**Intended Use**

For Research Use Only.

This antibody is intended for use in Immunohistochemical applications on formalin-fixed paraffin-embedded tissues (FFPE), frozen tissue sections and cell preparations. Interpretation of results should be performed by a qualified medical professional.

* The STAT6 antibody, clone EP325, has been manufactured using Epitomics RabMab® technology covered under Patent No.s 5,675,063 and 7,402,409.

**Immunogen**

Synthetic peptide corresponding to residues of human STAT6 protein.

**Summary and Explanation**

STAT6 is a human gene. The protein encoded by this gene is a member of the STAT family of transcription factors. In response to cytokines and growth factors, STAT family members are phosphorylated by the receptor associated kinases, and then form homo- or heterodimers that translocate to the cell nucleus where they act as transcription activators. This protein plays a central role in exerting IL-4 mediated biological responses. It is found to induce the expression of BCL2L1/BCL-X(L), which is responsible for the anti-apoptotic activity of IL4.

Recurrent somatic fusions of the NGFI-A–binding protein 2 (NAB2) gene and STAT6 gene, located at chromosomal region 12q13, have been identified in Solitary Fibrous Tumors (SFT). In one study, Fifty-nine of 60 SFT cases (98%) showed nuclear expression of STAT6, which was usually diffuse and intense. All other tumor types of soft tumor tissues were negative for STAT6, except for three dedifferentiated Liposarcomas and one deep Fibrous Histiocytoma, which showed weak staining. STAT6 is a highly sensitive and specific immunohistochemical marker for SFT and can be helpful to distinguish this tumor type from other soft tissue neoplasms. This antibody is intended for use in Immunohistochemical applications on formalin-fixed paraffin-embedded tissues (FFPE), frozen tissue sections and cell preparations. Interpretation of results should be performed by a qualified medical professional.

**Presentations**

<table>
<thead>
<tr>
<th>Catalog Num.</th>
<th>Antibody Type</th>
<th>Dilution</th>
<th>Volume/Qty</th>
</tr>
</thead>
<tbody>
<tr>
<td>BSB 3420</td>
<td>Tinto Prediluted</td>
<td>Ready-to-Use</td>
<td>3.0 mL</td>
</tr>
<tr>
<td>BSB 3421</td>
<td>Tinto Prediluted</td>
<td>Ready-to-Use</td>
<td>7.0 mL</td>
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<tr>
<td>BSB 3422</td>
<td>Tinto Prediluted</td>
<td>Ready-to-Use</td>
<td>15.0 mL</td>
</tr>
<tr>
<td>BSB 3423</td>
<td>Concentrated</td>
<td>1:50 - 1:200</td>
<td>0.1 mL</td>
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<tr>
<td>BSB 3424</td>
<td>Concentrated</td>
<td>1:50 - 1:200</td>
<td>0.5 mL</td>
</tr>
<tr>
<td>BSB 3425</td>
<td>Concentrated</td>
<td>1:50 - 1:200</td>
<td>1.0 mL</td>
</tr>
<tr>
<td>BSB 3426</td>
<td>Control Slides</td>
<td>Not Applicable</td>
<td>5 slides</td>
</tr>
</tbody>
</table>

**Precautions**

1. For professional users only. Ensure results are interpreted by a medical professional.
2. This product contains sodium azide (NaN₃), a toxic chemical which may react with plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent sodium azide build-up.
3. Ensure proper handling procedures are used with reagent. Always wear proper laboratory equipment such as laboratory coat and gloves when handling reagents.
4. Unused solution should be disposed of according to local and federal regulations.
5. Do not ingest reagent. If reagent ingested, contact a poison control center immediately.
6. For complete recommendations for handling biological specimens please refer to the CDC document, “Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories” (8).

**Storage**

Store at 2-8 °C. Do not use after expiration date listed on package label. Temperature fluctuations should be avoided. Store appropriately when not in use, and avoid prolonged exposure to room temperature conditions.

**Paraffin sections:** The antibody can be used on formalin-fixed paraffin-embedded (FFPE) tissue sections. Ensure tissue undergoes appropriate fixation to ensure best results. Pre-treatment of tissues with heat-induced epitope retrieval (HIER) is recommended using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020-B50 0023), ImmunoDNA Retriever with EDTA (BSB 0030-B50 0033) or ImmunoDNA Digestor (BSB 0108-0112). See reverse side for complete protocol. Tissue should remain hydrated via use of Bio SB Immuno/DNA Washer solutions (BSB 0029 & BSB 0042).

**Frozen sections and cell preparations:** The antibody can be used for labeling acetone-fixed frozen sections and acetone-fixed cell preparations.

**Presentation**

STAT6 is a rabbit monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.

<table>
<thead>
<tr>
<th>Antibody Type</th>
<th>Rabbit Monoclonal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isotype</td>
<td>IgG</td>
</tr>
<tr>
<td>Localization</td>
<td>Nuclear</td>
</tr>
<tr>
<td>Species Reactivity</td>
<td>Paraffin, Frozen</td>
</tr>
</tbody>
</table>

**Summary and Explanation**

STAT6 is a human gene. The protein encoded by this gene is a member of the STAT family of transcription factors. In response to cytokines and growth factors, STAT family members are phosphorylated by the receptor associated kinases, and then form homo- or heterodimers that translocate to the cell nucleus where they act as transcription activators. This protein plays a central role in exerting IL-4 mediated biological responses. It is found to induce the expression of BCL2L1/BCL-X(L), which is responsible for the anti-apoptotic activity of IL4.

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**Localization**

Nuclear

**Isotype**

IgG

**Presentation**

STAT6 is a rabbit monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.
Staining Procedure

1. Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positive charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028).
2. Air dry for 2 hours at 58° C.
3. Deparaaffinize, dehydrate and rehydrate tissues.
4. Subject tissues to heat epitope retrieval using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).
5. Any of three heating methods may be used:
   a. **TintoRetriever Pressure Cooker or Equivalent**
      Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA, and place in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 15 minutes. Open and immediately transfer slides to room temperature.
   b. **TintoRetriever PT Module or Water Bath Method**
      Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA at 95°-99° C. Incubate for 30-60 minutes.
   c. **Conventional Steamer Method**
      Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA in a Steamer, cover and steam for 30-60 minutes.
6. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
7. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer’s instructions.
8. Wash slides with IHC wash buffer or DI water.
9. Continue IHC staining protocol.

Recommended IHC Protocol

<table>
<thead>
<tr>
<th>Step</th>
<th>ImmunoDetector AP/HRP</th>
<th>PolyDetector AP/HRP</th>
<th>PolyDetector Plus HRP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peroxidase/AP Blocker</td>
<td>5 min.</td>
<td>5 min.</td>
<td>5 min.</td>
</tr>
<tr>
<td>Primary Antibody</td>
<td>30-60 min.</td>
<td>30-60 min.</td>
<td>30-60 min.</td>
</tr>
<tr>
<td>1st Step Detection</td>
<td>10 min.</td>
<td>30-45 min.</td>
<td>15 min.</td>
</tr>
<tr>
<td>2nd Step Detection</td>
<td>10 min.</td>
<td>Not Applicable</td>
<td>15 min.</td>
</tr>
<tr>
<td>Substrate-Chromogen</td>
<td>5-10 min.</td>
<td>5-10 min.</td>
<td>5-10 min.</td>
</tr>
<tr>
<td>Counterstain</td>
<td>Varies</td>
<td>Varies</td>
<td>Varies</td>
</tr>
</tbody>
</table>

Symbol Key / Légende des symboles/Erläuterung der Symbole

- **Storage Temperature** / **Limites de température** / **Zulässiger temperaturbereich**: 2°C-8°C
- **Read Instructions for Use** / **Consulter les instructions d’utilisation** / **Gebrauchsanweisung beachten**
- **Manufacturer / Fabricant / Hersteller**
- **Expiration Date / Utiliser jusque** / **Verwendbar bis**
- **Lot Number / Code du lot** / **Chargenbezeichnung**
- **Catalog Number / Référence du catalogue** / **Bestellnummer**

Product Limitations

Due to inherent variability present in immunohistochemical procedures (including fixation time of tissues, dilution factor of antibody, retrieval method utilized and incubation time), optimal performance should be established through the use of positive and negative controls. Results should be interpreted by a medical professional.

References