Maleimide Activated Alkaline Phosphatase

CATALOG #: M1242-1 (1 mg)

MOL. WEIGHT: 65 kDa

pH: 7.2 ± 0.2

ACTIVATION METHOD: Sulfo-SMCC

BINDING CAPACITY: ~2-4 mg of peptide (average MW of 1000 - 3000) per mg

alkaline phosphatase

FORM: Lyophilized powder

RECONSTITUTION: Reconstitute in distilled water at a concentration of 1-5 mg/ml

with gentle stirring.

STORAGE CONDITION: Lyophilized Maleimide Activated Alkaline Phosphatase should

be stored at -20 $^{\circ}$ C and is stable for 1 year. Reconstitute right before use. Note: Use the reconstituted protein

immediately. Do not store it.

DESCRIPTION: Maleimide Activated Alkaline Phosphatase is commonly used

to facilitate the conjugation of alkaline phosphatase to proteins, peptides and ligands containing sulfhydryl (-SH) groups for using it as an enzyme-linked detection reagent. Maleimide Activated Alkaline Phosphatase consists of alkaline phosphatase that has been modified by hetero bi-functional cross linker Sulfo-SMCC and contains several maleimide groups per alkaline phosphatase molecule while maintaining most of the enzyme activity. After conjugation, the activated alkaline phosphatase will form a covalent interaction with the peptides or ligands containing sulfhydryl groups and can be used as a probe for detection in various assays such as

ELISA, western blot etc.

APPLICATIONS: ELISA and Western Blot

PROCEDURE FOR PEPTIDE CONJUGATION:

12/20

Dissolve the sulfhydryl-containing peptide/hapten in 0.2-0.5 ml of phosphate buffer, pH
 7.2

Notes: For peptides/haptens with limited solubility in phosphate buffer, add DMSO (do not exceed DMSO 15% of total volume). Alkaline pH values (> 8.5) may hydrolyze the maleimide group or generate side reactions with amines. Peptides/haptens must contain cysteine or free sulfhydryl group in the reduced state and not disulfide bridges in order to react efficiently with the maleimide group. For peptides we recommend the use of 2 to 4 mg.

- Warm the Maleimide Activated Alkaline Phosphatase at room temperature (RT) and dissolve it in distilled H₂O right before use to yield a concentration of 1-5 mg/ml.
 Note: Do not vortex vigorously or heat the activated streptavidin.
- 3. Immediately mix the peptide (2-4 mg) with activated alkaline phosphatase and incubate at RT for 2-8 hr under occasional mixing.
- Peptide-conjugated alkaline phosphatase can be purified by gel filtration or dialysis to remove the unconjugated peptide.

Note: If the peptide-conjugated streptavidin is to be stored for > 2 weeks, it is recommended to store at -20°C. The peptide-conjugated streptavidin can be stored at -20°C for more than 6 months. Avoid repeated freeze-thaw cycles.

The coupling efficiency of conjugation can be determined by assaying the content of free sulfhydryl groups in the unreacted peptide using Sulfo-SMCC reagent.

RELATED PRODUCTS:

- Maleimide Activated HRP (Cat. No. M1322)
- Maleimide Activated KLH (Cat. No. M1317)
- Maleimide Activated OVA (Cat. No. M1318)
- Maleimide Activated BSA (Cat. No. M1316)
- Maleimide Activated Streptavidin (Cat. No. M1323)

For Research Use Only! Not to be used in humans.

Tel: 408-493-1800 | www.biovision.com |

Fax: 408-493-1801 tech@biovision.com