Korean J. of Pharmacology
Vol. 27, No. 2, pp. 99-108, 1991

Effects of Racemic Ketamine on Excitable Membranes of Frog

Jong Hwa Lee* and George B. Frank

Department of Pharmacology, University of Alberta, Faculty of Medicine, Edmonton, Alberta, Canada

ABSTRACT

The effect of racemic Ketamine HCl was observed on excitable membranes of sciatic nerve fibres and toe muscles from frog. Ketamine significantly depressed the amplitude of the action potential, maximum rate of rise and that of fall of action potentials of sciatic nerve by dose-dependent and time-course manner, and also it produced the inhibition of K^+-contracture in toe muscle. We used two different ways of sucrose gap method to to obtain the better results from sciatic nerve. We observed and compared the effect of ketamine on sciatic nerve with naloxone, 4-AP (4-aminopyridine) and TEA (Tetraethylammonium). Naloxone significantly but not totally blocked the effect of ketamine both on nerve and on skeletal muscle. 4-AP or TEA by itself had a significant depressant effect on the action potentials on nerve by central perfusion (extracellular perfusion), but both of these drugs did not much affect the action of Ketamine on nerve. The reversibility of effect of Ketamine (10 mM) was observed both on nerve and on skeletal muscles when exposed to drug for short duration. The effects of racemic ketamine described may provide to support that one of the mechanisms of the action of Ketamine on nerve and on muscles of frog might be related to non-specifically effect on receptors within the ion channels (K^+-channel, Na^+-channel or slow Ca^{++} channel)at higher dose which produces anesthetic effect and also it interacts specifically with one of the opioid receptors or subtype of these receptors which is sensitive to Naloxone at lower dose which produces analgesia.

Key Words: Ketamine, 4-AP, TEA, Action Potentials, K^+-contracture, Sucrose gap method

INTRODUCTION

Ketamine [2-(0-chlorophenyl)-2-(methylamino) cyclohexanone: CT-581] is used clinically as a cataleptic analgesics and anesthetic agent devoid of sedative and hypnotic properties which are different from any other general anesthetics (McCarry *et al.*, 1965), and classified as 'dissociative anesthetics' (Domino *et al.*, 1965). These unique combinations of characteristics produce profound dissociation of cortical and limbic system and psychomimetic episodes during recovery (White *et al.*, 1982).

Recently, several reports have suggested that Ketamine may interact directly with opiate receptor to produce its opiate-like actions (Zukin, 1982; Smith *et al.*, 1980) and also interact with excitatory amino acid receptors as a NMA antagonist (Thomson *et al.*, 1985; Anis *et al*, 1983; Martin and Lodge, 1985). Ketamine has been reported to mainly affect the inactivation system of the potential-activated sodium channel (Shrivastav, 1977; Benoit *et al.*, 1986; Maleque *et al.*, 1981) related to Na^+ current. It is well known that Ketamine produces analgesia at lower dose and also produces anesthetic effect at higher dose,

* To whom requests for reprints should be addressed.
Present address; Department of Pharmacy, Sahmyook University, Seoul, Korea

but little is known about the mechanisms of these effects. So we studied the effects of ketamine in several doses on different tissues using different methods to elucidate the mechanism of Ketamine on excitable cell membrane.

In our present study, we observed the effect of Ketamine with related to Naloxone because it is naturally assumed that if Ketamine had an agonistic effect on opiate receptor which would be reversed by Naloxone. We also observed and compared the effects of 4-AP and TEA on frog sciatic nerve in the concentration-dependent manner.

METHODS

Experiment on frog sciatic nerve fibers

The experiments were carried out on the isolated sciatic nerves from the leopard frog *Rana pipiens* at room temperature (18°C). The nerves were desheathed under a dissecting microscope and split longitudinally into two bundles from each side. The desheathed nerves were moved to place in sucrose gap apparatus similar to those modified by Hunter and Frank (1979) & Frank and Sudha (1987).

The nerve bundles were pulled through each hole of four rubber membranes in a five-chambered sucrose gap apparatus. For stabilization of split nerves, they were allowed to rest in a chambered bath for 1 hr. The experiments by sucrose gap method were designed two different ways to compare the difference of effect of drugs on the action potentials of the frog sciatic nerves.

Single sucrose gap experiment: After setting the nerve bundle in the bath, the central (the third) chamber was perfused with frog Ringer's solution at the rate of 1 ml/min and the one adjacent chamber (generally the second) was perfused with isotonic sucrose solution (214 mM) at the rate of 1 ml/min.

IsoKCl (123 mM) or drug in IsoKCl was applied to the end chamber (left side, the first one) and the other chambers (the fourth and fifth) were filled with frog Ringer's solution.

Double sucrose gap experiment: The second and the fourth chambers were perfused with isotonic sucrose solution (214 mM), the central chamber was perfused with frog Ringer's solution, and the other chamber (both ends) were filled with frog Ringer's solution.

Drugs in frog Ringer's solution were perfused into the central chamber via a threeway stopcock after stabilization of the nerve bundle in the bath.

Electric recording: The action potentials in this experiment were recorded between the two compartments seperated by the sucorse gap. Stimulating voltage (the first and the third chamber) and membrane potentials (the third and the fifth chamber) were conducted. The stimulating voltage was set to produce the maximal compound action potentials, and single rectangular pulses of superamaximal strength and 0.01~0.05 msec in duration were used for frog sciatic nerves.

The experiments were performed using a digit Oscilloscope (Nicolet 4094), and the action potentials were stored by the disk recorder (Nicolet XF-44) to analyse the data directly by computer (Hewlett Packard 9816) and to draw the pictures of real action potentials by X-Y Recorder (Hewlett Packard 7015B).

Solutions and drugs: The composition of the frog Ringer's solution was as follows (in mM): NaCl, 111.87: KCl, 2.57; $CaCl_2$, 1.08; NaH_2PO_4, 0.087; $NaHCO_3$, 2.38; and Dextrose 11.1.

Isotonic sucrose solution contained 214 mM sucrose, and IsoKCl solution contained 123 mM KCl. The drugs used in this experiment were ketamine HCl (Ketalar*; Parke Davis & Company), Naloxone HCl (Endo Laboratories), 4-AP (4-Aminopyidine; Sigma Chemical Company, St. Louis, Mo.) and TEA (Tetraethylammonium Chloride Hydrate; Aldrich Chemical Co. Inc., Milwaukee, Wis.).

All drugs were dissolved in either IsoKCl or in frog Ringer's solution for single surcrose gap experiment or double sucrose gap experiment, respectively.

All solutions and drugs were adjusted at pH 7.1 ~7.2.

The nerve bundle was stimulated and the action potentials were recorded at 2, 5, 10, 20, 30 and 60 min for the first hour and every 30 min thereafter which lasted 4 hr. The means of the effects recorded at each time were compared with each drug condition using Student's t-test, and $p < 0.05$ was taken as the level of significance.

Experiment on frog skeletal muscle fibers

All experiments were carried out at room temperature (18°C) using preparations isolated from the frog, *Rana pipiens*. Extensor longus digiti IV (toe) muscles of the frog were used in all contracture recording experiments. The muscles were allowed to equilibrate for a period of 40~60 min in Ringer's solution before an experiment was commenced.

Mechanical recordings: The muscles were dissected from the frog in a way similar to that described by Frank (1960) and were mounted vertically in a 5 ml bath containing choline-Ringer's solutin. The lower end of the muscle was fixed near the bottom of the bath and the upper end was attached to the arm of a strain gauge by means of a silk thread. 10 ml of the new solution was injected rapidly at the bottom of the both and the excess soultion was removed by suction at the top of the bath. To remove the high K^+ solution the bath was flushed twice about 10 ml apart with Ringer's solution. In all experiments both toe muscles from two feet of an individual frog were used. These muscles constituted the muscle pairs referred to below.

Isometric tensions were recorded with a strain gauge whose active elements consisted of two pixie transducers (Endevco model 8121A) in a wheatstone bridge configuration. The output was recorded with a digital stripchart recorder (Dianachart). The resting tension on the muscle was adjusted so that the muscle came back to a vertical position without assistance when the bath solution was put back into a previously drained bath.

The general procedure used for contractures was described previously (Frank, 1960). Briefly this usually consisted of determining the response of the muscle to an elevated K^+ concentration lasting 10 to 15 sec before and after treating the muscle with a solution of modified composition. After each test with elevated K^+, the muscle was kept in choline-Ringer's solution containing 2.47 mM K^+ usually for 30 min or more before another test was performed. At the start of each experiment at least two or three control responses to elevated K^+ were obtained before the muscle was placed in a modified solution.

Solutions and drugs: The choline-Ringer's solution used in these experiments had the following composition (in mM): choline chloride, 111.8; KCl, 2.47; $CaCl_2$, 1.08; $NaHCO_3$, 2.38; NaH_2PO_4, 0.087; and glucose, 11.1. A solution containing 123 mM of methane sulfonate-K^+ salt plus 1.17 mM $CaCl_2$ was used. The K^+ salt of the methanc sulfonate was made with KOH and methane sulfonate.

The drugs used in this study were Ketamine HCl (Parke Davis) and Naloxone HCl (Endo Laboratories). All drug solutions were prepared fresh for each experiment. The pH of all the solutions used was adjusted to 7.1 to 7.2.

RESULTS

Effects on frog sciatic nerve

Before observing the effect of Ketamine on frog sciatic nerve, control tests were carried out without drug for 4 hr both by single sucrose gap method (intracellular application) and by double sucrose gap method (extracelluar perfusion). In these control expriments, little difference was obtained from results between two different ways for 4 hr.

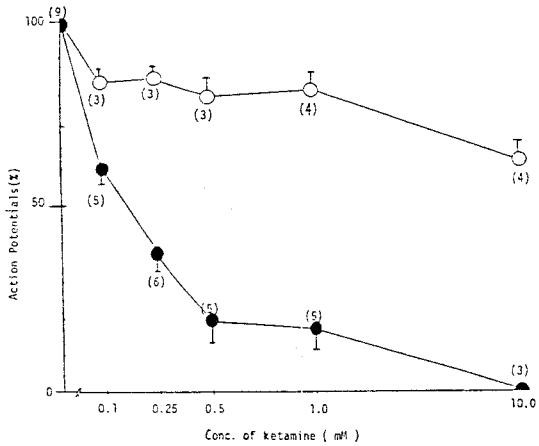

Fig. 1. Dose-response curve of ketamine on the action potentials of frog sciatic nerve at 4hrs.
Each point of the graph represents the mean ± S.E. from the number of experiments (n).
Experiments were done by single (○) sucrose gap method and double (●) sucrose gap method, seperately

When ketamine HCl (0.1 mM~10 mM) was perfused extracellularly to the nerve, the amplitude of the compound action potential were significantly depressed with dose-dependent and time-course manners ($p<0.01$). But intracelluar applications of Ketamine (0.1 mM~10 mM) were not shown significant difference from control (Fig. 1). Especially when higher dose of Ketamine (1 mM~10 mM) was perfused extracellularly, the action of Ketamine was significantly produced by depression of action potential and by changing the shape of action potentials (Table 1). The action potential of nerve started to be gone after 10 min perfusion of Ketamine (10 mM), eventually the action potentials were gone before 2 hr. When we washed out this nerve by perfusion of Ringer's solution at 10 min from starting of effect of this concentration, the recovery of the action potentials could be easily produced after 30 min of perfusion of Ringer's solution. In contrast, the recovery was not shown by perfusion (4 hr) of low dose of Ketamine (0.25 mM) after removal of this concentration (Fig. 3).

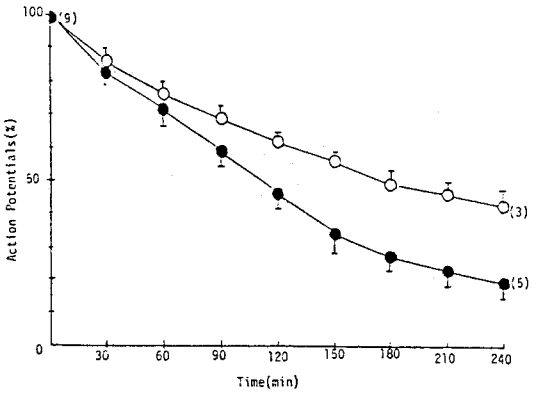

Fig. 2. Effect of Naloxone (0.01 μM) on Ketamine (0.5 mM) on the action potentials of frog sciatic nerve by double sucrose gap method.
Each point of the graph represents the mean ± S.E. of numbers of experiment (n), ketamine (●) and with naloxone (○), respectively.

Typical traces of the action potentials of frog sciatic nerve by Ketamine are shown in Fig. 3 and Fig. 6. We chose two concentrations (0.25 mM and 0.5 mM) from preliminary experiment for further study of the effect of Ketamine on nerve. A significant depression of the action potential of frog sciatic nerve was produced after 90 min perfusion and their maximal effect was observed by both of two concentrations at 4 hr. ($p<0.01$) (Table 1). To elucidate the mechanism of these effects of Ketamine, we used several drugs with Ketamine. At first various concentrations of Naloxone were tested on frog sciatic nerve from 0.01 μM to 100 μM. The 0.01 μM was chosen to demonstrate the antagonistic effect instead of other, because Naloxone 100 μM by itself produced the significant depression on action potentials of nerve like agonist (Fig. 4).

Naloxone blocked partially the depressant effects of Ketamine (0.25 mM and 0.5 mM) by simultaneous perfusion (Table 2 and Fig. 2). We compared the effect of Ketamine with the effects of 4-AP and TEA on nerve to observe the relationship to each other (Table 3). 4-AP by itself produced the depressant action of the action potential both by extracellular perfusion and by intracellular application, but 4-AP (10 mM) showed more significant depression on action potential by extracellular perfusion. Both of two K^+-channel blockers significantly depressed the am-

Table 1. Effect of Ketamine HCl on the action potentials of frog sciatic nerve by double sucrose gap method

Conc(mM)	n	30 min	60 min	120 min	180 min	240 min
Control	9	99.1±0.65	98.7±0.70	97.1±0.96	94.5±1.68	90.7±2.74
0.1 mM	5	95.5±0.29	90.7±1.45	79.0±0.98**	68.6±1.89***	60.8±3.23***
0.25 mM	6	85.6±3.16*	74.2±2.08**	58.2±3.24***	46.8±3.69***	37.4±4.46***
0.5 mM	5	82.3±4.67*	71.6±5.12***	46.2±5.94***	28.0±3.71***	19.8±2.72***
1.0 mM	5	55.9±6.69***	46.0±5.98***	33.2±4.92***	24.3±3.33***	17.2±4.18***
10 mM	3	14.1±7.34***	7.6±5.73***	0		

Mean±S.E n=number of experiments.
***$p<0.01$, **$p<0.02$, *$p<0.05$

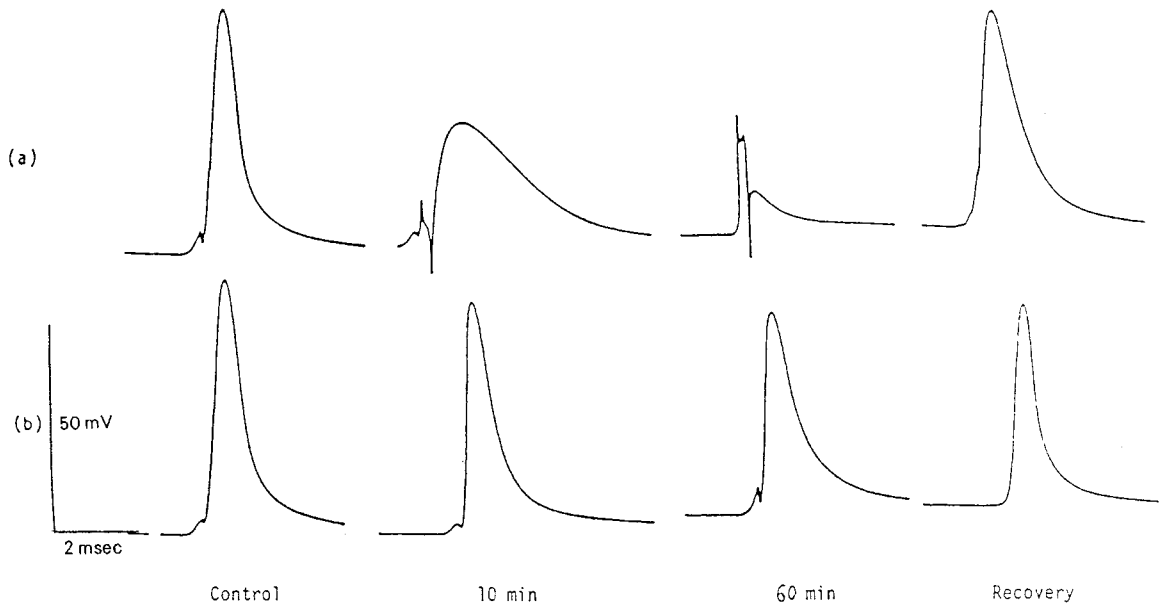

Fig. 3. Effect of Ketamine at higher dose (10 mM) and lower dose (0.25 mM) on action potentials of frog sciatic nerve by double sucrose gap method.
(a) 10 mM of ketamine (b) 0.25 mM of ketamine
To compare the effect of ketamine, (a) was recorded from the one split bundle of axon fibre and (b) recorded from the other split fiber of one side sciatic frog nerve.

Table 2. Effect of Naloxone (0.01 μM) on Ketamine HCl (0.25 mM)on the action potentials of frog nerve by double sucrose gap method

Group	n	30 min	60 min	120 min	180 min	240 min
ketamine	6	85.6±3.16	74.2±2.08	58.2±3.24	46.8±3.69	37.4±4.46
ketamine + Naloxone	8	85.4±1.40	80.2±1.20	69.2±2.34*	60.1±2.51**	51.9±3.32**

Mean±S.E n=number of experiments.
*$p<0.05$, **$p<0.01$

Table 3. Effect of 4-AP (4-Aminopyridine: 1 mM) and TEA (Tetraethylammonium: 10 mM) on Ketamine HCl (0. 25 mM) of the action potentials of frog sciatic nerve by double sucrose gap method

Group	n	30 min	60 min	120 min	180 min	240 min
Ketamine	6	85.6±3.16	74.2±2.08	58.2±3.24	46.8±3.69	37.4±4.46
Ketamine + TEA	3	81.6±6.53	69.0±4.20	48.3±3.50	41.1±3.29	34.0±4.64
Ketamine +4-AP	4	83.1±5.41	71.7±3.77	60.6±4.93	51.3±6.70	44.2±6.07

Mean±S.E. n=number of experiments.

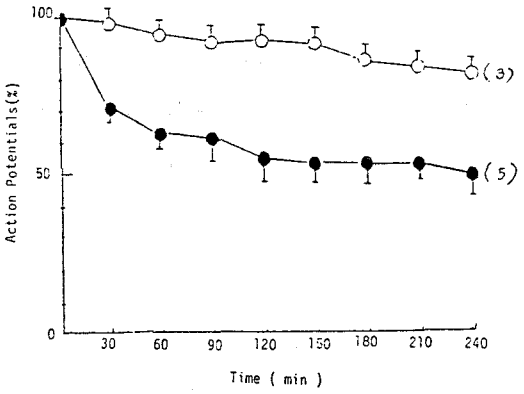

Fig. 4. Effect of Naloxone on the action potentials of frog sciatic nerve by sucrose gap method. Each point of the graph represents the mean±S.E. of numbers of experiment (n) from each group, naloxone (0.1 μM; ○) or Naloxone (0.1 mM; ●), respectively.

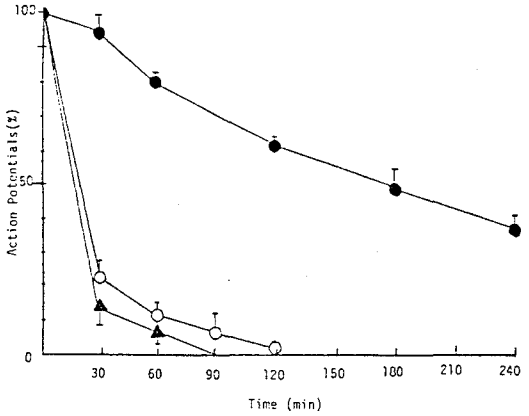

Fig. 5. Comparison of the effects of Ketamine, 4-aminopyridine & TEA on the action potentials of frog sciatic nerve in the concentrations of 10 mM by double sucrose gap method. Each point of the graph represents the mean±S.E. of 3 experiments from each group, ketamine (▲), 4-aminopyridine (○) & TEA (●), respectively.

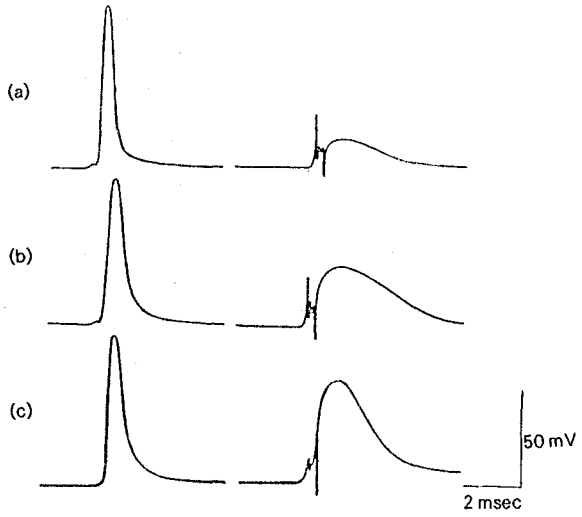

Fig. 6. Typical traces of the action potentials induced by Ketamine (a), 4-aminopyridine (b) and TEA (c) in the concentrations of 1 mM by double sucrose gap methods.
Left traces were controls and right traces were recorded 4 hrs after drug applications

Table 4. Effect of Ketamine HCl on K^+-contracture in toe muscle

Cone(mM)	n	30 min	60 min	120 min	180 min	240 min
Control	4	99.6±1.37	98.2±2.03	97.7±1.78	96.8±2.01	96.4±1.43
0.1 mM	2	98.8±1.26	95.6±1.35	94.5±2.06	94.8±0.60	93.5±1.96
0.25 mM	4	68.9±4.82**	62.4±5.98**	62.5±4.02**	58.0±6.02**	61.7±1.06**
0.5 mM	5	62.2±4.31**	53.8±6.03***	43.3±7.54***	39.8±6.02***	40.3±6.23***
1 mM	4	37.8±6.32***	24.7±7.45***	10.9±4.23***	0	

Mean±S.E, n=number of experiments.
***$p<0.01$, **$p<0.02$

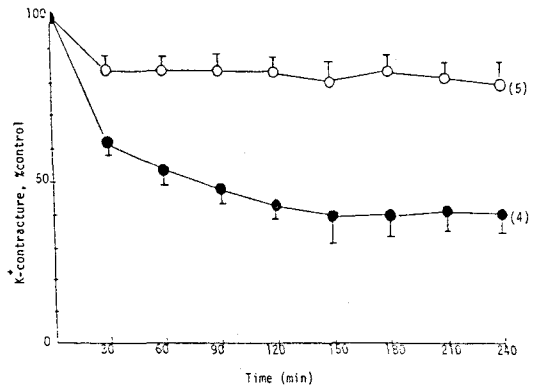

Fig. 7. Effect of Naloxone (0.01 μM) on ketamine (0.5 mM) on K^+-contracture in frog toe muscle. Each point of the graph represents the mean±S. E. of numbers (n) of experiments, Ketamine (●) and with Naloxone (○), respectively.

plitude of action potential, as Ketamine did, but ketamine was the most effective (Fig. 5). There was no significant difference observed when ketamine (0.25 mM) was simultaneously perfused with 4-AP or TEA (Table 3).

Effects on frog skeletal muscle

Inhibition of K^+-contracture on toe meuscle was observed with ketamine by dose-related and by time-course behaviours. Especially higher dose of Ketamine (1 mM) produced significantly the depressant effect on toe muscle less than 30 min after drug application,($p<0.01$) (Table 4), and also recovery after removal of drug was shown. Naloxone blocked the effect of Ketamine on toe muscle when simultaneously applied ($p<0.01$) (Fig. 7).

DISCUSSION

The results of this report demonstrate that Ketamine blocks the activities of nerve and also inhibits K^+-contracture in toe muscle by interaction with ionic channels and with opiate receptors. In our experiments, Ketamine depressed the amplitude of action potentials of frog sciatic nerve, and it produced the inhibition of K^+-contracture in toe muscle and also was showed the depression of rate of rise of action potential and prelongation of falling phase of action petential. Especially the concentration-dependent decrease in the amplitude of action potentials by Ketamine suggests that the blockade of activity must be related to ionic channels (Aronsta, *et al.*, 1982).

At higher doses (10 mM on nerve & 1 mM on toe muscle), the effects of ketamine were quickly shown and the reversibility was produced by rapid removal of ketamine. At lower doses (0.25 mM~0.5 mM), the effect of this drug was oberved the decrease on nerve activity and the inhibition of K^+-contracture by time-course manner, and also these effects was blocked by low dose of Naloxone (0.01 μM). Because Ketamine is very similar to PCP (phencyclidine) in its spectra of pharmacological effects, several investigators have provided the evidence that ketamine produces excitatory phenomena and behavioral changes whose actions are related to the subtype of opioid receptor (Brady and Balster, 1982; Corssen and Domino, 1966; Parsons *et al.*, 1988) which might be naloxone-insensitive receptor (Fratta *et al.*, 1980; Tam 1983; Arhem and Rydqvist, 1986) or naloxone-sensitive receptor (Smith *et al.*, 1980).

The mechanism of Ketamine action has been studied on frog sciatic nerve (Diamond *et al.*, 1986), on squid giant axon (Shrivastav, 1977), on vascular smooth muscle (Lundy and Frew 1981), and on frog skeletal muscle (Marwaha, 1980; Maleque *et al.*, 1981), nevertheless, little clear explantions for the mechanism of action of ketamine has been elucidated. Here we used the reacemic mixtures of Ketamine for this study even though several reports indicated the differences in the activity of the enantiomer (Brady and Balster, 1982; Zukin, 1982; Lundy *et al.*, 1986).

It was suspected at the onset of this study that Ketaminc might have opioid agonistic properties which are sensitive to Naloxone. This was based on earlier studies (Smith w*et al.*, 1980, 1982) that demonstrated a reduction conducted by electrically and mechanically. We observed the little effect of Ketamine with intracellular application on frog sciatic nerve but the effect was rapidly and fully showed by extracellularly perfused with Ketamine. The site of this action of Ketamine seems to be related to the extracellular perfused area and one can assume its site preferentially close to the outside of the excitable membrane and then might be diffused into

the axoplasm later which related to the stereospecific manner of opioid receptor. Because this mechanism of action, at higher dose (1 mM-10 mM) of Ketamine produced a significant action both on nerve and on skeletal muscle in a few minutes, and this effect could not be totally blocked by naloxone, but lower dose (0.25-0.5 mM) of this drug produced effective action on nerve and on skeletal muscle by time-course manner which was blocked by Naloxone. Ketamine produced the significant derceases in the amplitude, the rate of rise and the falling phase of action potential that suggested a decrease in sodium conductance both at higher and at lower dose, ketamine also produced the prolongation of its falling phase supposed to be more effective on potassium conductance at higher doses. Several reports have supported the effect of Ketamine was induced by NMA (N-methyl-Asopartate) receptor both in CNS and in PNS (Anis *et al.*, 1983; Thomson *et al.*, 1985) as manner of non-competitive action to NMDA-receptor (martin and Lodge, 1985). Also considering with related to structural formula, PCP and Ketamine are so similar that both appear to manifest that actions at PCP site-mediated kappa or sigma opioids (Parson *et al.*, 1988). We observed and compared Ketamine with 4-aminopyridine or TEA on frog sciatic nerve by sucrose gap method. We found the Ketamine has the most potent effect in the concentration of 10 mM, but the typical traces of action potential were so similar after perfusion of drug. 4-AP and TEA are potent K^+ channel blocker (Sherratt *et al.*, 1980; Hille, 1967) that prolong the action potential of demyelinated and unmyelinated fiber via blocking the valtage-dependent potassium current (Targ and kocisis, 1986; Bowe *et al.*, 1987). 4-AP selectively suppressed potassium current when applied to either internal (By single sucrose gap method) or external membrane surface (by double sucrose gap method) in our study which was same with the results from Yeh *et al.* (1976). 4-AP is more potent than TEA in the depressant effect which suggest 4-AP reduce both inward-and outward-going K^+-current, whereas TEA blocks only the outward movement of K^+ ions (Yeh *et al.*, 1976) and there is difference between 4-AP and TEA that TEA is more effective on nodal channels while 4-AP blocks internodal channel, and density of nodal versus paranodal K^+ channels in frog nerves depends on fiber diameter (Smith and Schauf, 1981; Schauf, 1987). In our data, it was observed that the mechanism of action of Ketamine at higher dose is likely to that of 4-AP but the depressant action of ketamine at lower dose seems to be partially related to opioid receptor which is sensitive to naloxone.

In conclusion, ketamine affects the electrical properties of the excitable membranes by decrasing the amplitude and the rate of rise of the action potentials, and by oprolongation of its falling phase. This mechanism of action of ketamine is likely related to the ionic channel on cell membrane which is firstly sensitive to ketamine. In fact, this drug suppressed sodium conductance both at higher dose and at lower dose, also it depressed the potassium conductance at higher dose(Frank, 1975; Inoue & Frank, 1962).

We also observed that the effect of ketamine was reversed and blocked by naloxone which has an atagonistic effect at lower dose(0.01 μM) by mechanism of interaction with sterospecific opioid receptor.

However ketamine has shown the non-specific depressant effect at higher dose just like local anesthetics, we observed the agonistic effect of Ketamine on opioid receptor at lower dose.

Sine the possibility still remains that the one of action of ketamine might be mediated by interactions with naloxone-sensitive opioid receptor, we are planning to investigate the action of Ketamine separatelyas(+)-isomer and (−)-isomer) to know stereospecifically the mechanism of action of this drug.

REFERENCES

Anis NA, Berry SC, Burton NR and Lodge D: *The dissociative anaesthetics, Ketamine and Phencyclidine, selectively reduce excitation of central mammalian neurons by N-methyl-aspartate. Br J Pharmacol 79: 565-575, 1983*

Arhem P and Rydqvist B: *The mechanism of action of Ketamine on the myelinated nerve membrane. Eur J Pharmacol 126: 245-251, 1986*

Äronstam R, Narayanan L and Wenger DA: *Ketamine inhibition of ligand binding to cholinergic receptors and ion channels. Eur J Pharmacol 78: 367-370, 1982*

Benoit E, Carratu MR, Dubois JM and Mitolo-Chieppa D: *Mechanism of action of Ketamine in the current and voltage clamped myelinated nerve fibre of the frog. Br J Phamacol 87: 291-297, 1986*

Bowe CM, Kocsis JD, Targ EF and Waxman SG; *Physiological effects of 4-Aminopyridine on demyelinated mammalinan motor and sensory fibers. Ann Neurol 22: 264-268, 1987*

Brady KT and Balster RL: *Discriminative stimulus properties of Ketamine stereoisomers in Phencyclidine-trained rats. Pharmacol Biochem & Behavior 17: 291-295, 1982*

Corssen G and Domino EF: *Dissociative anesthesia: further pharmocologic studies and first clinical experience with the phencyclidine derivative CI-581. Aneshth Analg 45: 29-40, 1966*

Diamond BI, Hovdala HS and Sabelli MD: *Differential membrane effects of general and local aneshtetics. Anesthesiology 43: 651-660, 1975*

Domino EF, Chodoff P and Corssen G: *Pharmacologic effects of CI-581, a new dissociative anesthetic in man. Clin Pharmacol Ther 6: 279-291, 1965*

Frank GB: *Effects of changes in extracellular calcium concentration on the potassium-induced contracture of frog's skeletal muscle. J Physiol(Lond.) 151: 518-538, 1960*

Frank GB: *Two mechanisms for the meperidine block of action potential production in frog's skeletal muscle: non-specific and opiate drug receptor mediated blockade. J Physiol(Lond.) 252: 585-601, 1975*

Frank GB and Sudha TS: *Effects of Enkephalin, applied intracellylary, on action potentials in vertebrate A and C nerve fibre axons. Neuropharmacology 26: 61-66, 1986*

Fratta W, Casu M, Balestrier A. Loviselli A, Biggio G and Gessa GL: *Failure of ketamine to interaction with opiate receptors. Eur J Pharmacol 61: 389-391, 1980*

Hille B: *The selective inhibition of delayed potassium currents in nerve by Tetraethylammonium ion. J Gen Physiol 50: 1287-1302, 1967*

Hunter EG and Frank GB: *An opiate receptor on frog sciatic nerve axons. Can J Physiol Pharmacol 57: 1171-1174, 1979*

Inoue F and Frank GB: *Action of procatine on frog skeletal muscle. J Pharmacol Exp Ther 136: 190-196, 1962*

Lundy PM and Frew R: *Ketamine potentiates catecholamine responses of vascular smooth muscle by inhibition of extraneuronal uptake. Can J Physiol Pharmacol 59: 520-527, 1981*

Lundy PM, Lockwood PA, Thompson G and Frew R: *Differential effects of Ketamine isomer on neuronal and extraneuronal catecholamine uptake mechanisms. Anesthesiology 64: 359-363, 1986*

Maleque MA, Warnick JE and Albuqerque EX: *The mechanism and site of action of Ketamine of skeletal muscle. J Pharmacol Exp Ther 219: 638-645, 1981*

Martin D and Lodge D: *Ketamine acts as a non-competitive N-methyl-D-aspartate antagonist on frog spinal cord in vitro. Neuropharmacology 24: 999-1003, 1985*

Marwaha J: *Electrophysiological studies of the action of Ketamine in frog skeletal muscle. Neuropharmacology 19: 765-772, 1980*

McCarthy DA, Chen G, Kaump DH and Ensor C: *General anaesthetic and other pharmacological properties of 2-(0-chlorphenyl)-2-methylamino cyclohexanone HCl(CI-581). J New Drugs 5: 21-33, 1965*

Parsons CG, Gibbens H, Magnago TSI and Headley PM: *At which 'sigma' site are the spinal actions of Ketamine mediated? Neurosci Lett 85: 322-328, 1988*

Schauf CL: *Differential sensitivity of amphibian nodal and paranodal K^+ channels to 4-Aminopyridine and TEA. Experientia 43: 405-408, 1987*

Sherratt RM, Bostock H and Sears TA: *Effects of 4-Aminopyridine on normal and demyelinated mammalian nerve fibres. Nature 283: 570-572, 1980*

Shrivastav BB: *Mechanism of Ketamine block of nerve conduction. J Pharmacol Exp Ther 201: 162-170, 1977*

Smith DJ, Pekoe GM, Martin LL and Coalgate B: *The interation of Ketamine with the opiate receptor. Life Sciences 26: 789-795, 1980*

Smith KJ and Schauf CL; *Size-dependent variation of nodal properties in myelinated nerve. Nature 293: 297-299, 1981*

Smith DJ, Westfall DP and Adams JD: *Assessment of the potential agonistic and antagonistic properties of Ketamine at opiate receptors in the guinea-pig ileum. Neuropharmacology 21: 605-611, 1982*

Tam SW: *Naloxone-inaccessible δ receptor in rat central nervous system. Proc Natl Acad Sci USA 80: 6703-6707, 1983*

Targ E and Kocsis JD: *Action potential characteristics of demyelinated rat sciatic nerve following application of 4-Aminopyridine. Brain Res 363: 1-9, 1986*

Thomson AM, West DW and Lodge D; *An N-methylaspartate receptor-mediated synapse in rat cerebral cortex: a site of action of Ketamine? Nature 313: 476-481, 1985*

White PF, Way WL and Trevor AJ: *ketamine-its pharmacology and therapeutic uses. Anesthesiology 56: 119-136, 1982*

Yeh JE, Oxford GS, Wu CH and Narahashi T: *Dynamics of Animopyridine block of potassium channels in squid axon membrane. J Gen Physiol 68: 518-535, 1976*

Zukin SR: *Differing stereospecificities distinguish opiate receptor subtypes. Life Sciences 31: 1307-1310, 1982*

=국문초록=

개구리 세포막에 대한 Racemic Ketamine의 영향

삼육대학 약학과* 및 알버타대학 의과대학 약리학교실

이종화* · George B. Frank

Racemic ketamine을 사용하여 개구리의 좌골신경 및 toe muscle에 대한 작용을 관찰하였다. 실험방법으로는 214 mM sucrose을 사용하여 서로 다른 두 종류의 투여 방법으로 세포막의 활동 전압에 대한 영향을 electric recording으로 관찰하였다. 즉, intracellular 투여는 single sucrose gap technique으로, extracellar 투여는 double sucrose gap technique을 사용하였으며 그 실험 결과는 아래와 같았다.

1. Racemic ketamine은 개구리의 좌골신경 및 toe muscle의 활동전압을 intracellular 및 extracellular 투여시 모두 의의 있게 억제하였다.
2. 개구의 toe muscle에서 K^+-수축을 억제하였다.
3. naloxone은 ketamine의 억제작용을 완전히 차단하지는 못하였다.