



Product Information

ExtrAvidin®-Alkaline Phosphatase

Product No. **E 2636**

Product Description

ExtrAvidin® is prepared from egg white avidin. It is a tetrameric protein containing four high affinity binding sites for biotin. ExtrAvidin® is a modified form of affinity purified avidin. ExtrAvidin® combines the high specific activity of avidin with the low background staining of streptavidin, a biotin binding protein produced by the bacteria *Streptomyces avidinii*. ExtrAvidin® binds biotin with the high affinity of egg white avidin, however, it does not exhibit the unwanted non-specific binding reported for egg white avidin at physiological pH, such as the staining of mast cells.

ExtrAvidin®-Alkaline Phosphatase may be used in conjunction with biotin in techniques such as ELISA, immunohistochemistry, protein blotting or with biotinylated-DNA probes. It provides a convenient yet highly sensitive and very specific detection system.

Reagents

The conjugate is provided as a solution in 0.05 M Tris-HCl, pH 8.0, containing 1 mM MgCl₂, 1% BSA and 15mM sodium azide as a preservative

Precautions

Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Product Profile

1. ELISA : Minimum 1:70,000
The working dilution of the conjugate for use as a reagent in enzyme immunoassay is determined by testing dilutions of the conjugate in microtiter plates coated with mouse IgG (1 µg/ml) and Biotin Rabbit Anti-Mouse IgG (Sigma Product No. B 8520). At

this dilution of the ExtrAvidin®-Alkaline Phosphatase conjugate, an absorbance of 1.0 at 405 nm after 30 minutes of substrate conversion (25 °C) is observed.

Substrate: *p*-Nitrophenyl Phosphate (pNPP), 1.0 mg/ml in 10% diethanolamine buffer, pH 9.8, containing 0.2% sodium azide.

2. Dot Blot
 - a. A minimum dilution of 1:300,000 was determined by indirect dot blot using human IgG (2.5 - 20 ng/dot) as the antigen, Rabbit Anti-Human IgG (Sigma Product No. I 8635) as the primary antibody and biotinylated anti-rabbit IgG as the secondary antibody.
 - b. A minimum dilution of 1:300,000 was determined in an indirect chemiluminescence assay using human IgG (2.5 - 20 ng/dot) as the antigen, Rabbit Anti-Human IgG (Sigma Product No. I 8635) as the primary antibody and biotinylated anti-rabbit IgG as the secondary antibody. 1,2-Dioxetane and enhancer was used as substrate.

3. Immunohistology
A working concentration of 10 - 30 µg/ml was determined using biotin conjugated anti-prostatic alkaline phosphatase and human prostate sections.

Conjugate Concentration: at least 1.5 mg/ml

Storage

Store at 2-8 °C. **DO NOT FREEZE**

JWM/PSS 12/02