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SHORT COMMUNICATIONS

ACUTE TOXICITY OF CAPSAICIN IN SEVERAL ANIMAL SPECIES

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CAPSAICIN is a major pungent agent present in various species of capsicum fruits. LILLE and RAMIREZ (1935) reported that i.p. administration of capsicum extract into dogs produced a fall in mean arterial pressure with an increase in salivary secretion and a small increase in gastric secretion. TOH *et al.* (1955) found similar effects of capsaicin in cats, with bradycardia, apnea and hypotension but no change in gastric secretion. However, the mean arterial pressure in dogs in response to capsicum extract was somewhat variable (BRENDER and WEBB-PEPLOE, 1969) and it caused only hypotension in the rabbits (TODA *et al.*, 1972). Rats given repeated doses of capsaicin were able to withstand a hot environment (JANCSO-GABOR *et al.*, 1970; CABANAC *et al.*, 1976).

The only reported acute toxicity study with capsaicin was in rats which were administered four increasing s.c. doses of capsaicin to a cumulative amount of 21·0–66·0 mg/rat; eight of 17 rats died (CABANAC *et al.*, 1976). Additionally, MOLNÁR and GYÖRGY (1967) reported that capsaicin administered i.v. at a dose higher than 10 μg/kg to cats caused a rapid fall in mean arterial pressure which was followed either by a pressor phase or by death. The acute toxicity of capsaicin in several animal species is reported in our study.

Capsaicin (8-methyl-*N*-vanillyl-6-nonenamide) was purchased from Sigma Chemical Co., St. Louis, Mo. Capsicum extract was obtained from the cold extraction of 100 g of capsicum fruits (*Capsicum minimum*) six times with acetone (250 ml each), filtered and evaporated to approximately 14·5 ml in a vacuum rotary evaporator at about 35°C. Remaining acetone in capsicum extracts was removed under nitrogen and vacuum in a desiccator. Capsaicin contents in the extracts and in various parts of capsicum fruits were determined by a method modified from that of KARAWYA *et al.* (1967).

Animals randomly selected from the Animal Production Center, Faculty of Science, Mahidol University, Bangkok, included weanling female (45–55 g) and adult female (130–150 g) Fischer derived strain rats; adult male and female Swiss albino mice (25–35 g); male Syrian golden hamsters (55–75 g); male albino guinea pigs (350–440 g), and male and female albino rabbits (500–600 g). Rats were housed individually in stainless steel wire cages and mice were housed in a group of 8–10 animals in aluminum tubs with galvanized steel wire covers. Hamsters, guinea pigs and rabbits were housed in groups of 4–6 animals in stainless steel wire cages. All animals were fed with a regular rat or rabbit chow (Gold Coin Ltd., Singapore) and water *ad libitum*.

Group of 8–10 rats or mice and 4–6 hamsters, guinea pigs or rabbits were used for each

dose of capsaicin. Capsaicin was dissolved in dimethylsulfoxide (Burdick and Jackson Laboratories, Muskegon, MI), propylene glycol, or in a mixture of ethanol–Tween 80 (1:1, v/v) and saline (1:25, v/v). Capsaicin was administered in dimethylsulfoxide to mice in volumes of 0·5 ml/kg (intratracheally), 1·67 ml/kg i.v., i.p., i.m., s.c., intragastrically, intrarectally and by dermal application and in ethanol–Tween 80–saline in a volume of 16·7 ml/kg (intragastrically). Capsaicin was also administered i.p. in dimethylsulfoxide or propylene glycol to rats and hamsters (1·67 ml/kg) and to guinea pigs and rabbits (0·2 ml/kg).

Intravenous administrations were made into the tail veins; intratracheal administrations into the tracheal lumen of rats under light anesthesia with ether. Capsaicin was administered into the stomach and rectum by intubation. The toxicity of capsaicin was also evaluated by i.m. (right thigh), s.c. (dorsal region of the skin) and dermal (shaved skin of the neck and the back) routes. All animals were observed during the first 3 hr and periodically over 12 hr for the onset of toxic signs and deaths. Survivors were observed daily for 1 week and then sacrificed. A necropsy was performed on all animals sacrificed at 7 days or found dead and their tissues fixed in 10% buffered neutral formalin. Paraffin sections were stained with Harris's hematoxylin and eosin, and also with PAS's Schiff stain. LD_{50} values were calculated by the method of LITCHFIELD and WILCOXON, (1949).

Adult female rats (130–150 g) were anesthetized with sodium pentobarbital (Nembutal, Abott Laboratories, North Chicago, Il, 50 mg/kg i.p.). The trachea and femoral artery were opened and cannulated. A strain gauge pressure transducer (P23AA, Statham Laboratories, Inc.) was connected to the femoral artery cannula (filled with heparinized saline, 100 units/ml) and to the Dynograph recorder (Type R, Beckman Instruments, Inc.). Respiratory rate was recorded with the pneumograph connected to the volumetric pressure transducer (PT 5A, Grass Instrument Co.). Electrocardiogram (EKG) was recorded using lead II of a Dynograph recorder. Mean arterial pressure, EKG and respiratory rates were recorded until death of the rat treated i.p. with a lethal dose of capsaicin.

Capsaicin contents (1·82 mg/g, dry wt) were highest in the whole fruits of *Capsicum minimum* (Prik Kee-nue). *C. annuum* (Prik Luyang) has a similar capsaicin content (1·67 mg/g, dry wt) as *C. minimum*, whereas much lower capsaicin contents (0·45 mg/g, dry wt) were found in *C. frutescens* (Prik Chee-fah). Capsaicin in various parts of *C. minimum*, core contained the highest amount of capsaicin (28·9 mg/g, dry wt) followed by the pericarp (4·35 mg/g, dry wt) and seed (1·16 mg/g, dry wt). Additionally, the core of *C. annuum* and *C. frutescens* also contained higher capsaicin contents than the pericarp and seed.

Capsaicin had an i.p. LD_{50} of 6·50 and 7·65 mg/kg in adult female and male mice (Table 1). The toxicity of capsaicin present in the capsicum extract was approximately 4 fold higher than that of pure capsaicin given i.p. to mice. Capsaicin had a slightly higher LD_{50} in weanling female rats administered i.p. in propylene glycol than LD_{50} of those observed when administered i.p. in dimethylsulfoxide ($P<0·05$). Guinea pigs are the most susceptible species to capsaicin toxicity with the LD_{50} of 1·10 mg/kg, whereas hamsters and rabbits are less susceptible.

Table 2 presents the relative lethality of capsaicin administered by various routes in the mouse. Capsaicin is very potent when administered i.v. in mice, while the LD_{50} was slightly increased ($P<0·05$) when administered intratracheally. The lethality when administered i.p. was similar to those observed when administered by i.m. and s.c. routes. Capsaicin was only about 1/340 as effective intragastrically as i.v. The rectal and dermal routes were used as a means of circumventing the gastric acid and or/ enzymatic activity, which may have contributed to the lower lethality when capsaicin was administered by the intragastric route.

TABLE 1. LETHALITY FOLLOWING INTRAPERITONEAL ADMINISTRATION OF CAPSAICIN AND CAPSICUM EXTRACT DISSOLVED IN DIMETHYLSULFOXIDE

Species*	Sex	Average body wt (g)	LD_{50}† (mg/kg)	Slope	Onset of convulsion (min)‡	Survival time (min)‡
Mouse (70)	M	29	7·65 (5·28–11·09)	2·32	1·08 ± 0·21	2·43 ± 1·08
Mouse (70)	F	30	6·50 (4·33–9·75)	2·08	1·09 ± 0·18	3·22 ± 0·99
Mouse (79)§	F	30	1·51 (0·91–2·52)	2·16	0·58 ± 0·25	2·50 ± 1·20
Rat (56)	F	51	10·40 (6·71–16·12)	2·27	0·56 ± 0·16**	4·57 ± 0·38
Rat (62)‖	F	54	13·20 (9·76–18·02)	2·26	1·35 ± 0·25	5·05 ± 0·32††
Rat (76)	F	141	9·50 (5·76–15·68)	3·08	1·19 ± 0·15	5·16 ± 0·40††
Hamster (20)	M	65	>120	—	—	—
Guinea pig (30)	M	405	1·10 (0·79– 1·52)	1·12	0·15 ± 0·05¶	5·50 ± 0·51††
Rabbit (12)	M, F	503	>50	—	—	—

*The numbers of animals used are shown in parentheses.

†Calculated by the method of LITCHFIELD and WILCOXON (1949). Figures in the parentheses are 95% confidence limits.

‡Time (mean ± S.E.) was recorded after capsaicin administration. Values marked ¶, ** and †† differ significantly from those female mice treated with capsaicin in dimethylsulfoxide: ¶$P<0·001$; **$P<0·01$; ††$P<0·05$.

§Mice were treated with capsicum extract. LD_{50} value was calculated according to the amount of capsaicin in the capsicum extract. The number of animals used are shown in parentheses.

‖These weanling rats were administered capsaicin dissolved in propylene glycol, not dimethylsulfoxide.

TABLE 2. RELATIVE LETHALITY OF CAPSAICIN DISSOLVED IN DIMETHYLSULFOXIDE WHEN ADMINISTERED BY VARIOUS ROUTES IN MALE MICE (25–35 g)

Route of administration*	LD_{50}† (mg/kg)	Slope	Onset of convulsion (min)‡	Survival time (min)‡
Intravenous (55)	0·56 (0·36–0·87)	2·23	0·05 ± 0·01‖	1·62 ± 0·28
Intratracheal (56)	1·60 (1·03–2·48)	1·55	—	2·22 ± 0·21
Intraperitoneal (70)	7·65 (5·28–11·09)	2·32	1·08 ± 0·21	2·43 ± 1·08
Intramuscular (84)	7·80 (5·53–10·99)	2·01	1·48 ± 0·37	6·53 ± 0·72**
Subcutaneous (50)	9·00 (5·08–15·93)	2·27	6·41 ± 0·88‖	9·47 ± 1·01‖
Intragastric (30)§	60–75	—	—	—
Intragastric (63)	190 (122–294)	2·81	3·38 ± 0·65¶	5·32 ± 0·77**
Intrarectal (12)	>218	—	—	—
Dermal (16)	>512	—	—	—

*The number of animals used are shown in parentheses.

†Calculated by the method of LITCHFIELD and WILCOXON (1949). Figures in the parentheses are 95% confidence limits.

‡Time (mean ± S.E.) was recorded after capsaicin administration. Values marked with ‖, ¶ and ** differ significantly from those mice treated i.p. with capsaicin: ‖ $P < 0{\cdot}001$; ¶ $P < 0{\cdot}01$; ** $P < 0{\cdot}05$.

§Capsaicin was dissolved in ethanol, Tween 80 and saline.

However, no deaths or toxic signs were observed in mice receiving rectal or dermal doses as high as 218 or 512 mg/kg, respectively.

After capsaicin administration, the rats became excited and convulsed within approximately 1–2 min. Limbs were extended, the rats became dyspneic and apparently died from respiratory failure within 2–5 min. Survivors of a single high dose of capsaicin administration demonstrated similar signs and recovered within half an hr. Intravenous administration of capsaicin induced the shortest onset time of convulsion (about 5 sec). The general characteristics of convulsion in the capsaicin treated rats are somewhat similar to the *grand mal* seizure in mice treated with a lethal dose of pentamethylenetetrazol, (GLINSUKON unpublished observation) except that there is a longer duration of excitation and jumping prior to seizure and tonic spasm of the musculature after death in the rats as well as other species treated with capsaicin (tonic spasm of the musculature was not observed in guinea pigs).

At autopsy, only hyperemia without hemorrhage was observed in the visceral organs and the muscular wall of the peritoneal cavity with a slight increase in the amount of peritoneal fluid in the rat treated i.p. with capsaicin. A similar observation was also found in mice treated i.p. with capsaicin. Histopathologic changes seen in the gastric mucosa of mice treated intragastrically with capsaicin revealed desquamatic necrosis with increased mucous material (PAS's Schiff stain). Some of the chief and parietal cells showed an appearance of pale basophilic cytoplasm and vacuolization. No significant histopathologic changes were observed in other organs.

The pattern of the electrocardiogram and heart rate did not change for 5 min after capsaicin administration. Respiratory rates were slightly increased during the first min, whereas a small increase in the tidal volume was also observed. The tidal volume then decreased to 10–20% of the control within 3–4 min and the respiration stopped. During this time, heart rate gradually decreased and electrocardiograph signals disappeared much later (about 6–14 min). Mean arterial pressure was somewhat variable in the rats treated with capsaicin. At the beginning, capsaicin causes a transient hypotension and then hypertension. Mean arterial pressure gradually decreased along with the decrease in the tidal volume. Convulsions were not observed in these anesthetized rats treated with a lethal dose of capsaicin. This finding was confirmed in mice anesthetized with sodium pento-

barbital and subsequently given a single lethal dose of capsaicin. All eight mice died without convulsion (STITMUNNAITHUM, M. S. Thesis, Mahidol University, 1977).

The LD_{50} values indicate a high susceptibility, in guinea pigs, rats and mice, whereas hamsters and rabbits are less susceptible to capsaicin. Capsaicin is a highly toxic compound when administered by all routes except gastric, rectal and dermal. CABANAC *et al.* (1976) published a report on the acute toxicity of capsaicin in which adult male rats were given four increasing s.c. doses of capsaicin (cumulative amount of 21·0–66·0 mg/rat). Unfortunately, the LD_{50} values were not determined. Thus, it is difficult to compare the acute toxicity of capsaicin to the present study because of the differences in age and strain of rat, and the repeated doses of administration.

The lethality of capsaicin in the capsicum extract was about 4-fold higher in mice injected i.p. than with pure capsaicin (Table 1), suggesting that capsaicin derivatives (MÜLLER-STOCK *et al.*, 1973) or other components in the capsicum extract have a synergistic effect. A similar finding on the greater inhibitory effect of an aqueous capsicum extract on the intestinal glucose absorption of the rat and hamster (MONSEREENUSORN and GLINSUKON, 1978) has also been observed. However, the lethality of capsaicin administered intragastrically about 1/30 that following i.p. or i.v. injection. This is probably attributable to the insolubility of capsaicin in aqueous solution (STECHER *et al.*, 1968; TANGKRISANAVINONT *et al.*, 1977) but not the instability in acid solution (TOH *et al.*, 1955; GLINSUKON, unpublished observation.)

Our results confirm previous reports on the apnoetic effect of capsaicin in the dog and cat (TOH *et al.*, 1955; COLERIDGE *et al.*, 1964; MOLNÁR and GYÖRGY, 1967) and suppression by sodium pentobarbital of the convulsions in the anesthetized rats (this study) and mice (STITMUNNAITHUM, M. S. Thesis, Mahidol University, 1977). However, the mechanism by which capsaicin acts is not yet known, however our results suggest that the possible cause of death is respiratory paralysis.

The lethality of capsaicin administered gastrically to the mouse is much less than that of the i.p. route. The minimum lethal intragastric dose of capsaicin per kg was about 100 mg which would be contained in about 32·4 g dry wt of fruits. For a 60-kg person, this toxic level would be comparable to the consumption of about 1·94 kg of dry wt of capsicum fruits, an extremely high amount. Additionally, the pain and pungent sensation of capsaicin in the fruits will prevent the over consumption of this spice (MOLNÁR, 1965). We conclude, therefore, that the acute toxicity of capsaicin as a food-borne substance in man would rarely occur, assuming that the mouse and man have a similar susceptibility.

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REFERENCES

BRENDER, D. and WEBB-PEPLOE, M. M. (1969) Vascular responses to stimulation of pulmonary and carotid baroreceptors by capsaicin. *Am. J. Physiol.* **217**, 1836.

CABANAC, M., CORMARECHO-LEYDIER, M. and POIRIOR, L. J. (1976) The effect of capsaicin on temperature regulation of the rat. *Pflugers Arch. ges. Physiol.* **366**, 217.

COLERIDGE, H. M., COLERIDGE, J. C. G. and HIDD, C. (1964) Role of pulmonary arterial baroreceptors in the effects produced by capsaicin in the dog. *J. Physiol.* **170**, 272.

JANCSO-GABOR, A., SZOLESANYI, J. and JANCSO, N. (1970) Stimulation and desensitization of the hypothalamic heat sensitive structures by capsaicin in rats. *J. Physiol.* **280**, 449.

KARAWYA, M. S., BULBAA, S. J., GIRGIS, A. N. and YOUSSEF, A. F. (1967) A micro method for the determination of capsaicin in capsicum fruits. *Analyst, Lond.* **92**, 581.

LILLE, J. DE and RAMIREZ, E. (1935) Pharmacodynamic action of the active principle of chilli (*Capsicum annuum, L.*). *Chem. Abstr.* **29**, 4836.

LITCHFIELD, J. T. and WILCOXON, F. (1949) A simplified method of evaluating dose-effect experiments. *J. Pharmac. exp. Ther.* **69**, 99.

MOLNÁR, J. (1965) Die pharmakologischen wirkungen des capsaicins, des schaf schmeckenden wirkstoffes im paprika. *Arzneimittel-Forsch.* **15**, 718.

MOLNÁR, J. and GYÖRGY, L. (1967) Pulmonary hypertensive and other haemodynamic effects of capsaicin in the cat. *Eur. J. Pharmac.* **1**, 86.

MONSEREENUSORN, Y. and GLINSUKON, T. (1978) Inhibitory effect of capsaicin on intestinal glucose absortion *in vitro*. *Fd. Cosmet. Toxic.* **16**, 469.

MÜLLER-STOCK, A., JOSHI, K. and BUCHI, J. (1973) Thin-layer and column chromatographic separation of capsaicinoids from the drug. 5. Study on the constituents of capsicum. *J. Chromat.* **79**, 229.

STECHER, P. G., WINDHOLZ, M., LEAHY, D. S., BOLTON, D. M. and EATON, L. G. (1968) In: *The Merck Index, An Encyclopedia of Chemicals and Drugs*. p. 203. Merck: Rahway.

TANGKRISANAVINONT, V., GLINSUKON, T. and BURANAWUTI, T. (1977) Solubility of capsaicin in various vehicles and solvents. *Bangkok Scientific Symposium Abstr.* p. 329.

TODA, N., USNI, H., NISHINO, N. and FUJIWARA, M. (1972) Cardiovascular effects of capsaicin in dogs and rabbits. *J. Pharmac. exp. Ther.* **181**, 512.

TOH, C. C., LEE, T. S. and KIANG, A. K. (1955) The pharmacological actions of capsaicin and analogues. *Br. J. Pharmac.* **19**, 175.