development. Caution is warranted on the interpretation of this hypothesis. There is, however, a possible analogy of endogenous regeneration following a colchicine treatment with the growth recovery which occurs when the tip of the coleoptile of intact wheat seedlings is decapitated[10]. Elongation of the tipless coleoptile resumes, and this is the result of biochemical changes arising during a period of reduced growth activity. This shows again that induction may well operate during regeneration of growth in the wheat coleoptile.

The assumption discussed above is incorrect if it is proved that during regeneration colchicine controls cell division exclusively at the level of the mitotic apparatus. That is to say, through structural changes of the centromeres or the protein monomers of the spindle fibers, so that these cell sites would no longer be available for the colchicine activity. Another hypothesis is that this substance might be restrained from interferring with the cell metabolism in a way similar to streptomycin resistance in bacteria[11]: the latter drug induces a change in a protein sub-unit of the bacterial 30 S ribosome. Protein synthesis is insensitive thereafter to streptomycin inhibition. This would seem to show that endogenous regeneration following a treatment with colchicine might result from the structural modification of specific cell constituents which affects either the binding of the colchicine molecules or the functioning of their complexes.

Résumé. Des doses de colchicine inférieures au seuil de son activité mutagénique sont à l'origine de la variation périodique du taux de croissance in vivo de la coléoptile de blé. En particulier, le comportement oscillatoire de cette régénération endogène a lieu dans un milieu qui contient la colchicine pendant toute la durée de l'incubation.

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Blood Chemistry of *Lamellidens corrianus*

Lamellidens is one of the commonest Indian fresh water bivalves, but almost nothing is known about its body functions. Since blood reflects upon the over-all metabolism of animal tissues, a preliminary biochemical study of the blood of *L. corrianus* has been undertaken here as an initial step towards the understanding of the functional organisation of this mussel.

The alkaline (pH 7.9) blood of the mussel comprises amoebocytes and plasma, the latter constituting its major volume and being transparent and colourless.

Presence of a respiratory pigment could not be definitely established in the mussel blood. Spectrophotometry revealed an insignificant absorption in the visible spectrum range with faint indication of maxima at 530 nm and 560 nm, suggesting the vascular pigment, if present, to be more like haemoglobin than haemocyanin. But the benzidine test for haemoglobin and the potassium ferro- and ferricyanide test for iron gave negative results both with plasma and blood cells. However, treatment of proteins precipitated from 10 ml of pooled blood with potassium ferrocyanide produced an extremely light blue colour, indicating the presence of ferric ions in traces. It appears therefore, that even if an iron-containing vascular pigment is present, it is in such small amounts as to be of little use in the oxygen carrying needs of the mussel.

Plasma protein concentration of the mussel, estimated to be 0.9 g/l, is, like the concentration observed in other bivalves and gastropods[1], appreciably lower than that reported for cephalopods[1]. Since plasma proteins chiefly facilitate fluid movement across capillary end-loops, their high level in cephalopods and low level in other molluscs may be justified by the presence in the former and absence in the latter of capillaries between arteries and veins.

Glucose content of the mussel plasma was found to be 8.2 mg/100 ml. Martin[2] has suggested that it is wasteful for molluscs to maintain more than the minimal required blood glucose because of its imperative loss during filtration of fluid from the heart for urine formation. The presently observed glucose concentration, if considered as the minimum required, would imply a very low energy demand and hence an equally low over-all metabolism in the mussel, even in comparison with other bivalves whose reported blood glucose levels are considerably higher[2].

Amylase activity was studied in the plasma as well as in a homogenate of amoebocytes separated from 10 ml of pooled blood of the mussel. Plasma was found to hydrolyse 0.082 mg and the amoebocyte homogenate 0.015 mg starch/h/mg plasma protein. However, while the amylase activity of plasma must be non-functional, since blood cannot be the site of starch digestion, that of the wandering amoebocytes may be functional and indicative of a probable digestive role of these cells. Leakage of enzyme from blood cells is, therefore, a likely source of plasma amylase, but the much higher amylase content of plasma in comparison with that of blood cells would indicate leakage from other sources also.

Acid and alkaline phosphatase activities of the mussel plasma were respectively found to release 0.0107 and 0.0116 mg phenol per hour per mg plasma protein. These activities, being extremely low, do not appear to be functional. Moreover, the possibility of their leakage from amoebocytes to plasma cannot be ignored, specially since phosphatases have been histochemically demonstrated in the mussel amoebocytes[3].

Glutamic oxaloacetic transaminase activity of the mussel plasma was found to release 0.0187 mg pyruvate per mg plama protein. Presence of this enzyme in the plasma may be indicative of the existence of citric acid cycle in the mussel.

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The mussel plasma does not contain fibrinogen, and incubation of a mixture of plasma and thrombin at 37 °C was not seen to produce clots even after 1 h. Absence of fibrinogen in the plasma confirms that the mussel is not in possession of a blood clotting mechanism comparable to vertebrates.

The mussel plasma also does not contain any human agglutinins, since it was found unable to agglutinate washed human red blood cells of A, B and O groups.

Résumé. Le sang de *Lamellidens corrianus* est alcalin et incolore. Il contient peu de pigment respiratoire et de proteines. Le plasma contient peu de glucose et pas de fibrogène ou d'agglutinines humaines. Une activité des enzymes amylase, GOT et des phosphatases a été constaté.

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[4] It is a pleasure to acknowledge the stimulation received from Mr. M. P. SINGH, Experimental Pathologist, King George's Medical College, Lucknow.

The Effect of Alloxan Diabetes on Skin Collagen Metabolism

The effects of alloxan induced diabetes on impaired wound healing was studied using granulation and skin tissue in the attempt to elucidate possible alterations in the metabolism of collagen. Although it is well recognized that diabetes results in defects in various connective and vascular tissues[1,2], the results reported here suggest that in the rate of formation and maturation of collagen may not be a significant factor.

Materials and methods. Diabetes was induced in 2 groups of male Sprague-Dawley rats weighing 40–50 g and 100–125 g by alloxan injection (120 mg/kg). Alloxan was administered either i.p. (40–50 g rats)[3] or by i.v.-injection (100–125 g rats)[4]. The presence of diabetes was indicated by marked glucosuria (> 0.5%), increased fluid intake and loss of body weight. Rats were maintained on a commercial stock diet through out the experimental period.

The effect of treatment on amino acid uptake by granulation and skin tissue was determined using a sponge implantation technique (40–50 g rats)[5] and the in vitro skin biopsy method of UITTO (100–125 g rats)[6]. In the implantation studies, polyvinyl sponges $1.5 \times 0.5 \times 0.3$ cm) were inserted in the subcutaneous space along the dorsum 10 days after initiation of alloxan treatment. The sponges remained 6 days before removal and decapitation. The sponges were then cut into small pieces and incubated in phosphate-free Krebs-Ringer solution (3 ml, pH 7.6) containing 20 m*M* N-2-hydroxyethylpiperazine-N^1-2-ethane sulphonic acid (HEPES), 20 m*M* glucose and 0.5 μCi U-^{14}C-lysine[6]. Incubations were carried out for 12 h at 37 °C in a metabolic shaker. After incubation, the sponges were completed, homogenized and aliquots were taken for counting and DNA analysis[7]. Dorsal skin biopsies (1 cm diameter, 100–150 mg) were taken 20 days after alloxan injection (100–125 g rats). The tissue was minced and incubated in the Krebs-HEPES solution (10 h, 37 °C) except U-^{14}C-proline was added at 0.1 μCi in place of radiolysine. The uptake of radioproline was linear for at least 18 h (Figure 1). Both tissues and sponges were dialyzed exhausively before radioactivity was determined by scintillation counting.

The diameters of wounds after biopsy were also measured daily until complete closure as an indirect index of wound healing (Figure 2). At the end of this period, a second biopsy was taken and total hydroxproline determined[8]. Urinary hydroxyproline was monitored at various time intervals with rats receiving alloxan injection. In addition, skin samples were taken from these animals and collagen solubility in neutral NaCl solution (1 *M*) was determined[9]. The extracted collagen was purified, electrophoresed on acrylamide gel, and the ratio of monomer (α) to dimer (β) collagen subunits was calculated after staining with amido black and scanning with a densitometer[10].

Results. The uptake of radiolysine and radioproline as cpm into granulation tissue from sponge implants and skin biopsy material is indicated in Tables I and II, respectively. In both cases, alloxan treatment stimulated

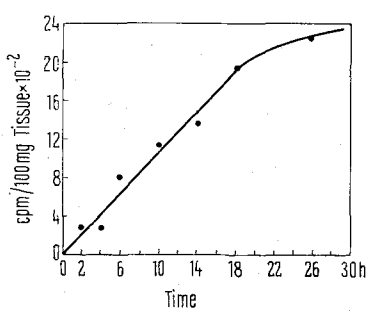

Fig. 1. The uptake of U-C^{14}-proline into rat skin. Radioproline was present at p. 1 μCi in Krebs-HEPES solution (3 mls)[7].

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