

Rabbit Anti-Human Protein Kinase C ζ [480-492] Serum

AS-2463G

Lot # 2584

The antiserum was raised in a rabbit which was immunized with synthetic Protein Kinase C zeta [480-492] covalently attached onto a carrier protein. The IgG fraction of the rabbit antiserum was prepared by precipitation, dialysis, and chromatography. Rehydrate the lyophilized antibody with 0.1 ml of 10 mg/ml BSA in PBS to achieve an IgG concentration equivalent to whole antiserum or with additional buffer for more dilute antiserum. This IgG fraction has been found to stain specifically fixed cultured cells by indirect immunofluorescence. The antibody was tested for recognition of other Protein Kinase C isozymes by ELISA techniques.

Antiserum Specificity

Polypeptide	% Cross Reactivity
Protein Kinase C ξ [480-492] (rat)	100
Protein Kinase C α [664-672]	0
Protein Kinase C β 1 [662-671]	0
Protein Kinase C β 2 [660-673]	0
Protein Kinase C γ [681-689]	0
Protein Kinase C δ [662-673]	0
Protein Kinase C ϵ [728-737]	0
Protein Kinase C θ [700-706]	0
Protein Kinase C η [676-683]	0
ACTH (human, 1-39)	0
ANP (human)	0
Calcitonin (human)	0
Somatostatin 28	0
Vasoactive Intestinal Peptide	0

Western Blotting Protocol

1. After SDS-PAGE (on either 4-15% gradient gels or single percentage gels, such as 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 IgG fraction after diluting to at least 500 ng/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.