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Binding properties of Staphylococcal protein A (SpA) to purified immunoglobulins tested by double immunodiffusion (Ouchterlony) technique.

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Protocol status: Working

We use this protocol and it's working

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Keywords: staphylococcal protein, purified immunoglobulin, immunoglobulin, double immunodiffusion, protein

- 1 The binding of SpA to purified IgG are investigated by double immunodiffusion.
- 2 Briefly, 1% agarose gels are prepared and wells cut into the gel using a template.
- 3 Initially, aliquots of 25 μ l each of SpA diluted 1:5 are applied to the centre well.
- 4 The peripheral wells are filled with 25 μ l each of purified IgG (1 μ g/ μ l) in PBS pH 7.4.
- 5 The gels are incubated at RT for 48–72 hours.
- 6 After that the gels are examined for precipitin lines.
- 7 Human IgG are included as positive control and turtle serum as a negative control.
- 8 The positive results are taken as the presence of precipitin line/s and negative results, the absence of precipitin lines.