Table 1. Adenosine Triphosphate, Creatine Phosphate, Inorganic Phosphorus Creatine and Glycogen Content of Rat Heart Tissue after Daily Administration of Thyroxin and other Substances

Lot	Adenosine triphosphate (μgm./gm. fresh tissue)	Creatine phosphate (μgm./gm. fresh tissue)	Inorganic phosphorus (μgm./gm. fresh tissue)	Creatine (mgm./100 gm. fresh tissue)	Glycogen (mgm./100 gm. fresh tissue)
A (6)	331 ± 27	308 ± 23	521 ± 46	408 ± 16	183 ± 11
B (6)	126 ± 11	< 10	316 ± 34	293 ± 33	149 ± 18
C (6)	224 ± 30	167 ± 19	409 ± 43	307 ± 26	166 ± 22
D (6)	121 ± 16	< 10	325 ± 29	367 ± 38	151 ± 17
E (6)	230 ± 28	165 ± 21	418 ± 46	376 ± 41	164 ± 14

Values ± standard error

were killed by bleeding and determinations of adenosine triphosphate, creatine phosphate, inorganic phosphorus[3], creatine[4] and glycogen[5] were carried out.

As shown in Table 1, administration of creatine phosphate partially prevents the fall of adenosine triphosphate and creatine phosphate content of the heart in animals given thyroxin at high dosage. Creatine alone cannot do this, and not only the adenosine triphosphate content but also creatine phosphate content remain low.

L. Angelucci
R. Boldrini
E. Mascitelli-Coriandoli

Istituto Biochimico Nazionale,
Via Cottolengo 12,
Rome.

[1] Mascitelli-Coriandoli, E., and Boldrini, R., *Nature*, **179**, 474 (1957).
[2] Mascitelli-Coriandoli, E., and Boldrini, R., *Nature*, **179**, 1196 (1957).
[3] La Page, G. A., "Manometric Methods and Tissue Metabolism" (Burgess, Minneapolis, 1951).
[4] Hawk, P. B., *et al.*, "Practical Physiological Chemistry" (Blakiston, Philadelphia, 1949).
[5] Kahan, J., *Arch. Biochem. Biophys.*, **47**, 408 (1953).

Phospholipids, Proteins and Platelet-Lipoid

The time taken for Russell viper venom to accelerate the clotting of fasting human plasma poor in platelets—the Stypven time—is relatively long. The addition of very small quantities of some active phospholipids[1,2] to this system produces a shortening of the Stypven time, and the shortening is proportional to the quantity added. It may be concluded that there is very little available active phospholipid in fasting platelet-poor plasma. A simple phospholipid extraction of fasting platelet-poor plasma yields a material that greatly accelerates the Stypven time of platelet-poor plasma. It is concluded that there are molecules of active phospholipids in plasma, but that they are 'unavailable' for this test and presumably are bound in the plasma proteins.

If a critical strength of an isolated phospholipid, active in the Stypven system, is added to platelet-poor plasma at 37° C. or 10° C. it is found that this activity has decreased at the end of 2 hr. (To 0·5 ml. of fasting platelet-poor plasma is added 0·5 ml. of a predetermined concentration of an active phospholipid in distilled water. 0·1 ml. of this mixture is transferred to another 0·1 ml. sample of stock plasma and the Stypven time determined. This should be about 14 sec. if the plasma alone has a Stypven time of 20 sec. At the end of 2 hr. a further 0·1 ml. of the mixture is transferred to 0·1 ml. of stock plasma and the Stypven time recorded. Another 0·5 ml. of plasma and 0·5 ml. of phospholipid are then mixed and 0·1 ml. transferred to 0·1 ml. of stock plasma and clotted as before to check that the plasma, Stypven and original phospholipid activity have not altered.) It may be concluded that less phospholipid is available after 2 hr. Plasma rich in heparin-clearing factor gives the same results as normal plasma. It is unlikely that any enzyme system has altered the chemical composition of the phospholipid since this reaction proceeds equally well at 10° C. as at 37° C. A possible explanation is that the phospholipid has become associated with the plasma proteins and is no longer 'available' for this test.

Intact platelets are inactive in accelerating the Stypven clotting time of plasma. If they are mechanically fragmented, a potent accelerator becomes available. The properties of fragmented platelets in all coagulation tests (except clot retraction) are identical with various cephalins including phosphatidyl-ethanolamine, phosphatidyl serine and phosphatidyl inositide[3] and it has been suggested that the activity of platelet-lipoid is due to its content of any one or a mixture of these or similar 'active' phospholipids.

If platelet fragments are incubated in a critical strength with platelet-poor plasma the acceleration of the Stypven time caused by their addition does not decrease in two hours. A protein-free phospholipid extract of platelets behaves like an active phospholipid extracted from any other source.

If the two previous conclusions are correct, this last observation may suggest that if platelet-lipoid is active in blood coagulation by virtue of a phospholipid prosthetic group, then there is another part of the molecule which prevents the whole molecule from becoming associated with the plasma protein and thereby losing its activity. In a natural state, platelet-lipoid is almost certainly associated with protein. The activity of platelets may, therefore, depend on a preferred orientation in the platelet-protein molecule of a prosthetic phospholipid group which is 'available', while the phospholipids in plasma proteins are differently orientated and inactive or 'unavailable'.

Further work is in progress to expand and confirm these tentative conclusions.

J. R. O'Brien

Central Laboratory,
Milton Road,
Portsmouth, Hampshire.
Sept. 12.

[1] Poole, J. C. F., and Robinson, D. S., *Quart. J. Exp. Physiol.*, **41**, 31 (1956).
[2] O'Brien, J. R., *J. Clin. Path.*, **9**, 47 (1956).
[3] O'Brien, J. R., *Brit. J. Exp. Path.*, **38**, 529 (1957).

Adrenocorticotrophic Hormone in the Blood of Adrenalectomized Rats

It is well known that stress results in increased activity of the adrenal cortex which is caused by an increased secretion of adrenocorticotrophin by the pituitary gland. However, the exact mechanism which controls the secretion of this hormone is not fully understood. Sayers and Sayers[1] suggested that stress causes an increased utilization of adrenocortical steroids by the peripheral tissues and the resulting low blood level of corticoids acts as a stimulus to the pituitary gland to increase its secretion