LETTERS TO THE EDITORS

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Chlorocruorin and Hæmoglobin

THERE are four respiratory pigments which circulate in the blood or are found in the body cavity of animals. Two of these, the red hæmoglobin and the dichroic red-green chlorocruorin, are close chemical relatives. Chlorocruorin is confined to certain marine polychæte worms and is characteristic of the group Serpulimorpha, comprising the families Serpulidæ and Sabellidæ. Hitherto it has been thought that all species in this group possess chlorocruorin in their blood. This, however, is not the case. I have found chlorocruorin in twenty-one species of the group, belonging to fifteen genera. One of the species with chlorocruorin is *Spirorbis borealis* Daudin, but another species of the same genus, namely, *S. corrugatus* (Montagu), has hæmoglobin as its blood pigment. Yet another species, *S. militaris* (Claparède), has neither pigment: its blood is colourless. That chlorocruorin and hæmoglobin are really absent here, not just dilute, is proved by the fact discovered by my colleague, Miss Jean Hanson, that the benzidine reaction is negative in the blood vessels of *S. militaris*. These several species of *Spirorbis* live in similar situations, so that the possession of either or neither respiratory pigment appears to be functionally unimportant.

In *Serpula* the blood contains both chlorocruorin and hæmoglobin. It is greenish-brown in colour and has three strong absorption bands in the visible region of the spectrum, namely, the α-band of oxychlorocruorin and the α- and β-bands of oxyhæmoglobin. MacMunn[1] saw this spectrum, but did not recognize the hæmoglobin. Thus for the first time two respiratory pigments have been found in one blood. This duplicity is characteristic of the genus *Serpula*, for it occurs in both the species which I have been able to examine, namely, *S. vermicularis* L. and *S. lobiancoi* Rioja. We have here a chemical generic character that will have to be taken into account by taxonomists; it is comparable with chemical specific characters which the respiratory pigments have already furnished[2].

In one genus only of the Serpulimorpha have I found a respiratory pigment in tissue cells: *Potamilla*, with only chlorocruorin in the blood, has hæmoglobin in its muscles. Just as in the case of mammalian myoglobin, the wave-length of the α-band axis of the hæmoglobin in *Potamilla* muscle is longer than that of the mammalian blood pigment. The muscles of *P. reniformis* (O. F. Müller) are pink with hæmoglobin, whereas those of *P. stichophthalmos* (Grube) have just a little of the pigment. As the two species live side by side in the same stones, again the pigment difference does not seem functional.

The hæm of chlorocruorin is spectroscopically different from protohæm, the hæm of hæmoglobin[3]. Most animal cells contain protohæm[4], whether or not the animals possess hæmoglobin. It is therefore of particular interest to know if animals with chlorocruorin in their blood have protohæm or chlorocruorohæm in the cells of the various tissues of the body. I have studied this question in the Serpulimorpha, particularly in the largest member of the group, *Sabella spallanzanii* (Viviani), previously known as *Spirographis*[5]. By adding pyridine and measuring the hæmochromogen α-band I find that protohæm alone is present in the tissues; it is found in muscles, tube-forming glands, gut wall, nephridia, eggs and sperm. Chlorocruorin, which is in solution in the blood, has never been found in a cell, and its hæm, too, is absent from the cells of Serpulimorpha. Evidently chlorocruorohæm must be formed from protohæm somewhere in the blood system. This conversion has been accomplished *in vitro*[6]. Chemically it is not a big change: one of the two vinyl groups attached to the 4-pyrrol ring of protohæm is oxidized to a formyl group[7]. Cytochrome is widespread in animals[8], and since the hæms of cytochrome *a* [9] and of cytochrome oxidase[6] are very close to chlorocruorohæm, a similar oxidation of the all-pervading protohæm may take place in many animal cells. Incidentally, in the Serpulimorpha I have found no cytochrome.

In the lumen of the gut of serpulids there is a pigment in solution which has a protohæmochromogen spectrum. It recalls helicorubin in snails, and the gut hæmochromogen of *Daphnia*[10]. Presumably it is derived from the protohæm compound in serpulid tissues. What, then, if any, is the breakdown product of chlorocruorin in the blood? Is the chlorocruorohæm again reduced to protohæm, to swell the gut hæmochromogen?

Protohæm is also present in the mucoprotein tube of *Sabella* and in the membranous lining of the calcareous tube of *Protula*. This recalls the presence of chlorocruorin in the mucous jelly tube of Myxicola[3]. What other cases, except pathological ones, are known of the excretion, either into the gut or in skin secretions, of hæm compounds as such—that is to say, not broken down to bile pigments or porphyrins?

A full account of this work, most of which was done at the Zoological Station of Naples, will be published soon.

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Sept. 24.

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Effect of 4-Chloro-2-Methyl Phenoxyacetic Acid on the Mineral Content and Growth of Plants

Templeman and Sexton[1] have shown that certain concentrations of the aryloxyacetic acids produce a differential effect on the growth of plants of different species. This effect, very marked in the case of 4-chloro-2-methyl phenoxyacetic acid, has now been utilized as a means of eliminating weeds from cereal crops.

We began a study of the mode of action of 4-chloro-2-methyl phenoxyacetic acid on plant growth in 1946. As a result of field experience, rape and spring oats were selected as examples of susceptible and resistant crops, and corn chamomile for intermediate response. Seedlings of the three species were grown in small pots containing silver sand, surface-watered with 40 ml. of nutrient solution (pH 6) per pot, once or