Reçu le 15 décembre 1982.

Effect of amineptine on the firing rate of central monoaminergic neurons in the rat

BY

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(3 figures)

The firing rate of central noradrenergic, serotonergic and dopaminergic neurons was recorded in rats anaesthetized with chloral hydrate. The i.v. perfusion of amineptine decreased the frequency of discharge of noradrenergic and dopaminergic neurons but had no effect on serotonergic cells. Quantitatively amineptine was a more potent inhibitor of dopaminergic than of noradrenergic neurons. These experiments *in vivo* confirm biochemical studies *in vitro* reporting a selective action of amineptine on dopaminergic systems.

Introduction

Amineptine, (S-1694) [*N*-(dibenzo a, d cycloheptadien-5-yl)-7-aminoheptanoic acid] is a new compound whose structure is related to the tricyclic antidepressants (Fig. 1). In laboratory animals, amineptine has "antidepressant-like" properties as it antagonizes signs of depression induced by reserpine such as hypothermia, ptosis and catalepsia (POIGNANT, 1979; BORSINI *et al.*, 1981). Biochemical studies however differentiate amineptine from the tricyclic antidepressants. Amineptine increases the turnover of dopamine (DA) without modification of noradrenaline (NA) or 5-hydroxytryptamine (5-HT) metabolism (SAMANIN *et al.*, 1977), while it is well known that the tricyclic antidepressants reduce NA and 5-HT turnover without modification of DA metabolism (SCHUBERT *et al.*, 1970). The action of the tricyclic antidepressants on the amine turnover can be related to their inhibitory effect on the neuronal reuptake of NA and 5-HT (GLOWINSKI & AXELROD, 1964; CARLSSON *et al.*, 1968; ROSS & RENYI, 1975). Amineptine does not block the uptake of NA or 5-HT but it blocks DA reuptake and increases DA release from presynaptic neuronal pools. At high concentrations, it also increases the release of 5-HT (OFFERMEIER *et al.*, 1977). These studies suggest a different action of amineptine from the classical tricyclic antidepressants. It must however be pointed out that nomifensine, a new antidepressant, is a potent blocker of NA and DA uptakes (SCHACHT *et al.*, 1977).

Using electrophysiological techniques, it is possible to investigate *in vivo* the influence of a systemic administration of centrally acting drugs on the firing rate of central monoaminergic neurons. In a previous quantitative study, we have shown that the tricyclic antidepressants reduce the firing rate of central noradrenergic and/or

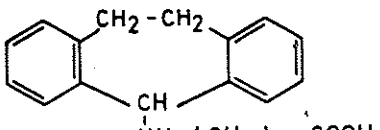

FIG. 1. *Chemical structure of amineptine.*

serotonergic neurons. This inhibitory effect is highly related to the blockade of NA and 5-HT uptake and can be explained by the intervention of different compensatory mechanisms (SCUVÉE-MOREAU & DRESSE, 1979). Nomifensine was also tested and appeared to reduce specifically the firing rate of noradrenergic and dopaminergic neurons without modification of the firing rate of serotonergic neurons (DRESSE & SCUVÉE-MOREAU, 1983).

New experiments were performed in order to compare amineptine with these various antidepressants. Its influence on the central noradrenergic neurons of the locus coeruleus (L.C.) and on the central serotonergic neurons of the dorsal raphe (D.R.) was investigated. These experiments were further completed by a study on the dopaminergic neurons of the area ventralis tegmenti of the mesencephalon (A10 group).

Methods

The experiments were performed on male Wistar rats weighing 200-300 g. The preparation of the animals and the recording procedure have been described in a previous paper (SCUVÉE-MOREAU & DRESSE, 1979). Briefly, the animals were anaesthetized with chloral hydrate (400 mg/kg i.p.) and a catheter was implanted in one jugular vein to make possible the perfusion of the drugs tested.

The firing rate of monoaminergic neurons was recorded by means of extracellular nickel-chrome microelectrodes stereotaxically implanted at the following coordinates: locus coeruleus : P 1.7-2.2 mm, L 1.0-1.3 mm from lambda, H 5.5-6.0 mm under the cerebellar surface (GRAHAM & AGHAJANIAN, 1971); dorsal raphe : A 0.2-0.5 mm, L 0.0 mm from lambda, H 4.5-5.0 mm under the cortical surface (AGHAJANIAN & HAIGLER, 1974); A10 : A 1.8-2.2 mm, L 0.6-0.8 mm from lambda, H 7.5 mm under the cortical surface (extrapolated from KÖNIG & KLIPPEL, 1963). The firing rate of the cells was recorded during a first control period of about 10 min, afterwhat the drug studied was perfused in the jugular vein by means of a perfusion pump (flow 6 ml/h). The perfusion was stopped when the frequency of discharge was reduced or increased of 50 % from the control rate. It was possible to make a quantitative comparison of the effect of the different drugs by calculating the total dose necessary to produce a 50 % decrease or increase of the firing rate. At the end of the experiment, the animals were perfused with a solution of formaldehyde 4 % and their brain was removed for the histological control of the position of the electrode. Amineptine sodium salt (*Servier*) was dissolved in NaCl 0.9 %. Doses refer to the base.

Results

(1) *Noradrenergic neurons*

Locus coeruleus neurons are characterized by a stable firing rate and a low frequency of discharge of 1 to 5 spikes/sec (GRAHAM & AGHAJANIAN, 1971; SCUVÉE-MOREAU *et al.*, 1979).

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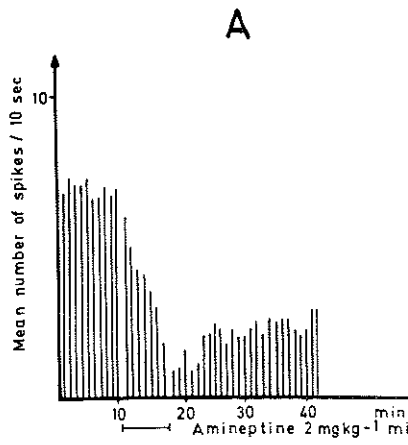

FIG. 2.

A. *Inhibitory effect of an intravenous per neuron of the locus coeruleus.*

B. *No modification of the firing rate of perfusion of a high dose of amineptine.* As repo clomipramine reduce the frequency of disch

(2) *Serotonergic neurons*

Raphe dorsal serotonergic neuron a low frequency of discharge of 0.2 to

The i.v. perfusion of amineptine d of discharge of D.R. neurons even a

(3) *Dopaminergic neurons*

A10 dopaminergic neurons fire at characterized by a progressive decre phasic positive-negative and have a lo non-dopaminergic neurons situated i vely faster and rhythmical firing rat 1973; YIM & MOGENSON, 1980).

As reported by BUNNEY *et al.* (19 dopaminergic neurons. In our experi sary to produce a 50 % decrease of t (M ± SE, $n = 4$). On the other han frequency of discharge of A10 neur dose required to produce a 50 % incr or haloperidol were used to confirm in this region.

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The i.v. perfusion of rather high doses of amineptine progressively decreases the firing rate of L.C. neurons without modification of the spike amplitude (Fig. 2 A). The mean total dose required to produce a 50 % reduction of the frequency of discharge is 12.7 ± 1 mg/kg (Mean ± SE; $n = 6$).

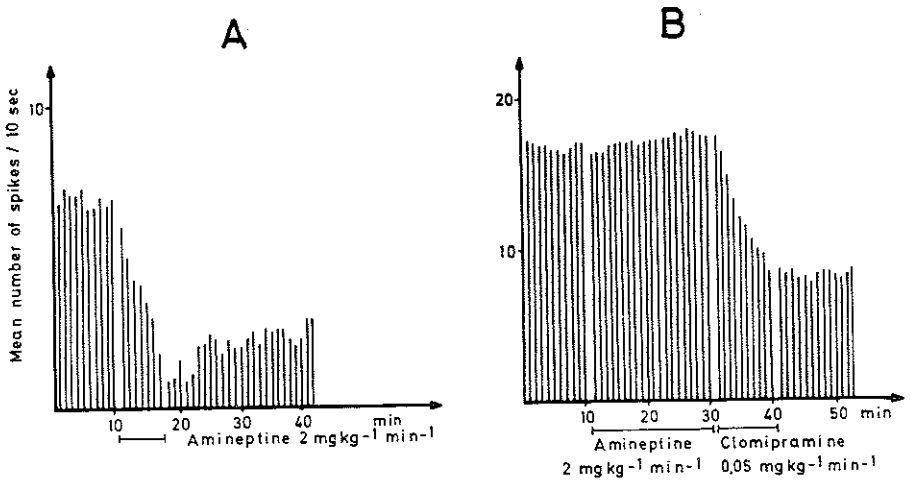

FIG. 2.

A. *Inhibitory effect of an intravenous perfusion of amineptine on the firing rate of a noradrenergic neuron of the locus coeruleus.*

B. *No modification of the firing rate of the serotonergic neurons of the dorsal raphe after i.v. perfusion of a high dose of amineptine.* As reported previously low doses of the tricyclic antidepressant clomipramine reduce the frequency of discharge of the serotonergic neurons.

(2) *Serotonergic neurons*

Raphe dorsal serotonergic neurons are characterized by a regular firing rate and a low frequency of discharge of 0.2 to 2 spikes/sec (AGHAJANIAN *et al.*, 1968 & 1978).

The i.v. perfusion of amineptine does not produce any decrease of the frequency of discharge of D.R. neurons even at the very high dose of 40 mg/kg (Fig. 2 B).

(3) *Dopaminergic neurons*

A10 dopaminergic neurons fire at a rate of 1 to 6 spikes/sec with occasional bursts characterized by a progressive decrease of the spike amplitude. The spikes are biphasic positive-negative and have a long duration. They could be confused with other non-dopaminergic neurons situated in the ventral tegmental area which have relatively faster and rhythmical firing rates and short spike durations (BUNNEY *et al.*, 1973; YIM & MOGENSON, 1980).

As reported by BUNNEY *et al.* (1973) amphetamine reduces the firing rate of the dopaminergic neurons. In our experimental conditions, the mean total dose necessary to produce a 50 % decrease of the frequency of discharge is 1.22 ± 0.2 mg/kg (M ± SE, $n = 4$). On the other hand, the antipsychotic haloperidol increases the frequency of discharge of A10 neurons. In our experimental conditions, the total dose required to produce a 50 % increase is about 0.05 mg/kg ($n = 4$). Amphetamine or haloperidol were used to confirm the dopaminergic nature of the cells recorded in this region.

The i.v. perfusion of amineptine induced a progressive decrease of the firing rate of A10 dopaminergic neurons (Fig. 3). A 50 % reduction of the frequency of discharge was obtained in four out of six animals at a mean total dose of 3.12 ± 1 mg/kg (M ± SE).

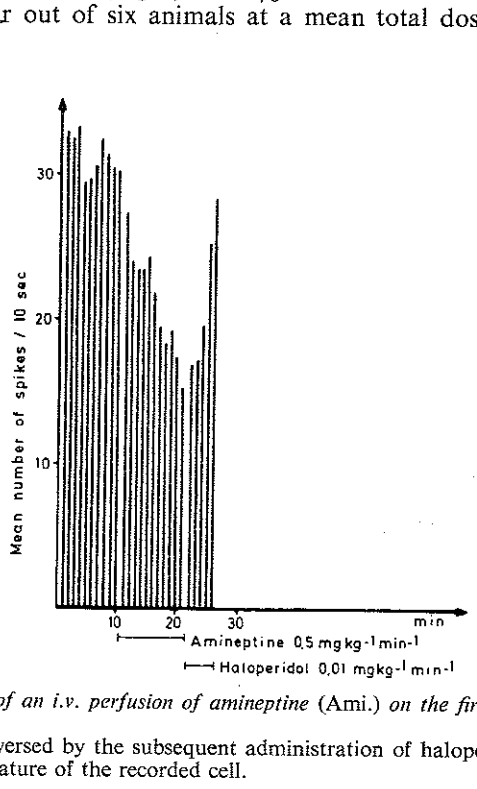

FIG. 3. *Inhibitory effect of an i.v. perfusion of amineptine* (Ami.) *on the firing rate of an* A_{10} *dopaminergic neuron.*

This inhibition is reversed by the subsequent administration of haloperidol (Hal.) which confirms the dopaminergic nature of the recorded cell.

Discussion

Electrophysiological studies *in vivo* confirm the selectivity of action of amineptine on dopaminergic neurons. In fact amineptine is devoided of effect on serotonergic neurons and its action on noradrenergic cells is four times less potent than its action on dopaminergic neurons.

As regards central *dopaminergic* neurons, the inhibitory effect of amineptine on the firing rate is probably related to the blockade of DA reuptake and to the consecutive increased availability of DA in the synaptic cleft, this effect being reinforced by the ability of amineptine to increase DA release. The decrease of the firing rate could be explained by two different compensatory mechanisms : (1) a postsynaptic mechanism implicating the intervention of uni- or multineuronal feedback loops originating from the postsynaptic elements innervated by A10 neurons, (2) a presynaptic mechanism implicating increased stimulation of inhibitory presynaptic receptors present on dopaminergic cell bodies (BUNNEY & AGHAJANIAN, 1978). In another study, we have shown that nomifensine also reduced the spontaneous firing of DA cells.

As regards central *noradrenergic* neurons, the inhibitory effect of amineptine is difficult to explain in view of the reported biochemical properties of this compound. Anyway this effect is very weak compared with the tricyclic antidepressants and nomi-

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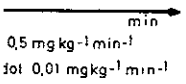

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As regards central *serotonergic* neurons, amineptine is completely devoided of effect on the firing rate even at very high doses. These results *in vivo* do not support biochemical studies *in vitro* showing that high doses of amineptine increase 5-HT release (OFFERMEIER *et al.*, 1977), which would be expected to produce a compensatory decrease of the firing rate of D.R. cells. Nevertheless the 5-HT release is not directly measured in our experimental conditions.

In conclusion, amineptine has a rather selective action on central dopaminergic neurons. This selectivity differentiates amineptine from the tricyclic antidepressants and from nomifensine.

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