Cell Culture Performance and Impurity Levels in Poloxamer 188

Kevin Patrick Kent, Scott Wilson, Sarah Trout, Andy Nikolas, Barry Drew and Chandana Sharma
SAFC, Cell Sciences and Development, 13804 W 107th St, Lenexa, KS 66062 USA

Abstract
Poloxamer 188 is used as a shear protectant in high shear stress cell culture media applications. Poor cell growth and viability for particular lots of poloxamer 188 was observed by several SAFC customers. A high shear stress cell based biological assay and analytical method that distinguish between good and poor performing lots of poloxamer 188 were developed by SAFC. Correlation between the two assays suggests that impurity levels observed in poloxamer 188 may decrease the performance of poloxamer 188 as a shear protectant.

Introduction
Poloxamer 188 is used in the biopharmaceutical industry as a shear protectant where sparging with gases or rapid agitation is required. The addition of poloxamer 188 to liquid media increases mammalian cell yield in agitated cultures, possibly due to association of poloxamer 188 with the cell membrane. SAFC customers reported poor cell growth associated with particular lots of poloxamer 188, which initiated investigations at SAFC into determining the difference between good and poor performing lots of poloxamer 188. Several good and poor performing lots of poloxamer 188 were verified to be within SAFC specifications (NF Grade). A cell based assay was developed to mimic the problems SAFC customers observed. In addition, an analytical method that distinguishes between the good and poor performing lots of poloxamer 188 were developed.

Analytical Method
Good and poor performing samples of poloxamer 188 were analyzed by UPLC-MS. In the poor performing samples of poloxamer 188 a more hydrophobic polymeric impurity was observed. The impurity differs from lot to lot and was identified as a polymer containing monomers similar to poloxamer 188.

Correlation Between Analytical and Biological
The correlation between the analytical and biological data is shown in Figure 4. The \( r^2 \) value suggests that only 60% of the variability in performance across poloxamer samples is captured by the impurity levels observed in the analytical method. In the inset of Figure 2 three distributor lots of the same manufacturer lot are shown with different levels of the impurity but the same retention time. Thus, the impurity in the three samples shown in Figure 2 inset is expected to be the same impurity. The inset in Figure 4 shows that the correlation coefficient for matched impurities is much higher, suggesting that much of the variability in the correlation comes from the structure of the impurity.

Conclusion
Poor performing lots of poloxamer 188 were associated with low viable cell densities in SAFC customer’s processes. A biological assay and an analytical method that distinguish between poor and good performing lots of poloxamer 188 was developed at SAFC. The correlation between the impurity levels and performance is low for all samples, but for expected matched impurity samples the correlation is high. This data suggests that the root cause for the poor performance associated with poloxamer 188 lots is a polymeric impurity composed partially of ethylene oxide and propylene oxide monomers.