a specific tissue component is involved, is supported by certain experimental data.

Thus Howell and Donahue[4] have advanced evidence to show that blood platelets are formed in capillaries of the lung, and are being discharged continuously into the blood stream. In 15 (88 per cent.) of 17 experiments they have found the number of platelets in the blood from the left side of the heart to be greater than in that from the right side of the heart, and that the increase averaged 13.3 per cent. in the 15 successful experiments. Further proof was adduced by perfusion experiments and by histological examination of the lung. In our experiments the lowering of the level of the plasma prothrombin as blood passes from the right to the left side of the heart by way of the pulmonary circuit corresponds both in frequency and in degree to the increase in blood platelets found by Howell and Donahue. The platelets disintegrate and in so doing, liberate thromboplastin. According to the present accepted mechanism of blood-clot formation, the action of thromboplastin and calcium on prothrombin changes it to thrombin. Platelets are the chief source of intravascular thromboplastin; they are fragile and are continuously undergoing disintegration. Loss of prothrombin is then dependent upon the thromboplastin and calcium. This mechanism accounts for the continued loss of prothrombin after the source of supply, the liver, has been removed.

SUMMARY

The level of the plasma prothrombin in the circulating blood is decreased during its passage through the pulmonary capillaries. In 85 per cent. of samples, plasma prothrombin has been found to be less in the blood of the left ventricle than in that from the right ventricle. This difference averaged 10.6 per cent. and ranged from 4 to 19 per cent. In no instance was the level in the right ventricle lower than that in the left. In samples of blood taken from the arterial and venous supply of the head, liver, spleen, intestine, kidney and hind limbs, no significant difference in plasma prothrombin levels was found.

A possible explanation of this role of the lung in the loss of plasma prothrombin is thought to be the production of blood platelets in this organ, as demonstrated by Howell and Donahue. Platelets, as they undergo disintegration, initiate the first stage of the clotting process by releasing thromboplastin, which, in the presence of calcium, changes prothrombin to thrombin.

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IMPEDANCE OF BIMOLECULAR FILMS

BIMOLECULAR films having water on both sides[1] were made by bringing together, in oil, a hanging drop and a sessile drop of water with a tanned protein monolayer on each.[2] The J tube holding the sessile drop and the I tube holding the hanging drop were each about 1 cm inside diameter and each had a cylindrical platinized lead electrode 1 cm long about 2 mm from the film end. The oil was amyl acetate, purified by shaking with water and filtering to remove surface active constituents, and the aqueous phase was a dilute solution of tannic acid in M/100 KCl acidified to about pH 3. A stable film of tanned egg albumin was obtained at each interface by carrying the dry powder through the oil on a moistened glass fiber. After the two monofilms were brought together, the resistance decreased as the intervening oil was forced out and did not reach a constant value for several hours.

Alternating current impedance data were obtained on one film as described and on a second in which lecithin was added to the oil before the films were brought together. There were only minor differences between these data. The impedances were measured with a Wheatstone bridge over the frequency range from 30 to 200,000 cycles per second.[3] The data of the second experiment have been plotted[4] as series resistance, R_s, *vs.* series reactance, X_s, in Fig. 1. The

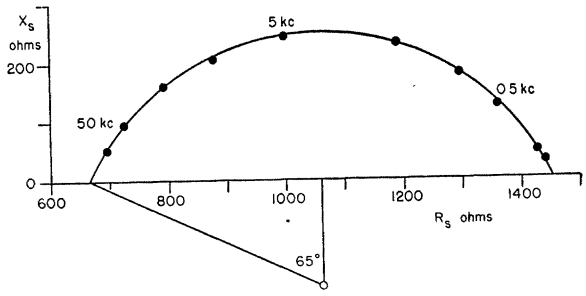

FIG. 1. Impedance locus, series resistance, R_s *vs.* series reactance, X_s, of a bimolecular tanned egg albumin and lecithin film. The frequencies indicated are in kilocycles per second. The film had an area of 0.07 cm², 790 ohms direct current resistance, 65° phase angle and 0.086 microfarads capacity at 1 kilocycle per second.

extrapolated infinite frequency resistance, R_∞, is the resistance of the electrolyte between the electrodes and the film. The difference between the extrapolated zero frequency resistance, R_0, and R_∞ is the direct current resistance of the film. From the film areas of

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approximately 0.07 cm² these film resistances were found to be 94 and 52 ohm cm², respectively. The changes of impedance with frequency between these two limits were due to the capacitative impedances of the films, which had constant phase angles of 68° and 65° and capacities of 1.07 and 1.23 microfarad/cm² at 1,000 cycles/second, respectively.

The resistance of these films is in marked contrast to the extremely low resistances found in untanned protein films.[1] This reopens the possibility that complex lipo-protein films may have an appreciable resistance and be able to produce diffusion potentials in biological systems. The constant phase angle and a capacity of about one microfarad/cm² is a striking characteristic of all living cell membranes so far measured[5] and we believe these to be the first artificial films produced between two aqueous phases which had these properties.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

MASS PRODUCTION OF VACCINE AGAINST TYPHUS FEVER OF THE EUROPEAN TYPE

IT is unnecessary to emphasize the importance at the present time of the development of prophylactic vaccination against typhus fever. Until 1925, it was supposed that immunization against the Rickettsiae diseases was possible only as a result of actual infection. Since then, however, various attempts to employ living, supposedly attenuated virus for prophylactic purposes have been made[1,2]—all of them, in our opinion, dangerous both for the individual and for the community. Spencer and Parker (1925)[3] were the first to demonstrate that active immunization against spotted fever could be achieved with killed, phenolized Rickettsiae obtained from infected ticks. Following this (1930), Weigl[4] reported analogous results with phenolized European typhus Rickettsiae harvested from the intestinal contents of lice artificially infected. Subsequently (1930), one of the writers with Batchelder[5] and then with Castaneda[6,7] and Macchiavello[8] demonstrated that active immunization against the murine and the European varieties of typhus fever could be produced in animals with formalinized suspensions of the respective micro-organisms. The principles were thus established, but difficulties still remained in the way of obtaining the relatively large amounts of Rickettsiae necessary for practical immunization. Numerous reports have been made by investigators who claim to have been able to cultivate Rickettsiae on media without tissue. The present writers, however, because of the negative results reported by other workers and their own failure, after long experience and persistent effort in similar attempts, feel that, up to the present, such tissueless cultivation of virulent Rickettsiae has not been achieved.

We may summarize the present state of affairs in regard to typhus vaccine production as follows:

As far as the murine variety of typhus fever is concerned, tissue culture methods of various kinds are easily carried out. By none of these procedures can such large quantities of Rickettsiae be produced as by the technique of intraperitoneal inoculation of x-rayed rats, a method sufficiently described in preceding communications. As first developed by one of us with Castaneda,[9] this method has been utilized in its original form and with modifications on a considerable scale by Castaneda in Mexico and Veintemillas in Bolivia.

Unfortunately, the rat methods have, in the course of years of effort, proved inapplicable to the classical European virus. In consequence, other procedures have been suggested.

The Weigl louse technique is effective, but entirely unsuitable for large-scale application.

Tissue culture methods based on the modified Maitland technique as first developed for European Rickettsiae by Nigg and Landsteiner[10] were used for immunization, first, by Kligler and Aschner[11] and

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