ON THE MUI'S VIEW OF "TRYPSIN ORIGIN" IN THE ASTRAZENECA'S COVID-19 VACCINE

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Let me start with a rather perplexing statement. When I read the MUI reports on findings on the use of trypsin of porcine origins in the AstraZeneca COVID-19 vaccine, I admit there is some confusion on my part when the MUI insisted that the vaccine contained porcine trypsin while AstraZeneca affirmed that their vaccine contained "no material of human or animal origin are used in the growth medium or feed (including no materials manufactured with animal-derived material)" (COVID-19 Vaccine Astra Zeneca Public Assessment Report, pg. 17.)

So, I went and did some research of my own. I read the COVID-19 Vaccine Astra Zeneca Public Assessment Report[1] submitted to the European Medicines Agency (EMA), which is also one of the documents referenced by the MUI in its own report. Unfortunately, I couldn't find the WHO dossier that was sent to BPOM (also referenced in the MUI report). Curiously, I found no reference and no mention of "trypsin", "porcine trypsin" or any of the tables the MUI report referenced in the document submitted to EMA (Table 2 and Table 3).

However, I did find a reference on how the vaccine was made and harvested from the host. AstraZeneca claimed that the harvesting process (page 16) involves a detergent-based cell lysis, which is just a fancy way of saying harvesting the cell's contents by destroying the cell membrane, then treating the cell's contents with nucleases (enzymes that degrade the host cell's DNA, but not the DNA that will be used as the vaccine) then clarified using depth filtration and then downstream processing to further purify and concentrate the vaccine. Here, Trypsin was not mentioned at all. Perhaps because the harvest process described previously negates the use of trypsin that is usually needed to detach cells from the petri dish. And if trypsin was used at all, it is in the process of passaging and maintaining the cell line they used to produce the virus seed.

Next, I looked up the use of T-REx™-293 Cell Line, which is the cell line used to produce this recombinant adenovirus[2] .This virus is a genetically modified version of a cell line that we use often in laboratories to produce proteins or expression vectors due to its efficiency in producing them. This T-REx™-293 cell line is propagated using normal culture media and under normal culture conditions. One of the necessary items to maintain this culture is Trypsin which is required to detach cells from the petri dish if the researcher wants to move the cells to a bigger dish or harvest the cells to do some experiments on them.

On the manufacturer's protocol, Trypsin-EDTA was listed as "Required item not supplied". But the manufacturers wrote it as "Trypsin EDTA or other trypsin solution", which means the user would have freedom of choice as to which type of Trypsin they utilize for this process, be it one sourced from an animal such as porcine trypsin or the synthetic version. The screenshot provided by MUI's report is of the "Related Materials" section of the protocol, located at the end of the

product manual which only listed Trypsin-EDTA[3]. This seems misleading to me as it gives the impression that this cell line can only be maintained using Trypsin-EDTA, while in reality Trypsin-EDTA or other trypsin solution would suffice. Since AstraZeneca never specified what type of Trypsin they use to maintain the T-REx™-293 cell line, **one cannot firmly say that what they used was Trypsin-EDTA (the trypsin variant of porcine origin)** and not one of the many trypsin replacement enzymes that have been developed and are free of animal components.

Biologics nowadays are required to be manufactured in accordance to current good manufacturing practices (cGMP)[4]. One of the tenets of cGMP is that materials that are destined for use in the human body (i.e drugs, vaccines, saline solution, vitamins) need to be sterile and free of animal components (xeno-free) or components derived from another species to avoid any adverse reactions by the immune system. As such, there have been many xeno-free reagents or synthetic reagents that have been developed to support this cGMP requirement to replace the conventional use of animal-derived reagents and enzymes. Reagents with animal components in them are usually only used during the research stage (because they're cheaper) and would not be used during any of the manufacturing process.

Even someone like me, who is a novice drug developer, know to avoid any reagents that have animal components during the manufacturing process, and if need be during the initial process development steps. Therefore, I'm quite sure that a multibillion-dollar company like AstraZeneca would know better than to use reagents containing animal products in their final manufacturing process. Or if they do, the initial step of trypsin usage is very far from the final product that has went several rounds of filtration and purification that the impurities are filtered out of the final product that is ready to be injected to the patients.

Drawing from the evidence gleaned so far, I remain unconvinced by MUI's claims that trypsin of a porcine origin was used in the manufacturing process. Perhaps more data will be forthcoming in the upcoming days, but for now I remain unconvinced as I'm unable to verify MUI's assertion using the submitted dossier to the EMA.

Footnotes:

1. https://www.ema.europa.eu/en/documents/assessment-report/covid-19-vaccine-astrazeneca-epar-public-assessment-report_en.pdf
2. https://www.thermofisher.com/order/catalog/product/R71007#/R71007
3. https://www.thermofisher.com/document-connect/document-connect.html?url=https%3A%2F%2Fassets.thermofisher.com%2FTFS-Assets%2FLSG%2Fmanuals%2Ftrexcells_man.pdf&title=VXNlciBHdWlkZTogR3Jvd3RoIGFuZCBNYWludGVuYW5jZSBvZiBULVJFeCBDZWxsIExpbmVz
4. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5609845/