Human Fibroblast Activation Protein α/FAP Antibody

Antigen Affinity-purified Polyclonal Sheep IgG
Catalog Number: AF3715

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Fibroblast Activation Protein α/FAP in direct ELISAs and Western blots.
Source	Polyclonal Sheep IgG
Purification	Antigen Affinity-purified
Immunogen	*S. frugiperda* insect ovarian cell line *Sf* 21-derived recombinant human Fibroblast Activation Protein α/FAP Leu26-Asp760 Accession # Q12884
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
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.

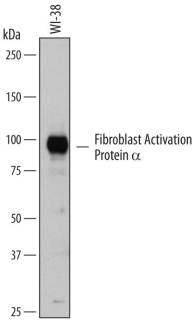
APPLICATIONS

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.*

	Recommended Concentration	Sample
Western Blot	0.5 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Immunoprecipitation	25 µg/mL	Conditioned cell culture medium spiked with Recombinant Human Fibroblast Activation Protein α/FAP (Catalog # 3715-SE), see our available Western blot detection antibodies
Simple Western	10 µg/mL	IMR-90 human lung fibroblast cell line and WI-38 human lung fibroblast cell line
Knockout Validated	Fibroblast Activation Protein α/FAP is specifically detected in WI-38 human lung fibroblast parental cell line but is not detectable in Fibroblast Activation Protein α/FAP knockout WI-38 human lung fibroblast cell line.	

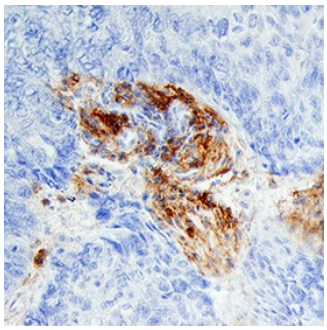
DATA

Western Blot



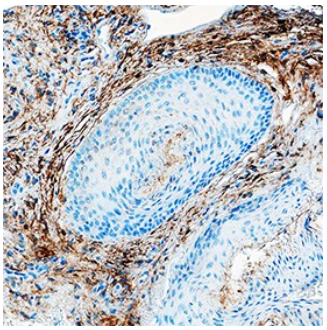
Detection of Human Fibroblast Activation Protein α/FAP by Western Blot. Western blot shows lysates of WI-38 human lung fibroblast cell line. PVDF membrane was probed with 0.5 µg/mL of Sheep Anti-Human Fibroblast Activation Protein a/FAP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3715) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # Catalog # HAF016). A specific band was detected for Fibroblast Activation Protein a/FAP at approximately 97 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



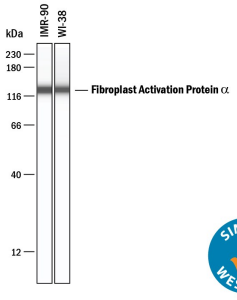
Fibroblast Activation Protein α/FAP in Human Squamous Cell Carcinoma. Fibroblast Activation Protein a/FAP was detected in immersion fixed paraffin-embedded sections of human squamous cell carcinoma using Sheep Anti-Human Fibroblast Activation Protein a/FAP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3715) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Sheep HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS019) and counterstained with hematoxylin (blue). Specific staining was localized to connective tissue. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Immunohistochemistry



Fibroblast Activation Protein α/FAP in Human Basal Cell Carcinoma. Fibroblast Activation Protein α/FAP was detected in immersion fixed paraffin-embedded sections of human basal cell carcinoma using Sheep Anti-Human Fibroblast Activation Protein α/FAP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3715) at 10 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Sheep HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS019) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western



Detection of Human Fibroblast Activation Protein α/FAP by Simple Western™. Simple Western lane view shows lysates of IMR-90 human lung fibroblast cell line and WI-38 human lung fibroblast cell line, loaded at 0.2 mg/mL. A specific band was detected for Fibroblast Activation Protein α/FAP at approximately 130 kDa (as indicated) using 10 µg/mL of Sheep Anti-Human Fibroblast Activation Protein α/FAP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3715) followed by 1:50 dilution of HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

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Human Fibroblast Activation Protein α/FAP Antibody

Antigen Affinity-purified Polyclonal Sheep IgG
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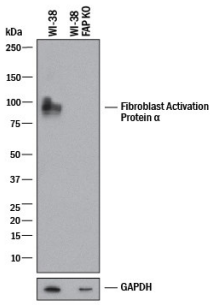
Knockout Validated



Western Blot Shows Human Fibroblast Activation Protein α/FAP Specificity by Using Knockout Cell Line. Western blot shows lysates of WI-38 human lung fibroblast cell line and human FAP knockout WI-38 human lung fibroblast cell line. PVDF membrane was probed with 0.5 µg/mL of Sheep Anti-Human Fibroblast Activation Protein α/FAP Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3715) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for Fibroblast Activation Protein α/FAP at approximately 97 kDa (as indicated) in the parental WI-38 human lung fibroblast cell line, but is not detectable in knockout WI-38 human lung fibroblast cell line. GAPDH (Catalog # AF5718) is shown as a loading control. This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

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

FAP (also known as seprase) is a Type II transmembrane serine protease that is structurally related to dipeptidyl peptidase IV (1). FAP has substrate specificity similar to dipeptidyl peptidase IV, which is specific for N-terminal Xaa-Pro sequences, but FAP is also an endopeptidase able to degrade gelatin and Type I collagen (2). The enzymatically active form of FAP is a dimer (3). FAP has a restricted tissue distribution. It is not detectable in normal tissues or resting fibroblasts, but is highly expressed on reactive stromal fibroblasts in epithelial cancers (4), in granulation tissue during wound healing, and in bone and soft tissue sarcomas (5). Because of its expression patterns and enzymatic activities, FAP is believed to play roles in tumor invasion, tissue remodeling, and wound repair.

References:

1. Scanlan, M.J. *et al.* (1994) Proc. Natl. Acad. Sci. USA **91**:5657.
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5. Rettig, W.J. *et al.* (1988) Proc. Natl. Acad. Sci. USA **85**:3110.

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