



**nanomag<sup>®</sup>-D, Ø 130 nm und 250 nm**

**Covalent binding of proteins/ antibodies by carbodiimide activation of COOH groups  
(EDAC-Method)**

Particle type:	nanomag <sup>®</sup> -D
<b>Product-code:</b>	<b>09-02-132 / 09-02-252</b>
Particle diameter:	130 nm / 250 nm
Particle surface:	COOH

Material:

- 10 ml nanomag<sup>®</sup>-D suspension (surface: COOH, c= 10 mg/ml)
- 0.5 M 2-(4-morpholino)ethanesulphonic acid buffer which was adjusted to pH 6.3 with 0.5 M Na<sub>2</sub>CO<sub>3</sub> (0.5 M MES-buffer)
- 0.1 M 2-(4-morpholino)ethanesulphonic acid buffer which was adjusted to pH 6.3 with 0.5 M Na<sub>2</sub>CO<sub>3</sub> (0.1 M MES-buffer)
- 12 mg 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide hydrochloride (EDAC)
- 24 mg N-hydroxysuccinimide (NHS)
- 1 mg Protein
- PBS buffer (pH=7.4)
- 25 mM glycine in PBS buffer

Device:

permanent magnet

Procedure:

- dissolve 12 mg EDAC and 24 mg NHS in 2 ml 0.5 M MES-buffer (pH=6.3) and add this solution to 10 ml nanomag<sup>®</sup>-D suspension (surface: COOH, c= 10 mg/ml),
- incubate the suspension with continuous mixing for 1 hour at room temperature,
- wash the activated particles with 10 ml 0.1 M MES-buffer at a permanent magnet,
- resuspend the particles in 10 ml 0.1 M MES-buffer (pH=6.3) containing 1 (or less mg protein/antibody),
- incubate the suspension with continuous mixing for 3 hours at room temperature,
- add 1 ml 25 mM glycine in PBS buffer
- incubate the suspension with continuous mixing for 30 min at room temperature,
- wash the particles with 10 ml PBS buffer at a permanent magnet,
- resuspend the particles in 10 ml PBS-buffer (pH=7.4).

The protein binding capacity of nanomag<sup>®</sup>-D particles (Ø 130 nm or 250 nm) is about 1.5 - 2.0 µg/mg for albumin (BSA), protein A, avidin and streptavidin.