trout) nor Houston *et al.*[2] (in investigations on rainbow trout) observed any consistent variation in this ion. Hickman *et al.* describe an increase with temperature in the chloride levels of 8° and 16° C-acclimated trout. A similar situation was noted by Houston *et al.* in summer fish of the same species, but not in winter animals. A relationship of this type does not appear in the brown trout[5]. Increases in plasma potassium were observed by both Gordon and Houston *et al.* at elevated temperatures. A slight drop in the plasma concentration of this ion was, however, recorded by Hickman *et al.* Similarly, the studies on rainbow trout disagree with respect to the relationship between calcium levels and temperature. Both rainbow[2,4] and brook trout[6] display increases in total phosphate levels at higher temperature.

The significance to be attached to the differences observed in these studies is uncertain. It would appear, however, that both the set-points and precision of ionic regulation can be altered by temperature acclimation. Nevertheless, recent studies on thermal shock effects[4,7] suggest that the animals are capable of some considerable degree of compensation. The functional importance of this is clear with regard to sensory-neuromotor activity, and may be important in terms of metabolic adjustment to temperature as well[1,8].

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## Pyrogenicity of 'Pyromen'

Wyllie, Limbosch and Nyhus[1], in commenting on the results of Baume, Nicholls and Baxter[2], state that the inhibition of gastric secretion by lipopolysaccharide "was clearly described in 1950" by Blickenstaff and Grossman[3]. Blickenstaff and Grossman used 'Pyromen', a commercially available pyrogen (Nesset *et al.*[4]), which is very impure and only partially characterized. It was for this reason that their results were not discussed by Baume *et al.*[2] in their brief communication.

'Pyromen' is an extract of *Pseudomonas* obtained from autoclaved cells by tryptic digestion[4]. The material represented 25–35 per cent of the cell mass. Analyses for nucleic acids, protein and ash accounted for approximately 80 per cent of cell dry weight and 50 per cent of the extract. The authors[4] then assumed that the remainder represented pyrogenic polysaccharide. From newer methods of extraction it is concluded that lipopolysaccharide represents 1–2 per cent of cell mass rather than the 20 per cent inferred by Nesset and co-workers.

The actual amount of lipopolysaccharide in the preparation of 'Pyromen' is difficult to deduce. From the chemical analyses no conclusions are possible. Studies on pyrogenicity indicated a three-fold increase in activity compared with whole cells which could mean the presence of 5 per cent lipopolysaccharide, but the pyrogenic dose was at least one thousand times greater than that for the purified lipopolysaccharide used by Baume *et al.*[2].

From its biological properties, we may presume that 'Pyromen' contains some lipopolysaccharide but, unlike Wyllie *et al.*[1], conclude that there is no evidence that the lipopolysaccharide component was responsible for the inhibition of gastric secretion. In contrast, Baume *et al.*[2] used a purified lipopolysaccharide which has been fully characterized[5], and was made available in a water soluble form[6]. This left no doubt that the effects on gastric acid secretion were caused by the lipopolysaccharide.

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## Permeability and Excitation of Insect Muscle

THE membrane of most nerve and muscle cells is assumed to be more permeable to potassium ions than to sodium ions in the resting condition and the potential observed approaches that of a potassium electrode as predicted by the Nernst equation. The plasma membrane becomes much more permeable to sodium ions during excitation, and the potential across the membrane reverses, producing a positive overshoot of the action potential. The skeletal muscles of herbivorous insects, however, have unusual bioelectric properties and have been studied by Wood[1–4] and Huddart[5–7]. Analysis of the haemolymph and myoplasm of Orthoptera, Dictyoptera and Lepidoptera reveals an unusual ionic distribution[8] and it is suggested that the observations of the resting and action potentials fail to agree with the values predicted by the Nernst equation and with the classical studies of excitable cells. For example, frequently no positive overshoot to the action potential has been recorded. It has been suggested that active transport of chloride ions may contribute to the resting potential[4], and that an inward movement of sodium[9] or magnesium ions[1,9] may participate in the action potential. Our present knowledge in this field has been reviewed by Aidley[9].

If sodium ions enter the cell during the action potential some system must be present to remove them. A sodium–potassium–magnesium–ATPase has recently been found in the plasma membrane of many tissues of other species and is believed to be associated with active cation transport[12]. A study of the enzyme properties of the muscle membrane of herbivorous insects could represent the first step in establishing which ions move and carry the ionic current during excitation.

Cockroach (*Periplaneta americana*) coxal muscle was therefore studied to determine whether a sodium–potassium–magnesium–ATPase was present in a microsomal preparation. The muscle microsomal fraction was prepared according to experimental details given elsewhere[10] except that the isolation medium contained 10 mM EDTA. ATPase activity was determined as the inorganic phosphorus liberated[11] in 15 min at 25° C, the reaction being linear during this period. Other conditions of incubation are given in Table 1, in which the results of two representative experiments are summarized. It can be seen that the magnesium–ATPase activity constitutes much of the total activity; addition of either sodium

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