Influence of apomorphine hydrochloride, dibutyryl-apomorphine and Lysenyl on plasma prolactin concentrations in the rat

In agreement with Kamberi, Mical & Porter (1970), Hökfelt & Fuxe (1972) postulated that dopaminergic tubero-infundibular neurons inhibit the release of prolactin from the anterior pituitary gland in rats by stimulating the secretion of prolactin inhibiting factor (PIF). This theory is supported by the fact that substances which inhibit the dopaminergic neurotransmission, e.g. neuroleptics or reserpine, stimulate the growth of mammary glands and lactation by raising the serum level of prolactin (for review see Lu, Amenomori & others, 1970; Meites & Clemens, 1972; Meites, Lu & others, 1972; Sulman, 1970). Conversely, L-dopa, a substance which raises the dopamine concentration in the cns, can lower the level of serum prolactin (Lu & Meites, 1971). The same effect can be achieved using ergot alkaloids (for review see Floss, Cassady & Robbers, 1973). In this context it is worth noticing, that among other mechanisms of action a stimulating effect on central dopaminergic receptors is attributed to ergocornine and to the potent prolactin inhibitor 2-Br-α-ergocryptine (CB 154) Hökfelt & Fuxe, 1972, Corrodi, Fuxe & others, 1973).

We investigated the influence of apomorphine hydrochloride, its dibutyrylester, and the synthetic ergot alkaloid Lysenyl SPOFA [*N*-(D-6-methyl-8-isoergolenyl)*N'*-*N'*-diethylcarbamide hydrogenmaleate, lisuride hydrogen maleate] on the plasma prolactin concentration in the rat.

Apomorphine hydrochloride, a substance with a central stimulating effect on dopaminergic receptors, in doses of 1 and 10 mg kg^{-1}, significantly lowered the plasma prolactin concentration in mature ovariectomized rats substituted with oestrogens, although the effect only lasted (nearly independent of the dose used) for a short time (see Table 1). Because of this, for further work we used the dibutyrylester of apomorphine (ZK 48 241). In contrast to apomorphine hydrochloride, the dibutyrylester is effective after subcutaneous application of high dosages for much longer both as a reserpine antagonist in the mouse [12·5 mg kg^{-1}, s.c. in sesame oil antagonized the effect of reserpine (4 mg kg^{-1}, i.v. 24 h previously) for 12 h] and as a stereotyped behaviour-inducing agent in the rat. A single dose of 30 mg kg^{-1} ZK 48 241 led to a reduction in the plasma prolactin concentration in ovariectomized rats, substituted with oestrogens, which lasted for 24 h (see Table 1). When the dose was reduced, the primary effect was a shortening of the duration of action. A secondary effect was a reduction of the strength of the lowering effect on plasma prolactin concentration. As well as using ovariectomized rats substituted with oestrogens, we investigated the effect of ZK 48 241 and of Lysenyl on the plasma prolactin concentrations of intact mature rats pretreated with reserpine.

Wistar rats, 200 g were pre-treated with reserpine (2 mg kg^{-1}, i.p.) the day before treatment (at about 10 a.m.). 24 h later the animals were treated with the test substances and decapitated (1 h later Lysenyl groups and saline control; 6 h later ZK 48 241 group and sesame oil control). Plasma prolactin was measured by radioimmunoassay. A single dose of 3·0 mg kg^{-1} ZK 48 241 led to a distinct decrease in plasma prolactin concentrations from 135 ± 28 ng ml^{-1} (s.e.m.) NIAMD-RP-1* to 87 ± 17, while both 0·1 and 0·5 mg kg^{-1} Lysenyl reduced the control values of 186 ± 21 to 45 ± 4 and 42 ± 3. This shows that Lysenyl too in this model has prolactin-inhibiting properties.

* National Institute of Arthritis and Metabolic Diseases—Reference Preparation—1.

Table 1. *The influence of apomorphine hydrochloride and dibutyrylapomorphine (ZK 48 241) on plasma prolactin levels in the ovariectomized rat substituted with oestradiol-17β.*

Substance	Dose mg kg^{-1}	Time after injection of the test substance when blood was collected	n	Concentration of prolactin in plasma* $\bar{x}\pm$ s.e.m.
Experiment 1**				
Apomorphine HCl	10·0	30 min	5	19·6±2·1
	1·0	30 min	5	19·2±3·7
	0·1	30 min	5	19·0±1·2
Controls (0·9% NaCl)	—	30 min	5	53·8±14·5
Experiment 2***				
Apomorphine HCl	10·0	30 min	5	13·4±1·3
	1·0	30 min	4	16·2±5·5
	0·1	30 min	5	27·6±4·3
Controls (0·9% NaCl)	—	30 min	4	61·0±21·8
Experiment 3****				
Apomorphine HCl	1·0	2 h	6	58·0±13·4
ZK 48 241	30·0	2 h	7	25·0±4·0
Controls (sesame oil)	—	2 h	6	77·8±12·5
Apomorphine HCl	1·0	24 h	7	75·0±15·3
ZK 48 241	30·0	24 h	7	45·1±2·1
Controls (sesame oil)	—	24 h	7	123·4±34·4

*Expressed as ng ml^{-1} of NIAMD–RP–1.

**Wistar rats, 200 g, ovariectomized and substituted for 3 days with 2 μg oestradiol–17β rat^{-1} day^{-1} in sesame oil s c. Apomorphine HCl was administered 1½–3 h after the last oestrogen injection, that is between 9·30–11.00 a.m. on the 3rd day. Blood was collected by puncture of the external jugular vein.

***The same rats and conditions as described under ** were used, but blood samples were taken one day later (on the 4th day of substitution with oestradiol–17β).

****Wistar rats, 200 g, ovariectomized and substituted with 10 μg oestradiol–17β rat^{-1} day^{-1} in sesame oil s.c. The test substance was applied on the 9th day of substitution between 9·30 and 11·00 a.m. All doses of ZK 48 241 refer to apomorphine base. Blood was collected in the same way as described under ** but 2 h and 24 h after injection of the test substance. Between the two blood sample collections the animals were given another 10 μg oestradiol–17β injection in the afternoon.

It is used therapeutically mainly because of its antiserotoninergic effect (Aušková, Rezabek & others, 1974; Podvalová, Votáva & Dlabač, 1970). It also shows reserpine antagonistic effects and produces stereotyped behaviour in mice [ED 50 Lysenyl after 30 min in reserpine-treated (4 mg kg^{-1}, i.v. 24 before) mice (with 95% confidence limits): 0·41 (0·22–0·59) mg kg^{-1} for motor activity and 0·57 (0·44–0·71) mg kg^{-1} for stereotypic activity]. Both effects are similar to those of apomorphine hydrochloride [ED 50 for apomorphine HCl, condition as for Lysenyl, 0·39 (0·22–0·65) mg kg^{-1} motor activity, 0·35 (0·28–0·48) mg kg^{-1} stereotypic activity]. They are thought to be the expression of a central dopaminergic effect (Andén, Strömbom & Svensson, 1973; Lal, Sourkes & others, 1972). Furthermore, Lysenyl is known to suppress lactation in man and rats (Střibrný, Hájek & others, 1973; Aušková, & others, 1974) and therefore its effect on plasma prolactin concentration is of special interest. The results described on plasma prolactin concentration might be explained by the stimulation of

central dopaminergic systems. With Lysenyl, its known antiserotonin activity may also be involved.

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Ganglion blocking action of indomethacin

Hedqvist (1970) has shown that prostaglandins modulate adrenergic transmission. There are also several instances of changes in sympathetic activity when prostaglandin synthesis is inhibited by drugs such as indomethacin or aspirin. These include increased responses of the spleen to adrenergic nerve stimulation (Ferreira & Moncada, 1971) and increased concentrations of noradrenaline in the urine produced by an isolated kidney perfused with indomethacin (Junstad & Wennmalm, 1972). In view of these findings, we anticipated that the sympathetically mediated pressor reflex to carotid occlusion would be potentiated as a consequence of treatment with indomethacin, and we were surprised to find that the reflex was inhibited. We therefore attempted to discover why this happened, by studying the carotid occlusion response (COR) and the responses of the cat nictitating membrane simultaneously.

Arterial blood pressure was recorded from the femoral artery of cats under chloralose anaesthesia at 80 mg kg^{-1}. Indomethacin was infused through the left femoral vein, and the the right femoral vein was cannulated for injection of noradrenaline at 1–1·5 $\mu g\ kg^{-1}$.

The COR was obtained at 15 min intervals by occlusion of both carotid arteries for 20 s. Contractions of the nictitating membrane were produced by pre- and post-ganglionic stimulation of the superior cervical sympathetic trunk for 10 s using square pulses of 2 ms at 20 Hz.

Indomethacin was infused at 100 $\mu g\ kg^{-1}\ min^{-1}$ (i.v.). The infusion lasted 60 min (total dose 6 mg kg^{-1}) in those experiments which were designed to study changes in COR and blood pressure response to bolus injections of noradrenaline, whereas the infusion was stopped after 30 min (total dose 3 mg kg^{-1}) in those experiments in which nictitating membrane contractions were studied.