

## Peripheral blood leukocyte (PBL) analysis by flow cytometry

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Non-fasted mice were terminally anaesthetised and blood was collected into anticoagulant coated tubes via the retro-orbital sinus. Whole blood or the extracted buffy coat was stained with titrated cocktails of antibodies prior to analysis on a flow cytometer (BD LSRII or Beckman Coulter FC-500). All analysis was performed using FlowJo software.

The assay was performed at either 14 or 16 weeks of age, dependent upon the pipeline, with either Avertin or Ketamine/Xylazine as the anaesthetic. The diet the mice were on was pipeline dependant.