The development and validation of QC analytical techniques for the study of peptides and proteins.

Case Study – Development of peptide immunogenicity for detection of anti-drug antibodies in rat serum.

David Jardine, Dr. Penny Davey, Laura Kelman and Niels Donnelly
Tepnel Pharma Services, Hologic Ltd., Livingston, UK

History

Our client was developing a novel drug product and required full development and validation of a peptide immunogenicity ELISA for the detection of anti-drug antibodies in samples generated in vivo toxicity studies. The development included establishment of assay format and optimisation, evaluation of precision and dilution linearity, determination of assay cut-off, and the effects of sample matrix. Benefits of the method included the demonstration of specificity together with selectivity.

The development was completed within a design protocol to build quality and reliability into the development activities which included the following goals:

• Detection of positive control rabbit anti-sera
• Optimisation of peptide, positive control and conjugate
• Assay cut-off value determination

The method was used for validation and in vivo toxicology GLP studies.

Benefits of development

• Intra-batch and inter-batch precision <11%CV values were obtained indicating a robust and reproducible assay
• Rat negative serum does not interfere with assay, % recovery 88 to 110%
• Increased temperature incubation results in decreased negative OD values by 10%
• Assay specificity achieved, no cross reactivity observed with scrambled peptides
• Method validated and used for sample analysis in in vivo toxicity GLP studies

Typical ELISA plate read at 450nm with reference filter 630nm

Typical ELISA plate read at 490nm with reference filter 630nm