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# Sphingomyelin Synthase and PKC Activation

In their review of sphingolipids as regulators of cell function, Y. A. Hannun and R. M. Bell (*1*) discuss the hypothesis that some of the reactions involved in sphingolipid metabolism may regulate protein kinase C (PKC) activity through the generation or removal of PKC-inhibiting metabolites. An important addition to the pathways considered in (*1*) is the reaction catalyzed by phosphatidylcholine:ceramide phosphocholine transferase (sphingomyelin synthase). This enzyme transfers the phosphorylcholine head group from the phospholipid phosphatidylcholine (PC) to ceramide, yielding sphingomyelin and diacylglycerol (DAG) (Fig. 1). Since DAG is a known activator of PKC, the action of sphingomyelin synthase allows for a mechanism by which metabolism of sphingolipids can result in stimulation of PKC (through production of DAG).

The sphingomyelin synthase reaction is the major route of sphingomyelin synthesis in a number of biological systems, and a substantial fraction of the enzyme activity appears to be associated with the plasma membrane (*2*). Hence, the high activity, products, and location of sphingomyelin synthase are all consistent with a possible role in PKC regulation. Two examples of how sphingomyelin synthase might physiologically affect PKC activity are as follows. (i) The conversion of sphingosine to sphingomyelin, by acylation and subsequent sphingomyelin synthase–catalyzed head group transfer, would cause the net loss of a PKC inhibitor (sphingosine) and the gain of a PKC activator (DAG) ( Fig. 1). Such bimodal regulation might result in steep activation curves and "on-off" stimulation of PKC. This two-step pathway should also be considered when one interprets data from long-term experiments (more than 6 hours) with pharmacological concentrations of sphingosine, where significant metabolism is known to have occurred (*1*). (ii) The sequential action of sphingomyelin synthase (*2*) and neutral- or acid-sphingomyelinase (*3*) yields a two-step cycle in which ceramide is consumed and regenerated with the concomitant conversion of PC into phosphorylcholine and DAG (Fig. 1). In some instances, DAG may stimulate sphingomyelinase activity (*4*) or sphingomyelin synthase activity (*5*). If these observations are generally true, then the cycle could amplify a transient increase in DAG caused by receptor-mediated events by generating more DAG from the abundant stores of PC. A growing conviction that PC is a source of DAG in numerous signaling events (*6*) warrants further consideration of this cycle.

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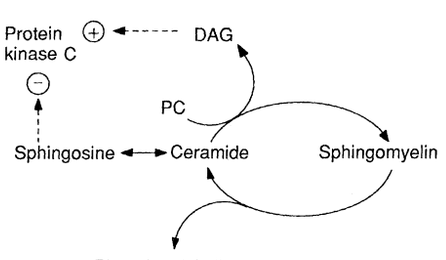


**Fig. 1.** Proposed dual role of sphingolipid metabolism in PKC regulation.

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*Response*: The "transferase" pathway of sphingomyelin synthesis is of proven significance for sphingomyelin. Its physiologic role, however, is at present unknown. The comment by Hampton and Morand raises the interesting possibility that sphingomyelin and diacylglycerol production are simultaneously controlled. A number of investigators have looked into this question [for example, (*1*)], and they have not found the "transferase" pathway to be physiologically regulated. However, since the field of physiologic studies of sphingolipid turnover is in its infancy, all options should be evaluated and merit scientific discussion.

*Note added in proof*: Recent studies on sphingomyelin turnover have defined metabolic pathways for regulated sphingomyelin hydrolysis and regeneration (*2*).

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# Rab 12 kD

C. E. Brinckerhoff *et al.* report the "Autocrine induction of collagenase by serum amyloid A–like and $\beta_2$-microglobulin–like proteins" (*1*). These authors have found that stimulation of rabbit fibroblasts with agents such as phorbol myristate acetate induces production of "autocrine proteins that, by themselves, can act on the fibroblasts to stimulate collagenase production." Isolation and $NH_2$-terminal amino acid sequencing of autocrine proteins of 14 kD and 12 kD revealed on "computer searching of the data base" respective homologies with human serum amyloid A and human $\beta_2$-microglobulin.

In the case of the "$\beta_2$-microglobulin-like" protein, additional insight is obtained by more extensive examination of the literature. The "Complete amino acid sequence of rabbit $\beta_2$-microglobulin" reported by Kindt and colleagues in 1979 (*2*) is identical at 16 of 18 positions determined for the Rab 12-kD autocrine protein. Furthermore, the residues at the two discrepant positions, 6 and 10, may also be identical. Deamidation of acid amides is a recognized problem in protein sequencing and could have resulted in assignment of aspartic acid at position 6 of Rab 12 kD (*1*) compared to asparagine in rabbit $\beta_2$-m (*2*). The other apparent difference is at position 10, which is tyrosine in