

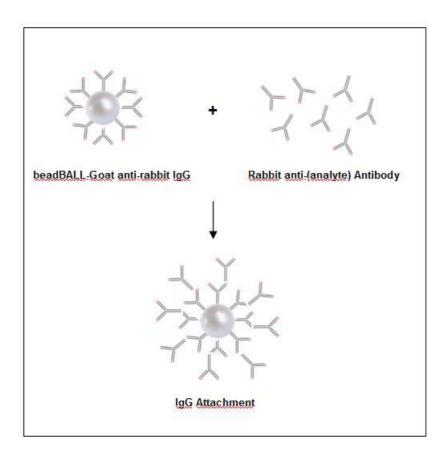
## Antibody binding protocol to beadBALL-Goat anti-rabbit IgG

## Introduction:

**beadBALL-Goat anti-rabbit IgG** is designed as a powerful tool for binding of rabbit IgG's.

The goat anti-rabbit IgG's are covalently coupled to the microspheres and can be used for an efficient method for the attachment of a rabbit antibody (analyte) and are applicable for cell enrichment from a heterogeneous cell suspension.

**beadBALL-Goat anti-rabbit IgG** is suitable in radio- and enzyme immunoassays which utilize a primary mouse IgG monoclonal antibody.





## **Equipment and reagents:**

- beadBALL-Goat anti-rabbit 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
- Microcentrifuge
- Binding capacity:
  - ~ 0.2 mg rabbit IgG / mg beadBALL-Goat anti-rabbit IgG

## **Protocol:**

The following protocol describes the coupling of antibodies on <u>10 mg</u> microspheres. The procedure can be scaled up by adjusting volumes of required reagents.

- Transfer 1 ml beadBALL-Goat anti-rabbit IgG microspheres in a 2 ml micro-centrifuge tube, add 1 ml W & B buffer and centrifuge for 1 minute at 500 x g. Remove the supernatant and repeat this step twice. Resuspend the microspheres in 0.5 ml W & B buffer.
- **2.** Add your antibody solution (max. volume 0.5 ml) with a required amount of the antibody based on the binding capacity of the microspheres.
- 3. Incubate at room temperature for one hour with gentle mixing.
- **4.** Add 1 ml **W & B buffer** vortex for 5 seconds, spin down, remove the supernatant and repeat the washing step three times.
- **5.** After the last wash, resuspend the antibody coated microspheres in desired volume of **W & B buffer.**
- **6.** Microspheres are now ready for the desired application or store at  $4^{\circ}$ C.
- 7. Optional: To recover the <u>rabbit antibody (analyte)</u>, suspend the antibody coated microsphere complex in **Elution buffer** and incubate at room temperature for 15 minutes with gentle mixing. Spin down and remove the supernatant, containing analyte, in a fresh tube.