Friday, July 17, 1981

Symposium XIII

Membrane Phospholipids

15:30–17:30 h

Grand Ballroom East

0997

REGULATION OF THE FATTY ACID COMPOSITION OF PLATELET PHOSPHOLIPIDS. M.L. McKean, J.B. Smith and M.J. Silver. Cardeza Foundation and the Department of Pharmacology, Thomas Jefferson University, Philadelphia, PA. 19107

The fatty acid composition of cell membrane phospholipids does not remain constant after de novo biosynthesis, but undergoes continual remodelling. One of the major routes for remodelling probably includes the deacylation-reacylation steps of the Lands Pathway. This has been shown to be important for the incorporation of long chain, polyunsaturated fatty acids into phospholipids by liver and brain. An understanding of the mechanisms involved in these processes in platelets is especially important in light of the large stores of arachidonic acid (AA) in platelet phospholipids and the role of AA in hemostasis and thrombosis. Previous results from this laboratory have shown that the turnover of radioactive AA, 8,11,14-eicosatrienoic and 5,8,11,14,17-eicosapentaenoic acids in the phospholipids of resting platelets is more rapid than the turnover of radioactive C_{16} and C_{18} saturated and unsaturated fatty acids. However, little is known about how fatty acids, especially AA and its homologues, are incorporated into platelet phospholipids during de novo biosynthesis or how they are exchanged during remodelling. At least three enzymes are involved in the deacylation-reacylation of phospholipids: phospholipase A_2; acyl CoA synthetase; and acyl CoA transferase. We have studied acyl CoA transferase and have found considerable activity in human platelet membranes. Experiments are in progress to determine the substrate specificity and other properties of this enzyme.

0998

CHANGES IN ARACHIDONATE-CONTAINING COMPLEX LIPID DURING PLATELET STIMULATION. S. Rittenhouse-Simmons. Departments of Hematology/Biochemistry, Brigham-Women's Hospital/ Harvard Medical School, Boston MA. U.S.A.

We have examined changes in the ^{3}H-arachidonic acid (^{3}HC20:4) content of phospholipid following stimulation of human platelets by thrombin, ionophore A23187, or collagen. Resolution of lipids and lipid metabolites was achieved using HPLC and thin-layer chromatography after extraction of platelets by a modified Bligh and Dyer technique. The most rapid changes, leading to formation of TXB_2, occur in response to thrombin and A23187. Both stimuli induce marked losses of ^{3}HC20:4 from phosphatidylcholine (PC) and phosphatidylinositol (PI) within 30 sec. Collagen also induces losses in PI and PC and formation of diglyceride (DG), which become appreciable only after 1-3 min. Whereas thrombin and A23187 exhibit similarities in terms of the rate of response, these agents differ with regard to the phospholipases activated. Exposure of platelets to A23187 causes some activation of Ca^{+2}-dependent PI-specific phospholipase C (PI-PLC); however, A23187 is an inefficient stimulus for this enzyme. Ionophore induces the formation of 1/4 to 1/6 as much PI-PLC-derived DG as does thrombin when comparable amounts of PI are hydrolyzed. A similar discrepancy is found with respect to generation of phosphatidic acid. Simultaneous addition of A23187 and thrombin does not impair the formation and metabolism of DG promoted by thrombin alone. Further, whereas indomethacin (1-100 µg/ml) exerts no inhibitory effects on PLC or the formation of DG in stimulated platelets, indomethacin does inhibit 1) the loss of ^{3}HC20:4 from PC in response to thrombin and 2) the loss of ^{3}HC20:4 from PC and PI in response to A23187. Indomethacin has been reported to inhibit platelet phospholipase A_2. It is clear from these studies that the activation of PI-PLC is not triggered merely by a general Ca^{+2} flux. In contrast, a shift in platelet Ca^{+2} stores appears to be a sufficient stimulus for phospholipase A.