

1-Step POLYMER HISTO-STAT™- Multivalent DAB Staining System

For No-Wash, Rapid IHC staining of MOUSE, RABBIT and RAT primary antibodies in less than 2- hours
Background-Free and Biotin-Free

Large volume kit: Product number: NB312LD

Small volume kit: Product number: NB312LD-20

PRODUCT DESCRIPTION

1-step Polymer HISTO-STAT Staining system is a **No wash, No Background** staining system formulated for **2-step IHC staining** of primary antibodies raised in mouse, rat and rabbit. The 1-Step POLYMER, HISTO-STAT is bio-engineered for high sensitivity, short incubation periods and no- background. Innovex HISTO-STAT Polymer Staining (detection) system only includes two-components; A multivalent anti Rabbit, anti Mouse and anti Rat **Polymer Second Step-peroxidase Reagent** and stable AEC OR DAB substrate/ chromogens. **1-step Polymer HISTO-STAT** staining system is universally applicable to staining all mouse, rat and rabbit primary antibodies. This system is also applicable to IHC and ICC staining of human and all animal tissues. This system is universally applicable to staining all tissues and cells regardless of their method of processing, e.g., paraffin sections, cryostat sections, cytocentrifuge preparations or cell smears.

"HISTO-STAT" 1-Step Polymer is an ideal detection system for staining of primary antibodies for paraffin and frozen sections as well as cytosmears. HISTO-STAT, 1-Step staining system is free of biotin, avidin and Streptavidin and use of this detection system provides sensitivity and speed as well as eliminating the need for biotin blocking step in tissues and cells that are rich in endogenous biotin. The use of **HISTO-STAT, 1-Step Polymer Staining system** is designed to eliminate the need for re-titration of primary antibodies upon the switch over to this system.

APPLICATION / INTENDED USE

This product is intended for IHC and ICC staining of MOUSE, RAT and RABBIT primary antibodies in tissues and cell preparations.

SYSTEM COMPONENTS and SPECIFICATIONS

Recommended incubation times for the system components are:

- Peroxide Block: **15-minutes**.
- Background Buster: **10-minutes**
- MOUSE or RABBIT or RAT primary antibody (**not provided**): 30-minutes for frozen sections and 30-minutes to 1-hour for paraffin sections. Observe manufacturer's recommended incubation time for the primary antibodies employed.
- Histo-Stat; 1-Step Polymer Peroxidase (HRP) solution: **45-minutes** incubation for paraffin sections and 30-minutes for frozen sections.
- Stable Turbo action two-component liquid DAB/substrate: **5-minutes**.
- Quick DAB Enhancer: **2-minutes**

INSTRUCTIONS

ALL INNOVEX PRODUCTS ARE DESIGNED TO BE IMPLEMENTED AT ROOM TEMPERATURE (NO HEAT IS REQUIRED).

No serum or protein blocking and NO extensive washes are required when staining with Innovex HISTO-STAT 1-Step Polymer

Following deparaffinization; Quench endogenous peroxidase enzyme activity by applying INNOVEX STABLE PEROXIDE BLOCK for **15-minutes** (provided in the kit) and then rinse with water for **30-seconds**.

1. Retrieve with INNOVEX Uni-Trieve OR with Retrieval of choice (for formalin fixed paraffin embedded sections).
2. Apply Background Buster for **10-minutes** (provided in the kit).
3. Rinse with PBS or Innovex HRP Enhancing Wash Buffer once for 5-seconds.
4. Incubate the section or smear for 30-minutes to 1-hour with Mouse OR Rat OR Rabbit primary antibodies (not provided); Observe the manufacturer's recommended incubation time for the primary antibody employed.
5. Rinse with PBS or Innovex HRP Enhancing Wash Buffer once for 5-seconds.

CONTINUED OVER

6. Incubate with the **HISTO-STAT; 1-Step Polymer** for **45-minutes** for paraffin sections and for frozen sections.
7. Rinse with PBS or Innovex HRP Enhancing Wash Buffer once for 5-seconds.
8. Incubate with mixed DAB/substrate solution for **5-minutes** and rinse with water.
9. Apply Quick DAB Enhancer for 2-minutes and rinse with water (DAB Enhancer is included in the kit); This step is optional.
10. Counterstain with hematoxylin (Innovex Product number NB305).
11. Rinse hematoxylin with tap water by filling and emptying the slide holder vessel with tap water until water runs clear.
12. Mount slides as usual OR mount from water with Innovex permanent "Advantage Mounting Media" Product number NB300).

Substrate/chromogen mixing protocol

Mix DAB chromogen by adding 2 drops of DAB chromogen (component 2) to 2 ml of Ready-To-Use substrate buffer (component 1) in the provided graduated mixing tube, cap the tube tightly and mix by inversion or mix by drawing the mixture in and out of transfer pipette a few times. **Left over mixed DAB solution is stable for 1-week when kept refrigerated and can be re-used within 7 days post initial mixing. This minimizes reagent waste and disposal costs.**

STORAGE CONDITIONS

Store in refrigerator at 2-8°C through expiration date noted on the vials.

Important Notes:

- Innovex HISTO-STAT 1-Step Polymer staining reagents and components are **no wash, no background** staining kits, a single 5-second washes in between incubation steps will be sufficient for rinsing.
- HISTO-STAT 1-Step Polymer kit is universally applicable to staining all antibodies; Follow manufacturer's primary antibody incubation time for primary antibody employed.
- Rinsing with Innovex Signal Enhancing Wash Buffers amplifies stains and produces sharper and more resolved staining.

FOR PROFESSIONAL AND RESEARCH USE