EFFECTS OF CENTRALLY ACTING MUSCLE RELAXANTS ON THE ANEMIC DECEREBRATE RIGIDITY IN RATS

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Effects of centrally acting muscle relaxants on the tonic and phasic tension of the forelimbs were studied in the rigid rats. Anemic decerebrate rigidity of rats was obtained by ligation of both basilar and common carotid arteries. Tension recorded from forelimbs was employed as measures of the intensity of the rigidity. In addition to the rigidity developed in forelimbs (tonic component), phasic tension (phasic component) was induced by mechanical stimulation given to the hindlimbs. On the basis of the effects on the tonic and phasic components of rigidity, the drugs examined were classified into three types of groups: (1) drugs which depressed both tonic and phasic components (mephenesin and tolperisone), (2) drugs which depressed tonic and augmented phasic components (orphenadrine and chlorpromazine), and (3) drugs which depressed phasic but had almost no effects on the tonic components (diazepam, nitrazepam and clonazepam).

Keywords—rigidity; muscle relaxants; mephenesin; tolperisone; orphenadrine; chlorpromazine; benzodiazepines

INTRODUCTION

Physiological and pharmacological studies have indicated that the experimental rigidity in cats due to anemic decerebration¹⁾ is dependent on hyperactivity of α -motor system without involvement of hyperactivity of γ -motor system.²⁻⁴⁾ Recently, it was reported that anemic decerebrate rigidity can be produced at a highly successful rate in rats, and the rigidity was also due to hyperactivity of α -motoneurons.⁵⁾ Landau and Clare⁶⁾ found no increase in fusimotor activity in H reflexes of

hemiplegic patients, and suggested that α -motoneurone remained the site of exaggerated synaptic function in such a type of spasticity.

In the patients with cerebral palsy, they seem to have "associated reactions" (tonic reflexes arising in and acting on the limbs) as well as tonic neck and labyrinthine reflexes.⁷⁾ In the anemic decerebrate rats, we obtained phasic tension of rigid limbs by mechanical stimulation of hindlimb or tail, and expected that such rigid animals provided a good experimental model for some type of

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spasticity.

The purpose of the present study is to examine the effects of some centrally acting muscle relaxants on muscle tension of rigid forelimbs (tonic component) and the phasic tension of forelimbs induced by mechanical stimulation of hindlimbs (phasic component), and to classify such drugs on the basis of the effectiveness on the tonic and phasic components.

METHODS

The procedure for preparing the anemic decerebrate rigidity was the same as that described in a previous report.⁵⁾ The experiments were performed on male Wistar rats (250–460g, Nippon Rat Co.). Rats were anesthetized with ether and fixed on their back. After trachea, oesophagus and bilateral common carotid arteries were exposed, the trachea was cannulated, and the oesophagus was then cut between two ligatures. The occipital bone was exposed by scraping off the adhering muscles. After the common carotid arteries were ligated bilaterally, a trephined hole (diameter 5 mm) was made in the central part of the bone to make the basilar artery visible. Trephining was carried out carefully to avoid injuring the dura mater. Under sufficient anesthesia, the dura mater was cut along the basilar artery, and then the artery was cauterized with a bipolar pincette coagulator electrodes (Mizuhoika Micro 1C). As soon as the procedure was finished, anesthesia was discontinued. Within 30 min after ligation of the common carotid arteries and cauterization of the basilar artery, rigidity developed. Marked extension of the forelimbs and rigidity of the neck occurred and followed by extension of the hindlimbs and tail.

When the extension of the forelimbs became marked and stable in the preparation, the tension recorded from forelimbs was employed instead of electromyographic response,⁵⁾ as a measure of the intensity of the rigidity. The rat with sustained rigidity was placed on its back, with the hindlimbs and head fixed (Fig.1). One end of a celluloid bar (10 cm) with a strain gauge (Shinko, M 102) was put on the forelimbs, and then the forelimbs were pushed down by letting down the other end of

celluloid bar to the level of the shoulder. The output of the strain gauge was linearly related to the compressing force applied to the rigid forelimbs, and was detected through bridge circuit and recorded on a DC-recorder (Rika Electronics B-381). Mechanical stimulation on the rigid hindlimb was given by stretching rostrally both hindlegs fixed at each ankle (5 mm, for 3 sec, once every 45 sec) (Fig. 1). All experimental drug administration to decerebrate rats was postponed until 50 min had elapsed following surgery and discontinuing of the ether anesthesia. At the end of experiments, the rat was killed by ether inhalation, and tension in the most relaxed status of the forelimbs was recorded.

The effects of drugs were obtained by comparing the mean of the tension measured for 10 min at maximum response, with the control measured just before the drug administration.

Drugs used in the study were mephensin (Myanol[®], Chugai), tolperisone-HCl (Nippon-Kayaku), orphenadrine-HCl (Dainippon), chlorpromazine-HCl (Wintermin[®] parenteral solution, Shionogi), diazepam (Sumitomo), clonazepam (Sumitomo) and nitrazepam (Sumitomo). Mephensin, tolperisone and orphenadrine were dissolved in 0.9 % saline. Chlorpromazine was prepared by diluting the parenteral solution in 0.9 % saline. Diazepam, clonazepam and nitrazepam were suspended in 0.5 % CMC solution. Drug solutions were injected through a cannula inserted either into a femoral vein or intraduodenally.

FIG. 1. Diagrams of Arrangements for recording Rigid Forelimb Tension and stimulating Hindlimbs to elicit Phasic Component of the Tension in Anemic Decerebrate Rats

RESULTS

1. Tonic and Phasic Components of Rigid Forelimbs

Rigid forelimbs had the tonically sustaining tension of about 20 g (tonic component); spontaneous phasic augmentation of the forelimb tension was often observed (Fig.2a). Phasic tension of the rigidity (phasic component) was clearly induced in the forelimbs by mechanical stimulation of the hindlimbs (Fig.2b). This phasic component of the forelimbs appeared in all anemic decerebrate rats and lasted for more than one hour. In C_1 spinalized rat, however, neither tonic nor phasic component was observed.

Since the mechanical stimulation of the hindlimbs involved noxious stimuli on the ankle skin and stretching of gastrocnemius muscle, we examined what kind of stimuli caused the phasic component of the forelimb tension. As shown in Fig.2c, the phasic component of the rigid forelimbs caused by the stretch given to gastrocnemius muscle in the hindlimb was not clear and intermittent. On the other hand, the phasic com-

FIG. 2. Effect of Various Stimulations of the Hindlimbs on the Rigid Forelimbs in Anemic Decerebrate Rats

- a: Tonic component of rigid forelimbs tension. Upward deflection: spontaneous phasic component of forelimbs tension.
 b: Tonic component plus phasic component induced by stretching hindlimbs. Upward deflection: phasic component of forelimbs tension.
 c: Phasic component induced by stretching gastrocnemius muscle (5 mm).
 d: Phasic component induced by pinching, pricking or puffing of the skin near an ankle. Ordinate: Tension (g).

ponent caused by pinching, pricking or puffing on the skin near an ankle was marked and regular (Fig.2d). This indicates that the phasic component of the forelimb rigidity is associated with a mechanical stimulation on cutaneous afferents.

2. Effects of Mephenesin and Tolperisone

Effects of mephenesin and tolperisone are shown in Fig.3(a,b). Dose-response relationships for two drugs are depicted in Fig.3b. Tonic as well as the phasic components were reduced by mephenesin (20–80 mg/kg, *i.v.*) and tolperisone-HCl (20 mg/kg, *i.v.*). Depression by mephenesin (20–40 mg/kg, *i.v.*) was short-lasting, and recovery was observed within 10 min after the administration. Mephenesin (80 mg/kg, *i.v.*) caused 85 % and 94

FIG. 3. Effects of Mephenesin and Tolperisone on Tonic and Phasic Components of Forelimbs in Anemic Decerebrate Rats

- a: Effects of mephenesin and tolperisone. Dots indicate when drugs were given *i.v.* Abscissa: Time after administration (min). Ordinate: Tension (g).
 b: Dose-response relationships for mephenesin (●) and tolperisone (○).
 ——— tonic.
 - - - - phasic.

Abscissa: Dose on a logarithmic scale. Ordinate: Tension at maximum response in percent of the control. Each point represents the mean of 5 separate experiments, with the S.E. indicated. *: Statistical difference between responses of tonic and phasic components ($p < 0.05$, calculated according to Student's *t*-test).

% depression on the tonic and phasic components, respectively. The effect of tolperisone was stronger than that of mephenesin; tolperisone-HCl (20 mg/kg, *i.v.*) caused 95 % and 72 % depression on the tonic and phasic components, respectively. Depression by tolperisone-HCl (5–20 mg/kg, *i.v.*) was also short-lasting, and a complete recovery from depression caused by 20 mg/kg was observed within 30 min after the administration.

3. Effects of Orphenadrine and Chlorpromazine

Orphenadrine-HCl (2.5–10 mg/kg, *i.v.*) and chlorpromazine-HCl (0.625–2.5 mg/kg, *i.v.*) depressed the tonic component dose-dependently

FIG. 4. Effects of Orphenadrine and Chlorpromazine on Tonic and Phasic Components of Forelimbs in Anemic Decerebrate Rats

a: Effects of orphenadrine and chlorpromazine. Dots indicate when drugs were given *i.v.* Abscissa: Time after administration (min). Ordinate: Tension (g).

b: Dose-response relationships for orphenadrine (●) and chlorpromazine (○).
—— tonic
----- phasic.

Abscissa: Dose on a logarithmic scale. Ordinate: Tension at maximum response in percent of the control. Each point represents the mean of 5 separate experiments, with the S.E. indicated. *: Statistical difference between responses of tonic and phasic components ($p < 0.05$, calculated according to Student's *t*-test).

(Fig. 4). These drugs, however, did not reduce, but rather enhanced the phasic component. Orphenadrine-HCl (10 mg/kg, *i.v.*) caused 75 % depression of the tonic component and 104 % enhancement of the phasic component. Effects of chlorpromazine on these two components were quite similar to those of orphenadrine. Chlorpromazine-HCl (2.5 mg/kg, *i.v.*) caused 80 % depression of the tonic component and 74 % enhancement of the phasic component. At each dose of these drugs, there was a tendency for both tonic and phasic components to recover within one hour after the administration.

FIG. 5. Effects of Diazepam, Nitrazepam and Clonazepam on Tonic and Phasic Components of Forelimbs in Anemic Decerebrate Rats

a: Effects of diazepam, nitrazepam and clonazepam. Dots indicate when drugs were given intraduodenally. Abscissa: Time after administration (min). Ordinate: Tension (g).

b: Dose-response relationships for diazepam (●), nitrazepam (○) and clonazepam (▲).
—— tonic.
----- phasic.

Abscissa: Dose on a logarithmic scale. Ordinate: Tension at maximum response in percent of the control. Each point represents the mean of separate experiments (diazepam: 5, nitrazepam: 3, and clonazepam: 3), with the S.E. indicated. *: Statistical difference between responses of tonic and phasic components ($p < 0.05$, calculated according to Student's *t*-test or Welch procedure whenever variances were unequal).

4. *Effects of Diazepam, Nitrazepam and Clonazepam*

Diazepam depressed the phasic component markedly, but its depressant effect on the tonic component was weak (Fig.5). Diazepam depressed the phasic component by 50 % at 2.5 mg/kg administered intraduodenally and completely depressed at 10 and 40 mg/kg. Tonic component was rather resistant to the drug and more than 80 % of the component remained even after the administration of 40 mg/kg of the drugs. Nitrazepam and clonazepam were used to study whether or not the depression by diazepam of the phasic component was a common property of benzodiazepine derivatives. These two drugs showed similar effects with diazepam. Nitrazepam and clonazepam, at a intraduodenal dose of 10 mg/kg, depressed the phasic component selectively (about 80 % depression). On the other hand, the tonic component was depressed about 20 % even at a dose of 40 mg/kg. Inhibitory effects of these three drugs on the phasic component appeared within 5 min after the intraduodenal administration and then developed gradually. Depressant actions of these drugs on the phasic component were long-lasting, and no recovery was observed within one hour after the administration.

DISCUSSION

In addition to the rigidity developed in rat forelimbs due to anemic decerebration (tonic component), phasic tension (phasic component) was obtained by mechanical stimulation given to the hindlimbs (Fig.2). Although the selective stretching of gastrocnemius muscle had little influence on the forelimb tension (Fig.2c), stimulation of cutaneous afferents (pinching, pricking or puffing on the hindlimb skin) induced phasic tension (Fig.2d). The phasic component of the forelimb tension could not be observed in spinalized rats. Thus, induction of the phasic component can be attributed to hyperactivity of a system involving cutaneous afferents and bulbar reticular formation which is hyperactive due to anemic decerebration.

Phasic component of the rigidity in rats seems to mimic the abnormal motor functions observed in

patients with spasticity; so the rigid rat may be regarded as a good experimental model for this type of muscle disorder. Those centrally acting muscle relaxants examined may be classified into three groups based on the mode of action on the phasic and tonic components developed in the rigid forelimb: Group I, which depressed both the tonic and phasic components (mephenesin and tolperisone), Group II, which depressed the tonic but augmented phasic components (orphenadrine and chlorpromazine), and Group III, which depressed the phasic component but had almost no effect on the tonic component (benzodiazepines). In man, mephenesin has been found to be effective against spasticity when injected intravenously, although the duration of action is very short.⁸⁾ Tolperisone is said to have a beneficial effect. Diazepam seems to be of value, although therapeutic results obtained by various authors show some variations.⁸⁾ Chlorpromazine has proved to be of no value in the relief of spasticity.⁸⁾ No reports have been found that orphenadrine is effective. Thus, it may safely be said that in general drugs which depress the phasic component of the experimental rigidity are effective in the clinical spasticity. However, a relation between experimental pharmacologic properties of drugs and clinical effects on spasticity has not been established, and remains to be clarified in the future.

Differences in the mode of action on the tonic and phasic components indicated that induction of each component includes different mechanism at least in part. Non-selective depressant actions of mephenesin and tolperisone (Fig.3) may be ascribed to their depressant actions of α -motoneurone to which forelimb belongs.⁹⁾

Chlorpromazine has been shown to cause a significant depression of both facilitation and inhibition of patellar reflex produced by stimulation of facilitatory and inhibitory areas of the reticular formation.¹⁰⁾ In contrast to the effects of chlorpromazine in the intact or the decerebrate animal, the drug even in very large doses produces no significant depressant effects on mechanically or electrically evoked reflexes in spinal animals.^{11,12)} Orphenadrine has effective depressant actions on patellar reflex in cats facilitated by stimulations of

the brain stem reticular formation,¹³⁾ but shows no appreciable effects on mechanically evoked spinal reflexes in spinal cats.¹⁴⁾ In the present study, the tonic component was depressed by chlorpromazine and orphenadrine, but the phasic component was rather facilitated (Fig. 4). Our results, together with the above-mentioned reports, suggest that the tonic component of rigidity is associated with the descending pathways from bulbar reticular formation, and that the phasic component may be related to a system involving cutaneous afferents facilitated by descending pathways different from those which cause tonic component.

Diazepam and other benzodiazepines depressed the phasic component selectively, but had almost no effect on the tonic component (Fig. 5). These interesting actions of benzodiazepines are in contrast with actions of chlorpromazine and orphenadrine. It was reported that propriospinal and spino-bulbo-spinal reflexes occurred in rats.¹⁵⁾ The selective depressant action of benzodiazepines on the phasic component may be ascribed to the inhibitory action on the hindlimb cutaneous afferent inputs which may facilitate α -motoneurons to which forelimbs belong, *via* ascending propriospinal or spino-bulbo-spinal reflex pathways. Diazepam enhances the presynaptic inhibition in the spinal cord¹⁶⁾ and the cuneate nucleus.¹⁷⁾ The depressant action of the drug on the phasic component observed in the present study might be the results of the enhanced presynaptic inhibition in the cutaneous afferents from hindlimbs.

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