

**Antibody Quality Control of non-phospho antibodies:**

Name of Antibody: GST-HAX mouse

Sheep Number: S868C

Bleed Number: 1<sup>st</sup> bleed, 2<sup>nd</sup> bleed, 3<sup>rd</sup> bleed - immunoblotting

Date Purified: 27/5/10

**Immuno Blotting:**

Recognises endogenous HAX in mouse embryonic fibroblasts

Can recognise HA tagged recombinant HAX in HEK 293 cell lines

Concentration used: 1 ug/ul

**Immunoprecipitation:**

Currently under evaluation with endogenous protein from Mouse embryonic fibroblast.

HEK293 cells were either transfected with HA empty vector (lane 1)/ HAX-HA (lane2) and 5 $\mu$ g of protein was subjected to SDS-PAGE analysis along with 25 $\mu$ g of whole cell lysates from Hax WT and KO mouse embryonic fibroblasts. Proteins were transferred onto nitrocellulose membrane and immunoblotted with anti-HAX mouse Ab at 1  $\mu$ g/ml overnight at 4°C.

Binding of the primary antibody was detected using rabbit peroxidase conjugated anti-sheep IgG antibody (1 in 2500 dilution) and enhanced chemiluminescence.

