

# Affinity-Purified Goat Anti-human/mouse/rat FKBP51 Antibody

## ORDERING INFORMATION

**Catalog Number:** AF4094

**Lot Number:** ZAW01

**Size:** 100 µg (sufficient for 330 mL of blotting solution)

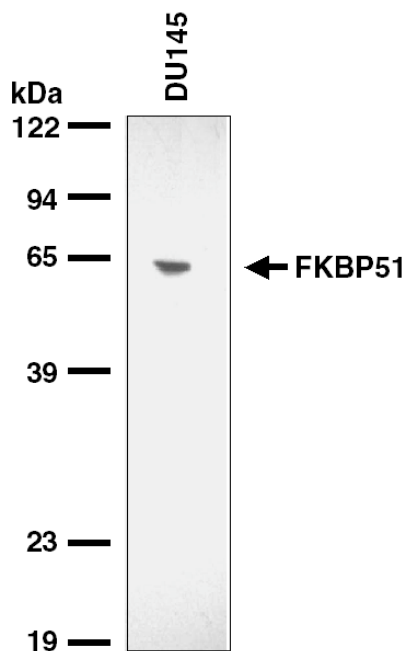
**Storage:** -20° C

**Specificity:** human/mouse/rat FKBP51

**Immunogen:** *E. coli*-derived recombinant human FKBP51

**Ig Type:** goat IgG

**Applications:** Western blot  
Immunohistochemistry



**Detection of FKBP51 with AF4094.** Lysate from mouse DU145 cells was transferred to an Immobilon-P membrane and immunoblotted with goat anti-human/mouse/rat FKBP51, as described in *Protocols for Immunoblotting*. A 5 second exposure to film is shown.

## Background

FK506 binding protein, 51 kDa molecular weight (FKBP51) is a peptidyl-prolyl isomerase that catalyzes the transition between *cis*- and *trans*- proline residues critical for proper folding of proteins. The macrolide immunosuppressants FK506 and Rapamycin are FKBP51 inhibitors. FKBP51 levels are induced by glucocorticoids. It associates with HSP90 complexes that are critical for the proper folding of steroid receptors. Single nucleotide polymorphisms in FKBP51 have been associated with major depression and hyper-responsiveness to antidepressants.

## Preparation

Goat antibodies were raised against purified, *E. coli*-derived, recombinant human FKBP51 (rhFKBP51; aa 2 - 457; Accession # NM\_004117). Polyclonal antibody was affinity-purified on a column derivatized with the recombinant protein and further purified by isolating the IgG fraction.

## Formulation

Lyophilized from a 0.2 µm-filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

## Reconstitution

Reconstitute with sterile PBS. If 0.2 mL of PBS is used, the antibody concentration will be 500 µg/mL.

## Storage

Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

## Specificity

The antibody detects endogenous human, mouse, and rat FKBP51 and does not cross-react with recombinant human FKBP12, FKBP13, FKBP38, or FKBP52 using Western blots.

## Applications

**Western blot** - An antibody concentration of 0.3 µg/mL is recommended.

### Protocols for Immunoblotting

Blotting Buffer	Blocking Solution	Antibody Solution
25 mM Tris, pH 7.4	5% nonfat dry milk in Blotting Buffer	2% nonfat dry milk in Blotting Buffer
0.15 M NaCl	Adjust pH to 7.4	Adjust pH to 7.4
0.1% Tween® 20		

1. Transfer the electrophoresed proteins to an Immobilon-P membrane (Millipore) and incubate the membrane for 1 hour at room temperature in Blocking Solution.
2. Incubate the membrane overnight at 4° C in antibody solution containing 0.3 µg/mL goat anti-human/mouse/rat FKBP51.
3. Wash the membrane at room temperature for 30 minutes with 5 or more changes of Blotting Buffer. Changing the membrane containers often reduces background.
4. Incubate the membrane for 1 hour at room temperature in Antibody Solution containing a 1:1,000 dilution of HRP-conjugated donkey anti-goat IgG (R&D Systems, Catalog # HAF109).
5. Wash the membrane for 30 minutes with 5 or more changes of blotting buffer.
6. Detect with WesternGlo™ Chemiluminescent detection reagents (R&D Systems, Catalog # AR004) or equivalent.

**Cell lysates for Western blots** - To prepare total cell lysates, cells are solubilized in hot 2X SDS gel sample buffer (20 mM dithiothreitol, 6% SDS, 0.25 M Tris, pH 6.8, 10% glycerol, 10 mM NaF and bromophenyl blue) at  $2 \times 10^6$  -  $1 \times 10^7$  cells per mL. The extracts are heated in a boiling water bath for 5 minutes and then sonicated with 3 - 4 bursts of 5 - 10 seconds each. Samples are diluted with 1X SDS sample buffer to the desired concentration.

**Immunohistochemistry** - This antibody will detect human FKBP51 in cells and tissues. The working dilution is 5 - 15 µg/mL. For chromogenic detection of labeling, use R&D Systems Cell and Tissue Staining Kits (CTS series).