cipitation of these compounds and γ-globulin. Apparently the albumin forms soluble complexes with either or both the dye and the γ-globulin.

These results are of interest in connexion with attempts to manufacture antibodies *in vitro*[2], and also with the observation that the reaction of polyhaptenic substances with purified hapten-homologous antibody is considerably different from that of these substances with antiserum[3].

This work was initiated in the laboratory of Prof. Linus Pauling, California Institute of Technology, whom I wish to thank for his assistance. A complete account of the work will be published elsewhere.

JOHN L. MORRISON

Department of Chemistry,
University of Alberta,
Edmonton, Alberta.
July 25.

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Influence of Temperature and Iron on Hæmoglobin Synthesis by *Daphnia*

Daphnia, in common with other branchiopod Crustacea, synthesizes hæmoglobin in response to a deficiency of dissolved oxygen in the surrounding water[1]. The hæmoglobin is in solution in the blood. In poikilothermal animals, chemical processes in the body normally proceed more rapidly at a high external temperature, and we have found that in *Daphnia* hæmoglobin synthesis is quicker in warm water. We wished, in addition, to know whether, at the lower of two temperatures, *Daphnia* would eventually acquire as much hæmoglobin as at the higher temperature (the low oxygen contents of the waters being maintained at the same level) or whether at the higher temperature a greater amount of hæmoglobin would be synthesized. The second alternative proved to be correct. In one experiment with *D. obtusa*, in water containing approximately 1 ml. dissolved oxygen per litre, the hæmoglobin content of the blood increased, in 28 days, five-fold at 17° C. but seven-fold at 28° C. After that there was no further increase. One cause of the greater synthesis at the higher temperature is probably the low oxygen content of the tissues, due to a high metabolic rate.

There are great differences in the iron content of natural fresh waters in which *Daphnia* lives. It has been found to vary between 50 mgm./l. and no iron detectable by dipyridyl (before or after reducing). The question thus arises as to whether the addition of iron salts to water containing little, or no, detectable iron would increase the quantity of hæmoglobin synthesized by *Daphnia* in response to oxygen deficiency. Our work has shown this to be the case. In one experiment, at 20° C., in water with no detectable iron and a low dissolved oxygen content of 0·9 ml./l., the blood hæmoglobin increased seven-fold in sixteen days (deriving its iron no doubt from algal food) and then remained steady for a further twelve days; whereas in similar water, to which 4 mgm./l. iron was added every other day, the increase in sixteen days was between nine-fold and ten-fold, with subsequent maintenance of this level for twelve days.

It was found that ferrous salts are more effective than ferric salts in augmenting hæmoglobin synthesis. This is curious for the following reason. A hæmochromogen is present in the gut lumen of *Daphnia*[2]. At the low oxygen content of the outside water necessary for hæmoglobin synthesis by *Daphnia*, this hæmochromogen can be seen with a spectroscope to be in the reduced state, the α-absorption band showing clearly. Now, the redox potential for the reduction of various hæmochromogens is known to be well below that at which the ferric ion is reduced to the ferrous state[3]. It is thus probable that in the gut lumen of *Daphnia* iron is in the ferrous state, even if swallowed as a ferric salt. The explanation of the greater effect of ferrous salts in increasing hæmoglobin synthesis seems to be as follows. The ferric salt added to the experimental water is gradually converted to ferric hydroxide and precipitated. The ferrous salt is also converted to ferric hydroxide, but in a more finely divided state which remains longer in suspension, and is thus available to *Daphnia* for a longer time.

A full account of these results will appear in the *Proceedings of the Royal Society*.

H. MUNRO FOX
ELIZABETH A. PHEAR
BARBARA M. GILCHRIST

Bedford College,
University of London.
Dec. 17.

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Role of Choline in the Oxidation of Fatty Acids by the Isolated Liver

THE lipotropic effect of choline has been interpreted most generally by assuming that fatty acids are carried out of the liver in the form of plasma phospholipides. However, the role of these compounds in the transport of fatty acids has been questioned[1]. The results presented here strongly suggest that choline acts by enhancing the oxidation of fatty acids in the liver.

In each experiment, slices or homogenates were prepared simultaneously from the liver of rats previously maintained on different diets, namely, (*A*) a choline-deficient diet (diet 26, containing 5 per cent casein[2] with added guanidoacetic acid 1 per cent), (*B*) a choline-supplemented diet (diet 26, with added choline-hydrochloride 0·4 per cent), or (*C*) a stock diet. In most experiments one or two animals received also 0·1 millimole of choline-hydrochloride, given in two intramuscular injections, 1·5 and 0·5 hr. before the rats were killed. The slices or homogenates were incubated with 1-^{14}C sodium stearate (obtained from U.S. Testing Co., Hoboken, N.J., on allocation from the U.S. Atomic Energy Commission) in a medium of Ringer-phosphate without calcium or magnesium[3]. Control flasks containing the same amounts of materials were kept at 100° for 10 min. before the addition of the isotopic stearate and then incubated. The carbon dioxide produced during the incubation was trapped in sodium hydroxide, precipitated as barium carbonate, and its radioactivity determined with a *Q* gas-flow counter. Total lipides, lipide phosphorus, and total nitrogen were determined on separate samples of the liver.