-viable spores or root fragments. A global evaluation looked at 28 commercial AMF inoculants in field or greenhouse conditions (Salomon et al., 2022). Out of 25 commercial inoculants tested in sterile soil under favorable conditions, only 4 resulted in root colonization, demonstrating that 84% of tested inoculants did not contain viable propagules.

Difficulty in identifying AMF strains compatible with local conditions and microbial communities is a challenge to the efficacy of AMF as a biostimulant. In non-sterile soil (realistic environment for producer or grower use), native AMF communities were already present, and the addition of the commercial product failed to increase colonization (Salomon et al., 2022). Due to lack of field evidence, more value would come from promoting indigenous AMF communities by implementing management practices that promote soil health rather than inoculation with biostimulant products (Basiru et al., 2025; Rubin et al., 2023).

FREE LIVING N FIXERS (FLNF)

Free living N fixers (FLNF) are single celled organisms that have the ability to convert atmospheric nitrogen (N_2) into bioavailable forms of N inside the cell. Here, we focus on the FLNF as these are available on the market as a soil additive, with some products being gene-regulated so N fixation will not be inhibited during the life cycle of the cell. It is important to note that FLNF are different than N fixers that form a symbiotic relationship with plants (e.g. *Bradyrhizobium* which associates with soybean). These would be considered seed inoculants, as opposed to a general soil additive.

N_2O EMISSIONS

Free-living N fixers will not reduce N_2O emissions through a direct effect. The potential benefit of FLNF from an indirect effect would occur if less N fertilizer was applied, reducing the amount of nitrate that may exist at any time. However, this claim is not well supported, as field experiments with proper controls are lacking, and the existing data show inconsistent results (Giller et al., 2024). In fact, Giller et al., (2024) after reviewing existing literature on N_2-fixing rhizobia, concluded **that there is not robust evidence that inoculation of crops with free-living and/or endophytic bacteria leads to fixation of an agronomically significant amount of atmospheric N_2 gas**. These results have been substantiated by a summary of field N response data across the North Central United States, where 59 out of 61 site-years of data had no yield increase with use of the product, nor any N supply of the product (Franzen et al., 2023). However, a recent publication by Woodward et al., (2023) suggests FLNF products do supply N, but the amount is relatively low (<10 lb/ac), and the effect will not occur every year.

From our review, we only identified one paper that measured N_2O after inoculation with FLNFs. This work included a N-fixing microbe treatment which included *Azotobacter vinelandii* and *Clostrium pasteurianum* as well other 'secondary organisms' including *Pseudomonas fluorescens* and

Nitrosomonas, Nitrococcus, Nitrobacter and *Bacillus* (Souza et al., 2019). The application of this biostimulant led to greater N_2O emissions when compared to urea alone, but when applied with an amino acid blend N_2O emissions were not significantly different than emissions from urea alone. **Since gene-regulated N fixers have potential to add more N in the soil environment that is available to crops, there theoretically could be direct potential for greater N_2O emissions**.

There is a fair amount of controversy in the market of FLNF, the authors of Giller et al. (2024) call for regulation to prevent sale of inoculant products with unsubstantiated claims and that products should be verified and that (i) the inoculant bacterium can fix N_2 from the atmosphere (i.e. that it possesses all the genes required to make nitrogenase), (ii) it has a clear mechanism to protect nitrogenase from poisoning by free oxygen, (iii) the bacterium is present in sufficient numbers throughout the growth cycle of the plant, (iv) that enhanced respiration can be detected from the putative N_2-fixing tissues, (v) that inoculation of the nonlegume growing in an N-free medium leads to prolific growth and accumulation of nitrogen, and (vi) more than one method is used to confirm quantitatively significant inputs from N_2-fixation in the field.

CARBON SEQUESTRATION

The potential for FLNF to increase C sequestration would entirely occur through indirect effects. While theoretically this indirect effect could contribute to carbon sequestration, we did not find any research directly quantifying the effect of FLNF application to SOC. The effectiveness of FLNF on increased crop growth has only been demonstrated in greenhouse pot studies, and a decrease in performance of FLNF is observed when pot grown plants are shifted to a field (Bhattacharjee et al., 2008).

OTHER PLANT GROWTH PROMOTING RHIZOBACTERIA (PGPR)

Plant growth promoting rhizobacteria generally refer to beneficial bacteria that enhance plant growth through improved nutrient adsorption and increased stress tolerance (Shah et al., 2021; Upadhyay et al., 2023). Here, we include bacteria used as biostimulants (not primarily N-fixers) include *Bascillus, Pseudomonas, Streptomyces, and Paenibacillus.*

N_2O EMISSIONS

The effect of this group of biostimulants on N_2O emissions is indirect, similar to the effect observed with free living N-fixers. In a controlled benchtop incubation study measuring N_2O emissions, Calvo et al. (2013) compared a commercially available biostimulant SoilBuilder to a *Bacillus* mixture with four strains. SoilBuilder with UAN fertilizer significantly reduced total N_2O emissions compared to UAN alone. When applied with urea, both Bascillus and Soilbuilder led to increases in total N_2O emissions compared to urea alone. The authors note the possibility that the microorganisms may not have survived the length of the study, further clouding results. The inconsistency in results across strain of inoculant and fertilizer source further highlights the variability in this effect.

CARBON SEQUESTRATION

PGPR have potential to increase SOC indirectly by specifically increasing belowground inputs of plant roots and exudates or by increasing carbon storage capacity of soils (Bamdad et al., 2022; Da Silva et al., 2021). A controlled growth chamber study found that inoculation with *Pseudomonas fluorescens* increased C storage per unit of N and increased root length (Nie et al., 2015). However, no long-term field evidence of SOC increases following PGPR inoculation was found. More work needs to be done investigating the viability of PGPR as biostimulants in agricultural landscapes.

MICROALGAE

Microalgae is a diverse group of photosynthetic microorganisms including cyanobacteria (e.g. *Nostoc, Anabaena)* and eukaryotic organisms like green algae and diatoms. Microalgae can be applied to agricultural systems as a biostimulant either living or dead. When applied dead, they have shown effectiveness as a biostimulant, promoting plant growth and tolerance to stress (Kollah et al., 2016; Sánchez-Quintero et al., 2023; Win et al., 2018). However, most work on microalgae as a biostimulant has focused on inoculation as live microorganisms into rice paddy fields where growing conditions allow for photosynthesis and nutrient acquisition into the cropping systems, promoting growth and yield.

N_2O EMISSIONS

We did not find any research that would demonstrate a direct effect of microalgae application on N_2O emissions. However, there is potential for microalgae to have an indirect effect on N_2O emissions. Most research on cyanobacteria is under rice paddy cropping systems and potential effects are based on greenhouse or benchtop studies rather than true field conditions. This indirect effect would occur as a function of less inorganic fertilizer applied to soil. When applied as a dry biostimulant, cyanobacteria (*nostoc specifically)* provided a slower release of nutrients than urea (Do Nascimento et al., 2019). Thus, when applied living, cyanobacteria have the ability to fix N (Dasgupta et al., 2021), 3-20 mg of N per 100 cubic cm of nutrient free medium (Rubin et al., 2023). One paddy field study in China found that application of living *Anabaena* provided N to the rice crop and allowed for a 50% reduction in urea fertilizer application without reducing yield (X. Song et al., 2021). Even under this specific rice paddy cropping system, it is important to note that cyanobacteria is sensitive to disturbance, and field operations such as planting, tillage, and herbicides could greatly reduce population and reduce effects of the biostimulant (Do Nascimento et al., 2019). If future field research shows that microalgae applications do not reduce fertilizer need, then the net result on N_2O could be an increase in direct emissions.

CARBON SEQUESTRATION

Microalgae have the capability to capture and store CO_2, potentially influencing soil carbon dynamics. A review by De Silva et al. (2024) synthesized the direct and indirect pathways of the contribution of microalgae to soil carbon sequestration (Figure 4). Microalgae also have several direct pathways of contributing to SOC, including capturing atmospheric CO_2, secretion of extracellular polysaccharides which supply nutrients to soil microbes, and creating stable aggregates. A review by De Silva et al., 2024 found that inoculation with living cyanobacterial strains can successfully increase microbial biomass carbon in a variety of cropping systems in both field and laboratory conditions. Microalgae can be very productive in terms of fixing C in laboratory and benchtop mesocosms, but field evidence is limited to paddy rice systems. In agricultural field studies, any increases in C would be reflected in the difference in SOC stocks comparing inoculated soils to control, after subtracting the C that was added from the inoculant (Rubin et al., 2023).

An indirect contribution of microalgae to carbon sequestration is through promoting plant growth and biomass (Alvarez et al., 2021). Cyanobacteria has demonstrated the ability to increase root length and total plant mass in wheat fields compared to non-inoculated controls (Hakkoum et al., 2025). While this increase in plant biomass could lead to potential increases in SOC, the study did not directly measure SOC.

Microalgae also play a significant role in soil aggregation, which is fundamental to retention of organic carbon, preventing SOC from breaking down by making it easier for carbon compounds to move from labile carbon pools to recalcitrant carbon pools (Mason et al., 2023). Specifically, cyanobacteria have trichomes with mucilaginous sheaths, allowing them to adhere to soil and promote aggregation (Falchini et al., 1996). Microalgae also play a significant role in soil aggregation, which is fundamental to retention of organic carbon, preventing SOC from breaking down by making it easier for carbon compounds to move from labile carbon pools to recalcitrant carbon pools (Mason et al., 2023). Although improved soil aggregation is known to enhance SOC levels, no studies have directly linked increased SOC stocks to microalgae biostimulant applications through the mechanism of soil aggregation.

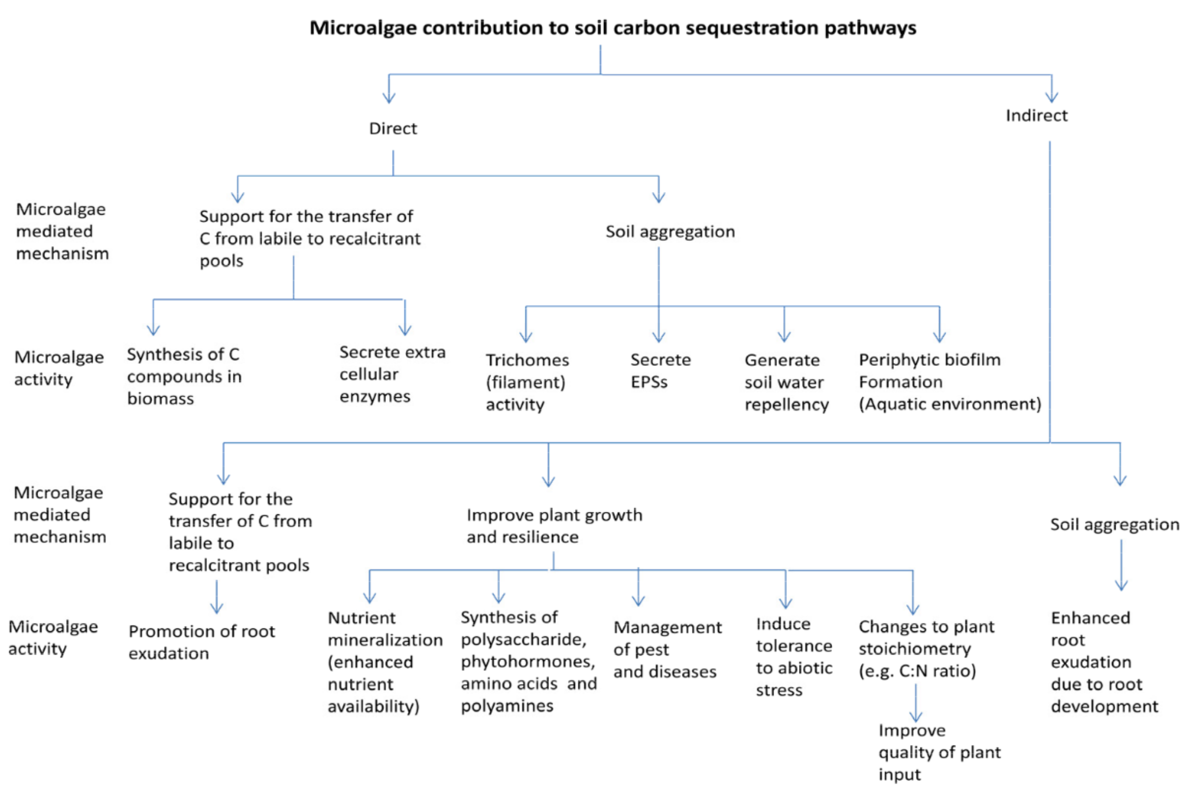

Figure 4. Indirect and direct mechanisms by which microalgae contribute to soil carbon sequestration (reprinted from De Silva et al., 2024).

HUMIC & FULVIC ACID

Humic and fulvic acids are a product of specific extraction methods. Humic acids (HA) are soluble at high pH ranges and Fulvic acids (FA) are soluble across the full range of pH. Humic acids are made up of larger molecules, with chemically reactive functional groups made up of hydrophobic compounds (Hayes, 2006). Fulvic acids (FA) are comprised of small hydrophilic molecules with the ability to chelate minerals, allowing for transport of minerals and nutrients through plant membranes and into the cells (Piccolo et al., 1992). There is much debate in the literature if these actually occur in nature or are in fact simply the product of the extraction method itself (Kleber and Lehmann, 2019). These methods were historically used to define soil organic matter pools, but more advanced techniques have replaced them. Regardless of this debate, humic acids and fulvic acids are soils as soil additives, although it is difficult to always understand from what material these products were extracted from. The literature is generally not clear on this aspect of HAs and FAs. Reviews address their role in soil and plant effects after inoculation but rarely explain the nature of the HAs and FAs that were applied, nor the methodological approaches used to obtain them, nor the chemical nature of the products themselves. There are many reviews of HAs and RAs if interested in understanding them better (Sen et al., 2020; Maffia et al., 2025); but here we simply review the studies that have demonstrated effects on N_2O and C sequestration as a response to application of these materials.

N_2O EMISSIONS

Humic and fulvic acids have the potential to reduce N_2O emissions by improving nitrogen retention and limiting denitrification in agricultural soils. HA can successfully inhibit urease activity, slowing the conversion of urea to ammonium, reducing the accumulation of nitrate and leaving less N susceptible to denitrification (Kong et al., 2022). In a greenhouse study, humic acid urea (humic acid combined with urea by hot fusion) reduced N_2O emissions by 22% compared to urea alone (Dong et al., 2006). A field study demonstrates a reduction in N_2O emissions when HA is applied with a controlled released N fertilizer compared to the same rate of the controlled release fertilizer itself (Guo et al., 2022).

While great potential exists for N_2O emission reduction with a humic acid applied with other N sources, research described above took place on field and greenhouse studies in China and product efficacy would need to be demonstrated in other climates and management systems. While the body of literature exploring humic acid impact on crop performance and yield is quite robust, field studies directly measuring N_2O emissions with proper controls are lacking (Guo et al., 2022 appears to be the only one).

CARBON SEQUESTRATION

Humic acid can promote root growth and development, increasing above and below ground crop health (Da Silva et al., 2021; Mora et al., 2010). However, this increase in root growth does not always directly correlate to increases in SOC stocks. In a greenhouse study with increased root growth facilitated by humic acid, SOC did not significantly increase, potentially due to triggering of microorganisms which increased microbial utilization of SOC (Dong et al., 2006). While potential for HA and FA indirect contributions to SOC exist, long term field measurements specifically measuring root exudates SOC contribution following humic or fulvic product application is lacking (Olk et al., 2018).

Humic acid facilitates formation of soil macroaggregates, improving SOC storage. Humic acids have further been shown to stabilize SOC in soils with this impact increasing after more and more years of product application (Maffia et al., 2025). To maximize benefits of humic acid on soil health and plant growth, Maffia et al. (2025) suggest once a year application for general soil health, two or more times a year for degraded or sandy soils, and two to three times a year for high-intensity farming or crops with a high nutrient demand. However, while this recommendation does not include specifics on product and rate, authors of Olk et al. (2018) warn that improvements to soil organic matter are unlikely to occur through recommended application rates of liquid HA alone. This review also points out that most field studies testing these products only take place for one or two years with a different field location each year, so the potential long-term benefits of continuous application are not well documented.

SEAWEED EXTRACTS

Seaweed extracts (SE) are derived from marine macroalgae through bioactive ingredient extraction. SE contain phytohormones and trace elements which have potential to increase plant growth (particularly root development), stress tolerance, soil microbial activity, and soil structure. SE have been used in agriculture throughout history, applied as a foliar spray, seed treatment, or to the soil.

N_2O EMISSIONS

Limited research has examined the impact of SE on N_2O emissions, and no studies have directly measured its effect. A study investigating extracts from brown seaweed (*Ascophyllum nodosum*) on barley indicated an increase in yield and nitrogen uptake by the plant which theoretically could impact N_2O emissions (Goñi et al., 2021). Similar to other biostimulants, seaweed as a fertilizer replacement has potential to indirectly impact GHG emissions, but evidence of this effect is lacking.

CARBON SEQUESTRATION

Similar to N_2O emissions, the effect of SE on SOC is not well understood. In theory, application of SE as a biostimulant increases plant growth and root biomass which could lead to increases in soil carbon (Zaman et al., 2016). Currently, there is no empirical evidence to support this effect.

SILICON

Silicon is not considered an essential element for plant nutrition but is applied as a biostimulant for benefits on crop growth, increased resistance to abiotic stress, and ability to control mobility of heavy metals (Z. Li et al., 2018). In rice production, silicon is commonly applied as slag or monosilicic acid because rice accumulates silicon in its biomass, leading to soil depletion when grown as a monoculture.

N_2O EMISSIONS

Silicon has the potential to reduce N_2O emissions through increasing crop productivity and N uptake or by altering the soil microbial population. In a 3-year field experiment on paddy soils, application of a slag-based silicate fertilizer decreased N_2O emissions (A. Song et al., 2017). This effect was also observed with soil applied monosilicic acid in barley and rice (Bocharnikova & Matichenkov, 2024; Włodarczyk et al., 2019). While this biostimulant has potential to reduce N_2O emissions, this effect is only observed in paddy soils with specific conditions constantly shifting from aerobic to anaerobic conditions. Reduction in N_2O emissions is attributed to not only by increasing N uptake by the plant, but also by lowering abundance of microbial denitrifier communities in the soil (A. Song et al., 2017).

CARBON SEQUESTRATION

Silicon biostimulants have the potential to contribute to carbon sequestration indirectly by increasing crop root biomass. Silicon amendments have been shown to increase root biomass in rice by 10-80% (Ma & Takahashi, 2002). Both a greenhouse study with miscanthus, rapeseed, and soybean and a field study with rice demonstrated an increase in root biomass with the application of silicon (Bocharnikova & Matichenkov, 2024).

PROTEIN HYDROLYSATE

Protein hydrolysates are a mixture of polypeptides and free amino acids resulting from enzymatic and chemical hydrolysis of agro-industrial plant or animal by-products. They are utilized as a biostimulants primarily in the specialty crop industry, and have been widely proven to improve both crop yield and quality (Ciriello et al., 2024; Sun et al., 2024). To date, no work has been done investigating the effect of protein hydrolysate use on N_2O emissions or carbon sequestration.

PHOSPHITE

Phosphite is a reduced form of phosphate that acts as a biostimulant rather than a fertilizer because it cannot be converted to phosphate and has no direct effect on plant nutrition. Phosphite is mostly utilized in the horticulture industry where it has been shown to improve crop yield and quality (Gómez-Merino & Trejo-Téllez, 2015). While phosphite has the potential to impact carbon sequestration indirectly by increasing plant biomass, no work has measured the direct impact of phosphite on carbon sequestration or N_2O emissions.

CHITOSAN

Chitosan is a polymer derived from chitin, a waste product of the shellfish industry. Research on chitosan's use as a biostimulant is extremely lacking across the board, with most research on use in specialty crops. A field study found a slight increase in N_2O emission increase when chitosan was applied to cauliflower and tomato crops (Xu et al., 2022). While not directly measured, chitosan has potential to increase SOC indirectly by increasing plant biomass, as demonstrated with an increase in root biomass on chrysanthemums (Ji et al., 2017). Chitosan also has potential use as a hydrogel to encapsulate nitrification inhibitors to increase efficacy, delaying the release of the nitrification inhibitor (Minet et al., 2013).

Summary

While biostimulants are widely explored in the literature on their use for plant function and beneficial interactions, research investigating the potential effects on N_2O emissions is widely lacking. The potential impacts of biostimulants on carbon sequestration is better explored, with AMF and cyanobacteria the most studied of the biostimulants.

Our final conclusions are as follows:

N_2O

- **AMF inoculation has some potential for direct N_2O reduction, low potential for indirect effects N_2O reduction.**
- **There is no evidence that other biostimulants will lead to a reduction in N_2O emissions and can even lead to increases in N_2O emissions.**

C sequestration

- **Cyanobacteria are the only product (if living) add C as well as bring in C (through CO_2) into the soil.**
- **All other products supply small amounts of C through their application, but their benefit would only occur through indirect pathways (continued increase in plant biomass C returned to the soil).**

Our conclusions coincide with a recent review by Rubin et al. (2023), which provided a low "confidence score" on the effect of biostimulants on these parameters (Table 3). From the primary literature included in this review, 75% of this work was greenhouse or benchtop studies. Field research measuring the efficacy of biostimulants across representative sites and cropping systems is lacking. **However, given the recent popularity and interest in biostimulants, we expect a tremendous number of papers will be published over subsequent years, thus this is a topic that needs to be addressed periodically.** As research in this area grows, it is essential this work follows proper guidelines for measuring product efficacy. Lyons et al. (2024) created a set of guidelines providing data and design requirements for trials designed to test product efficacy. This protocol aims to ensure consistency in data collection for research on this topic, which should be considered in future research on biostimulants. Clearly there is tremendous research need for each category above on their effects on yield, nitrogen supply to plant, N_2O emissions, and carbon sequestration. However, we also need to recognize the diversity of products within each category that need to be tested independently on a product-by-product basis. Unless there is substantial research to develop field-trials, and long-term field trials, it is our conclusion that these products alone will not lead to meaningful gains in C sequestration or N_2O emission reductions.

Table 3. Summary of biostimulants and their effects on N_2O emissions and carbon sequestration (adapted from Rubin et al., 2023).

Category	Subcategory	Biostimulant	Mitigation elements[1]	Magnitude of effect[2]	Confidence score[3]
Microbial	Bacteria	Cyanobacteria	1. Increased SOC	++	L

			2. Reduced soil N_2O 3. Life cycle emission reduction		
Microbial	Fungi	Fungal additives	1. Increased SOC 2. Reduced soil N_2O 3. Life cycle emission reduction	±	L
Bacteria	Bacteria	Rhizobacteria	1. Reduced soil N_2O 2.Increased SOC 3. Life cycle emission reduction	±	L
Non-microbial	Plant derived	Seaweeds & seaweed extracts	1. Increased SOC	±	L

When amendments involve multiple mitigation elements, these elements are arranged in order of theorized magnitude, with stronger effects first and weaker effects last. A qualitative assessment of unconstrained project-level potential for climate mitigation, assuming no social or scalability barriers, is provided next (+ = low magnitude; ++ = medium magnitude; +++ = high magnitude; ± = mixed results for mitigating or exacerbating climate change). A confidence score for the magnitude of the potential (L - low evidence, M - medium evidence H - high evidence) is then provided for each amendment, based on the quantity of papers and quality of measurements of the empirical studies for each amendment. Rubin et al., (2023) further explains the process behind determination of magnitudes and confidence scores.

Science Gaps and Recommendations

Critical science gaps need to be filled to improve GHG mitigation research, data collection, and modeling tools to better understand the potential of conservation agriculture practices to mitigate agricultural GHG emissions within and across agricultural sectors. The gaps and needs identified below and associated recommendations are oriented to achieving the goal of enabling GHG mitigation assessment capabilities at the granularity necessary to meaningfully guide agricultural practice adoption and implementation decisions to optimize GHG mitigation across a range of geographic and operational settings. Such assessment capabilities would inform and improve the development and refinement of public and private sector practice standards, policies, programs, and investments to incentivize practices to optimize achievement of GHG mitigation alongside profitability for farmers, cost-effectiveness for incentive and cost-share programs, and achievement of other conservation objectives.

There is a substantial and growing research on nutrient management strategies for increasing farm profitability, reducing nutrient losses, and supporting GHG mitigation goals. Specifically, the extent to which a 4R nitrogen management strategy (right rate, right source, right timing, and right placement) can simultaneously achieve farm profitability and environmental stewardship has been studied extensively in the context of soil fertility, water quality, and (to a lesser extent) GHG emissions. Despite these scientific advances, key knowledge and data gaps remain, limiting the potential usefulness of decision support frameworks. We outline several of these gaps below and potential steps for improving information, modeling tools, and nutrient management recommendations.

- Significant data and research gaps inhibit the understanding of GHG mitigation potential through an expanding array of nutrient management practices. However, fertilizer application and associated nutrient management practices account for a significant portion of GHG emissions from agriculture, and reduction in application rate is one of the most certain forms of GHG reductions available within agricultural systems. Therefore, a comprehensive scientific roadmap for GHG mitigation through nutrient management is crucial. This roadmap should:
    - Articulate the existing research and key gaps from on the ground science and how it links, or fails to link, with modeling and farmer decision making.
    - Determine the tools, guidelines, and information needed for a company to credibly claim GHG reductions and chart a pathway to achieve this.
    - Address/understand variation rather than identify the average effect with NM products to develop probability-based metrics.
    - Develop an understanding of how, when, and where NM products are used to help define what can be considered an Enhanced Efficiency Fertilizer in practice.
    - Outline proper design of contributing experiments to build the evidence base from in-field experiments.
        - Lyons et al., 2024 does this to some extent, but this paper stops short of recommending quantitative considerations for experimental designs (power analysis, etc.) so there is more to build on.
    - Develop a similar guide for meta-analyses to ensure meta-data are useful for models and policy, as discussed below.
- Field research measuring the efficacy of biostimulants across representative sites and cropping systems is lacking, especially research following proper guidelines for measuring product efficacy.

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