but there was poor agreement between the activator and apparent prothrombin content of different plasma fractions up to 40 per cent saturation with sulphate, at which level almost all of both constituents had been removed. This was in part due to removal of coagulase inhibitor which is present to some degree in all sera, and which is precipitated at the lower concentrations of ammonium sulphate. Likewise four Seitz filtrations of rabbit and human plasma removed prothrombin activity completely with only partial loss of activator. Quantitative comparisons after each filtration were again made difficult by the early removal of coagulase inhibitor, especially in the case of human plasma. This loss of prothrombin on Seitz filtration may be more apparent than real, since Tager and Hales found it to be partly restored by acid precipitation and resuspension. Like prothrombin, activator is removed from plasma by barium sulphate and is destroyed by crystalline trypsin and pepsin.

If prothrombin participates in coagulase clotting, it does so in a manner essentially different from that found in normal coagulation. Mixtures of coagulase and plasma show no loss in prothrombin content on clotting, as would be the case had there been conversion of prothrombin to thrombin. Likewise, mixtures of prothrombin, factor V and coagulase incubated at 37° C. show no progressive increase in the amount of coagulant present as occurs in the usual two-stage prothrombin conversion test. Coagulase itself disappears when clotting has taken place, and is presumably attached to the fibrin clot. Thus while coagulase activator is closely associated with prothrombin both in occurrence and physical properties, we cannot as yet conclude that they are identical, and the studies are being continued. The present results will be published in full elsewhere. We should like to thank Drs. R. G. Macfarlane, Rosemary Biggs and R. A. Kekwick for helpful advice and material.

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Production of Coombs's Serum

The production of a high-titre anti-human globulin serum is a matter of importance to a laboratory engaged in the detection of incomplete antibodies. Our experience with the usual methods of production has been disappointing; but we have obtained good results with the following course of injections.

Adult male and female rabbits of a mixed stock have been used. They are fed on bran mash and greenstuff. (1) Intramuscular injections of 5 ml. alum-precipitated globulin into each hind leg. The globulin is prepared by the method of Proom[1] as described by Mollison, Mourant and Race[2]. Interval of two weeks. (2) The intramuscular injections are repeated. Interval of four–eight weeks. (3) Intraperitoneal injection of 1 ml. sterile human serum. (4) Next day, intravenous injection of 1 ml. sterile human serum. Interval of 5 days. (5) The rabbit is bled, the serum inactivated and absorbed as usual.

This course of injections has now been carried through on six rabbits. No trouble from anaphylaxis has been encountered. Every rabbit has produced a reagent capable, when diluted 1 : 10,000, of agglutinating fully sensitized red cells. The strongest serum was reported by another laboratory as active when diluted about 1 : 256,000.

The sera are normally used diluted 1 : 50–1 : 100, since they are most active at this dilution. If sera of this potency are used at a greater concentration, their activity is less. It is hoped to give a fuller account of this finding elsewhere.

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Effect of Anions on the Respiration of Kidney Slices from Adult Rats

Slices of adult rat kidney have been found not to take up oxygen at a constant rate in media which are suitable for other tissues[1,2]. They were found to do so, however, in a medium containing physiological concentrations of sodium, potassium, calcium, magnesium, phosphate and chloride ions and glucose[3]. Because its ionic requirements are so critical, this was considered a suitable tissue to use in an investigation of the relative importance of the cations and anions in balanced media. Slices of adult rat kidney were therefore set up in Barcroft manometers under oxygen at 38·5° C. in media having the cation concentrations which had been found to support their steady respiration, but in which chloride was replaced by other anions.

Thus, an isotonic solution corresponding to the "Medium *A*.2" (ref. 3) was prepared by mixing 0·154 *M* sodium nitrate, 232 ml., 0·154 *M* potassium nitrate, 8 ml., 0·11 *M* calcium nitrate, 3 ml., and 0·154 *M* magnesium sulphate, 2 ml., and adding 24 ml. of phosphate buffer prepared by bringing *M*/15 disodium hydrogen phosphate to *p*H 7·4 with *N*/10 nitric acid. Glucose was added to give a concentration of 100 mgm./100 ml. In seven experiments using this medium, the oxygen uptake during the first hour was 4·2 (S.D. 0·4) µl. oxygen/hr./mgm. initial moist weight. Four control experiments set up at the same time in the chloride-containing medium *A*.2 gave a rate of 4·2 (S.D. 0·2) µl. oxygen/hr./mgm.

Another medium was prepared in which the sodium, potassium and magnesium ions were introduced as sulphates, and the calcium ion as nitrate. This medium had the same concentrations of all cations as *A*.2, and was therefore hypotonic on account of the divalence of the sulphate anion, its total osmolar concentration being about three-quarters that of the chloride and nitrate media. Two sets of slices in this medium gave respiratory rates of 4·7 and 4·8 µl.