R&D ® Ab

Research & Diagnostic Antibodies

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Rabbit Anti-D_2 Receptor Sera

AS-3526S Lot # 7047

The antiserum was raised in a rabbit which was immunized with D_2(272-282) covalently attached onto a carrier protein, and it has been characterized by immunocytochemical, western immunoblot and ELISA techniques. The antiserum has been found to be highly specific for this peptide sequence and is suitable for immunocytochemical detection of both the short and the long forms of the D_2 dopamine receptor. Rehydrate the antibody with 0.1 ml of PBS which contains 10 mg/ml BSA or with additional buffer (such as 10 mg/ml BSA in PBS) for more dilute antibody.

Antiserum Specificity

Polypeptide	% Cross Reactivity
D_2 Dopamine Receptor(272-282)	100
D_{2S} Dopamine Receptor	~ 70
D_{2L} Dopamine Receptor	~ 60
D_1 Dopamine Receptor(9-21)	0
D_1 Dopamine Receptor	0
D_3 Dopamine Receptor(2-10)	0
D_3 Dopamine Receptor	0
D_4 Dopamine Receptor(176-185)	0
D_4 Dopamine Receptor	0
D_5 Dopamine Receptor(23-35)	0
D_5 Dopamine Receptor	0

Immunocytochemical Staining

This antiserum has been found to stain specific cells in various regions of PLP fixed rat brain sections at 1:5000 dilution. For additional information on PLP fixative see the FAQ page on our web site at www.RDAbs.com.

Western Immunoblot

Western immunoblots using whole rat brain homogenate resulted in a pair of bands being detected at ~48 and ~51 kD at a 1:800 dilution.

Western Blotting Protocol

1. After SDS-PAGE (on either 4-15% gradient gels or single percentage gels, such at 12% gels) and electrophoretic transfer to PVDF membrane, block the membrane overnight with 2% normal goat serum in TBS/Tween-20 buffer.
2. Wash x 2 with TBS/Tween-20.
3. Apply the rabbit polyclonal antibody after dilution to at least 1:800 (Note: higher dilutions may be needed). Use 2% normal goat serum in TBS/Tween-20 as buffer for the primary antibody. Let the primary antibody bind for 2-4 hours.
4. Wash x 3 with TBS/Tween-20.
5. Apply affinity purified HRP-goat anti-rabbit IgG antiserum diluted 1:2500 (dilution may vary depending upon supplier) in 2% normal goat serum in TBS/Tween-20. Incubate 1-2 hours. Note: greater sensitivity may be achieved using ABC techniques.
6. Wash x 4 for 5 minute/wash with TBS/Tween-20.
7. Develop color using the enhanced DAB reaction.