ORT_19 - Immunoglobulin G micro purification using TRIM21 coated microplates

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Introduction: Antibodies are molecules extensively used in *in vitro* diagnostic and therapeutic applications. However, identifying new antibodies requires cost-effective and large-scale screening of binding molecules. Therefore, the development of improved microscale purification techniques may assist the rapid screening of molecules allowing the discovery of new and improved antibodies for diagnostic and therapeutic purposes TRIM21 is a soluble IgG-binding protein that has been shown to detect IgG from several mammalian species. In addition, two histidine residues mediate the molecular interaction between TRIM21 and IgGs. These findings suggest that the use of imidazole-containing buffers may disrupt the interaction between these molecules, generating a neutral pH-based elution method for IgG purification. Therefore, we investigated using TRIM21 immobilized into polystyrene plates to capture and purify IgGs on a microscale.

Objective: This project aimed to investigate the ability of TRIM21 to capture and purify IgGs from serum based on neutral pH conditions.

Methodology: TRIM21 was coated into polystyrene plates, followed by incubation with serum, removing nonbinding species, washing with PBS-based buffers, and elution with increasing imidazole concentrations. Eluted fractions were evaluated by ELISA and dot blots using anti-human, anti-rabbit, anti-horse, anti-mouse, anti-sheep, anti-cattle secondary antibodies.

Results: The use of 0.5M imidazole buffer allowed purification of IgGs from different species validating this method as a promising tool for antibody purification.

Conclusion: The promising results prove the principle of plate-based chromatographic applications—a good alternative for developing purification processes.

Keywords: Immunoglobulin G; Affinity chromatography