FcRn/antibody interactions
An Immunitrack/GE collaboration

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Imagination at work
Biacore™ T200 assay conditions for studies of FcRn antibody interactions

Fig 1. Assay principle. Using Biotin CAP Kit (GE Healthcare) a streptavidin conjugate is first bound to the chip followed by capture of biotinylated FcRn and binding of the antibody. At the end of the analysis cycle the surface is regenerated and ready for next determination. Running buffer: 20 mM phosphate, 150 mM NaCl, 0.05 % Surfactant P20, pH 6.0. The Biotin CAP kit contains Sensor Chip CAP, streptavidin conjugate and regeneration solution.
Antibody binding profiles for different species of FcRn at pH 6.0

Fig 2. Examples showing binding profiles for different types of antibodies to human, macaque and mouse FcRn. Binding behavior towards human and macaque FcRn was very similar. All antibodies showed highest binding towards mouse FcRn. Binding specificities for all antibodies were in agreement with published data*. Biotin CAP Kit (GE Healthcare) was used with FcRn capture levels 90-100 RU and antibody concentration series: 25, 74, 222, 666 and 2000 nM. Running buffer: 20 mM phosphate, 150 mM NaCl, 0.05 % Surfactant P20, pH 6.0.

*Abdiche, Y. N. et al. 2014, mAbs, 7, 331-338.
pH dependent binding of antibodies to FcRn facilitated using Dual inject

Fig 3. pH dependent binding of infliximab to different FcRn species using BiotinCAP Kit and Dual/Co-inject. Dual/Co-inject injects two solutions in immediate sequence. Infliximab was diluted into respective pH buffer and injected (Solution 1) followed directly by a second injection containing buffer of the same pH (Solution 2). Biotin CAP Kit (GE Healthcare) was used with FcRn capture levels 90-100 RU and infliximab concentration: 100 nM. Running buffer: 20 mM phosphate, 150 mM NaCl, 0.05 % Surfactant P20, pH 6.0.

Results showed almost identical infliximab binding behavior towards human and macaque FcRn, while the binding to mouse FcRn was significantly higher for all pHs. As expected there was no binding at physiological pH.