Use of an *ex vivo* granulocyte shape change assay to determine pharmacokinetic-pharmacodynamic relationship for CAM-3001, a new antibody therapy in development for the treatment of rheumatoid arthritis

Suzanne Cohen[1], Matthew McCourt[1], Phillip Monk[3], Tim Meyers[1], Emma Appleton[1], Bing Wang[4], Cathy Drinkwater[2], Andrew Nash[2], Ian Anderson[1], Matthew Sleeman[1]. [1]MedImmune Ltd, Cambridge, United Kingdom, [2]CSL, Melbourne, Australia, [3]Synairgen, Southampton, United Kingdom, [4]MedImmune, Hayward, United States.

CAM-3001, a monoclonal antibody to the GM-CSFRα chain, is currently in clinical trials for the treatment of rheumatoid arthritis (RA). Using granulocytes purified from peripheral blood, we demonstrate that CAM-3001 is equipotent against human and cynomolgus monkey (cyno) GM-CSFR. Following this, we determined pharmacokinetic (PK) and pharmacodynamic (PD) profile of CAM-3001 following a single 10mg/kg iv dose in this species. Here we describe the relationship between levels of free CAM-3001 and functional blockade of GM-CSFR in vivo.

2 male and 2 female adult cynomolgus monkeys were dosed iv with 10mg/kg CAM-3001. Samples for PK analysis were taken pre-dose and then at 1 hour and on days 2, 3, 5, 9, 15, 22, 29 and 36 post dosing. CAM-3001 levels in serum samples were measured by antigen capture ELISA. Samples for PD analysis were collected pre-dose, 1 hour and days 3, 9, 15, 22 and 36 after dosing. CAM-3001 receptor blockade on neutrophils and eosinophils was determined by changes in forward scatter by flow cytometry (shape change) after 1-4 hours GM-CSF stimulation of purified granulocytes (in vitro) or erythrocyte depleted blood samples (ex vivo).

Using purified granulocytes, Schild analyses were performed for human and cyno shape change assays. Using this technique, preliminary data showed that CAM-3001 had a pA2 of 10.6 (slope=0.96) and 10.8 (slope=1.1), which equate to apparent affinities of 27pM and 16pM for human and cyno GM-CSFR respectively. Using a whole blood assay, a significant shape change (67%) in response to GM-CSF stimulation ex vivo was observed in the eosinophil population. Following single 10mg/kg intravenous infusion of CAM-3001, responsiveness to GM-CSF but not eotaxin was lost. Lack of response to GM-CSF was apparent until Day 22, when 3 of 4 animals re-gained GM-CSF dependent shape change activity. The 4th animal did not regain responsiveness until Day 29. This functional GM-CSFR blockade correlated with serum concentrations of CAM-3001 and suggested that a loss of effective blockade occurred between serum concentrations of 12-71 mcg/mL. The terminal half-life of CAM-3001 was estimated as approximately 5.6 days.

CAM-3001 has similar affinity and functional activity at the human and cyno GM-CSF receptors. Therefore, cyno is a suitable species for pharmacology and toxicology studies. The PK profile of CAM-3001 was similar to expected for a human monoclonal antibody in cyno. In addition, CAM-3001 demonstrated functional blockade of the GM-CSFR after in vivo administration that correlated with serum concentrations. The results from this study have been used to help determine the PK/PD relationship of CAM-3001 and predict effective dosing for clinical trials.