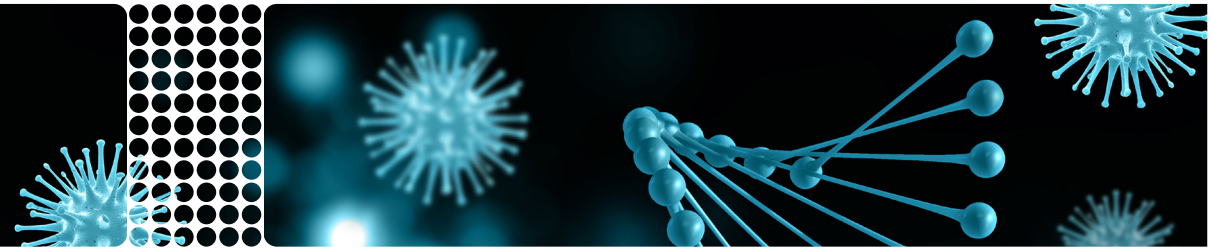

Case study

Improving Influenza virus production in HEK293 cell culture supplemented with animal-free insulin

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Background

Cell-culture based vaccines are a valuable alternative to egg-produced vaccines. HEK293SF-3F6 is a suspension cell line used to produce Influenza virus, in flasks and large bioreactors. HEK293SF-3F6 cells grow in commercially-available serum-free media but show weak growth profiles. Insulin has been proved to positively affect cell survival, by promoting anti-apoptotic and mitogenic pathways, and to interact with cellular signalling pathways exploited by the influenza virus.

This study addresses the effect of recombinant insulin supplementation on HEK293SF-3F6 cell growth and Influenza virus production.

Study Description

Different doses of insulin were supplemented to HEK293SF-3F6 cells grown in chemically-defined serum-free media and viable cell density was measured. Influenza virus production was measured as HA concentration, following HEK293SF-3F6 transduction with Influenza virus. Phosphorylated Akt was measured as indication of Akt pathway activation in response to insulin addition.

Results

Addition of 10-20mg/L of insulin every 72hrs resulted in up to 4-fold increase of viable cell density of HEK293SF-3F6 cells (fig. 1a). Addition of 25-100 mg/L insulin at time of infection increases the yield of influenza (H1N1 A/Puerto Rico 8/34) virus by almost 2-fold (fig. 1b, left panel), without affecting the total cell count at harvest (fig. 1b, right panel). Addition of insulin to HEK293SF-3F6 cells transfected with influenza virus showed increased Akt activity (fig. 1c).

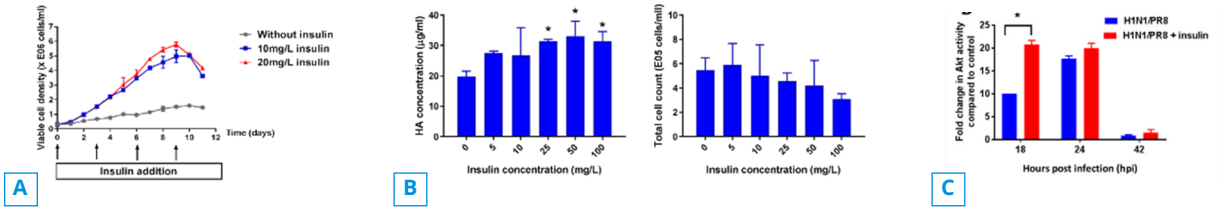

Figure 1: Effects of insulin supplementation to HEK293SF-3F6 cells. Viable cell density following supplementation of 10 and 20 mg/L insulin (A). Concentration of influena virus (left panel) and total cell number (right panel) at different insulin concentrations 5-200 mg/L (B). Akt acitivity measured in response to Influenza virus transfection with or without insulin supplementation (C).

Conclusion

- Growth limitations observed in a chemically-defined media are alleviated by insulin.
- Insulin increases the yield of influenza virus in a 24-well microbioreactor.
- Insulin is a strong activator of the PI3K/Akt pathway, which plays a key role in influenza production.

Further information

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