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DATASHEET

Biotin Actin

From rabbit skeletal muscle (α-actin)

For Use in Research Only.

Not for Use in Diagnostic Processes.

Quantity: 1x100μg Cat.#: 8109-01

Product Description

G-actin from rabbit skeletal muscle is a single chain polypeptide with a molecular weight of 42kD consisting of 375 amino acids. Biotin-Actin is chemically modified G-actin by esterification with Biotin. Choosing a biotinylation using a \sim 20 Å spacer, renders the Biotin group to be readily available for antibody detection. Biotin-Actin is >99% pure according to scanning densitometry from Coomassie Blue G-250 stained PAA-Gels.

Biotin Actin is validated to polymerize in the presence of 100mM KCl, 2mM MgCl₂. Immunoblotting using ExtrAvidin® alkaline phosphatase (Sigma, Cat. #: E2636) 1:10,000 with JetStain (Cat. #: 6022) detected 1ng of Biotin Actin.

Biotin-Actin is supplied as a lyophilized powder containing 2mM Tris-Cl pH 8.2, 0.4mM ATP and 0.1mM DTT, 1mM NaN₃, 0.2mM $CaCl_2$ and 0.5% disaccharides when reconstituted with H_2O to a 1mg/ml (23.8 μ M) solution.

For dilution of G-actin or exchange of ligand buffer into actin compatible buffer, MonoMix (Cat no.: 5100-0*) may be used. In the absence of nucleators, the polymerization of G-actin can be initiated by PolyMix (Cat no.: 5000-0*).

Product Handling

Preparation of a working stock

Example: Add 100μ l of H_2O to the tube containing 100μ g Biotin-Actin to obtain a working stock of 1mg/ml (23.8 μ M). Allow the G-actin solution to rehydrate for 5 minutes at room temperature and vortex mildly or and dissolve by pipetting up and down. Leave at RT for another 5 min for rehydration. The final concentration of Biotin G-actin should not exceed 3mg/ml. For standard applications the Biotin-Actin working stock is now ready to use.

For critical assays the working stock should be applied to a gel filtration column or dialyzed (100vol. of MonoMix, O/N) followed by high-speed centrifugation. For less critical assays the actin working stock may be pre-spun (15.000rpm, 10min).

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