Simple and rapid analysis of tocilizumab using HPLC - fluorescence detection method

Abstract

We developed a novel assay using high-performance liquid chromatography (HPLC) with fluorescence detection for the determination of tocilizumab (TCZ), after it has undergone a facile and rapid pretreatment. TCZ belongs to the same subclass as IgG1 (Immunoglobulin G subclass 1), and we could separate TCZ from IgG1 without antigen–antibody reactions, with the novel detection method. The separation of these antibodies was achieved by pretreatment with an organic solvent containing a base, such as trimethylamine and triethylamine. The effect of these bases on the separation of TCZ is related to the hydrophobicity of the base rather than the electrostatic charge. The results indicated that the surface charge of antibodies changed because of the structural change, even though the difference in the amino acid sequences of the antibodies was very low. Our method is available for the separation of the antibody subclasses, and it would be useful to assay TCZ in blood.