J. Pharm. Pharmac., 1972, **24**, 447–451 *Received February* 3, 1972

On the localization of the hypotensive effect of L-dopa

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Mean arterial blood pressure was recorded in anaesthetized rats before and after a mid-collicular transection of the brain (decerebration). Basal blood pressure was not changed by the decerebration. Injection of L-dopa (200 mg/kg, i.p.) after peripheral dopa decarboxylase inhibition by L-α-hydrazino-α-methyl-β-(3,4-dihydroxyphenyl) propionic acid (MK 486, 100 mg/kg, i.p.) resulted in a significant reduction of arterial pressure to the same level in both control (sham-operated) and decerebrated rats after 30 min. In other experiments, anaesthetized rats were spinalized at C7-Th1. Basal blood pressure became significantly lower than in control and decerebrated rats and L-dopa after MK 486 in the same doses did not affect blood pressure. Biochemical determinations of noradrenaline and dopamine showed that administration of L-dopa after MK 486 to decerebrated rats in the same doses as in the blood pressure experiments resulted in a pronounced increase of dopamine in both parts of the brain.

After inhibition of dopa decarboxylase in peripheral tissue, L-dopa and structurally related compounds induce a hypotensive effect which is most likely due to effects on the central nervous system (Henning & Rubenson, 1970a, b, 1971; Rubenson, 1971a, b). So far, the localization of the site at which the hypotensive effect is exerted within the central nervous system has not been examined.

The organization and functioning of central cardiovascular regulation is incompletely understood but the integration of cardiovascular reflexes is believed to occur in a network of neurons in the lower brain stem. These medullary cardiovascular stations are often collectively named the vasomotor centre and there is experimental evidence for the existence of both a pressor and a depressor area (Oberholzer, 1960; Folkow, Heymans & Neil, 1965). In the precise localization of the depressor area, one of the important structures appears to be the nucleus tractus solitarii (Alexander, 1946; Scherrer, 1966). This structure has a rich network of noradrenaline nerve terminals as revealed histochemically (Dahlström & Fuxe, 1965); this may suggest an involvement of this area in the hypotensive action of L-dopa as well as of other structurally related compounds.

In an attempt to assess the importance of the medullary vasomotor centres for the hypotensive effect of L-dopa, we have examined its effects in rats after transection of the brain stem at collicular level, which leaves the centres intact. Further, the effect of L-dopa was studied in animals in which the spinal sympathetic pathways were interrupted by spinalization at a high level. The results indicate that the blood pressure lowering effect of L-dopa may be exerted on a site in the lower brain stem.

METHODS

Male Sprague–Dawley rats, 180–250 g, were anaesthestized with pentobarbitone sodium (35 mg/kg, i.p.) and both common carotid arteries were ligated and a poly-

ethylene catheter introduced into the left carotid artery. A tracheal cannula was inserted. The skull was opened on both sides of the midline immediately rostral to the sutura coronaria. Blunt transection of the midbrain was performed at the level of the anterior colliculus (see Fig. 1). The level and completeness of the transection

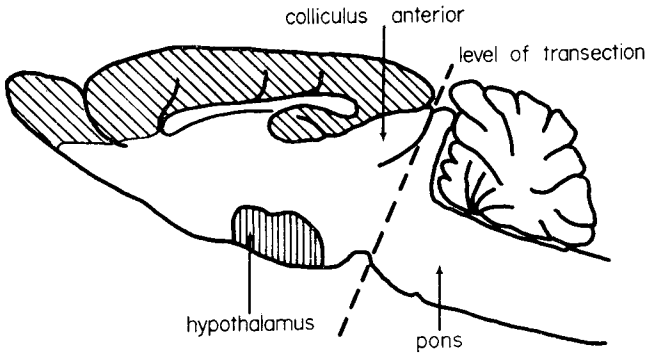

FIG. 1. Sagittal section through rat brain indicating the level of transection used in the present experiments. The part of the brain rostral to the transection is referred to as forebrain and the part caudal to the transection as the brain stem.

was always checked at the end of the experiments. To test the effect of midcollicular decerebration, animals were sham-operated (skull and dura opened) and thereafter transected as described above. Mean arterial blood pressure was recorded before and after decerebration by attaching the carotid catheter to a Statham P23Dc pressure transducer writing on a Grass Model 7 Polygraph. Pentobarbitone anaesthesia was maintained throughout the experiment.

In other experiments the spinal cord was transected at the level of C7-Th1 under pentobarbitone anaesthesia and blood pressure was recorded as described above.

The blood pressure values given represent averages of recordings for 5 min periods immediately before and at various time intervals after drug administration as reported in Results.

Biochemical determinations of dopamine (Bertler, Carlsson & Rosengren, 1958) and noradrenaline (Carlsson & Lindqvist, 1962) were made in rats treated as in the blood pressure experiments. The animals were killed by exsanguination while still under anaesthesia 30 min after the injection of L-dopa. Brain dissection was performed rapidly on an ice-cold glass plate.

The drugs used were: L-α-hydrazino-α-methyl-β(3,4-dihydroxyphenyl) propionic acid (MK 486) and L-3,4-dihydroxyphenylalanine (L-dopa). Drugs were administered intraperitoneally dissolved in warm saline (5–10 ml/kg), to which a few drops of N HCl had been added. Control animals received the vehicle. Doses and time intervals are given in Results. Tests of significance were by Student's *t*-test after analysis of variance by two independent criteria of classification. *P* values less than 0·05 were regarded as significant.

RESULTS

Blood pressure experiments

In one series of experiments the blood pressure was recorded before and after decerebration. The blood pressure of sham-operated rats was 125 mm Hg (s.e.

4·0, n=5) and this value was not significantly different from that observed in the same rats 30 min after decerebration (123 mm Hg, s.e. 4·7, n=5).

In two other series of experiments in decerebrated and sham-operated rats the effect of L-dopa (200 mg/kg) was examined 30 min after pretreatment with MK 486 (100 mg/kg); control decerebrate animals were given MK 486 and saline. The results are given in Fig. 2. Again, no significance in basal blood pressure was found between sham-operated and decerebrated animals (Fig. 2A and C). Administration of MK 486 did not lower blood pressure significantly in either group of rats. Administration of saline to decerebrated rats had no effect on blood pressure as recorded after 30 and 120 min ($P > 0{\cdot}1$, Fig. 2B). L-Dopa given after MK 486 lowered blood pressure after 30 min significantly both in decerebrated ($P < 0{\cdot}005$, Fig. 2C) and in sham-operated rats ($P < 0{\cdot}001$, Fig. 2A). Two h after L-dopa the blood pressure in decerebrated rats was still significantly depressed compared to basal blood pressure ($P < 0{\cdot}01$) but not compared to the pressure recorded before MK 486. In sham-operated animals the blood pressure had returned to control level 2 h after L-dopa. L-Dopa lowered blood pressure to the same level in both groups of rats.

In spinal rats initial blood pressure was 76 mm Hg (s.e. = 4·8, n=4), which is significantly lower than basal pressure of both sham-operated and decerebrated rats ($P < 0{\cdot}001$), Administration of MK 486 (100 mg/kg) did not change blood pressure after 30 min (Fig. 2D). L-Dopa (200 mg/kg) given 30 min after MK 486 caused no significant changes as recorded after 30 and 120 min.

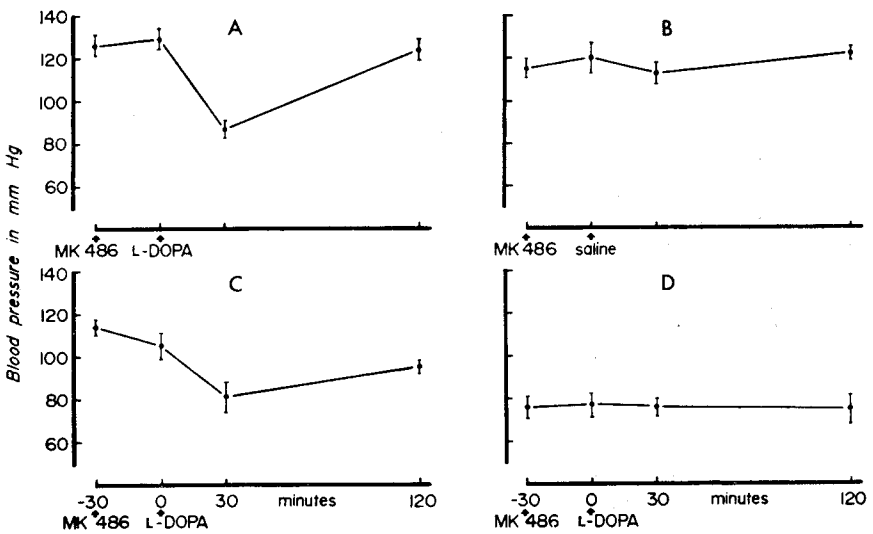

FIG. 2. Changes in mean arterial blood pressure in anaesthetized rats after i.p. injections of drugs as indicated. A. Sham-operated animals given L-dopa (200 mg/kg) 30 min after MK 486 (100 mg/kg) (n=4). B. Decerebrated animals given saline 30 min after MK 486 (100 mg/kg) (n=4). C. Decerebrated animals given L-dopa (200 mg/kg) 30 min after MK 486 (100 mg/kg) (n=8). D. Spinal animals given L-dopa (200 mg/kg) 30 min after MK 486 (100 mg/kg) (n=4). The values are means with s.e. in mm Hg.

Biochemical experiments

Concentrations of dopamine and noradrenaline were determined in the two parts of the brain in decerebrated rats after a combined treatment with MK 486 (100

mg/kg, i.p.) given 60 min before death and saline or L-dopa as given in the blood pressure experiments 30 min before death. Forebrain values for the saline group were 0·27 for noradrenaline and 0·80 for dopamine and for the L-dopa group 0·32 and 4·30 μg/g tissue. Brain stem values for the saline group were 0·42 and 0·04, and for the L-dopa group 0·35 and 1·54 (μg/g tissue) for the respective amines. Variance within groups in the dopamine series was 0·0977, and in the noradrenaline series 0·0029 (n=4).

In control rats, significantly more noradrenaline was found in the brain stem than in the forebrain. On the other hand, only insignificant amounts of dopamine were found in the brain stem. Administration of L-dopa resulted in a pronounced increase of dopamine in both parts of the brain in sham-operated as well as decerebrated rats. No significant changes in noradrenaline content was observed in either part of the brain ($P > 0·1$).

DISCUSSION

The present results show that separation of telencephalic and diencephalic brain regions by a transection at midcollicular level does not influence basal arterial pressure in anaesthetized rats, while a high spinal transection causes a severe fall in pressure. This is in keeping with observations by Alexander (1946) who found that brain transections as far caudally as the lower third of the pons had no significant effect on blood pressure in anaesthetized cats.

We further noticed that the hypotensive response to L-dopa after peripheral dopa decarboxylase inhibition is present after a midcollicular decerebration. The magnitude of the response was largely the same as that seen in conscious rats (Henning & Rubenson, 1970 a, b) and was not altered by the decerebration procedure. However, the duration of the hypotension after L-dopa appeared to be longer in transected animals. In rats spinalized at the level of C7-Th1 the initial blood pressure was similar to that attained at the maximal hypotensive effect of L-dopa in the other group of animals. L-Dopa failed to lower blood pressure further in spinal rats, indicating that its hypotensive action is mediated via the spinal sympathetic pathways.

The biochemical experiments demonstrated that the brain stem contains hardly any dopamine while noradrenaline concentrations were higher in this region than in the forebrain, which is in agreement with previous findings (see Holtz & Palm, 1966). Administration of L-dopa after MK 486 resulted in the formation of large amounts of dopamine in both parts of the brain but no change in noradrenaline levels occurred. This was also found in an analysis of whole brain (Henning & Rubenson, 1970). However, these findings do not necessarily mean that the depressor effect of L-dopa is mediated through a direct action of dopamine on catecholamine receptors. A detailed analysis of the depressor action of L-dopa in conscious animals points to the importance of a displacement of endogenous noradrenaline by dopamine accumulated in the central nervous system (Rubenson, 1971 a, b).

The experiments reported appear to localize the hypotensive effect of L-dopa to the lower brain stem but it should be emphasized that further work is needed to permit a definite localization of the action within this region. In addition, an action on spinal sympathetic centres cannot be excluded.

Acknowledgements

This work has been supported by grants from the Medical Faculty, University of

Göteborg, and from the Swedish State Medical Research Council (Nos B72-14X-255-08A and B72-14X-2863-03). MK 486 was kindly supplied by Merck Sharp & Dohme International. Skilful technical assistance was given by Miss Ann-Sofie Järnström.

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