Arachidonoyl Substrate Specificity Of Diacylglycerol Kinases

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Diacylglycerol kinases (DGKs) are an important class of lipid signaling enzymes. DGKε is unique among DGK isoforms in having specificity for DAG substrates with an arachidonate moiety. This contributes to the enrichment of lipid intermediates in the PI-cycle with arachidonate. The importance of DGKε in neuronal function has been demonstrated in studies with knockout mice.

We have shown that removal of 58 residues from the amino terminus of this protein, including the membrane-inserting segment, had a negligible effect on substrate specificity. It is intriguing that the domain, responsible for DGKε specificity for DAG substrates with an arachidonate moiety, still is unknown.

We have now identified a region of DGKε containing four conserved residues, very similar to those found responsible for recognition of arachidonic acid in lipoxygenases. We demonstrated that several mutations within this region of DGKε significantly impact enzyme activity, decreasing it to less than 2% of the activity of wild-type DGKε. Based on our results, we propose a model of the substrate-binding pocket located in the accessory domain of DGKε. The relationship of this finding to the substrate specificity of other isoforms of DGK is also explored.