

IgG Fraction of Rabbit Anti-hPTH(1-34) Serum

AS-717

Lot # 10140

This IgG fraction was prepared by precipitation, dialysis and chromatography from an antiserum that was raised in a rabbit which was immunized with hPTH(1-34) covalently attached onto a carrier protein. The antiserum has been shown to be specific for hPTH(1-34) and was characterized by western blotting, ELISA, RIA and cell staining techniques. This IgG fraction is suitable for immunocytochemical and western immunoblotting detection of hPTH. Dilute the lyophilized antibody with 0.1 ml of 10 mg/ml BSA in PBS for the equivalent concentration in whole antiserum, or with additional buffer for more dilute antibody.

Each vial contains 2.38 mg of purified IgG.

Antiserum Specificity

Polypeptide	% Cross Reactivity
PTH(1-34) (human)	100
PTH(1-84) (human)	100
PTH(1-84) (bovine)	80
PTHrp(1-35)	< 0.1
ANP	0
ACTH(1-39) (human)	0
VIP	0

Immunocytochemical Staining

This IgG fraction has been found to stain formaldehyde fixed sections known to express PTH at a concentration of 50 µg/ml.

Western Blotting Protocol

1. After SDS-PAGE on a 15% gel 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 IgG fraction after dilution to at least 25 µg/ml (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.