

IgG Fraction of Rabbit Anti-Human Protein Kinase C θ [700-706] Serum
AS-2483G Lot # 5837

The antiserum was raised in a rabbit which was immunized with synthetic Protein Kinase C theta [700-706] (human, rat) covalently attached onto a carrier protein. Rehydrate the lyophilized antiserum with 0.1 ml of 10 mg/ml BSA in PBS for the equivalent of whole antiserum, or with additional buffer for more dilute antiserum. This antiserum has been found to stain specifically fixed cultured cells by indirect immunofluorescence. The antiserum was tested for recognition of other Protein Kinase C isozymes by ELISA techniques.

Antiserum Specificity

Polypeptide	% Cross Reactivity
Protein Kinase C θ [700-706] (human, 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 ξ [480-492]	0
Protein Kinase C η [676-683]	0
ACTH (human, 1-39)	0
ANP (human)	0
Calcitonin (human)	0

Western Blotting Protocol

1. After SDS-PAGE on 10% percent gels and electrophoretic transfer to PVDF membrane, block the membrane overnight with 4% normal goat serum in TBS/Tween-20 buffer.
2. Wash x 2 with TBS/Tween-20.
3. Apply the rabbit antibody after preparing a 1:500 dilution. Use 2% normal goat serum in TBS/Tween-20. 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 for 5 min x 4 in TBS/Tween-20.
7. Develop color using the enhanced DAB reaction.