3D PRINTING TECHNOLOGY: SUPPLY CHAIN INDEPENDENT SINGLE-USE PLASTIC WARE AND BIOREACTORS FOR CELL CULTURE

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Key Words: Supply Chain, 3D Printing, Cell Culture, Printed Bioreactor, Sustainable

The worldwide supply chain issues for single-use (SU) shaking flasks prompted us to develop 3D printed counterparts to maintain common cell types in cell culture (HEK, CHO and Insect cells) intended for biologics production, including the production of viruses and virus like particles. We investigated different commercially available low-cost materials and their compatibility for cell culture. We identified poly lactic acid (PLA) as perfect candidate for 3D printed parts for cell culture applications and expanded the manufacture of shaking flasks to the manufacture of a 3D printed bioreactor vessel. As this vessel differs slightly in geometry and material from their respective commercial glass-vessel equivalent, k_La measurements and a temperature sensitivity study were performed to confirm sufficient oxygen supply and maintaining the process temperatures. For both process conditions used for mammalian and insect cell culture results are comparable between the 3D printed PLA and the commercially available glass vessel (***Figure 1***). Small 3D printed PLA parts, such as shaking flasks[1], can be sterilized by autoclaving and remain structurally stable. Despite extensive print file optimization, the PLA bioreactor vessel was too brittle after autoclaving. Alternatively, we tested the sterile printing performance of thermoplastic materials, as the printing process itself heats the material well beyond any point of survivability of contaminating organisms (~200°C). Moreover, the power consumption of sterile printing within a sterile work hood is also substantially lower than traditional autoclaving and no water is used. While 3D printed SU plasticware is unlikely to be universally adopted as a full replacement, especially not in GMP environment, it is a perfect solution to replace even reusable glass-ware from a sustainability standpoint, while additionally offering freedom of geometric design. It is also a welcome alternative whenever other options are not available, and an economic option for low-income countries, given the sharp decrease in prices of 3D printers in recent years (<500 € equivalent to 530 $) and low-costs of PLA filament (~15 €/kg equivalent to ~33 $/lb).

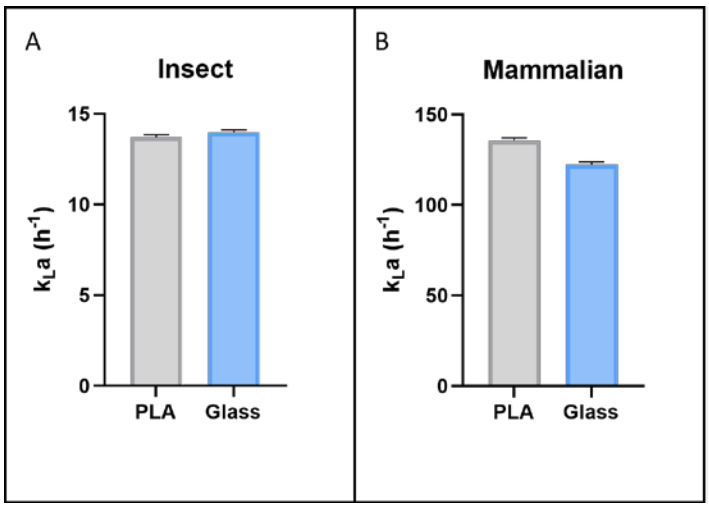

Figure 1: A) k_La measurements for 0.3 SLPM B) k_La measurements for 3 SLPM; In gray the 3D printed PLA vessel, in blue the commercially available glass vessel. Standard deviation (n=3)

1 Satzer, P. & Achleitner, L. 3D printing: Economical and supply chain independent single-use plasticware for cell culture. *N. Biotechnol.* **69**, 55–61 (2022).