

ImmunoCruz™ mouse ABC Staining System: sc-2017

PRODUCT

ImmunoCruz™ ABC Staining Systems include 1.0 ml normal blocking serum, 250 µg biotinylated secondary antibody, 0.5 ml each avidin and biotinylated horseradish peroxidase (AB reagents), 1.0 ml 50x peroxidase substrate, 1.0 ml 50x DAB chromogen and 3.0 ml 10x substrate buffer. Also included are mixing bottles for the preparation of reagent working solutions. One ImmunoCruz™ ABC Staining System contains sufficient reagent for 200 slides.

Solutions to be provided by the researcher are phosphate buffered saline (PBS) prepared in glass distilled H₂O; 0.1–1% hydrogen peroxide (H₂O₂) diluted in PBS, distilled H₂O or methanol (optional); primary antibody; any reagents needed to fix and/or deparaffinize specimens on slides; counterstain (optional); and mounting medium.

PREPARATION OF WORKING SOLUTIONS

Use only freshly prepared buffers. Prepare all working solutions in the mixing bottles provided. After preparation, insert the drop dispenser top (supplied in inverted position) into the cap in correct orientation. Place the entire unit on the bottle and twist the cap. The drop dispenser will snap into place. To remove the drop dispenser for refilling, press laterally with thumb until the top snaps off. To prevent evaporation, secure the caps on bottles when not in use. After completion of the staining procedure, working solutions should be discarded and mixing bottles washed with distilled H₂O.

- **Blocking serum:** In mixing bottle 1 (blue cap), combine 75 µl normal blocking serum stock with 5 ml PBS.
- **Biotinylated secondary antibody:** In mixing bottle 2 (green cap), combine 75 µl normal blocking serum stock, 5 ml PBS and 25 µl biotinylated secondary antibody stock.
- **AB enzyme reagent:** In AB mixing bottle (purple cap), combine 50 µl reagent A (avidin), 50 µl reagent B (biotinylated HRP) and 2.5 ml PBS. Mix and let stand for approximately 30 minutes.
- **Peroxidase substrate:** In substrate mixing bottle (yellow cap), combine 1.6 ml distilled H₂O, 5 drops 10x substrate buffer, 1 drop 50x DAB chromogen and 1 drop 50x peroxidase substrate. Sufficient for 15–20 slides.
- Refer to the Immunoperoxidase Staining Protocol at www.scbt.com for tissue section preparation and additional technique notes.

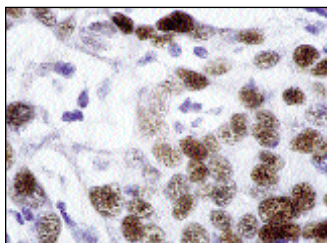
PROCEDURE

All steps are carried out at room temperature in a humidified chamber. Staining dishes or coplin jars may also be used. Apply sufficient volumes of reagents to completely cover the section; 100 µl is usually adequate, or 1–3 drops of working solutions. Use suction to remove reagents after each step, but avoid drying of specimens between steps.

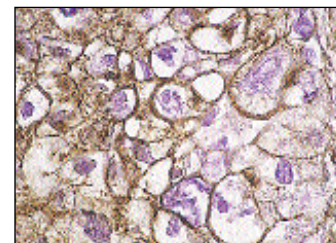
- **Optional:** After preparation of tissue sections, slides may be incubated for 5–10 minutes in 0.1–1% hydrogen peroxide diluted in PBS, deionized H₂O or methanol to quench endogenous peroxidase activity. Wash in PBS twice for 5 minutes each.
- **Optional:** Incubate sections for one hour in 1.5% blocking serum in PBS (mixing bottle 1). This step may be omitted if non-specific staining is not a problem. Blot excess blocking serum from slides.

- Incubate sections with primary antibody for 30 minutes at room temperature or overnight at 4° C. Optimal antibody concentration should be determined by titration; recommended range is 0.5–5.0 µg/ml, diluted in 1.5% blocking serum in PBS (from mixing bottle 1). Wash with three changes of PBS for 5 minutes each.
- Incubate sections for 30 minutes with biotinylated secondary antibody as prepared in mixing bottle 2 or at approximately 1 µg/ml. Wash with three changes of PBS for 5 minutes each.
- Incubate sections for 30 minutes with AB enzyme reagent (AB mixing bottle). Wash with three changes of PBS for 5 minutes each.
- Incubate sections in 1–3 drops peroxidase substrate (substrate mixing bottle) for 30 seconds–10 minutes or until desired stain intensity develops. The section may be checked for staining by rinsing with H₂O and viewing under a microscope. If necessary, add additional peroxidase substrate and continue to incubate. Wash sections in deionized H₂O for 5 minutes.
- **Optional:** Counterstain sections in Gill's formulation #2 hematoxylin for 5–10 seconds. Immediately wash with several changes of deionized H₂O.
- **Optional:** Destain with acid alcohol and bluing reagent. Wash with tap water.
- For paraffin-embedded tissue sections, dehydrate as follows: 2x 95% ethanol for 10 seconds each, 2x 100% ethanol for 10 seconds each, 3x xylenes for 10 seconds each. Wipe off excess xylenes.
- Immediately add 1–2 drops of permanent mounting medium and cover with a glass coverslip. Observe by light microscopy.

DATA



p53 (DO-1): sc-126. Immunoperoxidase staining of formalin-fixed, paraffin-embedded human breast carcinoma tissue. Note intense nuclear staining of ductal epithelia.



ICAM-1 (G-5): sc-8439. Immunoperoxidase staining of formalin-fixed, paraffin-embedded human colon carcinoma tissue showing membrane localization of ICAM-1.

STORAGE

Store all ABC Staining System components at 2–8° C. ****DO NOT FREEZE.**** Stable for one year from the date of shipment.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

MATERIAL SAFETY DATA SHEET

SECTION 1 - PRODUCT AND COMPANY INFORMATION

Component Name: Hydrogen Peroxide Solution

Supplier: Santa Cruz Biotechnology, Inc.
2145 Delaware Ave.
Santa Cruz, California 95060
800.457.3801 or 831.457.3800

Emergency: ChemWatch
Within the US & Canada: 877-715-9305
Outside the US & Canada: +800 2436 2255
(1-800-CHEMCALL) or call +613 9573 3112

SECTION 2 - COMPOSITION/INFORMATION ON INGREDIENT

<u>Substance Name</u>	<u>CAS #</u>	<u>SARA 313</u>
Hydrogen Peroxide-1802 (Solution in H2O) 90 atom% 180	-	NO

<u>Ingredient Name</u>	<u>CAS #</u>	<u>%</u>	<u>SARA 313</u>
Hydrogen Peroxide	7722-84-1	≤ 5	Yes
Water	7732-18-5	≥ 95	No

SECTION 3 - HAZARDS IDENTIFICATION

EMERGENCY OVERVIEW
Target organ(s): Eyes. Skin.

HMIS RATING
HEALTH: 0
FLAMMABILITY: 0
REACTIVITY: 1

NFPA RATING
HEALTH: 0
FLAMMABILITY: 0
REACTIVITY: 1

For additional information on toxicity, please refer to Section 11.

SECTION 4 - FIRST AID MEASURES

ORAL EXPOSURE
If swallowed, wash out mouth with water provided person is conscious. Call a physician.

INHALATION EXPOSURE
If inhaled, remove to fresh air. If breathing becomes difficult, call a physician.

DERMAL EXPOSURE
In case of contact, immediately wash skin with soap and copious amounts of water.

EYE EXPOSURE
In case of contact with eyes, flush with copious amounts of water for at least 15 minutes. Assure adequate flushing by separating the eyelids with fingers. Call a physician.

SECTION 5 - FIRE FIGHTING MEASURES

FLASH POINT
N/A

AUTOIGNITION TEMP
N/A

FLAMMABILITY
N/A

EXTINGUISHING MEDIA

Suitable: Water spray. Carbon dioxide, dry chemical powder, or appropriate foam.

FIREFIGHTING

Protective Equipment: Wear self-contained breathing apparatus and protective clothing to prevent contact with skin and eyes.
Specific Hazard(s): Emits toxic fumes under fire conditions.

SECTION 6 - ACCIDENTAL RELEASE MEASURES

METHODS FOR CLEANING UP
Absorb on sand or vermiculite and place in closed containers for disposal.
Ventilate area and wash spill site after material pickup is complete.

SECTION 7 - HANDLING AND STORAGE

HANDLING
User Exposure: Avoid inhalation. Avoid contact with eyes, skin, and clothing.
Avoid prolonged or repeated exposure.

STORAGE
Suitable: Keep tightly closed. Store at 2-8°C

SPECIAL REQUIREMENTS
Light sensitive.

SECTION 8 - EXPOSURE CONTROLS / PPE

ENGINEERING CONTROLS
Safety shower and eye bath. Mechanical exhaust required.

PERSONAL PROTECTIVE EQUIPMENT
Respiratory: Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU). Respiratory protection is not required. Where protection is desired, use multi-purpose combination (US) or type ABEK (EN 14387) respirator cartridges.
Hand: Protective gloves.
Eye: Chemical safety goggles.

GENERAL HYGIENE MEASURES
Wash thoroughly after handling.

EXPOSURE LIMITS	Country Source	Type	Value
Poland	NDS		1.5 MG/M3
Poland	NDS Ch		4 MG/M3
Poland	NDSP		-

SECTION 9 - PHYSICAL/CHEMICAL PROPERTIES

Form	liquid	
pH	N/A	
BP/BP Range	N/A	
MP/MF Range	N/A	
Freezing Point	N/A	
Vapor Pressure	23.3 mmHg	30 °C
Vapor Density	1.1 g/l	
Saturated Vapor Conc.	N/A	
SG/Density	1 g/cm3	
Bulk Density	N/A	
Odor Threshold	N/A	
Volatile%	N/A	
VOC Content	N/A	
Water Content	N/A	
Solvent Content	N/A	
Evaporation Rate	N/A	
Viscosity	N/A	

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Surface TensionN/A
Partition CoefficientN/A
Decomposition Temp.N/A
Flash PointN/A
Explosion LimitsN/A
FlammabilityN/A
Autoignition TempN/A
Refractive Index1.335
Optical RotationN/A
Miscellaneous DataN/A
SolubilityN/A
N/A = not available

SECTION 10 - STABILITY AND REACTIVITY

STABILITY

Stable: Stable.
Conditions to Avoid: Rust. Light.
Materials to Avoid: Brass Iron and iron salts., Finely powdered metals Copper, Copper alloys, Galvanized iron, Zinc, Nickel

HAZARDOUS DECOMPOSITION PRODUCTS

Hazardous Decomposition Products: Oxygen.

HAZARDOUS POLYMERIZATION

Hazardous Polymerization: Will not occur

SECTION 11 - TOXICOLOGICAL INFORMATION

ROUTE OF EXPOSURE

Skin Contact: May cause skin irritation.
Skin Absorption: May be harmful if absorbed through the skin.
Eye Contact: May cause eye irritation.
Inhalation: Material may be irritating to mucous membranes and upper respiratory tract. May be harmful if inhaled.
Ingestion: May be harmful if swallowed.

TARGET ORGAN(S) OR SYSTEM(S)

Eyes. Respiratory system. Skin.

SIGNS AND SYMPTOMS OF EXPOSURE

To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

SECTION 12 - ECOLOGICAL INFORMATION

No data available.

SECTION 13 - DISPOSAL CONSIDERATIONS

APPROPRIATE METHOD OF DISPOSAL OF SUBSTANCE OR PREPARATION

Contact a licensed professional waste disposal service to dispose of this material. Dissolve or mix the material with a combustible solvent and burn in a chemical incinerator equipped with an afterburner and scrubber. Observe all federal, state, and local environmental regulations.

SECTION 14 - TRANSPORT INFORMATION

DOT

Proper Shipping Name: None
Non-Hazardous for Transport: This substance is considered to be non-hazardous for transport.

IATA

Non-Hazardous for Air Transport: Non-hazardous for air transport.

SECTION 15 - REGULATORY INFORMATION

US CLASSIFICATION AND LABEL TEXT

US Statements: Target organ(s): Eyes. Skin.

UNITED STATES REGULATORY INFORMATION

SARA LISTED: No

CANADA REGULATORY INFORMATION

WHMIS Classification: This product has been classified in accordance with the hazard criteria of the CPR, and the MSDS contains all the information required by the CPR.

DSL: No

NDSL: No

SECTION 16 - OTHER INFORMATION

The above information is believed to be correct but does not purport to be complete and should be used only as a guide. The burden of safe use of this material rests entirely with the user.

2/15/2011

MATERIAL SAFETY DATA SHEET

PRODUCT AND COMPANY INFORMATION

Component Name: DAB 50X Concentrate Liquid Horseradish Peroxidase (HRP)
Substrate System

Supplier: Santa Cruz Biotechnology, Inc.
2145 Delaware Ave.
Santa Cruz, California 95060
800.457.3801 or 831.457.3800

Emergency: ChemWatch
Within the US & Canada: 877-715-9305
Outside the US & Canada: +800 2436 2255
(1-800-CHEMCALL) or call +613 9573 3112

1. HAZARDOUS INGREDIENTS

REAGENT: 3,3'-Diaminobenzidine Tetrahydrochloride
WEIGHT: 360.1 g/mole
FORMULA: C₁₂H₁₄N₄

TOXIC: MAY CAUSE CANCER. POSSIBLE MUTAGEN. AVOID ALL CONTACT.

NATURE OF HAZARD: Harmful if swallowed, inhaled, or absorbed through skin. May cause skin irritation.

2. PHYSICAL DATA

APPEARANCE: Clear amber solution.

MELTING/FREEZING POINT: 0° C (water)

BOILING POINT: 100° C (water)

SOLUBILITY: Water.

3. FIRE AND EXPLOSION HAZARD DATA

EXTINGUISHING MEDIA: Water, Foam, Dry Chemical, CO₂

SPECIAL FIRE FIGHTING PROCEDURES: Use respirator and protective clothing. Toxic fumes could be emitted when water is evaporated with fire.

4. HEALTH HAZARD DATA

ACUTE EFFECTS: Harmful if swallowed, inhaled, or absorbed through skin. may cause irritation.

CHRONIC EFFECTS: Suspected carcinogen. Lab experiments have shown mutagenic effects. To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

FIRST AID: In case of contact, immediately flush eyes or skin with copious amounts of water for at least 15 minutes while removing contaminated clothing and shoes. Discard contaminated clothing and shoes. If swallowed, wash out mouth with water provided person is conscious.

EFFECTS OF OVEREXPOSURE:

INHALATION: Adverse health effects from vapors and spray mists in poorly ventilated areas may include irritation of the mucous membranes of the nose, throat, respiratory tract and symptoms of headache and nausea.

SKIN CONTACT: Avoid all contact. May cause cancer.

EYE CONTACT: Avoid all contact.

5. EMERGENCY AND FIRST AID PROCEDURES

SEE FIRST AID ABOVE.

6. REACTIVITY DATA

STABILITY: Stable

HAZARDOUS DECONTAMINATION PRODUCTS: Carbon monoxide and carbon dioxide.

HAZARDOUS POLYMERIZATION: Will not occur.

INCOMPATIBILITY MATERIALS TO AVOID: Strong oxidizing agents and strong bases.

7. SPILL OR LEAK PROCEDURE

STEPS TO BE TAKEN IN CASE MATERIAL IS RELEASED OR SPILLED: Contain spill then clean up with copious amounts of soap and water containing 20% bleach. Avoid contact with skin or clothing.

WASTE DISPOSAL METHODS: Observe all Federal, State, and Local laws concerning health and pollution. Dilute with equal amounts of bleach and flush with copious amounts of water.

8. SPECIAL PROTECTION INFORMATION

Good housekeeping procedures and laboratory practice is best preventative. Use in well ventilated areas. Store in refrigerated conditions. Do not allow product to enter storm or sanitary sewers, lakes, rivers, streams, or public water supplies. Notify local authorities if this happens or is threatened.

9. OTHER INFORMATION

The above information is believed to be correct but does not purport to be complete and should be used only as a guide. The burden of safe use of this material rests entirely with the user.

2/15/2011