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## Anti-beta Actin, Clone 4F7

(mouse monoclonal IgG)

Catalog # 05-0079

Lot # 100195

**Immunogen:** Synthetic peptide corresponding to a sequence of human beta Actin.

**Specificity:** Recognizes beta Actin specifically.

**Species Cross-reactivity:** Human, mouse, and rat.

**Formulation:** 0.1mg of mouse monoclonal IgG in 0.1ml of 0.01M Tris-HCl, pH 8.0, 0.15M NaCl, 25% glycerol, 0.02% sodium azide. Protein was determined by OD<sub>280</sub> absorbance.

**Physical State:** Frozen liquid

**Storage:** Store the product at -20°C. Product is stable for about 6 weeks at 2-8°C as an undiluted liquid. Prepare working dilution fresh each day. Avoid repeated freezing and thawing.

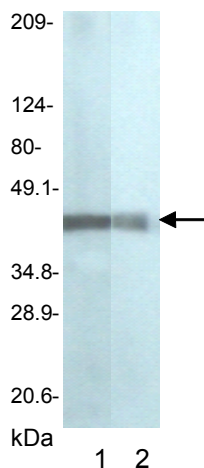
**Note:** For maximum recovery of product, centrifuge the original vial prior to removing the cap.

**Background:** Actins are highly conserved proteins that are involved in various types of cell motility and are ubiquitously expressed in all eukaryotic cells. Beta Actin is one of the two non-muscle cytoskeletal actins and is relatively stable and appears to remain at normal levels regardless of experimental treatment. It is generally used as an internal control for experiments.

**FOR IN VITRO RESEARCH USE ONLY**  
**NOT FOR USE IN HUMANS**

### Research Applications

**Immunoblot Analysis:** A working concentration of 0.5~2µg/ml is suggested.



A431 cell lysate was resolved by electrophoresis, transferred to PVDF membrane and reacted with the recommended dilution of Anti-beta Actin. The membrane was incubated using a goat anti-mouse secondary antibody conjugated to HRP and in ECL detection system to visualize results.

Arrow indicates beta Actin protein (~42kDa).

Lane 1: 1µg/ml of this lot antibody was used to detect beta Actin

Lane 2: 1:1000 dilution of a commercial antibody was used

## **Immunoblot Protocol**

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on samples and transfer the proteins to a PVDF membrane.
2. Block the blotted PVDF membrane in freshly prepared PBS containing 5% milk for 1 hour at room temperature with constant agitation.
3. Incubate the PVDF membrane with the recommended dilution of Anti-beta Actin diluted in freshly prepared 5% milk-PBS for 1 hour at room temperature or overnight with agitation at 2-8°C.
4. Wash the PVDF 3 times with PBS.
5. Incubate the PVDF in the secondary reagent of choice (a goat anti-mouse secondary antibody conjugated to HRP, Catalog # 02-0001, 1:3000 dilution was used) in 5% milk-PBS for 1 hour with agitation at room temperature.
6. Wash the PVDF 3-5 times with PBS.
7. Incubate the PVDF in ECL substrate for 1 min and cover the membrane by PE film.
8. In the darkroom, expose the PVDF to Autoradiography film for 5 s~ 5 min to visualize results.