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Anti-inflammatory, Analgesic and Ulcerogenic Properties of *S*-(+)-Ibuproxam, Racemic Ibuproxam-β-cyclodextrin and *S*-(+)-Ibuproxam-β-cyclodextrin

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Abstract

The anti-inflammatory, analgesic and gastric mucosal damage-inducing activities of *S*-(+)-ibuproxam, and *S*-(+)-ibuproxam-β-cyclodextrin, new propionic acid derivatives, and racemic ibuproxam-β-cyclodextrin were investigated in three animal models and compared with those of racemic ibuproxam, racemic ibuprofen and its optical enantiomer *S*-(+)-ibuprofen.

The anti-inflammatory activities of racemic ibuprofen, *S*-(+)-ibuprofen and racemic ibuproxam in carrageenan-induced paw oedema in rats were almost equipotent and slightly greater than those of *S*-(+)-ibuproxam and *S*-(+)-ibuproxam-β-cyclodextrin, and significantly greater than that of racemic ibuproxam-β-cyclodextrin. In abdominal constriction tests in mice, the analgesic effects of racemic ibuproxam, *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin were significantly less pronounced than those of racemic ibuprofen and *S*-(+)-ibuprofen. Ulcerogenic activity of *S*-(+)-ibuproxam-β-cyclodextrin in rats was found to be significantly weaker than that of racemic ibuproxam-β-cyclodextrin, racemic ibuproxam and *S*-(+)-ibuproxam and, most notably, weaker than those of racemic ibuprofen ans *S*-(+)ibuprofen.

These results indicate that *S*-(+)-ibuproxam-β-cyclodextrin could be a novel potent anti-inflammatory and analgesic agent with a therapeutic index more favourable than that of the classical non-steroid anti-inflammatory drugs ibuprofen and ibuproxam.

A review of marketed pharmaceutical products reveals that the majority of currently available chiral synthetic compounds are sold as racemic mixtures (Heydorn 1995). This is primarily because until recently it was not technically or economically feasible to separate racemic mixtures into their individual enantiomers. Several classes of non-steroidal anti-inflammatory drugs (NSAIDs), including the 2-aryl-propionic acid derivatives, are chiral compounds and exist in two stereoisomeric forms. Ibuprofen is an optically active 2-aryl-propionic acid derivative with potent anti-inflammatory, analgesic and antipyretic properties. However, it also exhibits gastric toxicity, which is related to its prostaglandin synthesis suppressing activity. Ibuproxam (*R*,*S*-2-(4-isobutylphenyl)-propiohydroxamic acid), a prodrug of ibuprofen, has some advantages over ibuprofen, such as reduced gastric irritation and better bioavailability (Orzalesi et al 1980). In this and other 2-aryl-propionic acids, the 2-position carbon is asymmetric and undergoes an in-vivo inversion of configuration. In both humans and rats, *R*-ibuprofen is metabolically and stereoselectively inverted to *S*-ibuprofen (Kaiser et al 1976; Nakamura et al 1981). Because the drug is administered as a racemic mixture and only *S*-ibuprofen possesses pharmacological activity, the chiral inversion reaction assumes a therapeutic significance. There are several advantages to be gained from the use of the *S* enantiomer, such as administration of smaller doses, reduced side-effects, and simplified pharmacokinetics. To minimize side-effects (especially gastric irritation) and reduce the effective dose, *S*-(+)-ibuproxam, a prodrug of *S*-(+)-ibuprofen, the pharmacologically active component of ibuprofen, was synthesized (Zmitek et al 1995) (Fig. 1).

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Since the clinical usefulness of NSAIDs may be limited by the appearance of gastrointestinal symptoms or other adverse effects at doses close to those effective against inflammation and pain, there has been increasing interest in optimizing the efficacy of drug activity through the use of rationally designed drug-carrier materials. Cyclodextrins are a strong candidate for such a role. Cyclodextrin complexation allows the modification of the physical and chemical properties of a drug and hence alteration of its pharmacokinetic and possibly pharmacodynamic properties. Owing to ibuproxam's low water solubility and previous findings that racemic ibuproxam complexed with β-cyclodextrin possessed better bioavailability than racemic ibuproxam alone (Zmitek et al 1992), we prepared some new cyclodextrin inclusion compounds with racemic or *S*-(+)-ibuproxam. Racemic ibuproxam-β-cyclodextrin is a complex of the established NSAID ibuproxam and an inert cyclic macromolecule, β-cyclodextrin (molar ratio 1·0 ibuproxam : 1·1 β-cyclodextrin : 4·4 H_2O).

Materials and Methods

Chemicals

Racemic ibuproxam, *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin *S*-(+)-ibuproxam-β-cyclodextrin were synthesized at Lek Pharmaceutical and Chemical Company d.d. (Ljubljana, Slovenia). Other drugs were obtained from the following sources: racemic and *S*-(+)-ibuprofen, Ethyl Corporation, Baton Rouge, LA; sodium carrageenan (Viscarin) Marine Colloids Inc., Springfield, NY; phenyl-*p*-quinone, Sigma, St. Louis, MO, USA.

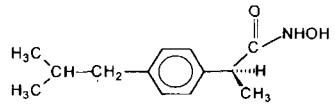

FIG. 1. The structure of *S*-(+)-ibuproxam.

Drug administration

Test compounds were suspended in a 10% gum-arabic solution and given orally by gastric tube, in the volume of 10 mL kg^{-1}. The dosage of racemic and *S*-(+)-ibuproxam-β-cyclodextrin, expressed as mg kg^{-1}, refers to the quantity of the active parent drug ibuproxam.

Animals

Male Han: WIST rats (150–170 g) and female Han: NMRI mice (18–25 g) were used. Animals were housed in air-conditioned rooms maintained at 20 ± 1°C. Unless otherwise indicated, the animals were given free access to food and water. All studies were performed between 0700 and 1500 h in a quiet room. The animals were used only once.

Anti-inflammatory activity (carrageenan-induced oedema)

The method of Winter et al (1962) with a slight modification was used. Groups of eight rats, fasted overnight, were injected with 0·1 mL of a 1·5% suspension of carrageenan in 0·9% NaCl solution into the sub-plantar region of the right hind paw 1 h after the oral administration of the test compound. An equal volume of 0·9% NaCl was injected into the left hind paw as a control. The volumes of both hind paws were measured by means of plethysmometer (Model 7150, Ugo Basile) 0, 1, 3, 4, and 5 h after the carrageenan injection. Controls had a suspension vehicle and for each dose of the test compound a separate control group (n = 8) was employed. Percentage of hind-paw swelling was calculated.

The data were used to compare the area under the time-course curve (from 0 to 5 h after carrageenan injection). ED30 is the dose calculated to have produced 30% inhibition of carrageenan-induced oedema. The ED30 value was determined by the method described at the end of this section (see statistical analysis).

Analgesic activity (abdominal constriction)

Groups of ten mice were given the test compound orally. After 0·5 h phenylquinone (0·02%, 0·25 mL) was injected intraperitoneally. The number of abdonimal contrictions was counted for 15 min, starting 5 min after the intraperitoneal injection. A control group was dosed with the suspension vehicle.

Ulcerogenic effect

A modification of the method of Cashin et al (1977) was employed. At the end of the experiment to test the anti-

Table 1. Effects of *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin (rac.ibx-β-CD, and *S*-(+)-ibuproxam-β-cyclodextrin (*S*-ibx.-β-CD) and reference drugs on carrageenan-induced paw oedema in rats.

Drugs	Dose (mg kg^{-1})	Hind-paw swelling (%) 1, 2, 3, 4 and 5 h after carrageenan injection				
		1	2	3	4	5
Racemicibuproxam	Control	15·5 ± 1·1	28·8 ± 1·5	39·0 ± 1·9	49·5 ± 2·8	57·6 ± 3·1
	10	16·5 ± 6·9	22·0 ± 8·3	35·2 ± 8·6	40·5 ± 10·6*	43·4 ± 15·7*
	25	16·1 ± 9·3	22·9 ± 7·8	32·0 ± 6·7*	34·2 ± 11·7*	43·6 ± 14·2*
	50	10·7 ± 6·8	16·6 ± 6·1*	19·9 ± 11·0*	22·4 ± 8·9*	30·1 ± 16·1*
	100	15·2 ± 7·9	13·4 ± 7·9*	15·9 ± 8·2*	13·1 ± 8·1*	15·2 ± 8·1*
	200	10·11 ± 4·2	11·9 ± 4·6*	14·9 ± 5·9*	18·1 ± 7·1*	25·9 ± 6·9*
S-Ibuproxam	Control	15·2 ± 0·9	28·2 ± 1·8	37·8 ± 2·2	50·0 ± 2·2	54·8 ± 2·8
	25	17·4 ± 6·5	26·4 ± 6·2	33·4 ± 3·6	37·7 ± 8·2*	44·2 ± 9·4*
	37·5	15·5 ± 5·5	24·25 ± 6·0	30·5 ± 11·6	33·1 ± 11·6*	51·9 ± 13·8
	50	14·4 ± 7·7	13·1 ± 6·4*	15·9 ± 8·9*	20·1 ± 10·1*	25·3 ± 9·4*
	100	15·1 ± 12·3	19·2 ± 8·5*	15·4 ± 8·1*	17·1 ± 8·9*	21·6 ± 9·4*
Rac.ibx.-β-CD	Control	13·1 ± 1·0	24·8 ± 1·3	37·0 ± 2·3	48·7 ± 2·6	56·1 ± 2·5
	10	7·9 ± 5·5*	14·7 ± 8·0*	21·2 ± 9·4*	32·7 ± 10·3*	41·2 ± 6·5*
	25	8·2 ± 5·1*	14·0 ± 5·7*	21·0 ± 6·7*	25·2 ± 8·4*	33·6 ± 8·6*
	50	10 ± 3·5*	13·7 ± 3·8*	20·1 ± 4·4*	26·5 ± 4·7*	39·7 ± 10·0*
	100	10·2 ± 3·1*	18·6 ± 5·4*	18·4 ± 4·3*	25·1 ± 7·5*	31·5 ± 12·2*
	200	8·0 ± 6·8*	12·9 ± 5·5*	15·4 ± 5·9*	19·5 ± 7·0*	27·0 ± 9·6*
S-ibx.-β-CD	Control	16·0 ± 1·3	26·5 ± 1·5	40·0 ± 1·8	50·8 ± 2·1	59·0 ± 2·2
	10	12·4 ± 7·0	18·5 ± 8·0*	26·7 ± 8·8*	36·6 ± 11·5*	45·9 ± 13·6*
	25	7·0 ± 3·3*	16·6 ± 4·4*	29·0 ± 6·9*	46·4 ± 11·9*	66·2 ± 11·8*
	50	15·4 ± 9·1	16·6 ± 9·0*	23·2 ± 9·3*	27·7 ± 11·9*	36·1 ± 10·6*
	100	8·5 ± 3·6*	10·5 ± 5·3*	12·5 ± 6·9*	17·5 ± 5·0*	24·2 ± 8·8*
	200	9·5 ± 4·1*	11·4 ± 3·8*	14·9 ± 5·2*	20·4 ± 5·7*	30·0 ± 7·6*
Rac.ibuprofen	Control	18·6 ± 1·1	31·2 ± 1·7	43·9 ± 2·5	52·1 ± 2·6	67·5 ± 3·2
	10	17·9 ± 9·2	20·7 ± 3·7*	30·2 ± 4·3*	39·2 ± 11·9*	53·4 ± 17·7*
	25	13·4 ± 5·1	19·2 ± 7·1*	21·0 ± 11·1*	23·37 ± 7·2*	32·5 ± 7·9*
	50	17·9 ± 7·0	19·2 ± 6·6*	24·1 ± 8·0*	22·1 ± 6·9*	26·1 ± 10·7*
	100	14·7 ± 6·1	17·9 ± 6·2*	18·9 ± 7·5*	22·2 ± 7·1*	26·0 ± 6·2*
S-Ibuprofen	Control	16·5 ± 0·6	31·2 ± 1·1	44·1 ± 2·1	55·6 ± 2·6	64·5 ± 2·6
	10	12·5 ± 4·2*	16·4 ± 5·7*	26·5 ± 9·9*	37·6 ± 19·4*	54·5 ± 25·3
	25	15·0 ± 10·3	15·7 ± 9·0*	21·2 ± 7·8*	29·4 ± 10·8*	40·2 ± 8·7*
	50	19·5 ± 6·3	22·5 ± 4·5*	23·0 ± 6·7*	27·7 ± 9·5*	34·5 ± 13·2*
	100	15·4 ± 7·2	15·6 ± 7·6*	19·9 ± 8·1*	21·2 ± 10·7*	26·6 ± 8·3*

Results are expressed as mean ± s.d. Drugs were administered orally 1 h before carrageenan injection. Eight rats per dose level were used in the test. Statistically significant differences with the corresponding control values are expressed as * $P < 0.05$ (*t*-test for grouped data).

Table 2. Effect of *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin and reference drugs on carrageenan-induced oedema and on gastric mucosa in rats.

Compounds	Anti-inflammatory activity (ED30)	Ulcerogenic effect (UD50)	UD_{50}/ED_{30}
S-(+)-Ibuproxam-β-cyclodextrin	35·23 (23·36–53·13)	90·22 (45·75–177·91)	2·56
Racemic ibuproxam	29·20 (18·36–46·45)	40·1 (21·8–73·9)	1·37
S-(+)-Ibuproxam	38·53 (23·04–54·43)	62·9 (38·5–102·9)	1·36
Racemic ibuproxam-β-cyclodextrin	45·10 (26·99–75·36)	57·8 (29·14–114·67)	1·28
Racemic ibuprofen	24·82 (15·37–40·06)	23·5 (14·1–39·1)	0·94
S-(+)-Ibuprofen	24·97 (16·24–38·39)	13·4 (5·4–33·2)	0·54

ED30 (mg kg^{-1}) is the dose calculated to have produced 30% inhibition of carrageenan-induced oedema; UD50 (mg kg^{-1}) is the dose that was effective in producing at least score 1 mucosal damage in 50% of the rats. Drugs were administered orally 1 h before carrageenan injection. Results are expressed as mean together with 95% confidence limits. Eight rats per dose level were used in the test.

inflammatory activity of each dose of each compound (6 h after dosing), the rat was decapitated, the stomach removed, washed with 0·9% NaCl, and opened along the lesser curvature. Ulceration of the mucosa was scored according to an arbitrary system: 0—no lesions; 0·5—hyperaemia; 1—one or two slight lesions present; 1·5—more than two lesions; 2—severe lesions; 3—very severe lesions; 4—lesions involving the whole mucosa. The observer was unaware of the treatment. A control group was dosed with the suspension vehicle. The ED50 was calculated according to the method of Litchfield and Wilcoxon (Tallarida & Murray 1987) at a dose that was effective to produce at least score 1 mucosal damage in 50% of the rats.

Statistical analysis

The ED50 values and 95% confidence limits of data from the ulcerogenic effects test were analysed on the basis of the number of animals that satisfied an all-or-none criterion. For this purpose the method of Litchfield and Wilcoxon was used (program 46, Pharmacologic Calculation system, version 4.2, Tallarida & Murray 1987).

The statistical method used to estimate ED30 and ED50 in the analgesic and anti-inflammatory tests was different from the standard ones (Krisch et al 1994). Logarithms of the ED30 and ED50 were estimated by fitting (continuous) regression model. In principle the logistic (Hill) function:

$$E = C_1 D^P/(D^P + C_2) \quad (1)$$

with dose D, response E and Hill coefficient P, was estimated directly. In both groups of tests the Hill coefficient P was equal to −1. In the case of the anti-inflammatory test, different controls were used for different doses, therefore the fitted Hill function was modified as follows:

$$E = (C_1 + U_1C_{31} + U_2C_{32} + U_3C_{33})/(1. + C_2D) \quad (2)$$

The additional (dummy) variables (U) were equal to 1 at doses with the same (corresponding) control and 0 in other cases. In the experiments of inhibition of phenyquionone-induced abdominal constriction, Hill function without modifications was used. So, logarithms of ED50 were estimated by fitting

$$E = C_1/(1. + C_2D) \quad (3)$$

All calculations were made using the NLP routine of the SPSS statistical package (SPSS Inc., Chicago, IL). For parametric data, statistical evaluation was performed by the *t*-test for grouped data.

Table 3. Effect of *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin, *S*-(+)-ibuproxam-β-cyclodextrin and reference drugs on phenylquinone-induced abdominal constriction in mice.

Compounds	Analgesic activity (ED50)
Racemic ibuproxam	43·0 (27·1–68·3)
S-(+)-Ibuproxam	73·0 (44·1–120·9)
Racemic ibuprofen	7·0 (4·3–11·3)
S-(+)-Ibuprofen	1·9 (1·27–3·2)
Racemic ibuproxam-β-cyclodextrin	31·65 (21·0–47·7)
S-(+)-Ibuproxam-β-cyclodextrin	30·36 (20·06–45·94)

ED50 (mg kg^{-1}) is the dose to have reduced the number of phenylquinone-induced abdominal constrictions by 50%. Results are presented as means together with 95% confidence limits. Drugs were administered orally 30 min before phenylquinone injection. Ten mice per dose level were used in each test.

Results

Anti-inflammatory activity (carrageenan-induced oedema)

The results are shown in Table 1 and Table 2. The ED30 value of *S*-(+)-ibuproxam was calculated to be 38·53 mg kg^{-1}, nearly equal to that of *S*-(+)-ibuproxam-β-cyclodextrin (ED30: 35·23 mg kg^{-1}), less potent than that of racemic ibuproxam (ED30: 29·20 mg kg^{-1}), racemic ibuprofen (ED30: 24·82 mg kg^{-1}) and *S*-(+)-ibuprofen (ED30: 24·97 mg kg^{-1}) and slightly more potent than that of racemic ibuproxam-β-cyclodextrin (ED30: 45·10 mg kg^{-1}). After the oral administration of all tested compounds, the limb volume was reduced as a function of the dose. The anti-inflammatory effect of oral racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin tested at the doses 10 and 25 mg kg^{-1} was significantly greater than that of the same doses of racemic and *S*-(+)-ibuproxam 2, 3 and 4 h after oral administration to rats in a carrageenan-induced paw oedema test (on average more than 30% vs no inhibition).

Analgesic activity (abdominal constriction test)

Table 3 summarizes the effects of tested compounds on phenylquinone-induced abdominal constriction in mice. The analgesic effect of *S*-(+)-ibuproxam was surprisingly weaker than that of racemic ibuproxam, racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin; the ED50 values were 73 mg kg^{-1}, 43 mg kg^{-1}, 31·65 mg kg^{-1} and 30·36 mg kg^{-1}, respectively. The ED50 value was calculated to be 7 mg kg^{-1} for racemic ibuprofen and 1·9 mg kg^{-1} for

Table 4. Effects of *S*-(+)-ibuproxam, racemic ibuproxam-β-cyclodextrin (rac.ibx.-β-CD) and *S*-(+)-ibuproxam-β-cyclodextrin (*S*-ibx.-β-CD) and reference drugs on gastric mucosa in starved rats.

Compounds	No. of rats with mucosal damage scored at least 1/group Dose (mg kg^{-1})					
	10	25	37·5	50	100	200
Racemic ibuproxam	0/8	0/8		6/8	7/8	8/8
S-Ibuproxam		0/8	3/8	4/8	5/8	
Rac.ibx.-β-CD	1/8	2/8		4/8	7/8	7/8
S-ibx.-β-CD	1/8	0/8		0/8	7/8	6/8
Racemic ibuprofen	0/8	6/8		6/8	6/8	
S-Ibuprofen	2/8	7/8		7/8	7/8	

Drugs were administered orally 1 h before carrageenan injection. Eight rats per dose level were used in the test.

S-(+)-ibuprofen. Thus the analgesic potency of racemic ibuproxam in mice was slightly weaker than that of racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin and significantly weaker than that of racemic and optically active ibuprofen.

Ulcerogenic effect

Table 2 and Table 4 illustrate the gastric mucosal damage-inducing activity of tested compounds. In the control group, no mucosal damage was found (n = 16). The gastric mucosal damage-inducing activity of *S*-(+)-ibuproxam was lower than that of racemic ibuproxam; the UD50 values were 62·9 mg kg^{-1} and 40 mg kg^{-1}, respectively. It can be noted that *S*-(+)-ibuproxam-β-cyclodextrin caused less injury to the gastric mucosa than did *S*-(+)-ibuproxam (UD50: 90·22 mg kg^{-1}) and that its gastric mucosal damage-inducing activity was much lower than its anti-inflammatory activity (UD50: 90·22 mg kg^{-1} vs ED30: 35·23 mg kg^{-1}) (Table 2). On the contrary, the reference compounds exert a relatively strong capacity for inducing gastrointestinal injury; the UD50 values were calculated to be 23·5 mg kg^{-1} for racemic ibuprofen and 13·4 mg kg^{-1} for *S*-(+)-ibuprofen. UD50 data in rats indicated that *S*-(+)-ibuproxam-β-cyclodextrin was less ulcerogenic than *S*-(+)-ibuproxam (1·4 times), racemic ibuproxam-β-cyclodextrin (1·6 times), racemic ibuproxam (2·2 times), racemic ibuprofen (3·8 times) and *S*-(+)-ibuprofen (6·7 times).

Discussion

In the present study some direct comparisons of anti-inflammatory, analgesic and ulcerogenic profiles of two new propionic acid derivatives, *S*-(+)-ibuproxam and *S*-(+)-ibuproxam-β-cyclodextrin, and racemic ibuproxam-β-cyclodextrin are made with racemic ibuproxam, racemic ibuprofen and *S*-(+)-ibuprofen which are considered to be the representatives of the anti-inflammatory propionic acid derivatives. Whenever a drug can be obtained in a variety of chemically equivalent forms (such as enantiomers), it is both good science and good sense to explore the potential for in-vivo differences between these forms (De Camp 1989). Ibuproxam is a derivative of ibuprofen and both possess an asymmetric carbon atom and can therefore occur as the *S* or *R* enantiomers. Ibuprofen is used clinically as the racemate although in-vitro (inhibition of prostaglandin synthetase system) and in-vivo, only the *S* enantiomer is effective (Adams et al 1976; Geisslinger et al 1989; Loew & Schuster 1989). However, Adams et al (1976) did not find this large difference in analgesic and anti-inflammatory potency between the two enantiomers in-vivo (mouse, rat, guinea pig) when they compared both enantiomers with the racemic substance, because inversion occurs. NSAIDs are among the most commonly prescribed drugs in the world but their use as anti-inflammatory, antipyretic, anti-thrombotic and analgesic agents continues to be primarily limited by their untoward effects on the gastrointestinal tract. Several strategies have been used to reduce the gastrointestinal injury caused by NSAIDs: coating to prevent absorption in the stomach; parenteral administration; and formulation of prodrugs (Wallace & Cirino 1994). Cyclodextrin complexation allows modification of a drug, and hence its pharmacokinetic and possibly pharmacodynamic properties (Szjetli 1994). The cyclodextrin molecule can be considered as a capsule of molecular size that is able to form an inclusion complex with the molecule of another substance (Szjetli 1994). A complexed drug may be less toxic than the free one, e.g. as in the case of local irritation, or more toxic because the solubility and bioavailability of poorly soluble toxic substances are increased by cyclodextrin complexation. From our studies on acute anti-inflammatory activity in rats it is clear that racemic ibuproxam is approximately as effective as racemic ibuprofen. These results are in accordance with that of Orzalesi et al (1977). In this model, *S*-(+)-ibuproxam is found to be weaker than racemic ibuproxam. The anti-inflammatory effect of oral racemic ibuproxam-β-cyclodextrin was somewhat weaker than that of racemic ibuproxam in a carrageenan-induced paw oedema test. We also found that *S*-(+)-ibuproxam-β-cyclodextrin was nearly equipotent to that of *S*-(+)-ibuproxam in reduction of carrageenan oedema in rats. The anti-inflammatory effect of oral racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin 10 mg kg^{-1} and 25 mg kg^{-1} was significantly greater than that of the same doses of racemic and optically active ibuproxam 2, 3 and 4 h after oral administration to rats. As described previously (Lee & Balfour 1994), oral piroxicam-β-cyclodextrin 3 mg kg^{-1} also produced a significantly greater anti-inflammatory effect and a faster onset of action than that of piroxicam 3 mg kg^{-1} 1 h after oral administration to rats in a carrageenan-induced paw oedema test. There was no difference in overall anti-inflammatory activity between the two compounds.

Limited data available on the analgesic and anti-inflammatory effects of piroxicam-β-cyclodextrin suggest that it is

equivalent to piroxicam in animal and human models of pain (Lee & Balfour 1994). Abdominal contriction responses induced with various algogenic agents in mice are used for evaluating the analgesic action of various compounds, but are not wholly reliable because most NSAIDs show slight inhibition (Tanaka et al 1992). The results of the effect of *S*-(+)-ibuproxam on phenylquinone-induced abdominal constriction show that its analgesic activity was rather weak in contrast to its relatively good anti-inflammatory activity. Unlike Orzalesi et al (1977), who caused abdominal pain in mice with acetic acid, we found that analgesic activity of racemic ibuproxam against abdominal constriction responses induced by phenylquinone was 6 times and 23 times weaker than that of racemic ibuprofen and *S*-(+)-ibuprofen, respectively. Inhibitory activities of racemic ibuproxam-β-cyclodextrin and *S*-(+)-ibuproxam-β-cyclodextrin against the constriction induced with phenylquinone were nearly equipotent and slightly stronger than that of racemic ibuproxam.

NSAIDs cause gastroduodenal mucosal damage by a combination of systemic inhibition of prostaglandin cycloxygenase and direct topical irritation (Rainsford 1989; Levi & Shaw-Smith 1994). Ulcerogenic activity of racemic ibuproxam was almost 2 times weaker than racemic ibuprofen, and this result is consistent with the findings of Orzalesi et al (1977). Furthermore, *S*-(+)-ibuproxam was 1·6 times less damaging to the gastric mucosa than was racemic ibuproxam. Indomethacin, flurbiprofen, biphenyacetic acid, naproxen, phenylbutazone and piroxicam are examples of compounds whose gastrointestinal mucosal irritating effects can be reduced by cyclodextrin complexation (Szejtli 1994). We also found out that ulcerogenic activities of complexed racemic and *S*-(+)-ibuproxam were weaker than those of racemic and *S*-(+)-ibuproxam. However, gastric mucosal injury can occur after absorption of the drugs, since it occurs following parenteral NSAID administration (Nuutinen et al 1993). Crystals of sparingly soluble NSAIDs can remain in contact with gastric mucosa for a longer period of time which can provoke an ulcer. Formulations of NSAIDs such as complexes with β-cyclodextrin that limit the contact of the drug with gastric mucosa may theoretically reduce the incidence of mucosal injury (Rainsford 1990). Studies in rats indicated that *S*-(+)-ibuproxam-β-cyclodextrin caused less injury to the gastric mucosa than the same doses of *S*-(+)-ibuproxam. The low water-solubility of *S*-(+)-ibuproxam, which prevents rapid dissolution and absorption from the gastrointestinal tract, was overcome by complexation with β-cyclodextrin. As assayed with the methods we used, a dose of *S*-(+)-ibuproxam-β-cyclodextrin 2·6 times greater than those necessary to exhibit anti-inflammatory actions, produced ulcerative lesions. Therefore, it can be concluded that *S*-(+)-ibuproxam-β-cyclodextrin is a potent anti-inflammatory and analgesic compound with the better therapeutic index (ratio UD50/ED30) than those of *S*-(+)-ibuproxam, racemic ibuproxam, racemic ibuproxam-β-cyclodextrin, racemic ibuprofen and its optically active enantiomer and is considered to be a promising therapeutic agent.

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