**Human/Rat α-Fetoprotein/AFP Antibody**

**Antigen Affinity-purified Polyclonal Chicken IgY**

**Catalog Number:** AF1369

**DESCRIPTION**

**Species Reactivity:** Human/Rat

**Specificity:** Detects human α-Fetoprotein in direct ELISAs and Western blots.

**Source:** Polyclonal Chicken IgY

**Purification:** Antigen Affinity-purified from egg yolks

**Immunogen:** Human umbilical cord serum

**Formulation:** Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

*Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.*

**APPLICATIONS**

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

<table>
<thead>
<tr>
<th>Recommended Concentration</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Western Blot</strong></td>
<td>0.2 µg/mL</td>
</tr>
<tr>
<td><strong>Immunocytochemistry</strong></td>
<td>8-25 µg/mL</td>
</tr>
<tr>
<td><strong>Immunohistochemistry</strong></td>
<td>5-15 µg/mL</td>
</tr>
<tr>
<td><strong>Simple Western</strong></td>
<td>2 µg/mL</td>
</tr>
</tbody>
</table>

**Knockout Validated**

α-Fetoprotein is specifically detected in HepG2 human hepatocellular carcinoma cell line but is not detectable in α-Fetoprotein knockout HepG2 cell line.

**DATA**

**Western Blot**

Detection of Human and Rat α-Fetoprotein/AFP by Western Blot. Western blot shows lysates of human liver tissue and H4-II-E-C3 rat hepatoma cell line. PVDF membrane was probed with 0.2 µg/mL of Chicken Anti-Human/Rat α-Fetoprotein/AFP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1369) followed by HRP-conjugated Anti-Chicken IgY Secondary Antibody. A specific band was detected for α-Fetoprotein/AFP at approximately 70 kDa (as indicated). This experiment was conducted under reducing conditions and using Immobilon Buffer Group 1.

**Immunocytochemistry**

α-Fetoprotein/AFP in BG01V Human Embryonic Stem Cells. α-Fetoprotein/AFP was detected in immunofluorescence fixed BG01V human embryonic stem cells using Chicken Anti-Human/Rat α-Fetoprotein/AFP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1369) at 10 µg/mL, for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Chicken IgY Secondary Antibody (red; Catalog # NL016) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

**Immunohistochemistry**

α-Fetoprotein/AFP in Human Liver cancer. α-Fetoprotein/AFP was detected in immunohistochemistry fixed paraffin-embedded sections of human liver cancer using 15 µg/mL of Chicken anti-Human/Rat α-Fetoprotein/AFP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1369) overnight at 4 °C. Tissue was stained (brown) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

**Simple Western**

Detection of Human α-Fetoprotein/AFP by Simple Western™. Simple Western lane view shows lysates of HepG2 human hepatocellular carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for α-Fetoprotein/AFP at approximately 70 kDa (as indicated) using 2 µg/mL of Chicken Anti-Human/Rat α-Fetoprotein/AFP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1369) followed by HRP-conjugated Anti-Chicken IgY Secondary Antibody. This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.
Knockout Validated

α-Fetoprotein/AFP Specificity is Shown by Immunocytochemistry in Knockout Cell Line. α-Fetoprotein/AFP was detected in immersion fixed HepG2 human hepatocellular carcinoma cell line but is not detected in α-Fetoprotein/AFP knockout (KO) HepG2 Human Cell Line cell line using Chicken Anti-Human/Rat α-Fetoprotein/AFP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1369) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 493-conjugated Anti-Chicken IgY Secondary Antibody (green; Catalog # NL018) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution
Reconstitute at 0.2 mg/mL in sterile PBS.

Shipping
The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

* Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C

Stability & Storage
Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

α-Fetoprotein is a major plasma protein in the fetus. Its concentration is normally low in the adult except when produced by certain tumors.