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Pharmacological properties of NK433, a new centrally acting muscle relaxant

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Abstract

The pharmacological properties of NK433 ((−)-(*R*)-2-methyl-3-(1-pyrrolidinyl)-4′-trifluoromethylpropiophenone monohydrochloride), a novel muscle relaxant, were investigated. NK433 inhibited intercollicular decerebrate rigidity (γ-rigidity) and anemic decerebrate rigidity (α-rigidity) dose dependently. NK433 was stronger in inhibiting γ-rigidity than α-rigidity. NK433 inhibited the increase in muscle spindle discharges induced by pinna pinching (γ-activity) without affecting muscle spindle discharges or neuromuscular transmission. At muscle relaxant doses in decerebrate rigidities, NK433 did not affect the muscle tone induced by morphine-HCl nor that of normal animals. These results suggest that NK433 selectively depresses the excessive muscle tone of decerebrate rigidities through its effects on the central nervous system, and inhibition of γ-activity causes a preferential depression of γ-rigidity in comparison to α-rigidity. In i.v. experiments, the effects of NK433 on decerebrate rigidities were similar to those of eperisone-HCl and tolperisone-HCl, but in p.o. experiments, NK433 was at least 3 times as potent as eperisone-HCl and tolperisone-HCl.

Keywords: Muscle relaxant, centrally acting; NK433; Decerebrate rigidity; Straub tail reaction; γ-Activity

1. Introduction

Tolperisone and eperisone, centrally acting muscle relaxants, are analogs of 2-methyl-3-aminopropiophenone and have been reported to be effective in the treatment of spasticity in patients (Černáček and Jágr, 1966; Kuroiwa et al., 1981); however, the potency and duration of their muscle relaxant activity are not satisfactory. Recently, we screened a series of 2-methyl-3-aminopropiophenone analogs and found that one of them, NK433 ((−)-(*R*)-2-methyl-3-(1-pyrrolidinyl)-4′-trifluoromethylpropiophenone monohydrochloride; Fig. 1), had more potent and longer-lasting centrally acting muscle relaxant activities than tolperisone and eperisone (Shiozawa et al., 1992). In this study, we investigated in more detail the pharmacological properties of NK433 on two types of decerebrate rigidity, which are used for animal models of human spasticity, in comparison with those of tolperisone and eperisone.

Intercollicular decerebrate rigidity (γ-rigidity), which has been used to evaluate skeletal muscle relaxants (Fujii et al., 1979; Tanaka et al., 1981; Ochiai et al., 1981), is produced by transection of the brain stem at the intercollicular level. Anemic decerebrate rigidity (α-rigidity) is produced by interruption of cerebral blood flow, which causes the destruction of the forebrain, the rostral brain stem and the anterior lobe of the cerebellum.

The effects on the muscle tone of normal animals and on the morphine-induced Straub tail reaction, another animal model showing excessive muscle tone (Ellis and Carpenter, 1974; Kameyama and Ukai, 1979; Novack, 1982; Pong et al., 1987), were studied.

As centrally acting muscle relaxants are given as oral preparations in clinical use, we adopted the oral route as well as intravenous route for drug administration in the study of decerebrate rigidity.

A preliminary account of this work was communicated to the XIth International Congress of Pharmacology (Sakitama et al., 1990).

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2. Materials and methods

2.1. Subjects and drugs

Male Wistar rats (215–597 g) and male ICR mice (28.2–42.1 g) were used.

The drugs used were NK433 (Nippon Kayaku, Japan), eperisone-HCl (synthesized by Nippon Kayaku), tolperisone-HCl (Nippon Kayaku), urethane (Wako or Tokyo Kasei, Japan), α-chloralose (Wako or Tokyo Kasei) and morphine-HCl (Sankyo, Japan). The drugs were dissolved in physiological saline for intravenous or intraperitoneal administration or dissolved in distilled water or suspended in 0.5% carboxymethylcellulose-Na for oral administration. Intravenous administration was performed via a catheter previously inserted into the femoral vein or the cervical vein. Oral administration was carried out via a catheter previously inserted into the gaster. The rectal temperature of animals after surgery was maintained at approximately 37°C with a heating pad and lamp.

2.2. γ-Rigidity preparations and measurement of muscle tone (rats)

Rigidity was produced and the muscle tone of the forelimbs was recorded according to the methods of Aoto et al. (1989). In brief, animals were anesthetized by ether, intubated with a tracheal cannula and fixed onto a stereotaxic apparatus. Intercollicular decerebration was performed by radiofrequency lesion of the midbrain (AP + 2, V +2, + 4, L ±1.5, Paxinos and Watson, 1982), using a Lesion Generator (Radionics, RFG-4, USA) and a lesioning electrode inserted into the midbrain. After lesioning, ether anesthesia was discontinued, and animals were placed on their backs with the neck bent backward to make the extension of the forelimbs marked. One end of a celluloid bar with strain gauges was put on the forelimbs, which were pushed down by letting down the other end of the celluloid bar to the level of the shoulder. The force with which the forelimbs pushed up this plate was detected as the output of the strain gauge, using a carrier amplifier (AP-5, Nihon Kohden, Japan), and recorded on a recorder (WTR-281, Watanabe, Japan). In the case of p.o. studies, artificial respiration was performed.

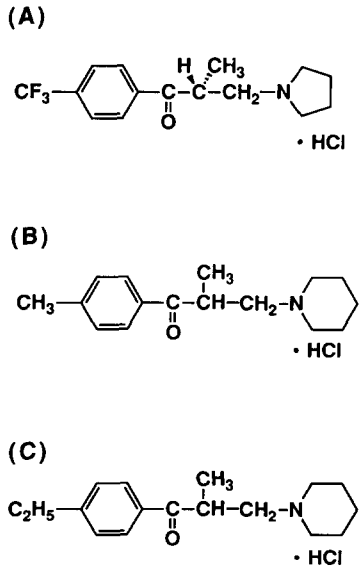

Fig. 1. Structure of NK433 ((−)-(*R*)-2-methyl-3-(1-pyrrolidinyl)-4′-trifluoromethylpropiophenone monohydrochloride) (A), tolperisone-HCl (B) and eperisone-HCl (C).

2.3. α-Rigidity preparations (rats)

Rigidity was produced according to the methods of Fukuda et al. (1974). Briefly, animals were anesthetized with ether and intubated with a tracheal cannula. After the esophagus was cut, the occipital bone was removed using a trepan to make the basilar artery visible. The basilar artery was cauterized by using the bipolar tweezer electrodes of a coagulator (Micro 1C, Mizuhoika, Japan). After anesthesia was discontinued, the common carotid arteries were ligated bilaterally. Recording of the muscle tone of the forelimbs was carried out by using the same methods as for γ-rigidity.

2.4. Measurement of muscle twitch tension (rats)

The animals were anesthetized by intraperitoneal administration of urethane (1.2 g/kg). The twitch tension of the gastrocnemius muscle, which was evoked by stimulation of the ipsilateral distal stump of the sciatic nerve (0.2 Hz, 0.05 ms in duration, supramaximal intensity), was recorded by a force transducer (SB-1T, Nihon Kohden) connected to the distal tendon with a thread. The output of the force transducer was amplified by a carrier amplifier and recorded on a recorder (WTR-281, Watanabe or RJG-3002, Nihon Kohden).

2.5. Recording of muscle spindle discharges (rats)

2.5.1. Ventral root transected preparations

The animals were anesthetized by intraperitoneal administration of urethane (1 g/kg) and α-chloralose (50 mg/kg). The hindlimb was denervated except for the nerves to the gastrocnemius muscle. The ipsilateral gastrocnemius muscle was isolated from the surrounding tissue, and its distal Achilles tendon was cut and loaded with tension (10–40 g). A laminectomy was performed in the lumbo-sacral region and the ventral roots were cut below L3 to eliminate motor innervation of the gastrocnemius muscle. Muscle spindle discharges were recorded from the fine dorsal rootlet

fibers of L3, L4 or L5 by bipolar platinum electrodes, displayed on an oscilloscope (VC-7, Nihon Kohden), transformed into square-wave pulses and fed into a staircase generator (EW-601, Nihon Kohden). The output of the staircase generator was recorded on a recorder (RJG-4124, Nihon Kohden). In some experiments, raw muscle spindle discharges were recorded on a thermal array recorder (RTA-1200, Nihon Kohden). A skin pouch was formed and the exposed tissues were covered with warm liquid paraffin.

2.5.2. Recording of γ-activity in ventral root intact preparations

γ-Activity was recorded from the dorsal rootlet indirectly as the increase in the frequency of muscle spindle discharges evoked by pinna pinching (Granit et al., 1952; Andrew et al., 1979). The animals were anesthetized by intraperitoneal administration of urethane (600 mg/kg) and α-chloralose (75 mg/kg). A similar preparation as outlined for muscle spindle discharges in ventral root transected preparations (Section 2.5.1) was adopted; however, the ventral roots were not cut in this series of experiments. The distal tendon of the gastrocnemius muscle was connected to a vibration generator (V-101, Akashi, Japan) and stretched for 20 s. Muscle spindle discharges were recorded from the fine dorsal rootlets of L3, L4 or L5, displayed on an oscilloscope, transformed into square-wave pulses and fed into an integrator (EI-601, Nihon Kohden). The output of the integrator was recorded on a recorder (RJG-4124, Nihon Kohden).

2.6. Morphine-induced Straub tail reaction (mice)

The animals were injected with morphine-HCl (15 mg/kg) subcutaneously 30 min after oral administration of the test compounds. The appearance of a rigid elevation of the tail (Straub tail reaction) was observed 30 min after morphine-HCl administration.

2.7. Traction test (mice)

When animals are hung by their forepaws on a horizontal wire 2 mm in diameter, they put their hindlimbs on the wire (traction response). The traction response was observed twice. The animals which failed

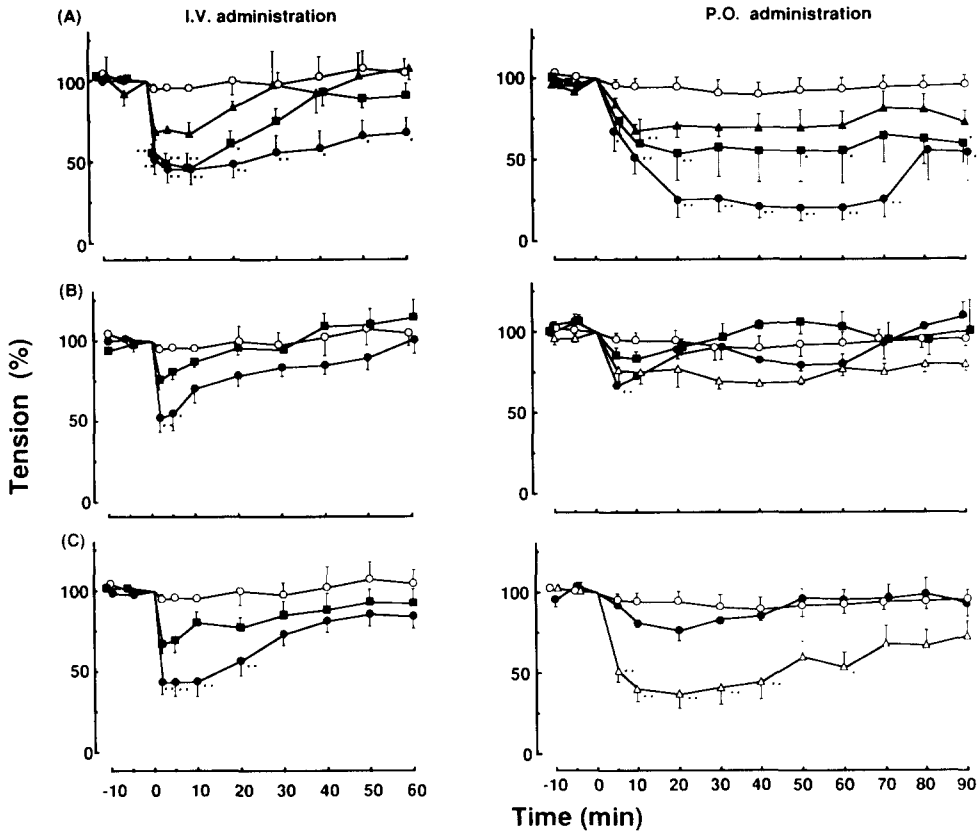

Fig. 2. Effects of NK433 (A), eperisone-HCl (B) and tolperisone-HCl (C) administered intravenously (left graphs: (○) control; (▲) 2.5 mg/kg; (■) 5 mg/kg; (●) 10 mg/kg) or orally (right graphs: (○) control; (▲) 25 mg/kg; (■) 50 mg/kg; (●) 100 mg/kg; (△) 200 mg/kg) on intercollicular decerebrate rigidity (γ-rigidity) in rats. Ordinates: mean tension of the rigid forelimbs, as percentages of the value just prior to drug administration, with S.E.M. indicated ($n = 3 \sim 9$). Abscissae: time in min after drug administration. * $P < 0.05$, * * $P < 0.01$: statistical significance from control group (Dunnett's test).

to show the traction response within 30 s in both trials were regarded as showing muscle relaxant activity. The traction reaction was observed 15, 30, 60, 90 and 120 min after oral administration of the test compounds.

2.8. Analgesic action (mice)

2.8.1. Acetic acid-induced writhing phenomenon

The animals were given 0.7% acetic acid-saline solution (0.1 ml/10 g body weight) intraperitoneally 30 min after oral administration of the test compounds. The number of writhings was counted for 5 min from 5 min after administration of the acetic acid-saline solution.

2.8.2. Tail-pinch test

When the base of the tail is pinched with an artery clip, the animal makes an effort to dislodge the clip by biting it (biting response). The animals that failed to show the biting response over 6 s after the tail-pinch were regarded as showing a positive antinociceptive effect.

2.9. Statistics

Data are expressed as means ± S.E.M. Comparison for significance between the control and drug-treated groups was performed with Dunnett's test or Fischer's exact test.

3. Results

3.1. Effects on γ-rigidity (rats)

As shown in Fig. 2A, left graph, intravenously administered NK433 dose dependently depressed the muscle tone of the forelimbs in the γ-rigidity preparation, and the depression reached a maximum 5 min after administration. Significant inhibition was observed at a dose of 5 mg/kg or more. At a dose of 10 mg/kg, the depression reached its maximum of more than 50%, and complete recovery was not observed within 60 min. When orally administered, NK433 also depressed the muscle tone of the rigid forelimbs in a dose-dependent manner (Fig. 2A, right graph). The depressant effect of NK433 at a dose of 100 mg/kg reached a peak of approximately 70% from 20 to 70 min after administration, and complete recovery was not observed within 90 min.

Eperisone-HCl and tolperisone-HCl intravenously administered dose dependently depressed the muscle tone of the rigid forelimbs, and the depression reached its maximum 2 min after administration (Fig. 2B and C, left graphs). Significant effects of eperisone-HCl and tolperisone-HCl were observed at a dose of 10 mg/kg. Although the maximal inhibition of eperisone-HCl and tolperisone-HCl at doses of 10 mg/kg was approximately the same as that of NK433, the effects of eperisone-HCl and tolperisone-HCl were eliminated within 30 min. When orally administered, eperisone-HCl depressed the muscle tone of the rigid forelimbs, but its maximal inhibition was approximately 30% even at a dose of 200 mg/kg (Fig. 2B, right graph). Tolperisone-HCl given orally at a dose of 100 mg/kg did not affect the muscle tone of the rigid forelimbs, but depressed it at a dose of 200 mg/kg. This depression reached a peak of approximately 60% 20 min after administration (Fig. 2C, right graph).

The dose-response relationships for the three drugs on the muscle tone of the rigid forelimbs in the γ-rigidity preparation are shown in Fig. 3. An intravenous dose of NK433 of 3.9 mg/kg produced a 50% inhibition of the muscle tone. The effects of eperisone-HCl and tolperisone-HCl were almost the same as that of NK433. An oral dose of NK433 of 33.8 mg/kg produced a 50% inhibition of the muscle tone. The inhibition elicited by eperisone-HCl did not reach 50% even at a dose of 200 mg/kg, and tolperisone-HCl did not affect the muscle tone at a dose of 100 mg/kg. Therefore, the effect of orally administered NK433 was more than 3 times and 5 times stronger than the effects of tolperisone-HCl and eperisone-HCl, respectively.

3.2. Effects on α-rigidity (rats)

When intravenously given, NK433, eperisone-HCl and tolperisone-HCl dose dependently depressed the muscle tone of the forelimbs in the α-rigidity preparation, and the depression reached a maximum at 10 min after administration. At a dose of 10 mg/kg, significant inhibition by NK433 was observed within 40 min, whereas inhibition by eperisone-HCl and tolperisone-

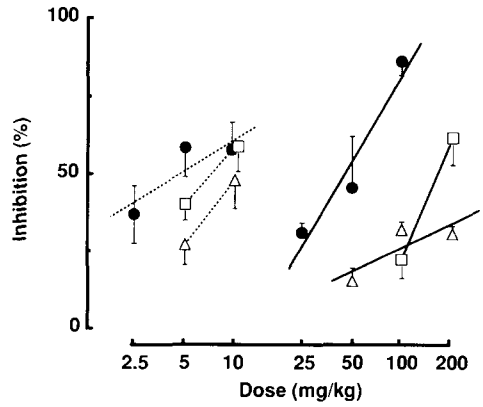

Fig. 3. Dose-response relationships for NK433 (●), eperisone-HCl (△) and tolperisone-HCl (□) administered intravenously (dotted lines) and orally (solid lines) on intercollicular decerebrate rigidity (γ-rigidity) in rats. Ordinate: maximal inhibition of γ-rigidity, as percentages of the value just prior to drug administration, with S.E.M. indicated. Abscissa: dose (mg/kg).

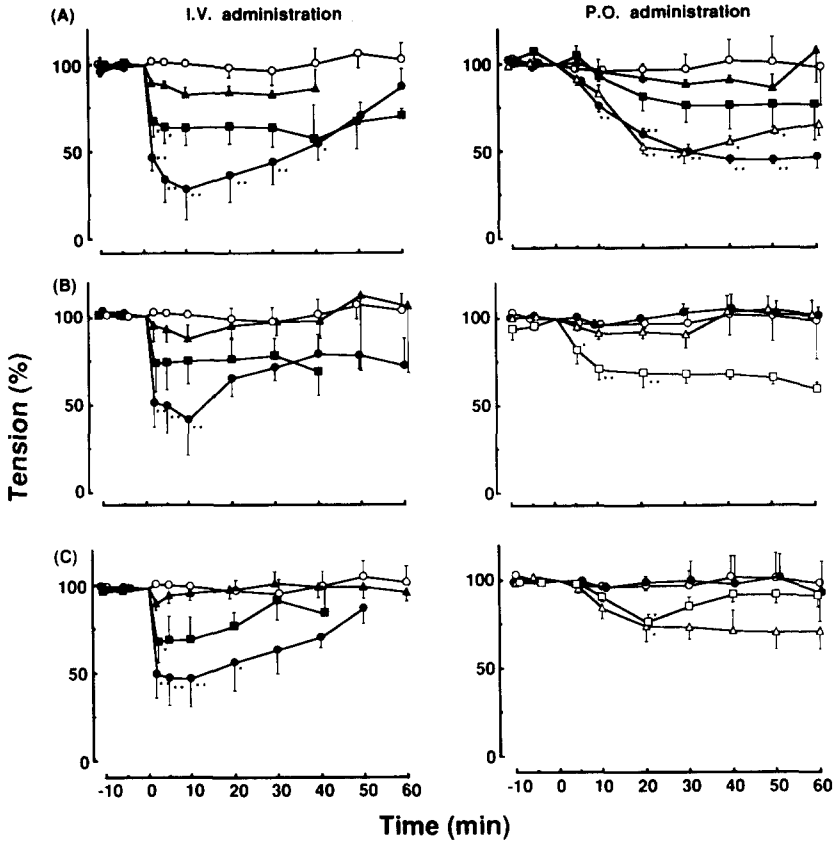

Fig. 4. Effects of NK433 (A), eperisone-HCl (B) and tolperisone-HCl (C) administered intravenously (left graphs: (○) control; (▲) 2.5 mg/kg; (■) 5 mg/kg; (●) 10 mg/kg) or orally (right graphs: (○) control; (▲) 25 mg/kg; (■) 50 mg/kg; (●) 100 mg/kg; (△) 200 mg/kg; (□) 400 mg/kg) on anemic decerebrate rigidity (α-rigidity) in rats. Ordinates: mean tension of the rigid forelimbs, as percentages of the value just prior to drug administration, with S.E.M. indicated ($n = 3 \sim 5$). Abscissae: time in min after drug administration. $^{*}P < 0.05$, $^{**}P < 0.01$: statistical significance from control group (Dunnett's test).

HCl was seen within 10 min and 20 min, respectively (Fig. 4, left graphs). When orally administered, NK433 also depressed the muscle tone of the rigid forelimbs in a dose-dependent manner. The depressant effect of NK433 at a dose of 100 mg/kg reached a peak of approximately 50% from 40 to 60 min after administra-

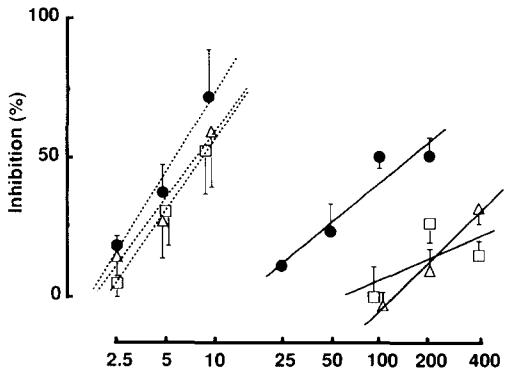

Fig. 5. Dose-response relationships for NK433 (●), eperisone-HCl (△) and tolperisone-HCl (□) administered intravenously (dotted lines) and orally (solid lines) on anemic decerebrate rigidity (α-rigidity) in rats. Ordinate: maximal inhibition of α-rigidity, as percentages of the value just prior to drug administration, with S.E.M. indicated. Abscissa: dose (mg/kg).

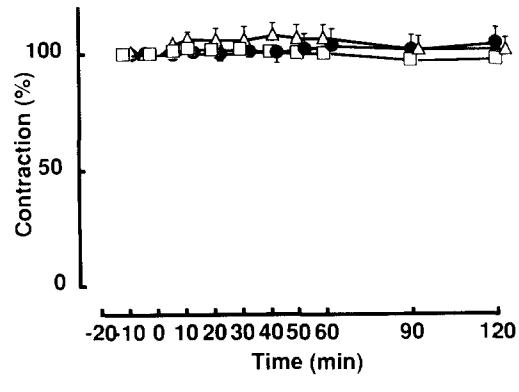

Fig. 6. Effects of NK433 (●), eperisone-HCl (△) and tolperisone-HCl (□) (200 mg/kg p.o.) on the twitches of the gastrocnemius muscle induced by electrical stimulation of the ipsilateral sciatic nerve in rats. Ordinate: mean contraction force of the gastrocnemius muscle, as percentages of the value just prior to drug administration, with S.E.M. indicated ($n = 6$). Abscissa: time in min after drug administration.

tion (Fig. 4A, right graph). Eperisone-HCl and tolperisone-HCl had little effect on the muscle tone of the rigid forelimbs even at a dose of 400 mg/kg (Fig. 4B and C, right graphs).

The dose-response relationships for the three drugs on the muscle tone of the rigid forelimbs in the α-rigidity preparation are shown in Fig. 5. Intravenous doses producing a 50% inhibition of the muscle tone for NK433, eperisone-HCl and tolperisone-HCl were 6.3, 8.4 and 9.3 mg/kg, respectively. An oral dose of NK433 of 107 mg/kg produced a 50% reduction of the muscle tone. Eperisone-HCl and tolperisone-HCl did not exhibit 50% inhibition even at a dose of 400 mg/kg. So the effect of orally administered NK433 was more than 4 times stronger than the effects of tolperisone-HCl and eperisone-HCl.

3.3. Effects on muscle twitch tension (rats)

To analyze the effects of NK433, eperisone-HCl and tolperisone-HCl on neuromuscular transmission, the muscle twitch tension evoked by stimulation of the ipsilateral sciatic nerve was recorded. NK433, eperisone-HCl and tolperisone-HCl, at doses of 200 mg/kg

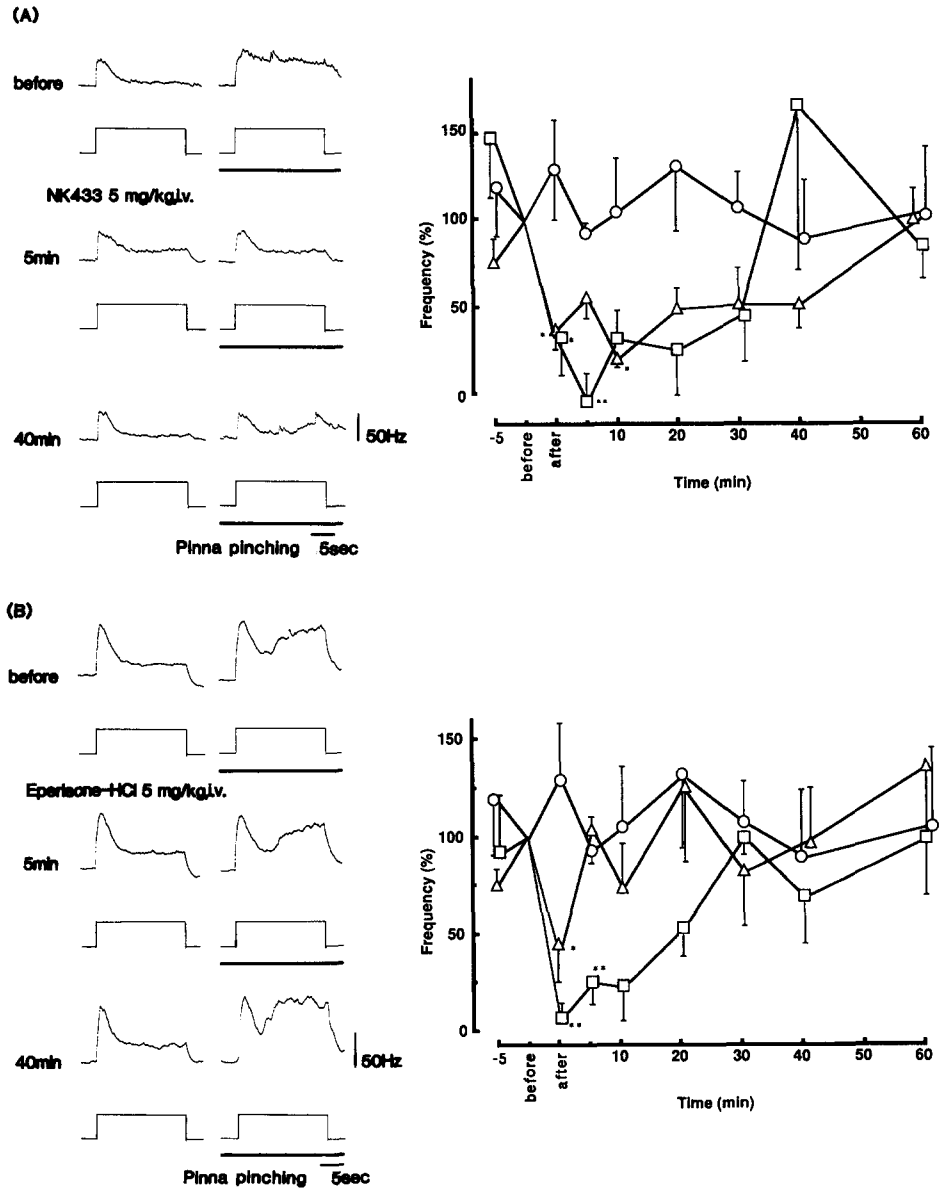

Fig. 7. Effects of NK433 (A) and eperisone-HCl (B) on static γ-activity in rats ((○) control; (△) 2.5 mg/kg; (□) 5 mg/kg). The left records of (A) and (B) show typical responses of the effect of NK433 or eperisone-HCl (5 mg/kg i.v.) on the integrated frequency of muscle spindle discharges (upper records) and period of stretching the gastrocnemius muscle (lower records), respectively. Horizontal bars indicate time for pinna pinching. The right graphs of (A) and (B) show the time courses of the effects of NK433 and eperisone-HCl. Ordinates: mean increase in muscle spindle discharges during pinna pinching, as percentages of the value just prior to drug administration, with S.E.M. indicated ($n = 4 \sim 6$). Abscissae: time in min after drug administration. * $P < 0.05$, ** $P < 0.01$: statistical significance from the control group (Dunnett's test).

Table 1
Effects of orally administered NK433, eperisone-HCl and tolperisone-HCl on the Straub tail reaction in mice

Drugs	Dose (mg/kg p.o.)	Observed activity [a]
Control		8/8
NK433	50	8/8
	100	8/8
	200	2/8 [b]
Eperisone-HCl	50	6/8
	100	7/8
	200	5/8
Tolperisone-HCl	50	5/8
	100	7/8
	200	7/8

[a] Number of mice showing tail reaction/number of mice tested.
[b] Significantly different from control, $P < 0.01$ (Fischer's exact test).

p.o., had no effect on the twitch tension of the gastrocnemius muscle (Fig. 6).

3.4. Effects on muscle spindle discharges (rats)

3.4.1. Ventral root transected preparations

NK433 and eperisone-HCl, at doses of 10 mg/kg i.v., had no effect on the frequency of muscle spindle discharges in the dosal rootlet under the conditions of ventral root transection, but tolperisone-HCl (10 mg/kg i.v.) decreased the frequency by approximately 30%.

3.4.2. Effects on increase in muscle spindle discharges evoked by pinna pinching in ventral root intact preparations

As shown in the left records of Fig. 7, muscle spindle discharges increased in the steady-state phase of the stretch following a temporal increase observed at the dynamic phase of the stretch. An increase in the frequency of muscle spindle discharges was induced by pinching the ear. NK433, at a dose of 5 mg/kg i.v., did not affect the integrated frequency of muscle spindle discharges induced by stretch of the gastrocnemius muscle, but reduced the increase in muscle spindle discharges evoked by pinna pinching (the left records of Fig. 7A). The right graph of Fig. 7A shows the time course of the effects of NK433. NK433 reduced the increase in muscle discharges evoked by pinna pinching in a dose-related manner. NK433, at a dose of 5 mg/kg, exerted a significant reduction within 5 min; thereafter the increase in muscle spindle discharges evoked by pinna pinching was restored. The effect of eperisone-HCl was almost similar to that of NK433 (the right graph of Fig. 7B).

3.5. Effects on morphine-induced Straub tail reaction (mice)

Morphine-HCl, at a dose of 15 mg/kg s.c., elicited the Straub tail reaction in all animals. NK433, at doses of 50 and 100 mg/kg p.o., had no effect on the Straub tail reaction, but at a dose of 200 mg/kg p.o. depressed it in 6 out of 8 animals, which was statistically significant. Eperisone-HCl and tolperisone-HCl did not affect the Straub tail reaction (Table 1).

3.6. Effects on traction test (mice)

NK433, at doses of 25, 50 and 100 mg/kg p.o., had no effect on the traction response, but at doses of 200 mg/kg p.o. or more significantly inhibited it within 60 min. Eperisone-HCl at a dose of 400 mg/kg inhibited the traction response in 3 out of 4 animals within 30 min. Tolperisone-HCl did not show statistically significant effects (Table 2).

Table 2
Effects of orally administered NK433, eperisone-HCl and tolperisone-HCl on the traction test in mice

Drugs	Dose (mg/kg p.o.)	Time after administration (min)					
		Before	15	30	60	90	120
Control		0/6 [a]	0/6	0/6	0/6	0/6	0/6
NK433	25	0/5	0/5	0/5	0/5	0/5	0/5
	50	0/6	1/6	0/6	0/6	0/6	0/6
	100	0/6	1/6	1/6	1/6	1/6	0/6
	200	0/5	5/5 [c]	4/5 [b]	5/5 [c]	3/5	2/5
	400	0/5	5/5 [c]	5/5 [c]	4/5 [b]	1/5	0/5
Eperisone-HCl	100	0/6	1/6	1/6	1/6	1/6	0/6
	200	0/6	2/6	0/6	0/6	1/6	0/6
	400	0/4	3/4 [b]	3/4 [b]	0/4	0/4	1/4
Tolperisone-HCl	100	0/6	0/6	0/6	0/6	0/6	0/6
	200	0/6	3/6	1/6	0/6	1/6	0/6
	400	0/6	3/6	3/6	1/6	1/6	1/6

[a] Number of mice showing muscle relaxation/number of mice tested. [b,c] Significantly different from control, $P < 0.05$ and $P < 0.01$, respectively (Fischer's exact test).

Table 3
Effects of orally administered NK433, eperisone-HCl and tolperisone-HCl on acetic acid-induced writhing in mice

Drugs	Dose (mg/kg p.o.)	n	Number of writhings
Control		11	9.5 ± 1.3 [a]
NK433	100	6	8.8 ± 2.2
	200	6	6.3 ± 2.3
Eperisone-HCl	100	6	11.8 ± 2.2
	200	6	5.7 ± 2.2
Tolperisone-HCl	100	6	12.3 ± 2.8
	200	6	7.3 ± 2.5

[a] Mean ± S.E.M.

3.7. Analgesic activity (mice)

3.7.1. Effects on acetic acid-induced writhing

All animals which were given 0.7% acetic acid-saline solution intraperitoneally showed writhing (9.5 ± 1.3 times within 5 min). NK433, eperisone-HCl and tolperisone-HCl did not change the number of writhings (Table 3).

3.7.2. Effects in tail-pinch test

In the control group, the biting response was observed in all animals 30 and 60 min after administration of distilled water, and only one out of 8 animals failed to show the biting response after 120 min. NK433 at doses of 50 and 100 mg/kg p.o. did not affect the biting response, but at a dose of 200 mg/kg p.o. significantly inhibited it 30 and 60 min after administration (6 out of 8 animals). Eperisone-HCl did not affect the biting response. Morphine-HCl dose dependently suppressed the biting response. At doses of 8 and 16 mg/kg s.c., the biting responses disappeared for 60 and 120 min, respectively (Table 4).

Table 4
Effects of orally administered NK433 and eperisone-HCl on the biting response in mice

Drugs	Dose (mg/kg)	Time after administration (min)		
		30	60	120
Control		0/8 [a]	0/8	1/8
NK433	50	1/8	0/8	0/8
	100	1/8	2/8	2/8
	200	6/8 [b]	6/8 [b]	3/8
Eperisone-HCl	50	0/8	1/8	1/8
	100	3/8	1/8	0/8
	200	2/6	3/6	1/6
Morphine-HCl [d]	4	3/8	3/8	3/8
	8	8/8 [c]	8/8 [c]	5/8
	16	8/8 [c]	8/8 [c]	8/8 [c]

[a] Number of mice which did not bite the clip/number of mice tested. [b,c] Significantly different from control, $P < 0.05$ and $P < 0.01$, respectively (Fischer's exact test). [d] Morphine-HCl was administered subcutaneously.

4. Discussion

NK433 administered by an intravenous or oral route decreased the muscle tone of the forelimbs in γ- and α-rigidity preparations in a dose-dependent manner. Magoun and Rhines (1946) proposed that an imbalance produced by decerebration that favors a facilitatory influence over the extensor γ-motoneuron causes an increase in the tone of the antigravity muscles. Although there are some dissimilarities between γ-rigidity and human spasticity (Burke et al., 1970), γ-rigidity is an appropriate animal model to evaluate exaggerated stretch reflexes because it has many features in common with human spasticity, such as the release of stretch reflexes of the antigravity muscles, the disappearance of the increased muscle tone caused by deafferentation and the presence of the clasp-knife phenomenon (Wiesendanger, 1985; Carew, 1985). Inhibitory effects of NK433 on γ-rigidity suggest that NK433 can be expected to exert a strong ameliorating effect on spastic patients.

The inactivity in neuromuscular transmission and muscle spindle discharges indicates that NK433 exerts its muscle relaxant activity not through its peripheral actions but through its effects on the central nervous system.

As NK433 did not affect muscle spindle discharges, we investigated the effect of NK433 on γ-activity as an increase in the muscle spindle discharges evoked by pinna pinching. NK433 inhibited the increase in the muscle spindle discharges evoked by pinna pinching, which indicates an inhibitory effect on γ-activity. As the inhibition of γ-activity was exerted at a dose of 2.5 mg/kg i.v., a lower dose than that causing a significant depression of α-rigidity, this inhibitory effect is involved in the preferential depression of γ-rigidity in comparison with α-rigidity.

NK433 orally given exerted a marked depression on α-rigidity as well as γ-rigidity. In anemic decerebrate rigidity, the hyperactivity of Deiters' nucleus results in overdriving of the extensor α-motoneuron. As for human spasticity, some observations suggest gamma hyperactivity, whereas others show that there are spastic patients who do not show gamma hyperactivity (Landau and Clare, 1964; Hagbarth et al., 1973). In the latter case, α-motoneuron hyperexcitability seems to be responsible for spasticity, as Landau and Clare (1964) postulated. These results might indicate that NK433 has an ameliorating effect not only on the spasticity caused by gamma hyperactivity, but also on spasticity without hyperexcitability of the γ-motoneuron.

NK433 at a dose of 100 mg/kg, which was enough to cause an apparent depression of the enhanced muscle tone of the forelimbs in γ- and α-rigidity, did not affect either the morphine-induced Straub tail reaction or the muscle tone of normal animals in the traction

test. These results suggest that NK433 selectively depresses the excessive muscle tone evoked in decerebrate animals and not the muscle tone induced by morphine-HCl or that of normal animals.

The pharmacological properties of eperisone and tolperisone were almost the same as those of NK433. Although eperisone did not have a peripheral action, as reported by Tanaka et al. (1981), tolperisone showed a weak inhibition of muscle spindle discharges, which was consistent with the results reported by Ono et al. (1984). The inhibitory effects of tolperisone on muscle spindle discharges might participate in its muscle relaxant activity.

In the i.v. experiments, the inhibitory effects of NK433 on γ- and α-rigidity were similar to those of eperisone and tolperisone, but in the p.o. experiments NK433 was at least 3 times as potent as eperisone and tolperisone. Although the underlying mechanisms are unknown, one of its metabolites, which was reported to show centrally acting muscle relaxant activity (Shiozawa et al., 1992), might contribute to the potent activity in the p.o. experiments. The centrally acting muscle relaxants are administered as oral preparations, therefore NK433 is expected to exert a stronger effect on human spasticity than eperisone and tolperisone.

NK433 and tolperisone, at a dose which causes an apparent depression of the enhanced muscle tone of the forelimbs in γ-rigidity, did not affect the morphine-induced Straub tail reaction induced by excessive excitation of the sarcococcygeus dorsalis muscle (Bilbey, 1960). These results indicate that NK433 and tolperisone predominantly depress the excessive muscle tone of γ-rigidity in comparison with that of the morphine-induced Straub tail reaction, which has been reported to be caused by dopamine release in the central nervous system (Gupta et al., 1988).

There are some clinical reports showing alleviating effects of centrally acting muscle relaxants on pain accompanied by muscular hypertonia (Gálos, 1992), so we investigated the analgesic activities of NK433, eperisone and tolperisone. NK433 inhibited the biting response caused by tail pinching at a dose similar to that inhibiting the muscle tone of normal animals in the traction test without affecting acetic acid-induced writhing. The time course of the inhibition of the biting response was similar to that of the traction test. The inhibitory effects of NK433 on the biting response seem to be caused by the muscle relaxant activity seen in normal animals.

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