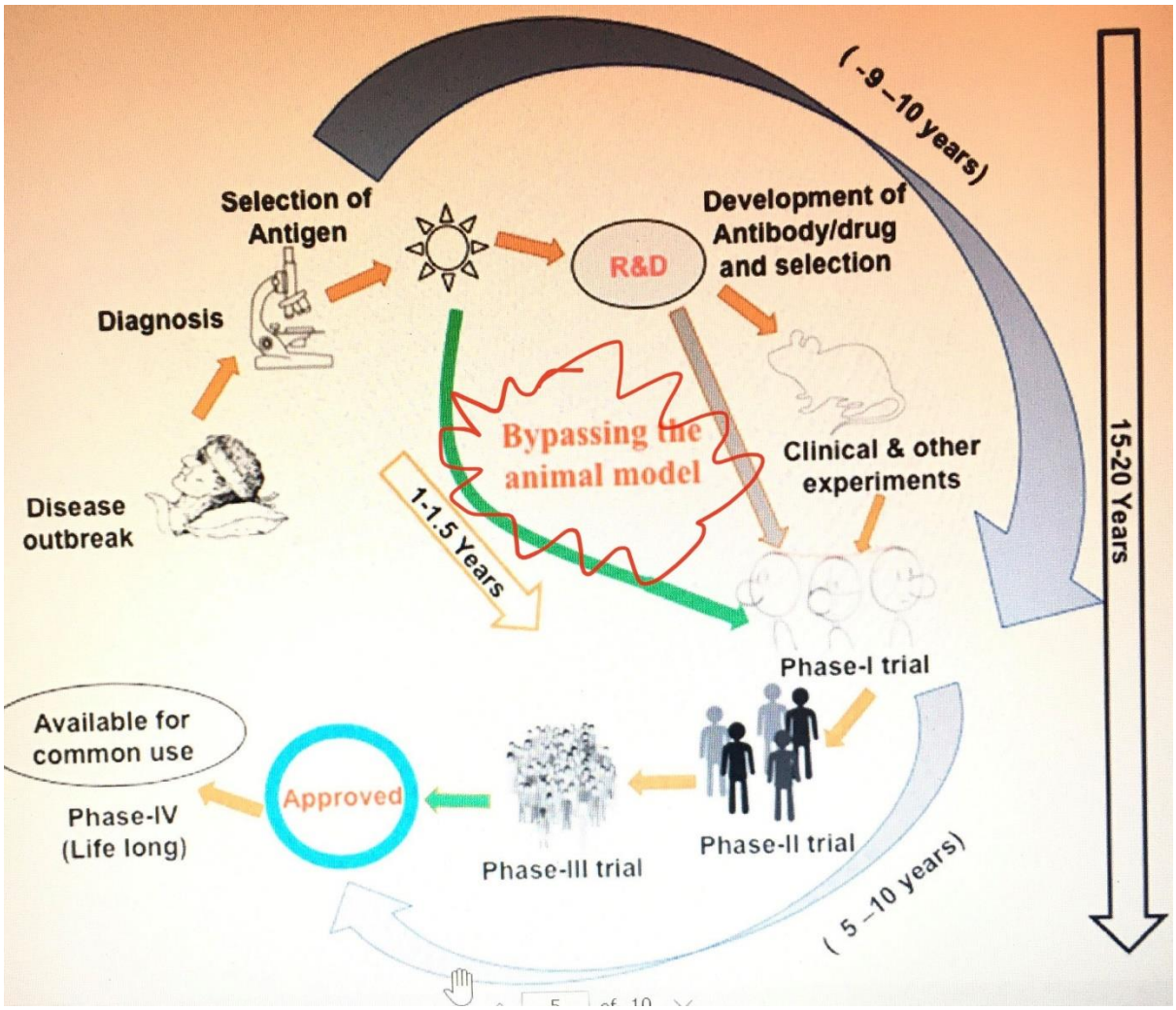
SARS-Cov-2 Vaccine and Bypassing Traditional Animal Trial Sequences and Timescales.

Published in January 2021.

As various vaccines for Covid-19 (SARS-CoV-2) are being released, many biotech companies have skipped traditional R&D animal studies and moved directly to Phase I human clinical trials. **"As various models for SARS-CoV-2 are under testing phase, biotech companies have bypassed animal studies and moved to Phase I clinical trials. In view of the present outbreak, this looks a justified approach, but the problem is that in the absence of animal studies, we can never predict the outcomes in humans".** (Deb et al. 2020).

Vaccine development typically takes at least 15–20 years and passes through six phases of assessment. (Deb et al 2012). See Diagram below from Deb et al (2020):



Not all biotech companies have completely bypassed all animal trials. The three vaccines released in the UK have utilised animal studies in their vaccine development, however they all deviated from the traditional vaccine-development lifecycle. Perhaps, initial plans to completely bypass animal studies raised concerns amongst the scientific community. Jonathan Kimmelman, director of McGill University's biomedical ethics unit stated in March 2020 that "Outbreaks and national emergencies often create pressure to suspend rights, standards and/or normal rules of ethical conduct. Often our decision to do so seems unwise in retrospect". (Boodman 2020).

Whilst animal experiments have been conducted with some of the vaccines, they have deviated from the traditional protocol. The diagram above shows that animal studies take place before human Phase I trials and that the whole vaccine development lifecycle can take 15-20 years. None of the vaccines released for emergency use in the UK (as of January 2021) have utilised the usual time-scales nor preceded their human trials with animal studies.

Pfizer and BioNTech vaccine released in the UK have published data on the success of their preclinical trials with animal studies on mice and macaque monkeys. However, rather than conducting these studies in the pre-human testing to ensure safety, they conducted these studies concurrently with phase I trials on humans. (Vogel et al. 2020). Furthermore, the whole vaccine development took less than one year.

AstraZeneca is another vaccine currently released for temporary emergency use in the UK. Whilst its animal studies on rhesus macaques took place before phase I trials and showed successful immunity against pneumonia, animal study times were a lot shorter than usual, (Doremalen et al. 2020). In the MHRA's (Medical and Healthcare Regulatory Agency's) recent advice document to health professionals administering the vaccine, they state "definitive animal studies have not been completed yet. The full relevance of animal studies to human risk with vaccines for COVID-19 remains to be established". Also "Non-clinical data reveal no special hazard for humans based on a conventional study of repeat dose toxicity. Animal studies into potential toxicity to reproduction and development have not yet been completed". (MHRA Reg 174).

The third and final vaccine available in the UK at this time (January 2021) is the Moderna vaccine. The animal studies also deviated from the traditional pre-human trial sequence of events and took place simultaneously with the human trials. A Moderna report claimed that the animal studies it conducted in developing the vaccine offers "full protection of mice, hamsters and non-human primates from SARS-CoV-2 and it does not lead to vaccine-associated enhanced respiratory disease", (Miller 2020). However, it should be noted that this information is not published on a peer review journal. Furthermore, it should be noted that the MHRA states "definitive animal studies have not been completed yet. The full relevance of animal studies to human risk with vaccines for COVID-19 remains to be established", (MHRA Reg 174).

In the absence of using traditional sequential animal studies with longer traditional timescales with the SARS-CoV-2 vaccine, it is important to examine studies on previous SARS vaccine animal trials that have followed the traditional timescales and sequences to look for clues for safety concerns, (Deb et al 2012).

Research dates as far back to 2002, when coronavirus outbreaks such as SARS first emerged. Studies showed that whilst some types of studies have been successful in the long-term, sometimes the vaccines actually made the subject worse. "For certain diseases, patients who have been previously infected by one strain of a virus and who are later infected by another strain can suffer outcomes that are worse than those infected only once". (Eroshenko et al 2020). SARS, aka 'Severe acute respiratory syndrome', emerged in China in 2002 and vaccines developed to combat the illness sometimes resulted in a worsening condition of the subject. (Tseng et al 2012). This event is called 'antibody-dependent enhancement' or 'antigen sin' and can be a common reaction. Whilst these SARS coronavirus vaccines initially helped with the disease and produced antibodies, the animals (ferret and primates) later developed hypersensitivity to the virus causing complications and more damage than the original infection. (Tseng et al 2002).

Thus, instead of protecting the subject, it also sometimes led to higher complications and death of the subject when the subject was reinfected with the original pathogen (Huisman et al. 2009). As a result, severe damage of the liver and the lungs led to death. (Zelleger et al 2020). Lung pathology was often characterized by diffuse alveolar damage caused by inflammation. Liver pathology also included inflammation from infection and includes steatosis, hepatitis and portal inflammation.

Antibody-dependent enhancement (ADE) has also been observed with the human MERS virus and with feline coronavirus (f-Cov). What is of particular concern is that the structure of the SARS-Cov-2 virus is of high risk for antigenic sin because it contains an S protein that exists in other coronaviruses such as SARS and MERS. The spike (S) protein on the surface of SARS-CoV is where the virus attaches to gain entry to the host cells. (Kassmy et al 2020). This is a good reason to

compare the animal studies of these other coronaviruses to SARS-Cov-2, because they all contain the (S) spike protein.

Animal trials are very important for vaccine trials as the vaccinated animals can be reinfected with the original pathogen to see if any reaction occurs. At a very minimum, in the absence of testing animals, individuals should satisfy themselves that the vaccine they are being encouraged or mandated to take, has been investigated for antibody-dependent enhancement (ADE) at multiple stages in its development. (Wang et al 2020). However, in terms of safety this does not compete with animal trials or long-term 10+ years clinical trials or indeed with long-term epidemiological studies.

Governments should not recommend the SARS-COV-2 vaccine because the evidence for safety has been for such a short period of observation. More time should be allowed for potential of immunopathological (ADE) reactions occurring among vaccinated individuals on exposure to infectious SARS-CoV-2. Tseng el al (2012) also recommended the same in relation to SARS-CoV. More time for observation is also recommended by other researchers. Eroshenko (et al 2020) reports, "ADE is often observed when antibody concentrations decrease as a result of waning immunity". Ideally waiting on immunity levels to drop in vaccinated individuals is recommended especially since ADE has already been reported with some SARS-CoV-2 vaccines. (Eroshenko et al 2020).

Vaccine-specific variations in ADE could occur for many reasons, including differences in vaccine adjuvants used and whether prior exposure to other CoV strains has occurred. Wang (et al 2020) therefore recommends further clinical studies to assess this risk in both vaccinated and infected individuals. Individuals and governments should consider this as such data may become even more critical as SARS-CoV-2 virus mutates or becomes seasonal.

The lives of billions of people are dependent upon the safety of SARS-Cov-2 vaccines. Therefore, scientists need more time to develop techniques in immunology to create vaccines that reduce or avoid ADE altogether. (Eroshenko et al 2020). The gold standard of safety for drug companies is double-blind placebo studies to occur over 5-10 years of clinical trials. The same should be mandated for vaccines, especially since they are injected directly into the circulatory system, as opposed to the digestive system as with pharmaceutical drugs. The digestive system provides a natural barrier to toxins and protects the organs. Eroshenko et al (2020) reports that epidemiological studies investigating ADE in individuals with multiple SARS-CoV-2 infections or cross-reactivity to common-cold-causing CoVs will likely take several years.

In summary, in the rush to release a vaccine, traditional sequences of events in vaccine development have been modified. Animal studies have occurred simultaneously with human trials and overall, the study timescales have been drastically reduced. Whilst there may be some data to approve safety and prove that immune responses do not occur when vaccinated subjects are reinfected. This cannot be confirmed until more time has passed. Even if vaccine manufacturers have taken steps along the way to test and mitigate for Antibody Dependent Enhancement (ADE), more time is still needed to evaluate properly. This is especially true because as Eroshenka et al (2020) reports, it is with the decreasing immunity levels that ADE often occurs. Vaccines have just begun to be released and immunity levels with the vaccinated against SARS-CoV-2 are at their highest. At the very least, governments and individuals should wait at least 3-5 years to determine potential ADE when immunity levels drop and reinfection occurs with new strains. It is therefore premature to recommend the vaccine to anyone on these grounds alone. Further, vulnerable individuals wishing to take the vaccine should be made aware of the potential long-term consequences.

References

Deb BIJAYEETA, HEMAL SHAH and SUCHI GOEL (2020)45:82 **"Global vaccine and drug efforts against COVID-19: Pros and cons of bypassing animal trials"** *Indian Academy of Sciences.* Indian Institute of Science Education and Research (IISER)

Boodman, Eric (2020) **"Researchers rush to test coronavirus vaccine in people without knowing how well it works in animals".** 11th March 2020. *STAT News.* https://www.statnews.com/2020/03/11/researchers-rush-to-start-moderna-coronavirus-vaccine-trial-without-usual-animal-testing/

Doremalen Neeltje Van et al. (2020) **"ChAdOx1 nCoV-19 vaccine prevents SARS-CoV-2 pneumonia in rhesus macaques"** *Nature.* 586, pages578–582(2020) https://www.nature.com/articles/s41586-020-2608-y

Eroshenko Nikolai, Taylor Gill, Marianna K. Keaveney, George M. Church, Jose M. Trevejo & Hannu Rajaniem (2020:38), pages789–791 **"Implications of antibody-dependent enhancement of infection for SARS-CoV-2 countermeasures"** *Nature Biotechnology*

Kassmy Jawad Al-, Jannie Pedersen, Gary Kobinger Vaccine (2020, *12*(8), 861) **"Candidates against Coronavirus Infections. Where Does COVID-19 Stand?"** *Viruses* https://doi.org/10.3390/v12080861

Miller Jacqueline (2020) PowerPoint Presentation **"Overview of Moderna's COVID-19 Vaccine (mRNA-1273)".** https://www.cdc.gov/vaccines/acip/meetings/downloads/slides-2020-12/slides-12-19/02-COVID-Miller.pdf

MHRA REG 174 **"COVID-19 Vaccine AstraZeneca - INFORMATION FOR UK HEALTHCARE PROFESSIONALS"** https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/951851/uk-clean-spc-covid-19-vaccine-astrazeneca-reg174.pdf

MHRA REG 174 **"COVID-19 Vaccine Moderna- INFORMATION FOR UK HEALTHCARE PROFESSIONALS"** https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/950661/Moderna_Information_HCP.pdf

MHRA REG 174 **"COVID-19 mRNA Vaccine BNT162b2-INFORMATION FOR UK HEALTHCARE PROFESSIONALS" Pfizer and BioNTech**
https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/948517/Information_for_Healthcare_Professionals_on_Pfizer_BioNTech.pdf

Tseng Chien-Te, Elena Sbrana, Naoko Iwata-Yoshikawa, Patrick C. Newman, Tania Garron, Robert L. Atmar, Clarence J. Peters, Robert B. Couch (2012. April) "**Immunization with SARS Coronavirus Vaccines Leads to Pulmonary Immunopathology on Challenge with the SARS Virus"** *Plus One* https://doi.org/10.1371/journal.pone.0035421

Vogel et al (2020) **"A prefusion SARS-CoV-2 spike RNA vaccine is highly immunogenic and 2 prevents lung infection in non-human primates"** *BioRxiv preprint.* https://www.biorxiv.org/content/10.1101/2020.09.08.280818v1.full.pdf

Wang Jiong and Zand Martin (2020) 10.1017 **"The potential for antibody-dependent enhancement of SARS-CoV-2 infection: Translational implications for vaccine development".** *Journal of Clinical and Translational Science*.

Zellweger Raphaël M, T. Anh Wartel, Florian Marks, Manki Song & Jerome H. Kim. (2020)19(8) 691-698 **"Vaccination against SARS-CoV-2 and disease enhancement – knowns and unknowns".** *Expert Review of Vaccines.* https://doi.org/10.1080/14760584.2020.1800463