Antihypertensive Effects of a Novel Calcium Antagonist, Semotiadil Fumarate (SD-3211), Alone and in Combination with Enalapril or Trichlormethiazide in Spontaneously Hypertensive Rats

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Semotiadil fumarate, a novel benzothiazine calcium antagonist, was given alone or in combination with either enalapril or trichlormethiazide to conscious, spontaneously hypertensive, rats daily for 2 weeks. Systolic blood pressure and heart rate were recorded 24 h before the start of the regimen and then every 2 and 24 h after the 1st, 3rd, 7th, 10th and 14th doses. When given alone, the antihypertensive effects of semotiadil (10 mg/kg, *p.o.*) and enalapril (5 mg/kg, *p.o.*) first became apparent after the 3rd dose and thereafter the effects appeared to develop daily although this effect had waned by the time of the next dose. When given in combination, however, these drugs appeared to potentiate each other and after the 7th dose, the antihypertensive effect persisted. Trichlormethiazide (30 mg/kg, *p.o.*) alone failed to exert any significant antihypertensive effect and in combination was not always additive to that of semotiadil. In contrast to the effect on blood pressure, the heart rate remained resistant to all these drugs. These results indicate that combined daily dosing of semotiadil, especially with enalapril, each at relatively low doses may be able to control hypertension in a continuous manner.

Keywords semotiadil; calcium antagonist; antihypertensive; enalapril; trichlormethiazide; spontaneously hypertensive male rat (SHR)

Semotiadil (semotiadil fumarate; SD-3211), (+)-(R)-2-[5-methoxy-2-[3-[methyl[2-[3,4-(methylenedioxy)phenoxy]ethyl]-amino]propoxy]phenyl]-4-methyl-2H-1,4-benzothiazin-3(4H)-one hydrogen fumarate, is a recently developed calcium antagonist, with a novel chemical structure and pharmacological profile.^{1–9)} So far, this non-dihydropyridine agent has been shown to interact with a novel Ca ion channel site which is distinct from the sites specific for 1,4-dihydropyridines (*e.g.*, nifedipine), phenylalkylamines (verapamil) or 1,5-benzothiazepines (diltiazem).^{4,5)} In addition, it also exhibits a slower onset and longer duration of action and is more vasoselective than verapamil and diltiazem but less selective than 1,4-dihydropyridines.^{2,6–9)}

A recent trend in clinical use of Ca antagonists is to combine them with other types of antihypertensives, in the expectation of obtaining a more effective control of severe and resistant hypertension with less unwanted effects than when used separately. In the present study, we attempted to see if such an advantage could be obtained from a combination of this novel drug with a typical angiotensin-converting enzyme (ACE) inhibitor, enalapril, or with a typical antihypertensive diuretic, trichlormethiazide, since clinically such combinations would be highly pertinent and realistic.

MATERIALS AND METHODS

Adult spontaneously hypertensive male rats (SHR, Okamoto strain) aged 21 weeks were obtained from Santen Pharmaceutical Co. (Osaka, Japan) and maintained on a special diet with a high salt content (sodium content 0.4%, Funabashi Farm, Chiba, Japan) and given water *ad libitum* throughout the experiment. Systolic blood pressure and heart rate were measured by the tail cuff method

(NARCO, PF-300, Houston, Tx, U.S.A.). Based on the measurements on day 0, animals were divided into six groups, I–VI, in such a manner that the average blood pressure did not differ among groups. A two-week regimen of daily administrations began on day 1 as follows: group I: twice with vehicle alone, group II: semotiadil (10 mg/kg, *p.o.*) supplied from Santen Pharmaceutical Co. and vehicle, group III: enalapril (Sigma Chemicals Co., St. Louis, Mo., U.S.A.; 5 mg/kg, *p.o.*) and vehicle, group IV: trichlormethiazide (Tokyo Kasei Co., Tokyo, Japan; 30 mg/kg, *p.o.*) and vehicle, group V: semotiadil and enalapril, and group VI: semotiadil and trichlormethiazide. Each drug was suspended in 0.5% methylcellulose (#400, Nakarai Tesque, Kyoto, Japan) as the vehicle before daily administration and given orally at a dose of 5 ml/kg/d. Subsequent measurements of blood pressure and heart rate were made every 2 and 24 h after each scheduled dosing on day 1, 3, 7, 10 and 14. Results were expressed as means $\pm$ S.E.M. Statistical significance was evaluated by Duncan's *t* test and $p < 0.05$ was taken as significant.

RESULTS AND DISCUSSION

The dose of semotiadil was selected using an ED₄₀ (12.3 mg/kg, *p.o.*) estimated in SHR by Takada *et al.*⁸⁾ which would produce a decrease in systolic blood pressure of 40 mmHg. At this dose of enalapril it was expected to inhibit vascular ACE consistently and to produce an antihypertensive action at least equal to that of semotiadil.¹⁰⁾ When separately given, the antihypertensive effect of each drug was first apparent on day 3 in our recording schedule (Tables I and II) and, thereafter, continued to increase, although every daily effect had waned by the time of the next dosing. When co-administered, however, it was found that the drugs potentiated

TABLE I. Effect of Semotiadil on the Blood Pressure of SHR in the Presence and Absence of Other Antihypertensive Agents

	<i>n</i>	0 d	1st day		3rd day	
			2 h	24 h	2 h	24 h
Vehicle	6	178 ± 7	171 ± 10	173 ± 9	171 ± 5	159 ± 4
Semotiadil 10 mg/kg	5	171 ± 5	164 ± 18	170 ± 4	148 ± 7 ^{b)}	161 ± 3
Enalapril 5 mg/kg	6	175 ± 6	155 ± 11	156 ± 9	147 ± 4 ^{b)}	156 ± 4
Trichlormethiazide 30 mg/kg	5	168 ± 5	169 ± 6	185 ± 10	167 ± 7	168 ± 3
Semotiadil + enalapril	6	171 ± 5	141 ± 10	159 ± 4	129 ± 2 ^{b-d)}	156 ± 4
Semotiadil + trichlormethiazide	5	169 ± 6	156 ± 14	164 ± 7	150 ± 6 ^{a,f)}	162 ± 3

Each value is the mean ± S.E. a) $p < 0.05$ and b) $p < 0.01$ vs. vehicle, c) $p < 0.05$ vs. semotiadil, d) $p < 0.05$ and e) $p < 0.01$ vs. enalapril, f) $p < 0.05$ vs. trichlormethiazide.

TABLE II. Effect of Semotiadil on the Blood Pressure of SHR in the Presence and Absence of Other Antihypertensive Agents

	<i>n</i>	7th day		10th day		14th day	
		2 h	24 h	2 h	24 h	2 h	24 h
Vehicle	6	162 ± 3	163 ± 5	161 ± 6	168 ± 6	166 ± 4	163 ± 7
Semotiadil 10 mg/kg	5	137 ± 9 ^{a)}	169 ± 3	142 ± 5	169 ± 4	146 ± 6 ^{b)}	168 ± 6
Enalapril 5 mg/kg	6	142 ± 3	162 ± 7	129 ± 4 ^{b)}	143 ± 7 ^{b)}	132 ± 4 ^{b)}	156 ± 5
Trichlormethiazide 30 mg/kg	5	161 ± 11	153 ± 4	156 ± 7	166 ± 2	155 ± 5	155 ± 4
Semotiadil + enalapril	6	113 ± 7 ^{b,c,f)}	140 ± 6 ^{a,d,e)}	117 ± 7 ^{b,c)}	145 ± 4 ^{b,d)}	102 ± 3 ^{b,d,f)}	139 ± 3 ^{b,d,e)}
Semotiadil + trichlormethiazide	5	136 ± 5 ^{a,g)}	159 ± 6	138 ± 10 ^{a)}	160 ± 7	120 ± 6 ^{b,d,h)}	156 ± 8

Each value is the mean ± S.E. a) $p < 0.05$ and b) $p < 0.01$ vs. vehicle, c) $p < 0.05$ and d) $p < 0.01$ vs. semotiadil, e) $p < 0.05$ and f) $p < 0.01$ vs. enalapril, g) $p < 0.05$ and h) $p < 0.01$ vs. trichlormethiazide.

TABLE III. Effect of Semotiadil on the Heart Rate of SHR in the Presence and Absence of Other Antihypertensive Agents

	<i>n</i>	0 d	1st day		3rd day	
			2 h	24 h	2 h	24 h
Vehicle	6	361 ± 20	359 ± 29	368 ± 26	355 ± 17	338 ± 19
Semotiadil 10 mg/kg	5	365 ± 18	357 ± 20	370 ± 19	362 ± 19	357 ± 18
Enalapril 5 mg/kg	6	374 ± 13	356 ± 14	353 ± 13	375 ± 11	376 ± 18
Trichlormethiazide 30 mg/kg	5	377 ± 19	385 ± 17	389 ± 32	369 ± 18	372 ± 13
Semotiadil + enalapril	6	392 ± 11	375 ± 22	368 ± 17	402 ± 13	389 ± 18
Semotiadil + trichlormethiazide	5	343 ± 11	348 ± 15	345 ± 12	353 ± 16	339 ± 7

Each value is the mean ± S.E.

TABLE IV. Effect of Semotiadil on the Heart Rate of SHR in the Presence and Absence of Other Antihypertensive Agents

	<i>n</i>	7th day		10th day		14th day	
		2 h	24 h	2 h	24 h	2 h	24 h
Vehicle	6	315 ± 17	340 ± 11	335 ± 22	323 ± 15	354 ± 23	362 ± 29
Semotiadil 10 mg/kg	5	339 ± 11	360 ± 9	355 ± 11	339 ± 11	361 ± 13	352 ± 11
Enalapril 5 mg/kg	6	344 ± 17	336 ± 14	345 ± 8	342 ± 15	363 ± 12	335 ± 12
Trichlormethiazide 30 mg/kg	5	344 ± 12	345 ± 22	357 ± 17	360 ± 14	367 ± 8	333 ± 12
Semotiadil + enalapril	6	347 ± 15	327 ± 12	394 ± 19 ^{a)}	339 ± 7	392 ± 17	327 ± 7
Semotiadil + trichlormethiazide	5	337 ± 7	339 ± 15	340 ± 11	360 ± 4	366 ± 11	337 ± 10

Each value is the mean ± S.E. a) $p < 0.05$ vs. vehicle.

each other and a continuous control of hypertension was gradually achieved.

Trichlormethiazide alone was ineffective throughout the experiment even though the dose level¹¹⁾ was higher than the diuretic dose (0.01–0.04 mg/kg, *p.o.*).¹²⁾ This appears to be in accord with a report that benzothiadiazine diuretics such as trichlormethiazide do not clearly exhibit antihypertensive effects in SHR.¹³⁾ In combination with

semotiadil, the antihypertensive effect was first apparent after the final treatment (see the 14th day in Table II) and it appears that no other interaction had taken place between this pair of drugs.

Compared with blood pressure, the heart rate was highly resistant to any of the drug treatments (Tables III and IV). Even when a strong antihypertensive action was continuously exerted by the combination of semotiadil

with enalapril, only a mild tachycardia was tentatively noted on one recording day (see day 10 in Table IV). Such resistant heart rate might have been caused by the direct suppressing effect of semotiadil on heart rate in addition to its antihypertensive effect, as previously reported.^{6,9)}

The results suggest that semotiadil co-administered with a typical ACE inhibitor, both at moderately antihypertensive doses, is able to continuously control the blood pressure in SHR, possibly with less untoward effects than used separately. Calcium antagonists have been reported to first elevate plasma renin which would then counteract their intrinsic antihypertensive effect.^{14,15)} In such a situation, the inhibition of ACE is expected to potentiate the antihypertensive effect of Ca antagonists. Although we did not monitor plasma renin in this experiment, it is highly likely that such a mechanism worked, making possible the 24 h control of hypertension. Previously, Kageyama *et al.*¹⁶⁾ have reported in conscious renal hypertensive dogs that semotiadil in combination with propranolol but not alone, was able to produce an antihypertensive effect which lasted 24 h. We thus conclude that semotiadil is a useful antihypertensive calcium antagonist not only on account of its intrinsic pharmacological profile but also the possible advantage offered by its compatibility with propranolol¹⁶⁾ and, now, with enalapril. Such combination therapy should be able to control severe and resistant hypertension more efficiently and safely.

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