

50 per cent of the enzyme catalysed production of reducing groups is lost on heating at 65° C for 5 min, again without loss of iodine binding-power; only α -amylase activity can now be demonstrated. For want of a better name, these activities have been designated polysaccharidase *A* and *B*. It is interesting that the total saccharification power of the applied extract can be accounted for by summing up the activities of band regions *a*, *b* and *c-e*. Experiments are in progress to provide a better understanding of the relationships between the polysaccharidases and α -amylase.

In order for this electrophoretic method to be valuable in work concerning internal changes of protein pattern with morphogenesis and differentiation, recoveries of protein and, indeed, enzyme activity from the gel column after electrophoresis must be put on a quantitative basis. The results in Table 1 show that recoveries of protein are excellent for all concentrations of protein applied but become essentially complete when 50 μ g of protein or more are applied. One would then expect that enzyme activities should be similarly recovered, and as seen in Table 2 the results confirm this expectation. Although experiments with carbohydrases only are presented at this time, there is good evidence that certain amino-acid activating enzymes present in *Physarum* can be recovered in the same manner.

Table 1. RECOVERY OF PROTEIN AFTER ELECTROPHORESIS

Protein applied to gel (μ g)	(No. of experiments)	Protein recovered from gel (μ g)	(No. of experiments)
10 \pm 2	(3)	9 \pm 2	(3)
7 \pm 3	(3)	6 \pm 3	(3)
12 \pm 3	(4)	10 \pm 2	(4)
50 \pm 3	(3)	48 \pm 4	(3)
48 \pm 3	(3)	48 \pm 3	(3)
48 \pm 2	(3)	45 \pm 2	(3)
100 \pm 1	(3)	100 \pm 1	(3)
99 \pm 2	(3)	99 \pm 3	(3)
95 \pm 2	(3)	94 \pm 2	(3)
350 \pm 4	(3)	348 \pm 5	(3)
345 \pm 3	(3)	340 \pm 5	(3)
350 \pm 4	(5)	348 \pm 4	(5)

No. of experiments shown in parentheses.

Table 2. RECOVERY OF ENZYME ACTIVITY AFTER ELECTROPHORESIS

Enzyme	Specific activity* of extract	(No. of experiments)	Specific activity* of band regions after electrophoresis†	(No. of experiments)	Specific activity* recovered from whole gel column after electrophoresis	(No. of experiments)
Amylase	35 \pm 2	(5)	175 \pm 3	(5)	35 \pm 2	(5)
Polysaccharidase <i>A</i>	12 \pm 1	(5)	100 \pm 2	(5)	11 \pm 1	(5)
Polysaccharidase <i>B</i>	10 \pm 1	(5)	130 \pm 1	(5)	13 \pm 1	(5)

* mg maltose per mg protein per 20 min.

† α -Amylase was obtained from band *c* to *e*, polysaccharidase *A* from band *a*, and polysaccharidase *B* from band *b*.

Total protein applied to each tube equals 350 μ g.

No. of experiments shown in parentheses.

It is apparent that this method is suitable for quantitative studies on protein patterns in *Physarum*. The application of this method can only result in further information which may lead to a biochemical definition of morphogenesis in this organism.

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PHYSIOLOGY

A Spinal Anaesthetic with Long Duration of Action

It is often desirable to deliver pregnant sheep by Caesarean section under epidural or spinal anaesthesia so that the respiratory and cardiovascular functions of the new-born lamb will not be depressed by a general anaesthetic. A single injection of the local anaesthetics at present available is sometimes not enough to provide anaesthesia of sufficient duration for experimental purposes. A new local anaesthetic, 2(2,6 dichlorophenoxy) ethyl dimethylamine hydrobromide (*SK* and *F 90,054*)¹ has proved to have a substantially longer duration of action.

Table 1. DURATION OF ACTION OF EPIDURAL AND SPINAL ANAESTHETICS IN YOUNG SHEEP: MEANS \pm S.E.

Time (min) from injection until	Epidural		Intrathecal	
	<i>SK</i> and <i>F 90,054</i>	Lignocaine	<i>SK</i> and <i>F 90,054</i>	Lignocaine
Loss of sensation	2.9 \pm 0.7	3.7 \pm 0.5	1.0 \pm 0.5	1.6 \pm 0.5
Return of sensation	205 \pm 23	92 \pm 22	156 \pm 36	59 \pm 9
Abolition of muscle movements	8.5 \pm 4.1	11.2 \pm 3.3	3.7 \pm 0.7	—
Return of muscle movements	128 \pm 31	67 \pm 13	123 \pm 38	—
Able to stand again	255 \pm 32	137 \pm 25	176 \pm 35	95 \pm 25

Ten Clun ewe lambs 9–10 months of age, weighing 30–39 kg with a mean crown-rump length of 78 cm, were used. Intrathecal (spinal) or epidural injections were given through the lumbosacral space in doses which anaesthetized the spinal nerves from L2–3 caudally. Lignocaine and *SK* and *F 90,054* (20 mg/ml.) were administered as a hyperbaric solution in 6 per cent (w/v) glucose. Identical volumes (1.5 ml. intrathecally or 4.5 ml. epidurally) of either solution were injected into each lamb at intervals of a week. The withdrawal of the hind-limbs in response to pinpricks below the hock was used as an index of sensory block; loss of tonus and inability to move the limbs on change of posture were used to assess motor block. As Table 1 shows the duration of anaesthesia caused by *SK* and *F 90,054* was greater than that caused by lignocaine after both epidural and spinal administration. There was no significant difference in the time of onset of anaesthesia after injection. After intrathecal injection lignocaine failed to abolish limb movements completely in several of the lambs, whereas *SK* and *F 90,054* caused complete loss of tonus. All the lambs fully recovered after both anaesthetics; there were no neurological abnormalities 12 h later or afterwards.

It is concluded that *SK* and *F 90,054* is a local anaesthetic with a duration of action more than twice that of lignocaine, which is suitable for spinal and epidural anaesthesia and the use of which may prove to be much wider than the present application.

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¹ British Patents 765,849 and 765,850.

A New Local Anaesthetic with a Long Duration of Action

We have previously reported that choline 2,6-xylyl ether bromide (*TM 10*, 'Xylocholine') is a potent and long-lasting local anaesthetic, and that its tertiary congener (*SK* and *F 90,033*) is also a local anaesthetic with a potency and duration of action similar to cocaine¹.

We have since prepared and tested some substituted 2-phenoxyethylalkylamines of the type shown in the general formula: