

STRONTIUM IONS STIMULATE THE METABOLISM OF PHOSPHOINOSITIDES IN HUMAN BLOOD PLATELETS. L.C. Best, E.A. Bone and R.G.G. Russell, Dept. Human Metabolism & Clinical Biochemistry, University of Sheffield Medical School, Beech Hill Road, Sheffield S10 2RX, UK.

We have previously shown that strontium ions (Sr^{2+}) cause platelet 5-hydroxytryptamine secretion and thromboxane (TxB_2) production in parallel. Here, we have further investigated the mechanism by which Sr^{2+} causes platelet TxB_2 production and platelet activation by examining phospholipid metabolism in washed human platelets in response to Sr^{2+} . Platelet phospholipid₃ pools were labelled by preincubation with ^3H -glycerol. Phospholipids and neutral lipids were extracted with chloroform-methanol and resolved by two-dimensional thin layer chromatography. Sr^{2+} (1-8mM) produced a marked depletion in platelet phosphatidyl inositol (PI) with a maximal effect at 10 minutes. The major products corresponded to polyphosphoinositides (PPIs) and lysophosphatidyl inositol (LPI). No significant changes in other phospholipids were detected. Similarly, no consistent effects were observed on mono or diglyceride levels. PI break-down in response to Sr^{2+} was inhibited by Ca^{2+} , prostaglandin E_1 , and trifluoperazine, suggesting the possible involvement of calmodulin in Sr^{2+} action. PI break-down was also partially inhibited by aspirin and indomethacin but not by imidazole. We suggest that Sr^{2+} induces platelet secretion and TxB_2 production by causing the specific break down of PI, presumably by activating a phospholipase. The significance of the observed effect upon PPI metabolism is unknown, although PPI turnover may be closely associated with the control of calcium fluxes. Thus, Sr^{2+} may mimic a rise in cytosol Ca^{2+} in the platelet which is thought to accompany platelet activation.

CHANGES IN TRIPHOSPHATIDYLINOSITOL METABOLISM DURING PGE_1 -INDUCED SHAPE CHANGE OF WASHED RABBIT PLATELETS. J.D. Vickers, R.L. Kinlough-Rathbone, and J.F. Mustard, McMaster University, Hamilton, Ontario, CANADA.

Since the inositol phospholipids are present in small amounts in platelets and turn over rapidly during platelet shape change, aggregation and release, they are thought to have a functional rather than structural role in platelets. We have previously reported that within 10 sec of stimulation of prelabeled, washed rabbit platelets with ADP, the amount of triphosphatidylinositol (TPI) is significantly reduced while the specific radioactivity of its [^{32}P]phosphate is increased. One explanation of this result is that ADP-stimulation may divert ATP required for phosphorylation of diphosphatidylinositol (DPI) to TPI, leading to a decrease in the amount of TPI. PGE_1 (10 μM) causes conversion of ATP to cAMP and induces a transient platelet shape change. The shape change may be due to the reduction in ATP with a concomitant fall in TPI. We have therefore studied whether PGE_1 -stimulation of washed rabbit platelets prelabeled with [^{32}P] causes a change in TPI. Within 10 sec the amount of TPI in PGE_1 -treated platelets was reduced from 2.22 nmoles/ 10^9 platelets to 1.98 nmoles/ 10^9 platelets ($p < 0.05$) although neither the [^{32}P] labeling (51.1×10^3 dpm/ 10^9 platelets) nor specific radioactivity (24.1×10^3 dpm/nmole) were significantly changed. These results are compatible with the theory that diversion of ATP by PGE_1 -stimulation of cAMP formation from ATP, may reduce the amount of TPI. A similar effect was observed previously with ADP-stimulation. PGE_1 caused no change in the [^{32}P] labeling of phosphatidic acid (PA) (ADP caused a 290% increase) and caused only a small increase in its specific radioactivity (16% compared to 270% with ADP). If the rates of turnover of TPI and PA which are reflected in their specific radioactivities are Ca^{2+} -dependent, Ca^{2+} sequestration due to increased cAMP levels induced by PGE_1 would, after the initial effects, terminate these changes. The results further support the suggestion that reduction in the amount of TPI may be involved in platelet shape change and initiation of aggregation.