

(Strept)avidin reagents

Product Description

Uptima (Strept)avidin conjugates are high quality reagents to be used with biotinylated probes (notably antibodies) in various immunotechnologies (ELISA, Blotting, Cytometry, Immunohistology).

	Unlabelled	FITC	Peroxidase	Alkaline Phosphatase	R-Phycoerythrin
Streptavidin Conjugates	UP51558C, 5mg UP51558O, 2mg	UP277130, 1 mg	UP395880, 1mg	UP518490, 1mg	Inquire
Neutralized Avidin Conjugates	UP25527A, 5mg	UP73578A, 1 mg	UP36570A, 1mg	UP38592A, 1mg	UP31259A, 1mg
Avidin Conjugates	UP39860D, 5mg	Inquire	Inquire	Inquire	Inquire

Form: powder (unlabelled), or ready-to-use solution (labelled Streptavidins)

Storage: +4°C
(long term storage: -20°C for unlabelled products)
Shipping: Room temperature or with blue ice

Associated Products:

Streptavidin Magnetic Beads *Uptibeads*, (UPR09030, UPR09031, UPR09020, UPR09021)

Immobilized biotins ([FT-UP88722](#)) and avidins ([FT-UP29337](#))

Scientific and Technical Information

Both Streptavidin and Neutralized Avidin are offered labelled by enzymes and fluorophores for ELISA, blotting, FCM, and IH techniques. Uptima (Strept)avidin conjugates are of very high quality, and offer advantages when compared with directly labelled primary antibodies :

- ▶ Lower background
 - ▶ Amplified detection signals
 - ▶ Easier to calibrate than different primary or secondary antibodies
 - ▶ More convenient for rarely used antibodies
 - ▶ Increased flexibility
- **Streptavidin** is isolated from *Streptomyces avidinii*, and has a very high affinity for biotin ($>10^{-14} \text{ M}^{-1}$). This makes the streptavidin-biotin interaction an ideal tool for many research applications. Streptavidin does not have any carbohydrates and has a lower ionic charge than avidin, resulting in a lower non-specific background. This makes streptavidin a preferred choice for many biotin-based applications.
 - **Avidin** is purified from eggs and has a even higher affinity for biotin ($>10^{-15} \text{ M}^{-1}$). It does not contain the RYD sequence found in Streptavidin, that is homologous of some integrins, giving the advantage of no unspecific binding that is observed in some detection systems with Streptavidin. Avidin presents however glycones and a higher ionic net charge, that's generated in some application higher background. Uptima offers to that point a chemically modified avidin, gathering the advantage of high specificity and affinity of avidin, but lowest background like Streptavidin. This **neutralized avidin** gives unsurpassed detection of biotinylated molecules.
 - **Labels:** HRP Peroxidase, AP Phosphatase, FITC

Contact your local distributor

Uptima, powered by



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FT-UP51558

Fluorescein (FITC) is a commonly used fluorescent label with an excitation wavelength of 495nm (argon laser)(max at 491nm), and an emission at 528nm (max at 518nm). Uptima FITC labelled antibodies are conjugated with 4-8 fluorophores per molecule to achieve the best signal to noise ratio.

UP75578A

Supplied in 0.01M phosphate buffered saline (PBS) pH 7.2, containing 1.0% BSA and 0.09% sodium azide as a preservative. The specific antibody concentration of this conjugate is 1.0 mg/ml. Molar ratio of fluorescein to avidin is between 3 - 8.

Suggested working concentration: one microgram to stain 1.0×10^6 cells in flow cytometric applications. However, each investigator should determine their own optimal working dilution for each specific research application

Applications: flow cytometry or immunohistochemistry (Vigier et al, 1988)

Storage +4°C, do not freeze

R-Phycoerythrin

UP 31259A

Supplied in 0.01M phosphate buffered saline (PBS) pH 7.2, containing 1.0% BSA and 0.09% sodium azide as a preservative. Suggested concentration of use is approximately 0.2-0.5 micrograms of product UP31259 to stain 1.0×10^6 cells in flow cytometric applications. However, each investigator should determine their own optimal working dilution for each specific research application

R-phycoerythrin to avidin molar ratio is between 0.7 and 1.3. R-phycoerythrin has an absorbance maximum at 565.5nm and an emission maximum at 578nm.

Storage +4°C, do not freeze

Horseradish peroxidase (HRP) is selected for its high activity and conjugated to the antibodies following an optimized process, which results in highly sensitive and stable antibodies. Peroxidase is one of the most commonly used enzymes as it is cheap and versatile, with an extensive range of soluble and insoluble substrates available. Recommended colourimetric substrates for HRP are TMB for ELISA (cat #UP664780) and TMB for blotting (cat # UP15426D). Higher sensitivity can be achieved by using a chemiluminescent substrate (UptiLight #UP99619A). One of the primary problems associated with HRP is non-specific staining that results from endogenous peroxidase activity within immunocytochemistry applications.

UP36570A

Supplied in 0.01M (3-[N-morpholino]propanesulfonic acid) (MOPS) pH 7.2, containing 1.0% BSA and .01% thimerosal as a preservative at a concentration of 1.0 mg conjugate protein per ml.

Working dilutions of approximately 1:4000 for protein blotting, 1:1000 for immunohistology and 1:20,000 for ELISA.

However, each investigator should determine their own optimal working dilution for each specific research application.

Storage +4°C, protect from light, do not freeze

Applications: blotting, immunohistochemistry or ELISA

Alkaline Phosphatase (AP) is an enzyme, which is isolated from calf intestines. It gives a more linear activity than peroxidase, and is suitable for most immunodetections. Alkaline phosphatase is especially recommended for applications, where high levels of endogenous peroxidase are present. Because reaction rates remain linear when using AP, the sensitivity can be increased by just allowing the reaction to proceed for longer periods of time. Recommended substrates for alkaline phosphatase are: BCIP/NBT for blotting and immunohistochemical applications (cat # UP096051) and pNPP for ELISA (cat # UP732500). Endogenous alkaline phosphatase activity found in some samples can be inhibited by levamisole. The reaction with pNPP allows kinetic readings.

UP38592

Supplied in 0.05M Tris pH 8.0, containing .001M MgCl₂, .001M ZnCl₂, 1.0% BSA and 0.09% sodium azide as a preservative. The specific antibody concentration of this conjugate is 1.0 mg/ml.

Working dilution for immunohistology is approximately 1:1000. However, each investigator should determine their own optimal working dilution for each specific research application.

Applications: blotting, immunohistochemistry or ELISA

Storage +4°C, do not freeze

Directions for Use

- Streptavidin and NeutralizedAvidin are classically used for:
 - coating of microplates and other supports
 - creating conjugates (of peptides, antibodies, any biological biomolecules)

Guidelines for coating protocols: 0.1 to 20µg/ml concentration in 0.1M carbonate pH9.6 or any other suitable buffer is recommended depending on application.

- Labelled Streptavidin and NeutralizedAvidin are classically used for:
 - direct immunodetection of bound biotin (ELISA sandwich)
 - indirect detection of biotin (ELISA inhibition)

Guidelines for detection protocols: PBST (150mM NaCl, 20mM phosphate, 0.05% Tween20) is a good buffer for most applications. However, TBS (150mM NaCl, 20mM Tris, pH7.5) is recommended for alkaline phosphatase-conjugates. A saturating agent may also be added. The dilution of use should be determined in each detection technic (ELISA or Blotting, FCM or IH...) and application, depending on assay conditions (saturating agent, nature of enzymatic substrate, duration of incubation...). 1/1000-1/50000 dilutions ordinarily suits for chromogenic substrates (TMP #UP66478, pNPP #UP66479...), and up 1/100 000 for chemiluminescent substrates (UptiLight #UP99619). Suggested dilutions may be given in batch certificates.

Other Information

For use *in vitro* only, not for diagnostic.

For any information, please contact Uptima, or your local distributor.

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Associated document:

FT-UP2557A Avidin and Neutralized avidin
NT-UP51558 Literature

Literature:

- Alon, R., Bayer, E. A., and Wilchek, M.¹ "Streptavidin Contains An RYD Sequence Which Mimics The RGD Receptor Domain of Fibronectin" (1990) *Biochemical and Biophysical Research Communications* **170**:1236-1241
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- Hiller, Y., Gershoni, J.M., Bayer, E.A. and Wilchek, M. (1987). Biotin binding to avidin. Oligosaccharide side chain not required for ligand association. *Biochem. J.* **248**, 167-171.
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