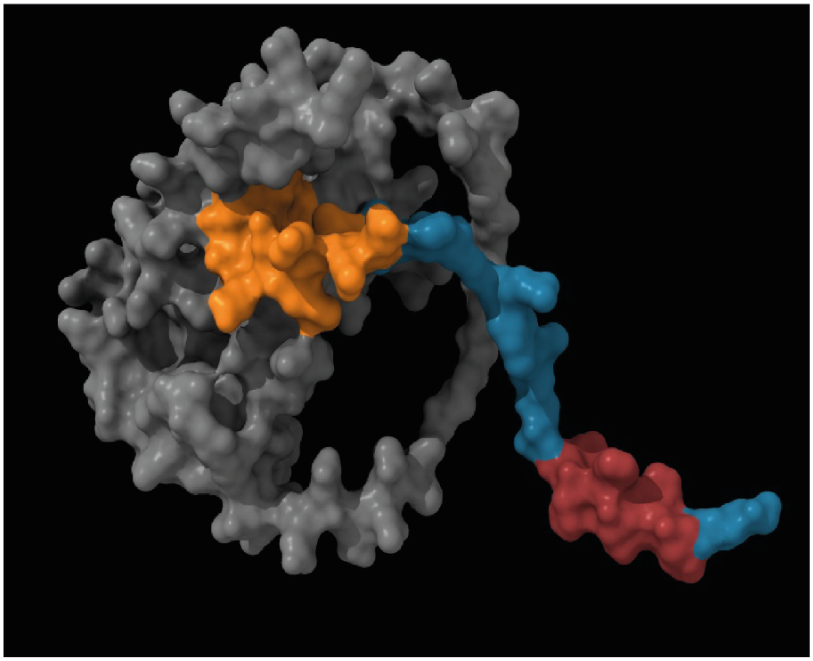

Production of recombinant NS2B-NS3 in *E. coli* and inhibition studies

Popular science summary

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The Next Chapter in Treating the Dengue Virus: A Novel Approach to NS2B-NS3 Production and The Pursuit of Naturally Derived Antiviral Agents

In the realm of virology, the dengue virus has long posed significant challenges to global health. With rising world temperatures and migrating host organisms the threat posed by this virus will only grow larger as time goes by. This study presents a pioneering approach to producing a key enzyme from the dengue virus - the NS2B-NS3, as well as a delve into the world of naturally derived antiviral agents for a brighter and more sustainable future.

The dengue virus is one of the most widespread viral diseases in the world today, being spread by mosquitoes present in regions where ~4 billion people live. The dengue virus is generally quite a mild disease, but frequent infections or a weakened immune system can lead to the more serious dengue hemorrhagic fever (DHF), or in the worst of cases, dengue shock syndrome (DSS). Two highly severe conditions resembling that of the infamous Ebola virus. When exploring the details of how dengue gets transmitted one of the key enzymes involved is called NS2B-NS3, playing a prodigious role in the replication of the virus. Having lived through the recent covid-19 pandemic we are all very familiar with the devastation caused by a viral outbreak. It is therefore extremely important that we develop new medicines that can treat these diseases - before it is too late. One way to stop the dengue virus would be to disrupt the function of the NS2B-NS3 – no enzyme, no virus, no infection. Imagine a set of dominoes with the NS2B-NS3 being that initial push required for setting off the chain of events required for infection. In this study, a novel approach was evaluated when producing the NS2B-NS3 resulting in an increase of the enzyme activity, making it behave more similarly to how it would in the actual host organism. Apparently, a small tag added to the enzyme during the production process actually inhibited the activity of the enzyme and removal of that tag yielded a more active NS2B-NS3. But wait, wasn't the key to stopping the disease to *decrease* the activity of the NS2B-NS3? That is correct, but it is also very important to have a fully functional enzyme that is as similar as possible to the one found in nature to use in future experiments. These new findings could lead to better outcomes when studying new pharmaceuticals that interferes with the function of the NS2B-NS3. But the exploration doesn't stop there. Three naturally derived inhibitors were also studied; a protein from Quinoa, carvacrol from the aromatic oregano-herb that is commonly found on top of pizzas, and chlorogenic acid, a compound that the majority of the world ingest daily through their morning coffee. The compounds were first analyzed through computer simulations using 3D models, which showed that all three of these compounds seemed to interfere with the function of the NS2B-NS3. The oregano and coffee compounds where then tested in the lab to see if they had an impact on the activity of the dengue enzyme. The oregano-compound did not inhibit the enzyme; however, the coffee-compound did decrease the activity of the enzyme by 24%. Hopefully these results can be a steppingstone for continuing our pursuit towards preventing the spread of the dengue virus and aid us in our fight against these insidious invisible threats.