

Anti - Desmin

Rabbit clonal antibody

(1 ml)

CAT#

DB 148-1

CONCENTRATED READY TO USE (RTU)

DB 148-0.1 DB 148-RTU-7 $(100 \mu l)$ (7 ml) DB 148-0.2 $(200 \mu I)$ DB 148-RTU-15 (15 ml) DB 148-0.5 (500 µl)

STORAGE AND APPLICATION

CONCENTRATED READY TO USE (RTU)

+4°C +4°C, Do not freeze! Storage: Storage:

Application: IHC-P, Application: IHC-P, dilution 1:100

ready to use

PRODUCT INFORMATION

Clone: F18-V

20 mM Tris-HCl, pH 8.0 Buffer: Stabilizer: 20 mg/ml BSA Preservative: 0.05% NaN₃

Specificity: Human

Expiration: 24 months from the shipping date

Peptide derived from the C-terminal sequence of human Immunogen: desmin. Antibody recognizes the epitope between

Thr453 - Val469.

Cellular localization: cytoplasm

smooth and skeletal muscle, leiomyoma tissue Positive control:

Protein accession number: P17661

IHC-P PROTOCOL - INSTRUCTION MANUAL

- Deparaffinize the section in 3 changes of xylene, 10 minutes each.
- Wash the section in 96%, 80% and 70% ethyl alcohol for 10 minutes each.
- Rinse in distilled water, 2 x 5 minutes.
- Block the endogenous peroxidase by incubating the tissue in 3% hydrogen peroxide (H₂O₂) for 10 minutes.
- Wash in distilled water, 2 x 5 minutes.
- For antigen retrieval: Immerse the slide in citrate buffer, pH 6.0* and incubate at 95-97°C in water bath for 25 minutes.
- Remove the staining to room temperature and let the slide to cool (in citrate buffer, pH 6.0) for 15 minutes
- Rinse in distilled water, 2 x 5 minutes.
- Wash in PBS (phosphate buffer saline, pH 7.0-7.5) supplemented with 0.05% of Tween-20 (buffer A), 2 x 5 minutes
- CONCENTRATED:

Incubate the section with primary antibody at the dilution 1:100 for 1 hour in the closed wet chamber.

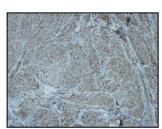
READY TO USE (RTU):

Incubate the section with primary antibody (ready to use) for 1 hour in a closed wet

- 11. Wash 3 x 5 minutes with buffer A.
- 12. Apply the secondary antibody (the protocol depends on the supplier), and proceed to standard immunohistochemistry protocol (HRP - Peroxide - DAB). Micropolymer-HRP detection kit rabbit/mouse dual of DB Biotech is suggested (http://www.dbbiotech.com/products/detection-system.html).
- 13. Wash 3 x 5 minutes with buffer A.
- 14. Apply the chromogen (DAB), 1 3 minutes
- Wash in water, 2 x 5 minutes.
- 16. Stain in hematoxylin for 5 minutes.
- Wash in distilled water, 3 x 2 minutes.
- 18. Mount the slide for observation.

* Citrate Buffer (10mM Citric Acid, 0.05% Tween-20, pH 6.0):

Citric acid (anhydrous) ------ 1.92 g; Distilled water ------ 1000 ml Mix to dissolve in 700 ml of distilled water. Adjust pH to 6.0 with 1M NaOH and then add $0.5 \ \text{ml}$ of Tween-20 and mix well. Adjust the final volume to 1 liter with distilled water. Store this solution at room temperature for 3 months or at +4°C for longer storage.



Desmin expression detected in the uterine leiomyoma tissue with DB 148 monospecific antibody. Formalin fixed, paraffin embedded human tissue (4 µm section) stained according to related DB Biotech datasheet.

VENTANA PROTOCOL - INSTRUCTION MANUAL

SHORT APPLICATION PROTOCOL FOR VENTANA BENCHMARK SLIDE STAINING SYSTEM

PROCEDURE: U ultraView DAB

- Deparafinization
- Heating (72 °C) at the medium temperatures. Deparafinization.
- Cell conditioning
- ULTRA conditioner #1 4.
- Heating glass (95 °C), incubation 8 min. (Cell conditioner #1; buffer CC1).
- ULTRA CC1 solution application 20 min. 6.
- Antibody incubation temperature 7.
- 8. Heating glass (36 °C), incubation 4 min.
- 9. Titration
- 10. Hand apply primary antibody 100 μl. Incubation 36 min.
- 11. ultraWash
- 12. Nuclear stain
- 13. Hematoxylin II application - one drop (nuclear stain). Cover and incubate 12 min.
- After nuclear stain
- Bluing reagent application, one drop. After nuclear stain, cover and incubate 4 min

LEICA BOND MAX PROTOCOL - INSTRUCTION MANUAL

SHORT APPLICATION PROTOCOL FOR LEICA BOND MAX SLIDE STAINING SYSTEM

Protocol F:

- Visualization system: BOND Refine DS9800
- Epitope retrieval / heating time / temperature: ER2 / 20 min. / 100 °C
- Incubation of primary antibody / temperature: 30 min. / 20 °C

PRECAUTIONS

- We strongly recommend to use DB Primary Antibody Diluent (catalog number DB D-125, or DB D-250), eventually alternative diluent (containing protease free BSA at the concentrations ≥ 1mg/ml) for dilution of concentrated antibodies, otherwise the warranty might be voided.
- Centrifuge the vial before use.
- Intended for professional In Vitro Diagnostic use in laboratories.
- Do not use after expiration date stamped on vial label.
- Avoid contamination of the reagent.
- Any discrepancies in the recommended procedures stated in the working protocol may
- The reagent contains sodium azide (NaN₃) which is highly toxic in higher concentrations. The concentration in the reagent (0.05%) is not considered as hazardous.
- Disposal of waste material must be conducted in accordance with local regulations.
- Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.

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