HALT CORONAVIRUS REPLICATION

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ABSTRACT

SIRT1 inhibitors can reduce replication of many viruses with certain similar characteristics to those of Coronaviruses, while p53 protein is another important factor in down-regulation of growth. Tenovin is a class of small molecules that inhibit Sirtuin 1 and 2, in addition to activate p53 protein, by means of regulation of the interactions used by Coronaviruses as self-defense mechanism. By blocking virus growth and continuous replication, associating the already tested Antiviral medicines, Covid-19 can be definitively eradicated.

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INTRODUCTION

1. How Coronavirus enters in the host

To date, it is known that the **Novel Coronavirus penetrates** into the host via the ***enzyme converting receptor for angiotensin II*** **(ACE2),** by **binding** to **glycoprotein S**, with some differences from what happens with Sars-Cov.[1]
From here, you can draw up a **list of interventions** to **reduce the expression of ACE2 activity**, in order to avoid (in whole or in part) the entry of the virus into the subject under attack, but not only. It is also possible to **reduce the ability of the virus to reproduce** and **lower its endoribonucleic activity**. So, let us see what can be done.

2. ACE2 inhibitors: direct and indirect way

We assume that **ACE inhibitors** (Lisinopril, Quinapril, Captopril) **don't reduce ACE2 activity**, despite being structurally related and traceable in the same organs, ACE2 has opposite biological effects (vasodilatation, bronchodilatation, activity to compensate for physical stresses, as some particular conditions, like hypoxia); some **direct ACE2 antagonists** are:

- The small molecule **MLN-4760-B** and its isomeric **MILN-4760**, much more selective and effective;
- **DX600** peptide (and **DX-512**) having a nanomolar affinity for ACE2, much more than for ACE (almost null), competitive and non-competitive inhibition.

The effectiveness and selectivity between these two elements are concentration-related, but at the concentration of 10μM, **MLN-4760 was found to have a better activity profile**.[2]

3. SIRT1 inhibitors as ACE2 and virus replication suppressor

There is also the possibility to act indirectly, through **a *Sirtuine* group protein**, **7 enzymes NAD+ dependent**, known as **SIRT1**.
Expression of ACE2 activity varies with binding to SIRT1 protein, so **inhibiting SIRT1, we'll lower ACE2 activity**;[3]

Sirtuine are located in the **cellular nucleus** (SIRT1, SIRT6 and SIRT7), in the **cytoplasm** (SIRT2), or in the **mitochondria** (SIRT3, SIRT4 and SIRT5) and are involved in many **cellular functions**, such as metabolism, the cell cycle, l'**apoptosis**, DNA repair, etc. Recent studies have found that sirtuine **may also have enzymatic functions**. They are sensors of changes in the intra- and extracellular environment, generally involved in maintaining human health, but are also implicated in some viruses replication.

METHOD OF INVESTIGATION

4. Examples of virus replication reduced by Sirtuines inhibitors

- Think, for example, of the **Tat protein** of the **HIV virus**: its **activity is regulated by SIRT1**, whose activity of deacetylase, vice versa is inhibited by the protein Tat. The **transcription** of HIV **is regulated by SIRT1**, by means of **Tat deacetylase**. **SIRT1 preserves the defenses of the virus, throughout its evolution**, allowing the **recycling of the protein tat**, which **binds to TAR**, and the continuous **prolongation of the mRNA transcription**;[4]

- **SIRT1 inhibition** (and sometimes even SIRT2) by minor **changes to siRNA**, is known to cause the **decrease in replication of additional viruses**: flu strains, VSV *(Vesicular Stomatitis Virus*), KSHV (*Kaposi's Sarcoma-associated Herpesvirus*), Hepatitis B, HCMV, adenovirus, polyomavirus and in some diseases counteract: some types of cancer and Huntington's chorea, sometimes in combination with antivirals, for the best results;

- Among the various other viruses that respond by **slowing their growth**, there is just **Mers-Cov**. It has been observed that **SIR2** (SIRT1 is the human correspondent) acts as the **proviral of Mers-Cov** in the **yeast**, due to interactions between **ORF4a** and eukaryotic cells. **YDL042C/SIR2** yeast gene is a **suppressor of ORF4a** function. **When SIRT1 is inhibited** by either chemical or genetic manipulation, **there is a reduction of MERS-CoV replication**. Moreover, **ORF4a inhibited SIRT1-mediated modulation of NF-κB signaling**, demonstrating a functional **link between ORF4a and SIRT1 in mammalian cells**. It been identified a functional link between the MERS-CoV ORF4a proteins and the YDL042C/SIR2 yeast gene.[5]

- **VSV-SARS-St19 infection is mediated by SARS-CoV-S protein in an ACE2-dependent manner**. VSV-SARS-St19 will be useful for analyzing the function of SARS-

CoV-S protein and for developing rapid methods of detecting neutralizing **antibodies** specific for **SARS-CoV** infection.
SIRT1 inhibitors stop the growth of VSV, since that **cells' apoptotic response is reduced in cells affected by VSV**.[6]

5. Coronavirus self-defense mechanism

INTERACTION WITH SARS-UNIQUE DOMAIN (SUD) AND PAPAIN-LIKE PROTEASE (PL^{PRO}) AGAINST P53

The **strategy developed** from Sars.Cov and other **coronaviruses** against host immune recognition system is expressed by a **Sars-Unique Domain (SUD)** that **interacts** with his partner cellular **E3 ubiquitin ligase** ring-finger and **CHY** zinc-finger domain-containing 1 **(RCHY1)** and with **papain-like protease (PL^{PRO})**. The consequence is a **down-regulation of p53, involved in reduction of virus replication**. The SARS-CoV papain-like protease is encoded next to SUD within **nonstructural protein** 3. **A SUD–PL^{PRO} fusion interacts with RCHY1 more intensively** and causes **stronger p53 degradation** than SARS-CoV PL^{PRO} alone.[7]

6. How p53 is very important in Coronavirus replication

P53 protein **reduce coronaviruses replication**, because normally activates genes that are involved in the cell's non-specific **antiviral defense system**. In cells which p53 is too lower, the rate of coronavirus **replication** is **several orders of magnitude higher** than that observed in cells in which the p53 is present.[7]

CONCLUSION

7. Tenovin-1 as hypothetic perfect molecule in the halt of replication

Tenovin-1 [Formal name: N-[[[4-(acetylamino)phenyl]amino]thioxomethyl-4-(1,1-dimethylethyl)]-benzamide] is a small molecule **inhibitor** of **sirtuin 1** and **sirtuin 2**,[8] an **activator** of **p53**, less toxic of Tenovin-6. Tenovin-1 at concentration of 10μM protects p53 from mdm2-mediated degradation with little effect on p53 synthesis. Histone deacetylase inhibitors like tenovin-1 can also **indirectly affect** the function of some **E3 ubiquitin ligases**.
Tenovin-6 is an analog of tenovin-1 At 10 μM, this compound is slightly **more effective** than tenovin-1 **at elevating p53** activity, but it's **more toxic**.

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