through muscarinic receptors or acetylcholine release. Our results show that one of the mechanism of cholinomimetic activity of prolactin is by inhibition of ChE. This anti-ChE action may be responsible for the earlier reported cholinomimetic activity of prolactin.

It is well known that anti-ChE agents like di-isopropyl fluorophosphate and physostigmine are significant analgesics in various experimental procedures (Koehm & Karczmar 1978). It is possible that the anti-ChE activity of prolactin we have found might play a part in the analgesic action. This view is supported by our observation that a significant correlation exists between anti-ChE and analgesic activity. However, although earlier studies on prolactin analgesia showed an involvement of the opioid system (Ramaswamy et al 1983) the absence of any effect of naloxone on ChE activity and its failure to reverse the anti-ChE activity of prolactin in the present work suggest that opioid mechanisms may not be involved in that action of prolactin.

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J. Pharm. Pharmacol. 1988, 40: 307-308
Communicated December 18, 1987

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Dopamine and apomorphine do not modulate the uptake of [^{3}H]D-aspartate in the rat striatum in-vitro

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Abstract—Sodium-dependent [^{3}H]D-aspartate uptake was measured in rat striatal homogenates. The uptake was inhibited by both L- and D-glutamate, with IC50 values of 5·6 and 224 μM, respectively. Dopamine (10^{-7}–10^{-4} M), apomorphine (10^{-7} M), sulpiride (10^{-6} M) or a combination of dopamine and sulpiride were found not to affect the observed uptake of [^{3}H]D-aspartate. Thus, the in-vitro dopaminergic modulation of high affinity glutamate uptake reported in the literature is not found when [^{3}H]D-aspartate is used instead of [^{3}H]L-glutamate.

There are many studies in the literature demonstrating that the release of a neurotransmitter from its nerve endings can be modulated by other neurotransmitter systems (for review, see Chesselet 1984). In the case of glutamatergic neurons, it has been demonstrated that potassium-stimulated release of tritium from striatal slices preincubated with [^{3}H]L-glutamate is inhibited by dopamine and its agonists in a manner that can be blocked by dopamine receptor antagonists (Mitchell & Doggett 1980; Rowlands & Roberts 1980). A dopaminergic modulation of the high affinity uptake of [^{3}H]L-glutamate into rat striatal (but not frontal cortical) homogenates has also been reported, with 10^{-7} M dopamine, apomorphine and bromocriptine producing 34, 19 and 20% inhibition, respectively, of the uptake (Nieoullon et al 1982, 1983).

One of the disadvantages associated with the use of [^{3}H]L-glutamate in these types of studies is the rapid metabolism of the amino acid, a problem that can be avoided by the use of [^{3}H]D-aspartate, which uses the same neuronal uptake system as glutamate (see Logan & Snyder 1972; Taxt & Storm-Mathisen 1984). In the present study, the effects of dopamine, apomorphine and sulpiride on the uptake of [^{3}H]D-aspartate into rat striatal homogenates has been investigated.

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Materials and methods

St. Mary's bred male and female Fisher rats, 200–300 g, were used in the study. The rats were decapitated and their brains rapidly removed, placed on ice, and the striata dissected. The uptake experiments were undertaken by a method based on that of Storm-Mathisen (1977) and similar to that used by Nieoullon et al (1982, 1983), with the exceptions that the whole striatum (rather than the rostral third) was used, [^{3}H]D-aspartate was used in place of [^{3}H]L-glutamate, the incubations were at 37 °C rather than at 25 °C, and the reactions were terminated by filtration rather than centrifugation. Briefly, the striata were homogenized 1:20 (w/v) in 0·32 M ice-cold sucrose, and 50 μL of homogenate added to incubation wells containing the dopamine agonist/antagonist under test dissolved in 460 μL Krebs phosphate buffer, pH 7·4. Reactions were started by the addition of 50 μL [^{3}H]D-aspartate (final concentration 65·7 nM at a specific activity of 14 Ci mmol^{-1}). After a 3 min incubation at 37 °C, the reactions were terminated by rapid filtration through Whatman GF/B filter papers, using a Brandel Cell Harvester. The filters were washed four times with isotonic saline and then allowed to dry before determination of the tritium content by liquid scintillation spectroscopy. Sodium-free uptake of [^{3}H]D-aspartate was determined by replacement of sodium salts in the Krebs buffer by iso-osmotic Tris HCl, pH 7·4.

[^{3}H]D-Aspartate was obtained from Amersham International plc, Amersham, UK. Dopamine hydrochloride, apomorphine

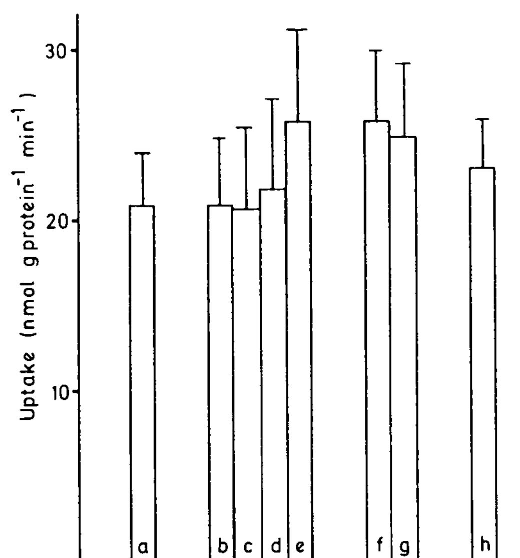

FIG. 1. Effect of dopamine (DA, 10^{-7}–10^{-4} M), apomorphine (Apo, 10^{-7} M), sulpiride (Sulp, 10^{-6} M) or a combination of sulpiride and dopamine on Na^+-dependent [^{3}H]D-aspartate uptake into rat striatal homogenates. Values are means ± s.e.m., n = 4–9. In no case were the values significantly different from the control values ($P > 0{\cdot}05$, two-tailed *t*-test). Key: a, control; b, DA 10^{-7}; c, DA 10^{-6}; d, DA 10^{-4}; e, Apo 10^{-7}; f, DA 10^{-7} + Sulp; g, DA 10^{-4} + Sulp; h, Sulp.

hydrochloride and sulpiride were obtained from the Sigma Chemical Co., Poole, UK. All other reagents were standard laboratory reagents of analytical grade wherever possible.

Results and discussion

Initial experiments indicated that in the absence of Na^+, there was little or no uptake of the [^{3}H]D-aspartate into the homogenates, in agreement with the literature (Davies & Johnston 1976). In addition, the Na^+-dependent uptake of [^{3}H]D-aspartate was inhibited by both L- and D-glutamate, with IC50 values (calculated from the mean (n = 10) competition curves with 11 concentrations of the inhibitors) of 5·6 and 224 μM, respectively, a finding in agreement with the literature (Balcar & Johnston 1972; Davies & Johnston 1976).

The effects of dopamine (10^{-7}–10^{-4} M), apomorphine (10^{-7} M), sulpiride (10^{-6} M) and a combination of sulpiride and dopamine on Na^+-dependent [^{3}H]D-aspartate uptake are shown in Fig. 1. None of the conditions significantly affected the rate of uptake. Such a finding differs from the reports of Nieoullon et al (1982, 1983) that dopamine and apomorphine, at concentrations of 10^{-7} M, are able to significantly reduce the high affinity uptake of [^{3}H]L-glutamate. Whether or not the difference between the present results and the results of Nieoullon et al (1983) reflect the different uptake substrates used is not yet elucidated, although in a recent study by the same group it was reported that 10^{-8} M dopamine did not affect striatal high affinity glutamate uptake, whereas this concentration of dopamine significantly reduced the enhanced high affinity glutamate uptake produced by prior electrical stimulation of the frontal cortical areas (Kerkerian et al 1987).

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