

# STORAGE AND HANDLING



## PURE ACTIN

Catalog Number	Mass	Quantity
190101 - 5 mg	15 mg total powder	5 mg actin protein
190101 - 10 mg	30 mg total powder	10 mg actin protein

STORE IN A COOL,  
DRY ENVIRONMENT

**For research use only.**

**Shipping:** shipped at ambient temperatures

**Storage Conditions:** store in a cool, dry environment

**Form:** desiccated powder (1 mg actin protein is supplied as 3 mg powder with extra mass attributed to trehalose, a lyoprotectant)

**Source:** rabbit skeletal muscle

**Molecular Weight:** ~43 kDa

**Purity:** >90% (SDS-PAGE)

**Buffer Conditions Upon Reconstitution:** 2 mM Tris-HCl, 0.2 mM  $\text{CaCl}_2$ , 0.2 mM ATP, 1 mM DTT, and 0.25 M Trehalose (pH 8.0)

**Shelf Life:** check product label for expiration date

### Technical Notes

- store desiccated at 4°C, avoid hot and humid storage conditions
- reconstitute only when ready to use, keep on ice
- perform clarifying spin to remove protein aggregates
- freeze in liquid Nitrogen after reconstitution if desired, avoid repeated freeze-thaw cycles
- visit [www.PureSoluble.com/protocols](http://www.PureSoluble.com/protocols) for common actin polymerization protocols

### Storage and Handling

Store Pure Actin desiccated at 4°C. Reconstitute only when ready to use by resuspending in ice-cold ultrapure water to 9 mg/ml [actin protein]. Note that 1 mg actin protein is supplied as 3 mg powder (extra mass attributed to trehalose, a lyoprotectant), and reconstitution/dilution should be based on the actin protein concentration. Dilute to 0.4 mg/ml [actin protein] with Actin Working Buffer (5 mM Tris-HCl, 0.2 mM  $\text{CaCl}_2$ , pH = 8.0) and add ATP to 0.2 mM and DTT to 0.5 mM. Incubate on ice for 1 hour and mix occasionally with gentle vortexing. Next, clarify the actin to remove any protein aggregates. Centrifuge the actin protein at 14k rpm (21k x g) for 15 minutes at 4°C in a microcentrifuge and recover the supernatant on ice. Add sodium azide to 0.05% and store at 4°C. The actin protein is now ready for experimental use and stable under these storage conditions for several weeks.

If desired, the reconstituted actin protein can be aliquoted into smaller experimental batches and flash frozen for later use. Working on ice, aliquot the actin protein into experimental batches not less than 5 ul in volume. Flash freeze the experimental aliquots in liquid Nitrogen and store at -80°C. Note that it is not recommended to dilute the actin protein prior to freezing. Thaw only when ready to use by placing briefly in a 37°C water bath. Once the actin protein is approximately halfway thawed, remove from the water bath and thaw to completion with gentle flicking. Immediately place thawed actin protein on ice and continue to work on ice. Discard any unused portion of the experimental aliquots to avoid repeated freeze/thaw cycles.

### Applications

Pure Actin will polymerize into filamentous F-actin when supplemented with KCl and  $\text{MgCl}_2$ , and kept above its critical concentration. Visit [www.PureSoluble.com/protocols](http://www.PureSoluble.com/protocols) for common actin polymerization protocols.



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