Product datasheet **MON-APP209**

AEC Single Solution

Reagents

Instructions for use

Product name	AEC Single Solution
Intended Use	AEC Single Solution is a ready-to-use solution intended for immunohistochemical and in situ-hybridisation staining procedures with horse radish peroxidase (HRP). AEC (3-Amino-9-ethylcarbazol) leads to the formation of a red-brown precipitate at the location of the target antigen or target nucleic acid. The precipitate is insoluble in aqueous mounting media and can be observed by light microscopy. AEC Single Solution is especially useful when a high sensitivity is desired.
Applications	IHC-P, IHC-Fr, IF
Summary and explanation	-

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES

Instructions for use

Principle of method -

Reagents provided

100 ml AEC Single Solution (ready-to-use)
2 Dropper Bottles

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES

Instructions for use

Storage and handling

The solution should be stored at 2-8°C without furt her dilution. Please store the reagent in a dark place and do not freeze it. Under these conditions the solution is stable up to the expiry date indicated on the label. Do not use product after the expiry date. AEC Single Solution is a ready-to-use solution. Preparation of a working solution as in other chromogenic substrates is not necessary. A positive and a negative control have to be carried out in parallel to the test material. If you observe unusual staining or other deviations from the expected results which could possibly be caused by the kit reagents please contact our technical support.

Reagent preparation

The solution is ready-to-use. AEC Single Solution can be used directly from the refrigerator and should be stored again at 2-8°C aft er use. When using the small package (8 ml) please directly drop from the bottle. When using the 100 ml package (MON-APP209) please transfer up to 8 ml of the AEC Single Solution into one of the provided dropper bottles. The transferred solution is stable for many weeks if stored at 2-8°C. The volume required for several staining runs should be transferred so that the 100 ml stock bottle has to be opened only a few times. If you would like to pipette the

Procedure

1) Rinse the slide with wash buffer after the previous incubation step. 2) Apply the AEC Single Solution to the slide. Incubate for 3-6 minutes. (Incubation time can be extended up to 30 minutes, if desired.) 3) Rinse with distilled H2O. 4) Counterstain with haematoxylin for about 30 seconds up to 5 minutes (depending on the desired staining intensity). 5) Rinse with distilled H2O. 6) Blueing in tap water for at least 5 minutes. 7) Mount with an aqueous mounting medium.

Expected results

During the reaction of the substrate with horse radish peroxidase in presence of the chromogen AEC, a red-brown precipitate is formed at the location of the target antigen or nucleic acid. The precipitate is insoluble in aqueous solvents and can be observed by light microscopy.

Trouble shooting

If you observe unusual staining or other deviations from the expected results please read these instructions carefully, contact our technical support. Also refer to the instructions of the detection systems for guidance on general troubleshooting.

Instructions for use

Quality control

We recommend carrying out a positive and a negative control with every staining run. The positive control permits the validation of appropriate processing of the sample. If the negative control has a positive result, this points to unspecific staining. Please refer to the instructions of the detection system for guidance on general quality control procedures.

Performance

Studies have been conducted to evaluate the performance of the kit reagents. The product has been found to be suitable for the intended use

Limitations of procedure

Immunohistochemistry is a complex method in which histological as well as immunological detection methods are combined. Tissue processing and handling prior to immunostaining, for example variations in fixation and embedding or the inherent nature of the tissue can cause inconsistent results (Nadji and Morales, 1983). In some tissues endogenous peroxidase activity may cause non-specific staining. The enzyme activity should be blocked by incubation with hydrogen peroxide solution (H2O2 solution). The step is carried out before incubation with primary antibody but after dewaxing and rehydration. Background staining due to endogenous biotin can be blocked through an avidin-biotin blocking step prior to the primary antibody incubation step. The coloured precipitate formed by AEC is soluble in organic solvents. The tissue sections therefore have to be counterstained with aqueous solutions (e. g. Gill's or Mayer's haematoxylin) and mounted with aqueous mounting media. The colour intensity of the reaction product can decrease with time, especially when exposed to light. The staining reaction itself can be influenced in the same way when carried out in strong light. Sanbio guarantees that the product will meet all requirements described from its shipping date until its expiry date, as long

Precautions

Use by qualified personnel only. AEC (3-Amino-9-ethylcarbazol) and the solvents used are considered hazardous materials. Material safety data sheets (MSDS) are available upon request. Wear protective clothing to avoid contact of reagent or specimen with eye, skin or mucous membrane. In case of reagent or specimen coming into contact with a sensitive area, wash the area with large amounts of water. Oxidising substances, e. g. metals, dust, bacteria or glass devices can influence the stability of AEC Single Solution. Such contaminations have to be avoided. Non-consumed solution needs to be discarded as dangerous substance.

References

1. Elias JM Immunohistopathology – A practical Approach to Diagnosis ASCP Pr
2 Nadji M and Morales AR Ann N.Y. Acad Sci 420:134-139, 1983
3. -

FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES