The Chase for the Perfect Vaccine

Abstract:

 As another flu season is gripping the world, the search for a more efficient vaccination is an extremely high priority. With many different strains of the influenza virus, paired with frequent genetic drift; crafting a perfect vaccination has posed a challenge to say the least. These viruses, as well as many others, have developed mechanisms to infect the host while eluding the immune systems fist line of defense. In order to be able to utilize these virus’ as an effectual attenuated vaccination, they must stimulate this response in order for the body to recognize the pathogen as well as create memory cells. A successful vaccination needs to meet a variety of different criteria including: high immunogenicity, low virulence, high attenuation, and protection against homologous and heterologous viral encounters. Du, Yushen, et al. have begun researching a new possible option for the influenza vaccination that focuses on identifying and incorporating certain mutations to the virus that greatly increase the capacity to stimulate an immune response. Previous attenuated vaccinations have low virulence and acceptable attenuation; but are still capable of avoiding the critical initial immune response.

Introduction:

When the influenza virus enters the host, it makes its way to the respiratory tract, using effective and adaptive mechanisms to avoid the bodies interferon expression (IFN); which serves as the bodies initial defense line against the introduction of a pathogen. By avoiding stimulation, the virus is given time to infect the host’s cell and replicate in vivo. The virus takes over the cell, replicates, and kills the cell releasing the virus to continue replication. It is not until the virus moves into the bloodstream that symptoms become visible, as the immune system finally identifies the pathogen and begins the immune response. Responsible for recognizing these pathogens are toll like receptors (TLRs) located on many immune cells, which identify certain patterns on the pathogens such as lipids or nucleic acids (Akira, Shizuo, et al.). Influenza, since using RNA, must contain nucleic acids that can be recognized by these TLRs. Once there is acknowledgement of a specific pattern the cell is activated, usually a dendritic cell (DC) or a macrophage by way of a TLR ligand (Akira, Shizuo, et al.). The triggering of this pathway directly induces the IFN, as well as leads to the production of cytokines, and the maturation of the dendritic cells. These DCs are very important as they obtain the antigen material and display it on the cells surface for the T cells to identify; bridging innate and adaptive immunity. These T cells, with knowledge of a specific antigen, will differentiate into various other immune assisting cells including memory cells, which are responsible for protection against subsequent, specific antigen exposure. Given the importance of TLR’s role in immune response, it serves as a very prominent target of viral immune hindrance. Disturbing the TLR will therefore suppress the IFN and delay immune response allowing viral replication. These viral mechanisms are considered IFN restricting modulators and eliminating these poses a possibility for future vaccinations (Du, Yushen, et al.).

Recent Progress:

Targeting these specific genetic segments that allowed IFN suppression, and mutating them with very IFN sensitive mutations was the goal of Du, Yushen, et al. In order to do this, they first developed an advanced system so they could sequence the entire influenza genome and identify the problematic modulators within the genome. They profiled the entire genome both with and without the use IFN selections, allowing them to locate these regulating modulators. They then overlapped these results with the same profile for viral replication; in hopes of discovering segments responsible for IFN suppression independent of viral replication. Once the desired modulators were chosen one of the seven plasmids, that together holds the influenza genome, was isolated for mutation. They then created 52 sub libraries of mutations on the desired modulators within this isolated plasmid. Through a process called cotransfection, the isolated plasmid (with a specific mutation) that was chosen of the 8-wild type (WT) plasmids, would be reintroduced with the other seven WT plasmids in the virus. Once reintroduced, all the sub libraries were tested for IFN sensitivity through the use of an exogenous IFN treatment; a treatment used to determine the actual sensitivity of the different mutations. The resulting mutations could either lead to an effected protein, have no/neutral effect, or terminate production of the protein. It is already known that the viral NS1 protein plays an important role for its interaction with the IFN pathway; so (the authors) chose to investigate mutations outside the NS1 protein. Of the tested mutations, several increased IFN sensitivity comparably to the WT virus. They then selected the 8 mutations that maximized IFN sensitivity and production, two of which were previously known mutations on the NS1 protein, and combined them into their new virus called the hyper interferon-sensitive (HIS) virus. This new, extremely IFN sensitive virus, is their potential new influenza vaccination.

To first assess their new virus, they compared the IFN sensitivity of HIS to the WT and as well as the previously known NS1 mutated virus. Each virus was placed in identical cells and tested for IFN sensitivity. The HIS virus showed much greater IFN sensitivity than both the WT and NS1 virus; while remarkably doubling the already know IFN sensitive NS1 mutations. With IFN sensitivity as high as hoped, next attenuation needed to be tested. HIS showed comparable replication to the WT virus in vitro, but when tested in mice and ferrets, the results showed much lower viral replication than the WT; at a similar level to current vaccination FluMist. This is sought in vaccinations as the virus used should flourish in the desired lab environment, but when introduced to the new host environment, should not excessively replicate and thrive; just provide enough exposure to elicit an immune response. Producing a significantly more IFN sensitive and attenuated virus was accomplished, the next factor to be assessed is how effective of an immune response does the HIS virus generate. Mice were used as samples and vaccinated with equal dosage of either HIS, WT, or FluMist and samples were collected 28 days later and then tested for specific antibody production. While the production of antibody hemagglutinin (HA) induced by HIS was less than the WT production, it was much greater than the HA antibody production of FluMist. When testing for immunoglobulin G (lgG), which is linked to restricting replication, HIS actually stimulated levels of production very close to that of the WT. After this 28-day frame post vaccination, the mice and ferrets were exposed to a WT virus of influenza, such to record the vaccines ability to protect against the exposure. Those vaccinated with HIS showed less viral replication, as well protected the host from the exposure; as shown upon no virus present in the lungs or nasal washes. The samples were then subjected to lethal doses of three differing strains of influenza, in which they showed much more resilience in survival, T cell memory response, as well as less weight loss.

Discussion:

These results by Du, Yushen, et al. yield a great potential to the advancement of not only the influenza vaccination, but of all live attenuated vaccines. With the capability of outlining the effects of certain mutations across the entire genome scientists can test these mutations under varying conditions in order to analyze these mutations in such condition. By removing certain modulators within the genome that allow for the viral capacity of IFN suppression, these vaccinations will produce that initial immune response that allows the host to build a memory to protect itself in the future. One problem found when attenuating virus’ though, is if mutated, what are the possible effects this could have on the rest of the virus when it replicates in the future. For instance, it could possibly make mutations in future replication more frequent. Another concern is what is the possibility that this live attenuated virus retains its virulence or mutates to increase replication. When it comes to replication, the HIS does not replicate once in vivo, while it does yield a greater immune response than previous vaccines, if the HIS could be programmed to replicate once and then terminate; this would allow for more antigen recognition and greater immune responses.

References

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