α-Amylase and its Release by Prostaglandin $F_{2\alpha}$ in Barley Endosperm Slices

3′5′ cyclic AMP (cyclic AMP) has been implicated[1,2] as an intermediate in the action of a number of mammalian hormones. It may also have a role in gibberellic acid triggered release of α-amylase during cereal grain germination[3,4]. It now appears that prostaglandins may be concerned in regulation of cyclic AMP levels in a variety of mammalian tissues. Most of the work reported has concentrated on the action of prostaglandins E_1 and E_2. In general they decrease cyclic AMP concentration in adipose tissue and increase it in most other tissues studied[5]. Prostaglandin $F_{2\alpha}$ has been reported to have no effect on adenyl cyclase in thymic lymphocytes[6] or intact uterus strips[7].

The present work indicates that prostaglandin $F_{2\alpha}$ can trigger release of α-amylase in barley endosperm slices. The barley used was *Hordeum vulgare* L. var. Maris Otter, dehusked by treatment with 50% H_2SO_4 and stored at room temperature. 2 mm endosperm slices in groups of 10 were incubated for 24 h at 25 °C with 4 ml of solution as indicated below. 1 ml *M* NaCl was added to the solutions before homogenizing in a Potter type homogenizer. The homogenates were left to stand for 1 h at room temperature before centrifuging (MSE bench centrifuge, 5 min, speed 10). α-amylase activity in the supernatant was assayed at 25 °C by the iodine-dextrin colour method of BRIGGS[8] and expressed in arbitrary units (AU) per 10 slices as described by DUFFUS[9].

Prostaglandins E_1 E_2 A_1 and $F_{2\alpha}$ prepared by Dr. J. E. PIKE were made available by the Upjohn Company, Kalamazoo, Michigan 49001. Prostaglandin $F_{2\alpha}$ supplied as the tromethamine salt was easily soluble in water. Solutions of prostaglandins E_1 E_2 and A_1 were prepared by dissolving 1 mg of prostaglandin in 0.1 ml of 95% ethanol and making up to 1.0 ml with sodium carbonate solution (0.2 mg/ml). The final pH was between 6 and 7.5.

The results show that prostaglandin $F_{2\alpha}$ at a concentration of $10^{-5}M$ can trigger α-amylase release in barley endosperm slices. The effect is small compared to that of gibberellic acid at a concentration of $10^{-5}M$ but is similar to that reported previously[3] for cyclic AMP at the same concentration. It does not appear to have an additive (or any) effect on the response to gibberellic acid. Prostaglandins E_1 E_2 and A_1 had no effect, either alone or in combination with gibberellic acid. A control experiment showed that no inhibitory effect on gibberellic acid triggered α-amylase release was observed with ethanolic sodium carbonate solution at the appropriate concentration. Polyunsaturated acids such as linolenic and linoleic acid, naturally occurring in mature barley seeds and thought to be precursors of prostaglandin in animals also had no effect.

The amount of α-amylase released appears to be finite and does not increase with long incubation. It may be suggested, therefore, that prostaglandin $F_{2\alpha}$ may bring about the release of a small amount of preformed α-amylase, possibly through the mediation of cyclic AMP.

Zusammenfassung. Prostaglandin $F_{2\alpha}$ kann die Freisetzung von α-Amylase im Endosperm von Gerste (*Hordeum vulgare* L.).

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Relative activities of α-amylase released by the action of gibberellic acid and prostaglandin $F_{2\alpha}$

Addition	α-amylase activity in AU/10 slices
Gibberellic acid ($10^{-5}M$)	0.34 ± 0.08 (14)[a]
Prostaglandin $F_{2\alpha}$ ($10^{-5}M$)	0.020 ± 0.013 (14)
Distilled water	< 0.0020 (14)
Prostaglandin E_1 ($10^{-5}M$)	< 0.0030 (4)
Prostaglandin E_2 ($10^{-5}M$)	< 0.0010 (2)
Prostaglandin A_1 ($10^{-5}M$)	< 0.0005 (2)

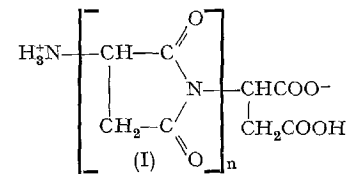
[a] The number of experiments is given in brackets.

1 E. W. SUTHERLAND, I. OYE and R. W. BUTCHER, Recent Prog. Horm. Res. *21*, 623 (1965).
2 G. A. ROBISON, J. Reprod. Fert. Suppl. *10*, 55 (1970).
3 C. M. DUFFUS and J. H. DUFFUS, Experientia *25*, 581 (1969).
4 A. G. GALSKY and J. A. LIPPINSCOTT, Pl. Cell Physiol. *10*, 607 (1969).
5 J. R. WEEKS, Naunyn Schmiedebergs Arch. Pharm. *269*, 347 (1971).
6 D. J. FRANKS, J. P. MCMANUS, J. F. WHITFIELD, Biochem. biophys. Res. Commun. *44*, 1177 (1971).
7 S. HARBON and H. CLAUSER, Biochem. biophys. Res. Commun. *44*, 1496 (1971).
8 D. E. BRIGGS, J. Inst. Brew. *67*, 427 (1961).
9 J. H. DUFFUS, J. Inst. Brew. *75*, 252 (1969).

The Hydrolysis of Polyimides[1]

Thermal polymerization of aspartic acid produces a polysuccinimide (I), a chain of aspartoyl residues[2,3]. We have investigated the alkaline hydrolysis of the imide rings of (I) which converts the polyimide to a polypeptide. The hydrolysis of imide rings has also interested investigators of the biological action of α-phthalimido-L-glutarimide (Thalidomide). The chemical reactivity of the phthalimide ring of Thalidomide has been established; for example, at pH 7 and 37 °C hydrolysis of the phthalimide ring proceeds at a significant rate[4].

The alkaline hydrolysis of polyimides can be expected to be kinetically complex due to increasing negative charge generated by carboxylate groups[5]. For this reason, a diimide, phthaloyl-DL-aspartoyl-β-alanine (IIA) was synthesized for a progressive study of the hydrolysis of polyimides. In addition, this diimide (IIA) can be related



(I)