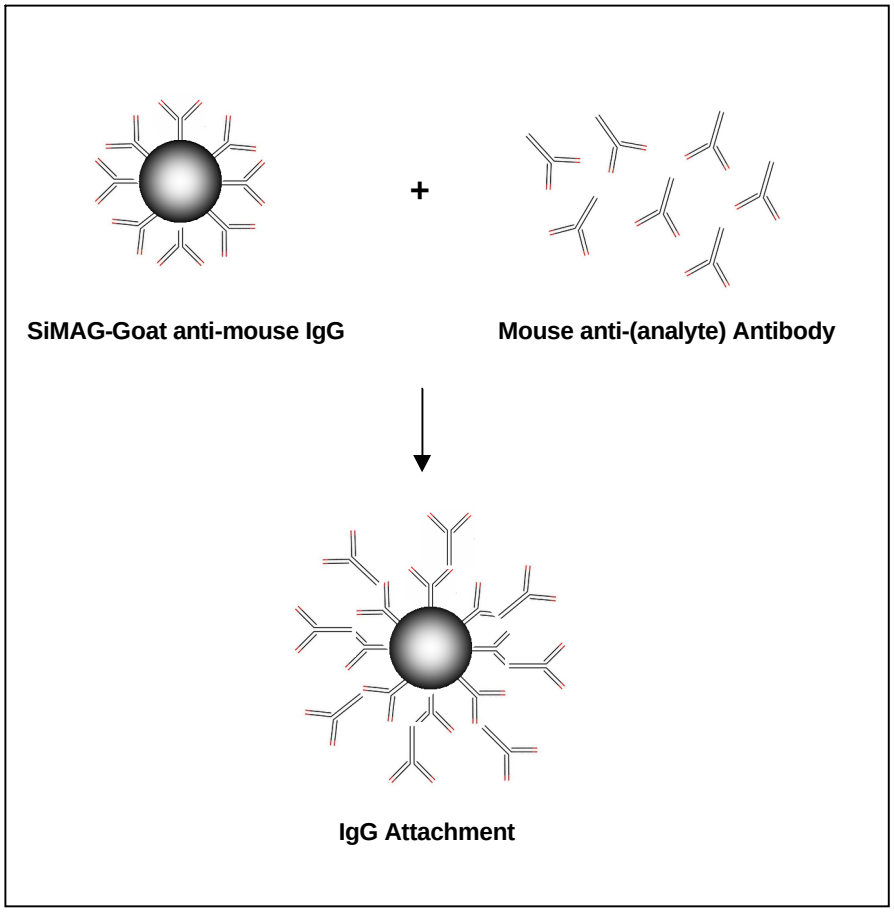
Protocol for binding of an antibody to SiMAG-Goat anti-mouse IgG

Introduction:

SiMAG-Goat anti-mouse IgG is designed as a matrix for immunomagnetic separation and purification of mouse IgG's.

The goat anti-mouse IgG's are covalently coupled to the magnetic silica particles and can be used for an efficient method for the attachment of an analyte and are applicable for cell separation from a heterogeneous cell suspension.

SiMAG-Goat anti-mouse IgG is suitable as a second antibody in radio- and enzyme immunoassays which utilize a mouse IgG primary monoclonal antibody.



Equipment & Reagents:

- **SiMAG-Goat anti-mouse IgG** (10 mg/ml in PBS, 0.05% sodium azide)
- **Wash & Binding buffer (W&B buffer):** PBS, pH 7.4
- **Elution buffer:** 0.1 M Glycine-HCl, pH 2.5
- **Magnetic separator** (**e.g. MagnetoPURE,** Product Number: MP-10)
- **Binding capacity:** ~ 0.2 mg mouse IgG / mg SiMAG-Goat anti-mouse IgG

Protocol:

The following protocol describes the coupling of antibodies on *10 mg* particles. The procedure can be scaled up by adjusting volumes of required reagents.

1. Wash the **SiMAG-Goat anti-mouse IgG** (10 mg/ml) particles three times with 1 ml W & B buffer by magnetic separation and resuspended the beads in 0.5 ml W & B buffer by vortexing.

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2. To this suspension, calculate and add your antibody solution (final volume of 0.5 ml) with a required amount of the antibody based on the binding capacity of the particles.

3. Incubate at room temperature for 1h with gentle mixing.

4. Add 1 ml W & B buffer vortex for 5 seconds, collect the beads for 30 seconds with the magnet, remove and discard the supernatant and repeat the washing step three times.

5. After the last wash, resuspend the antibody coated beads in desired volume of W & B buffer.

6. Beads are now ready for the desired application or store at 4°C.

7. Optional: To recover the analyte suspend the antibody coated bead complex in elution buffer and incubate at room temperature for 15 minutes with gentle mixing. Collect the beads with magnet and remove the supernatant, containing analyte, in a fresh tube.