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Purification of anti-anti-SpA antibody (Ab-2) using SpA-bearing Staphylococcus aureus cells.

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Protocol status: Working
We use this protocol and it's working

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Disclaimer

Abstract

A novel method was designed for the purification of Ab-2 from undiluted WSF of hyperimmune eggs. A total of six preparations were made.

Materials

MATERIALS

- Centrifuge 5424 R refrigerated with Rotor FA-45-24-11 rotary knobs 120 V/50–60 Hz (US) **Eppendorf Catalog #**5404000537
- Eppendorf Safe-Lock Tubes 1.5 mL PCR clean colorless 500 tubes **Eppendorf Catalog #**022363212
- SpA-bearing Staphylococcus aureus cells.

Troubleshooting

1 Mix in an Eppendorf microtube 0.9 ml of undiluted WSF from immunized birds with 25 µl of SpA-bearing Staphylococcus aureus cells (Sigma-Aldrich).

2 Incubate the microtube at 37^{O}C for 30 min.

3 After the incubation period, centrifuge the microtube in an Eppendorf 5424 centrifuge for 5 min.

4 Observe the microtube that presents a pellet of Ab-1 bound-cells at the bottom ,and the supernatant, where Ab-2 is present.

5 Decant the Ab-2 containing supernatant to another tube.

6 . Store it at -20^{O}C until further use.