

ANTI-CONJUGATED ROTENONE ANTIBODIES
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CATALOG NUMBER : AP115

TARGET : Conjugated Rotenone

IMMUNOGEN : Synthetic Rotenone conjugated to protein carriers

RAISED IN : Rat

CLONALITY : Polyclonal

ISOTYPE : IgG

PURITY : Antiserum previously absorbed on protein carriers, and purified

FORM : Lyophilized

RESEARCH AREAS : Environment and food processing

STORAGE INSTRUCTIONS: After reconstitution with 50µl of distilled water and 50µl of glycerol, the aliquot can be repeated freezed (up to five times), and stable at least 2 years.

CORRESPONDING ANTIGEN :

Gemacbio sells the corresponding antigen : Rotenone conjugate (code number : AG115)

EXAMPLES OF MATERIAL AND METHODS

• Example of immunohistochemistry protocol

Perfusion (Example for Adult male Sprague Dawley (weight around 0.5 kg)).

- 1-The animals can be deeply anaesthetized (for example with urethane-0.5-1.5g/kg, intraperitoneal).
- 2-Perfused via the ascending aorta with 50 ml of NaCl 9g/l (Heparinized) and pass through the system 800-1000 ml of cold 4% paraformaldehyde (Merck) in 0.1 M PB, pH 7.2-7.4, (ten minutes).
- 3-Dissect out the organs and place in a solution of 4% paraformaldehyde in 0.1M PB, pH 7.2, at 4°C for twelve to sixteen hours.

Example of immunohistochemical protocol

- 1-In order to avoid possible interference with endogenous peroxidase, free-floating sections will be treated with distilled water containing NH₃ (20%), H₂O₂ (30%) and NaOH (1%) for 20 min (other method is using a solution with 33% of H₂O₂ and 66% of methanol).
- 2-Then, wash the sections for 20 min in 0.15 M phosphate-buffered saline (PBS) (pH 7.2)
- 3-Pre-incubate for 30 min in PBS containing 10% of normal horse serum and 0.3% of Triton X-100 (mixed solution).
- 4-Incubate at room temperature (1h 30min) and overnight at 4° C in the same mixed solution containing anti-conjugated Rotenone antibodies (diluted 1/1,000–1/5,000; as recommended dilutions).
- 5-Then, the sections will be wash in PBS (30 min).
- 6-After that we will incubate for 60 min at room temperature with biotinylated anti-rat immunoglobulin (Vector) diluted 1/200 in PBS.
- 7-Wash during 30 min with PBS.
- 8-Sections will be incubated for 1 h with a 1/100 diluted avidin-biotin-peroxidase complex (Vectastain).
- 9-After that we will wash the sections in PBS (30 min)
- 10-Wash with Tris-HCl buffer (pH 7.6)(10 min).
- 11-The tissue-bound peroxidase will be developed with H₂O₂ using 3, 3' diaminobenzidine as chromogen.
- 12-Finally the sections will be rinsed with PBS and coverslipped with PBS/Glycerol (1/1).