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LETTERS TO THE EDITOR

Decrease in homovanillic acid as evidence for dopamine receptor stimulation by apomorphine in the neostriatum of the rat

Substances known for their ability to block the central catecholamine receptors, for example chlorpromazine or haloperidol, increase the concentration of homovanillic acid (HVA) in the neostriatum of various species (Andén, Roos & Werdinius, 1964; Laverty & Sharman 1965; Juorio, Sharman & Trajkov 1966; Sharman 1966). This has been suggested to depend on a feed-back mechanism resulting in an increase in the synthesis of dopamine to compensate for the effect of blocking the receptors (see Carlsson & Lindqvist 1963). The opposite phenomenon, a decrease in HVA after stimulating the catecholamine receptors with some dopamine-like transmitter, i.e. a compound with an effect similar to that of dopamine, has not been reported. However, in 1967, Ernst suggested that the compulsive gnawing induced by apomorphine in rats is not mediated via the release of catecholamines, as it is after treatment with amphetamine, but caused by the action of apomorphine on the dopamine receptors themselves. Further, evidence of a direct stimulating effect of the dopamine receptor in the rat brain by apomorphine was recently presented by Andén, Rubenson & others (1967) and Butcher & Andén (1969). These authors also demonstrated that apomorphine retarded the depletion of dopamine caused by a tyrosine hydroxylase inhibitor (H 44/68) and explained this by a negative feed-back mechanism due to activation of the dopamine receptor. I now report the effect of apomorphine on the level of HVA in the rat brain.

In four experiments, 12 male Sprague-Dawley rats were treated with apomorphine 15 mg/kg i.p. twice, with an interval of 1 h and killed 2 h after the first injection. All rats showed the typical compulsive gnawing. Four groups of 12 rats received no treatment. The level of HVA in the neostriatum of untreated rats was $0{\cdot}47 \pm 0{\cdot}049$ and of treated rats $0{\cdot}10 \pm 0{\cdot}025$ (μg/g; mean $\pm$ s.e.). The decrease in the levels of HVA is statistically highly significant ($P < 0{\cdot}001$, Student's t-test). When this finding is considered in the light of the unchanged levels of dopamine after apomorphine found by Andén & others (1967) in the whole brain of the rat, a pattern, seen in earlier studies with 5-hydroxytryptamine (5-HT) emerges. 5-HT and its corresponding acid, 5-hydroxyindoleacetic acid (5-HIAA), were measured at the same time after lysergic acid diethylamide and there was a decrease in the synthesis of 5-HT (Diaz, Ngai & Costa, 1968). The lack of significant change in the level of the dopamine found by Andén (1967) and the highly significant decrease in the HVA that I found, suggests a reduced activity in the dopamine neurons after apomorphine. If the action of apomorphine were to be mediated by release of dopamine, the level of dopamine might possibly remain unchanged but the HVA level would be increased significantly. Further, inhibition of monoamine oxidase would not leave the dopamine level unchanged. The decrease in HVA after apomorphine that I observed, correlates with the α-methyltyrosine-induced disappearance of dopamine shown by Andén & others (1967), and offers further biochemical evidence for the stimulating effect of the apomorphine on the dopamine receptor.

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Department of Pharmacology,
University of Goteborg, Sweden,
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BJÖRN-ERIK ROOS

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Activation of pepsinogen by sulphated glycosaminoglycans: a possible role in peptic ulcerogenesis

Sulphated glycosaminoglycans (SGS) occur in the gastrointestinal tract at lumenal and intracellular levels, and it has been suggested that they may be secreted by gastric zymogen cells (Schrager, 1964; Spicer, 1965; Spicer & Sun, 1967; Gerald, de Graeff & others, 1967). This association of SG and the source of gastric pepsin has led to the suggestion of an antipeptic role for SG at intracellular level (Gerald & others, 1967). Our experiments now suggest that a more subtle relation may exist between the zymogen pepsinogen and SGS.

Back diffusion of hydrogen ion in the abnormally permeable gastric mucosa present in gastric ulcer has been shown (Davenport, 1965; Overholt & Pollard, 1968); there is therefore a distinct possibility that at least regions of specific gastric cells may attain an abnormally low pH. With the chief cell, absorption of hydrogen ion would give rise to a system containing pepsinogen, hydrogen ion and SG. While the intracellular concentration of hydrogen ion may not reach a level sufficient to activate the pepsinogen at a significant rate, any marked increase in the rate of activation could, in conjunction with the autocatalytic nature of the acid activation of pepsinogen (Herriott, 1938), cause significant pepsin production and possibly consequent intracellular proteolysis.

Pepsinogen (Sigma Chemical Co.) was activated at various pH values and constant ionic strength for varying periods of time in the presence and absence of the SGS chondroitin sulphate-A or heparin, and the resulting pepsin was destroyed by raising the pH to 8. The pepsinogen remaining (unactivated) was measured by rapid activation at pH 1·6 followed by assay of the peptic activity using the haemoglobin digestion method. This technique (after Herriott, 1938) allows the effect of the SGS on the activation of pepsinogen, determined by difference, to be observed.

Pepsinogen (10 mg) was dissolved in 0·002 M phosphate buffer pH 6·9 (25 ml), and the varying amounts of SG added* to this solution which was kept at 37° for 10–15 min. Acetate buffer (9 ml) (Long, 1961), ionic strength 0·05, at the pH of activation (pH 3·6, 4·0, 4·3) and 37° was added at zero time to start activation. Aliquots (2 ml) were removed at suitable time intervals, mixed with saturated sodium tetraborate solution (2 ml) which had been diluted so that the pH was thereby raised to 8–8·5, and allowed

* Heparin 0·2–5 mg; chondroitin sulphate 1–20 mg.