PLATLETTY PROSTAGLANDIN ENDOXYRIDE FORMATION IN HYPERLIPIDEMIAS. R. Bisog, L.K. Wong, R. 
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To evaluate the role of prostaglandin endoperoxide formation in the mechanism(s) of platelet
hyperactivity in hyperlipoproteinemia, a study was undertaken in 8 type II patients, 3 type
IV, and 9 normal volunteers. Platelet aggregation and release were studied in all subjects.
Simultaneously, ESR-platelets were washed by centrifugation and suspended in modified Ringer’s
solution. One ml suspensions (0.8-1X10^8/pl) were added to 4.5ug of 138-Arachidonic Acid (AA;
sp. act. 50 mCi/mmol) and incubated for 30 sec and 5 min at 37°C. The methylated reaction pro-
ducts were separated by thin layer chromatography. AA, PGD2, PGF2A, and thromboxane B2 (THB2)
(Uppih) were used to identify and locate the respective bands. The products were analyzed by
mass spectrometry. TLC-plates were then scanned and corrected for radioactive counting. The
chromatograms displayed a pattern similar to that reported by Samelsum et al. Compared with
normal, hyperactive platelets of type II patients generated 25% more thromboxane A2 (TXA2)
after 30 sec incubation with AA, while at 5 min TXA2 was increased by 30% and TXA2 was still ele-
vated by a factor of 2. The behavior of type IV platelets was similar to normal.

Aspirin-treated platelets produced one product (PDE). Among the AA derivatives studied,
only eluted TXA2 induced platelet release.

Our data suggest that platelet hyperactivity in type II patients may be mediated by the
increased production and long survival of thromboxane A2.

HUMAN ENDOTHELIAL CELL INJURY MECHANISMS IN VITRO. R.T. Wall, I.A. Harker, G. Striker and
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Since endothelial cell desegmentation constitutes the initial event leading to acute and chronic vascular
diseases, we have developed an in vitro system for studying injury mechanisms using cultured cells. The
system involves the incubation of test material with confluent, 14C-labeled, cultured human umbilical vein
endothelial cells in 100% pooled human serum; injury was measured as % endothelial cell 14C release
(ECR) into the supernatant media. Baseline spontaneous ECR was 0.0% - 1.5% while specific rabbit
antihuman endothelial cell antibody (1:8 dilution) in the presence of fresh complement released 90% - 3 of
the total releasable 14C activity. Consistent with in vivo predictions, homocysteine in concentrations of
0.5-40 mM induced dose response ECR to 20%. Neither methionine nor homocystine increased ECR over
controls. Also as expected from experimental work endotoxin (S. enteritidis) caused dose response ECR,
ies. 24% - 3 at 10 ug/ml. Control studies with cultured human smooth muscle cells (SMC) demonstrated no
ECR with homocysteine but significant release occurred with endotoxin.

Specific complement dependent cytotoxic anti-endothelial cell antibody was demonstrated in the serum
of two thrombotic thrombocytopenic purpura (TTP) patients at 1:1 dilution, inducing ECR of 23.1% -1.4
and 31.0% - 0.5. The antibody was also demonstrated by immunofluorescent techniques and was absorbed
from the serum using human endothelial cells. One disease-free, six month survivor showed no cytoxic
activity. Serum from a patient with adult hemolytic-uremic syndrome demonstrated antibody dependent
cell mediated cytotoxicity with release of 22% - 1.1 when normal non-immune lymphocytes were added to
heat inactivated serum. Control studies with SMC showed no ECR with TTP sera. We conclude that assays
of endothelial cell 14C release are stable, reproducible and useful in the characterization of injury
mechanisms.