# abcam

## Product datasheet

# **AEC Substrate Kit ab64252**

13 References 3 Images

Overview

**Product name** 

**Tested applications** 

**General notes** 

AEC Substrate Kit

Suitable for: IHC-P

AEC substrate kit for use in IHC using HRP / peroxidase detection. Ready to use formulation.

Previously called AEC Substrate System (Ready to Use).

#### IHC protocol suitable for use with AEC Substrate Kit ab64252:

- 1. Deparaffinize and rehydrate formalin-fixed paraffin-embedded tissue section.
- 2. Use appropriate <u>antigen retrieval buffer or enzyme</u> (primary antibody dependent) to treat sections. Wash 3 times in buffer.
- 3. Add enough <u>hydrogen peroxide blocking solution</u> to cover the sections. Incubate for 10 minutes. Wash 2 times in buffer. If necessary, use <u>avidin biotin blocking</u>.
- 4. Apply <u>protein block</u> (or <u>normal serum</u> from same species as secondary antibody) and incubate for 5 minutes at room temperature to block nonspecific background staining. Wash once in buffer.
- 5. Apply primary antibody in **antibody diluent** and incubate.
- 6. Wash 4 times in buffer. Incubate slide with <u>biotinylated secondary antibody</u> (or <u>HRP</u> <u>polymer secondary antibody</u> and skip step 7). Wash 4 times in buffer.
- 7. Apply **streptavidin-HRP** and incubate for 10 minutes at room temperature.
- 8. Rinse 4 times in buffer. Place slide in <u>DAB substrate</u> or AEC Substrate ab64252 and incubate until desired color is achieved (1-10 mins, recommended 10 mins for AEC). Rinse 4 times in buffer.
- 9. Add enough drops of **hematoxylin** to cover the section. Incubate for 1 minute.
- 10. Rinse 7-8 times in tap water. Add **mounting medium** to cover the section.

Find complete IHC kits, and reagents for antigen retrieval, blocking, signal amplification, visualization, counterstaining, and mounting in the IHC kits and reagents guide.

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Pι	ro	pe	rti	es

Form Liquid

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Storage instructions

Store at +4°C.

Storage buffer

pH: 4.7

Constituents: 0.03% Hydrogen peroxide, 0.1% 3-amino-9-ethyl carbazole, 3% Dimethyl

formamide

stabilizers

Relevance

AEC (3-Amino-9-Ethylcarbazole) is a widely used chromogen for immunohistochemical staining. AEC produces a red end product that is soluble in alcohol and must be used with an aqueous counterstain and mounting media. AEC produces a clean, very intense stain that is free of

unwanted background.

#### **Applications**

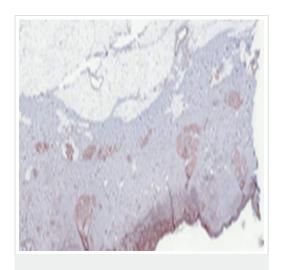
#### The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab64252 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use at an assay dependent dilution.

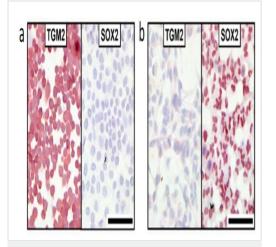
#### **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - AEC Substrate Kit (ab64252)

Image from Kryza et al. PLoS One; 11(2):e0149387. doi: 10.1371/journal.pone.0149387. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

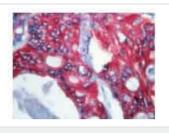
Immunostaining using anti-hMMP-9 murine monoclonal antibody. Slides of superficial spread melanoma tumors were incubated with the hMMP-9 antibody (ab58803, 1:300). Alkanie phosphatas esecondary antibody was stained with AEC (3-Amino-9-EthylCarbazole, ab 64252, Abcam, Cambridge) for 15 min and slides were counterstained with hematoxylin for examination.



Immunostaining for TGM2 and SOX2 staining in GSC 158 and GSC 157 cells.(a) Positive TGM2 and negative SOX2 staining in GSC 158 cells. (b) Negative TGM2 and positive SOX2 staining in GSC 157 cells. Cells were incubated with primary monoclonal Abs for 1 h at +37 °C (for SOX2, **ab6734**, 1/200) following the Lab Vision Ready-To-Use AEC Substrate System (ab64252) according to the manufacturer's instructions.

Immunohistochemistry (Frozen sections) - AEC Substrate Kit (ab64252)

Image from Trus et al., Viruses; 12(5):579. doi: 10.3390/v12050579. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - AEC Substrate System (Ready to Use) (ab64252)

Human colon carcinoma stained with anti-Keratin 20 antibody using AEC Substrate System (Ready to Use) (ab64252).

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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