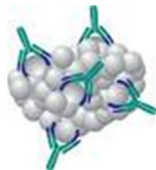


CYP450-GP



PRODUCT Hu-A007

ANTI-HUMAN CYP4A11 IgG

Polyclonal Antibody Developed in Rabbits, IgG Fraction

LOT RaR/B#3-7

Antiserum was developed in rabbits using purified human liver CYP4A11 as immunogen. The whole IgG fraction was purified from antiserum using caprylic acid/ammonium sulfate fractionation. Anti-human CYP4A11 IgG is provided as a powder after lyophilization from 100 mM potassium phosphate buffer (pH 7.4), 150 mM KCl, and 2.5 μ M thimerosal (added as a preservative).

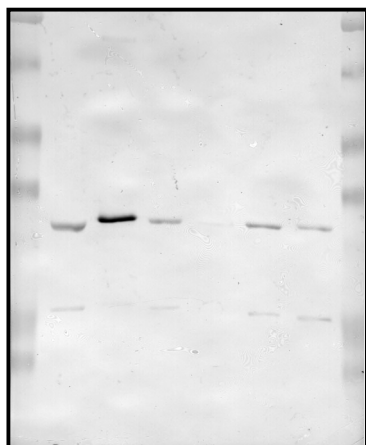
◆ **Specificity and Purity**

Specificity has been determined by Western blotting. Anti-human CYP4A11 IgG reacts exclusively with its corresponding 53 kDa antigen in human liver microsomes. In addition, the antibody recognizes the homologous CYP4A proteins in rat liver microsomes.

Antibody purity has been established by SDS-PAGE run under denaturing conditions. Two protein bands with molecular weights of 50 kDa and 25 kDa can be visualized by Coomassie blue staining, which correspond to the heavy and light chains, respectively, of rabbit IgG.

◆ **Reconstitution of Lyophilized Product and Storage**

Store lyophilized product at 0-5°C. For Western blotting, anti-CYP4A11 IgG should be reconstituted to 1 mg protein/ml by adding the appropriate amount of PBS/50% glycerol to the vial, and mixing gently until dissolved. The solution can then be stored at -20°C, as the presence of 50% glycerol prevents freeze/thawing. For immunoinhibition studies, reconstitute anti-CYP4A11 IgG in an appropriate buffer (e.g., 100 mM potassium phosphate, pH 7.4) at a concentration of 10-20 mg IgG/ml, and store at -20°C. The number of freeze/thaw cycles should be kept to a minimum.



Immunoreactivity of Anti-CYP4A11 IgG with human liver proteins

Lane 1 = Liver microsomes from Subject A (15 μ g)

Lane 2 = Purified CYP4A11 (0.1 μ g)

Lane 3 = Liver microsomes from Subject B (15 μ g)

Lane 4 = Purified CYP2E1 (0.1 μ g)

Lane 5 = Liver microsomes from Subject C (15 μ g)

Lane 6 = Liver microsomes from Subject D (15 μ g)

◆ **Use for Western Blotting**

Incubate blots overnight with 2.5 - 5.0 μ g rabbit anti-human CYP4A11 IgG/ml of appropriate blocking solution. After washing to remove unbound antibody, incubate with an anti-rabbit IgG conjugate of choice (e.g, anti-rabbit IgG-peroxidase), and develop accordingly. A detailed Western blotting method can be found in the [Protocols](#) section.

◆ **Use for Immunoinhibition**

Incubation of anti-human CYP4A11 IgG with human liver microsomes at a ratio of 5 mg IgG/nmol microsomal P450 (1.7 mg IgG/mg microsomal protein) before reaction initiation will typically give 70-85% inhibition of an exemplary CYP4A11-catalyzed reaction (e.g., laurate 12-hydroxylation; **see attached**). Methodology for conducting P450 immunoinhibition assays is given in the [Protocols](#) section.

