Spring 5-9-2016

Engineering, expression screening, and production cell line development of hetero Ig molecules using charge pair mutations

Jennitte Stevens
Amgen

Guna Kannan
Amgen

Trent Munro
Amgen

ENGINEERING, EXPRESSION SCREENING, AND PRODUCTION CELL LINE DEVELOPMENT OF HETERO IG MOLECULES USING CHARGE PAIR MUTATIONS

Jennitte Stevens[1], Guna Kannan[1], Trent Munro[2]

[1]Biologics Optimization and [2]Process Development, Amgen, Thousand Oaks

In recent years, there has been an increase in therapeutic indications that require bispecific targeting. Bispecific Hetero Ig antibodies that can target two antigens have long been considered as an attractive approach to drive synergistic biologic activity while maintaining the structure and stability of a traditional antibody. However, clinical development of such molecules has been hampered by CMC related challenges relating to product heterogeneity. During the development of a Hetero Ig molecule targeting the Wnt pathway antagonists Dkk-1 and SCL-1, we employed a novel strategy to drive the heterodimerization of IgG antibodies through the addition of charge pair reside mutations (CPM) at both the heavy chain and light chain surface interface. Through electrostatic interactions, these CPMs drive appropriate chain pairing, while minimizing undesired side products. During engineering and expression in transient expression systems, we identified combinations of single residue pair mutations that promoted correct chain pairing. However, the combination of antibody pairs and expression balance is important to enable reduction in undesired side products. These findings extend to stable cell line development, where vector design and appropriate analytics enable the identification of pools and then clones with desired product quality. We have expanded this strategy for the development of a platform approach toward the efficient development of HeteroIg molecules.