A Rapid Method for Bioburden Testing in Live Attenuated Influenza Vaccine Intermediates – From Concept to Implementation

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Bioburden levels are routinely evaluated throughout the viral vaccine manufacturing process. A rapid method for monitoring bioburden in in-process influenza vaccine material has been developed through a collaborative effort between MedImmune Vaccines, Inc. (Mountain View, CA) and Advanced Analytical (Ames, IA). The live, attenuated influenza virus vaccine material is produced by inoculating fertile, specific pathogen-free (SPF) chicken eggs. Following incubation and propagation of the virus in the SPF eggs, the allantoic fluids are harvested, which have natural potential for harboring microbial contamination. One-liter sub-lots of allantoic fluids are collected and sequestered until bioburden test results are obtained, at which time the sub-lots with satisfactory bioburden results are pooled and further processed. The sub-lot quarantine and delay in bioburden results prevents a continuous process, thus warranting a rapid bioburden detection method. Initial feasibility and proof-of-concept were successfully completed on the *Micro PRO*™ detection system using allantoic fluids spiked with *Staphylococcus aureus* and *E. coli*. Subsequently, a novel pre-treatment procedure was developed to significantly reduce background contribution from cellular debris and other allantoic fluid components. This collaboration has yielded a procedure for rapid screening of bioburden levels in in-process egg-based vaccine material.