20, 125518-62-9; **21**, 125518-63-0; **22**, 125518-64-1; **23**, 125518-65-2; **24**, 125518-66-3; **25**, 125518-67-4; **26**, 125518-68-5; **27**, 125518-69-6; **28**, 125518-70-9; **29**, 125518-71-0; **30**, 125518-72-1; **31**, 125518-73-2; **32**, 125518-74-3; **33**, 125518-75-4; **34**, 125518-76-5; **35**, 125518-77-6; **36**, 125518-78-7; *p*-$BrC_6H_4SO_2Cl$, 98-58-8; $H_3C(CH_2)_7SH$, 111-88-6; 2,4-thiazolidinedione, 2295-31-0; 5-bromo-1-naphthalenesulfonyl chloride, 50638-04-5; 1-bromo-2-mercaptonaphthalene, 90767-26-3; 2-naphthol, 135-19-3; (1-methylcyclohexyl)methanol, 14064-13-2; 5-[(4-fluorophenyl)thio]-2,4-thiazolidinedione, 125518-79-8; 2-(bromomethyl)naphthalene, 939-26-4; 2-(methylthio)naphthalene, 1076-67-1; 1-naphthalenesulfonyl chloride, 85-46-1; 5-(trifluoromethyl)-6-methoxy-1-naphthalenesulfonyl chloride, 113699-68-6; 8-methoxy-1-naphthalenesulfonyl chloride, 56875-58-2; 5-methyl-2,4-thiazolidinedione, 3805-23-0; 5-[(5-bromo-6-methoxy-2-naphthalenyl)thio]-2,4-thiazolidinedione, 125518-80-1; 5-[(6-(benzyloxy)-2-naphthalenyl)thio]-2,4-thiazolidinedione, 125518-81-2; 2-naphthalenethiol, 91-60-1; 6-[(ethoxycarbonyl)oxy]-2-mercaptonaphthalene, 125518-82-3; 6,7-bis[(ethoxycarbonyl)oxy]-2-mercaptonaphthalene, 125518-83-4; 6-methoxy-2-mercapto-5-(trifluoromethyl)naphthalene, 125518-84-5.

Molecular Design, Synthesis, and Antiinflammatory Activity of a Series of β-Aminoxypropionic Acids[1]

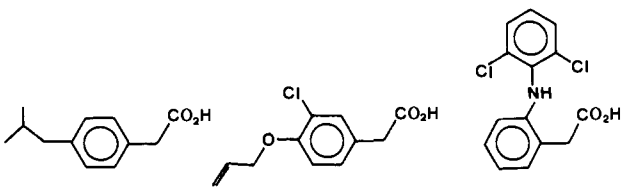
B. Macchia,*[†] A. Balsamo,[†] A. Lapucci,[†] F. Macchia,[‡] A. Martinelli,[†] S. Nencetti,[†] E. Orlandini,[†] M. Baldacci,[§] G. Mengozzi,[∥] G. Soldani,[∥] and P. Domiano[⊥]

Istituto di Chimica Farmaceutica, Istituto di Chimica Organica, and Laboratorio di Farmacologia Veterinaria, Universita' di Pisa, Pisa, Italy, Laboratori Baldacci, S.p.A., Pisa, Italy, and Centro di Studio per la Strutturistica Diffrattometrica del CNR, Istituto di Strutturistica Chimica, Universita' di Parma, Parma, Italy. *Received April 21, 1989*

Previous experimental and theoretical studies carried out on the mechanism of action of adrenergic drugs have shown that the (methyleneaminoxy)methyl moiety (C=$NOCH_2$, MAOMM) can be considered as a "bioisostere" of an aryl group (Ar). On this basis, a series of substituted β-aminoxypropionic acids (AOPAs) were synthesized as analogues of antiinflammatory arylacetic acids (ArAAs), in which the Ar portion is substituted by the MAOMM, with the aim of evaluating whether any antiinflammatory activity could be obtained from this class of drugs after the substitution of the Ar with the MAOMM. The antiinflammatory activity of the AOPAs synthesized was determined by carrageenan-induced rat paw edema, using diclofenac as the reference drug. The pharmacological data showed that most of the AOPAs examined exhibit a significant antiinflammatory activity, which in the case of the (*E*)-3-(benzylideneaminoxy)propionic acid (**7q**) is very close to that of the reference drug. Structural and theoretical studies were carried out in order to compare the conformation and the molecular reactivity of the AOPAs with those of the ArAAs. Pharmacological results showed that the ArAAs also generally exhibit an antiinflammatory activity after the substitution of the Ar with the MAOMM, thus supporting the hypothesis of a bioisosterelike relationship between these two moieties in this class of NSAIDs.

Nonsteroidal antiinflammatory drugs (NSAIDs) are a family of compounds which are chemically not very homogeneous. A large number of chemical structures have been found to exhibit antiinflammatory activity. Even if a general correlation between chemical features and biological activity is not found, drugs of this type can be subdivided into large classes of compounds with some general features in common.[2]

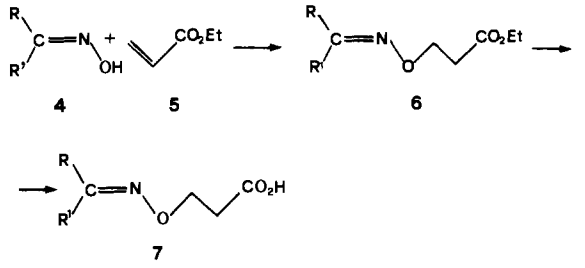
The arylacetic acid (ArAA) class has become one of the most widely developed and investigated over the past 2 decades. Many agents of this class, e.g. ibufenac (**1**), aclofenac (**2**), and diclofenac (**3**), are at present widely used in therapeutic practice.[2]



1 ibufenac **2** aclofenac **3** diclofenac

Previous experimental and theoretical studies[3] in the field of adrenergic drugs have indicated that, at least in the case of these type of drugs, the (methyleneaminoxy)-methyl moiety (C=$NOCH_2$, MAOMM) can be considered as a "bioisostere"[4] of either aryls (Ar) or other aromatic groups (see Figure 1).[3] These results suggested that it might be possible to effect the substitution of an Ar with

Scheme I



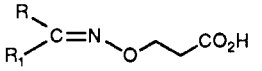
4 **5** **6** **7**

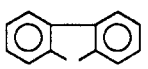
[†] Istituto di Chimica Farmaceutica.
[‡] Istituto di Chimica Organica.
[§] Laboratori Baldacci.
[∥] Laboratorio di Farmacologia Veterinaria.
[⊥] Istituto di Strutturistica Chimica.

(1) A preliminary account of this work was presented at the Joint Meeting on Medicinal Chemistry, Rimini, May 1985, Abstract T12.
(2) (a) Shen, T. Y. In *Burger's Medicinal Chemistry*; Wiley Interscience: New York, 1981; Chapter 62. (b) Cullen, E. *J. Pharm. Sci.* **1984**, *73*, 579. (c) Lombardino, J. G. In *Nonsteroidal Antiinflammatory Drugs*; Wiley-Interscience: New York, 1985, Chapter 4.
(3) Macchia, B.; Balsamo, A.; Lapucci, A.; Martinelli, A.; Macchia, F.; Breschi, M. C.; Fantoni, B.; Martinotti, E. *J. Med. Chem.* **1985**, *28*, 153 and references therein cited.
(4) (a) Thornber, C. W. *Chem. Soc. Rev.* **1979**, *8*, 563. (b) Burger, A. *A Guide to the Chemical Basis of the Drug Design*; Wiley-Interscience: New York, 1983; pp 28–84.
(5) Balsamo, A.; Broccali, G.; Lapucci, A.; Macchia, B.; Macchia, F.; Orlandini, E.; Rossello, A. *J. Med. Chem.* **1989**, *32*, 1398.
(6) Buchwald, P.; Domnariu, F. *Rev. Roum. Chim.* **1977**, *22*, 263.
(7) Gilon, C.; Knobler, Y.; Sheradsky, T. *Tetrahedron* **1967**, *23*, 4441.

0022-2623/90/1833-1423$02.50/0

Table I. Chemical and Pharmacological Data of β-Aminoxypropionic Acid Derivatives **7a–t**

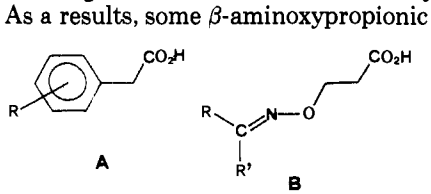


compd	R	R^1	mp or bp (mmHg), °C	recrystn solvent[a]	% yield[b]	formula[c]	inhibn activity on carrageenan paw edema[d] dose, mg/kg ip	inhibn activity on carrageenan paw edema[d] % inhibn at 4 h
7a	Me	Me	104–105 (j)[e]		70		50	21 (10)
							100	35* (10)
7b	c-C_3H_5	Me	oil	*f*	67	$C_8H_{13}NO_3$	50	16 (10)
7c	c-C_4H_8		60–62[e]	A	72		50	16 (10)
7d	*i*-Pr	Et	120–122 (0.8)		75	$C_9H_{17}NO_3$	25	21 (10)
							50	35* (20)
							100	27* (20)
7e	c-C_5H_{10}		38–39[e]	A	73		50	24* (10)
							100	53* (10)
7f	*n*-Pr	*n*-Pr	120–121 (0.3)[e]		81		50	0 (10)
7g	*i*-Pr	*i*-Pr	115–118 (0.3)		52	$C_{10}H_{19}NO_3$	50	12 (10)
7h			170–175 (2.0)		55	$C_{10}H_{15}NO_3$	50	16 (10)
7i	c-C_6H_{11}	Me	oil	*f*	55	$C_{11}H_{19}NO_3$	50	0 (10)
7j			oil	*f*	64	$C_{13}H_{19}NO_3$	50	19 (10)
7k	Et_2CH	H	oil	*f*	40	$C_9H_{17}NO_3$	50	0 (10)
7l	c-C_6H_{11}	H	70–73 (0.3)		61	$C_{10}H_{17}NO_3$	50	0 (10)
7m	Ph	Me	70–71[g]	B	82		25	23 (10)
							50	40* (20)
							100	31* (20)
7n	Ph	Et	33–35	A	65	$C_{12}H_{15}NO_3$	25	19 (10)
							50	28* (20)
							100	20 (20)
7o	Ph	Ph	104–105[h]	C	36		50	11 (10)
7p			127–129[e]	D	68		50	0 (10)
7q	Ph	H	66–68[e]	B	84		25	15 (10)
							50	66* (10)
7r	H	Ph	161–163	E	42	$C_{10}H_{11}NO_3$	50	0 (10)
7s		H	181–183	F	8	$C_8H_9NO_4$	50	15 (10)
7t		H	164–165	C	60	$C_8H_9NO_3S$	50	0 (10)
diclofenac (3)							25	31 (10)
							50	67* (10)

[a] A = petroleum ether; B = hexane; C = Et_2O; D = $CHCl_3$–petroleum ether; E = Et_2O–petroleum ether; F = AcOEt. [b] No efforts were made to optimize yields. [c] Anal. C, H, N. [d] A Student's *t* test was carried out. Results marked with an asterisk are percentage reductions which are significant (* = $p < 0.05$). The number of experiments is shown in parentheses. [e] Compound previously described by us as acylating agent for the synthesis of new β-lactam antibiotics (see ref 5). [f] Purified by column chromatography on silica gel: 9:1 petroleum ether–AcOEt (**7b**); 4:1 hexane–acetone (7i); 1:1 AcOEt–MeOH (**7j**); 1:1 AcOEt–hexane (**7k**). [g] Literature[6] mp 73–74 °C (AcOH–H_2O). [h] Literature[7] mp 106 °C (AcOH).

the MAOMM in drugs other than adrenergics, in which the Ar group seems to be a prerequisite for eliciting biological activity.

The group of antiinflammatory ArAAs (A) appeared to offer a promising substrate on which to verify our hypothesis. As a results, some β-aminoxypropionic acids (B,



AOPAs) were synthesized as analogues of the ArAAs (A), in which the Ar is substituted by the MAOMM. The choice of the substituents R and R^1 on the methyleneaminoxy portion of the new acids of type B was made in such a way as to have completely aliphatic, cycloaliphatic, aliphatic hydrogen, aromatic aliphatic, aromatic hydrogen substituted, and completely aromatic structures.

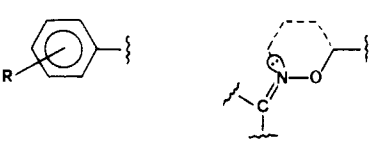


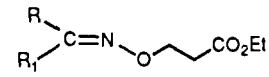
Figure 1. Representation of the bioisosterism between Ar and MAOMM.

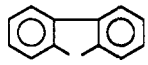
Chemistry

AOPAs **7** (Table I) were prepared as outlined in Scheme I, by Michael-type reaction of the appropriate oxime **4** with ethyl acrylate (**5**), followed by alkaline hydrolysis of the ethyl esters **6** (Table II). The structures of compounds **6** and **7** were established on the basis of their spectral data.

The geometry of the β-methyleneaminoxy group of compounds **6** and **7**, in cases where the cis–trans isomerism is possible, was assumed on the basis of the known configuration of the starting oximes **4**. The configuration around the C=N double bond of oximes **4** should remain

Table II. Chemical Data of Ethyl β-Aminoxypropionate Derivatives 6a–t



compd	R	R¹	mp or bp (mmHg), °C	% yield[a]	formula[b]
6a	Me	Me	66–70 (1.5)[c]	40	
6b	c-C_3H_5	Me	oil[d]	60	$C_{10}H_{17}NO_3$
6c	c-C_4H_8		102–105 (1.5)[c]	38	
6d	i-Pr	Et	oil[d]	48	$C_{11}H_{21}NO_3$
6e	c-C_5H_{10}		120–124 (1.2)[c]	41	
6f	n-Pr	n-Pr	75–78 (2.0)[c]	48	
6g	i-Pr	i-Pr	90–94 (2.5)	52	$C_{12}H_{23}NO_3$
6h			117–121 (0.2)	47	$C_{12}H_{19}NO_3$
6i	c-C_6H_{11}	Me	oil[d]	51	$C_{13}H_{23}NO_3$
6j			oil[d]	46	$C_{15}H_{23}NO_3$
6k	Et_2CH	H	oil[d]	41	$C_{11}H_{21}NO_3$
6l	c-C_6H_{11}	H	oil[d]	47	$C_{12}H_{21}NO_3$
6m	Ph	Me	122–126 (2.5)[e]	60	
6n	Ph	Et	91–94 (0.15)	75	$C_{14}H_{19}NO_3$
6o	Ph	Ph	oil[d]	54	$C_{18}H_{19}NO_3$
6p			198–200[c,f]	30	
6q	Ph	H	79–80 (0.5)[c]	45	
6r	H	Ph	oil[d]	54	$C_{12}H_{15}NO_3$
6s		H	oil[d]	32	$C_{10}H_{13}NO_4$
6t		H	oil[d]	34	$C_{10}H_{13}NO_3S$

[a] No efforts were made to optimize yields. [b] Anal. C, H, N. [c] Compound previously described by us (see ref 5). [d] Purified by column chromatography on silica gel: 9:1 petroleum ether–AcOEt (**6b**, **6j**, **6k**, **6o**); 7:3 petroleum ether/AcOEt (**6d**, **6l**, **6l**); 3:1 petroleum ether–AcOEt (**6r**); 3:2 AcOEt–benzene (**6s**, **6t**). [e] Literature[6] bp 118–120 °C (3). [f] Crystallized from $CHCl_3$–hexane.

unmodified in the nucleophilic addition of **4** to the double bond of ethyl acrylate (**5**), leading to **6**. Oximes **4** have been proved to be configurationally stable under the reaction conditions which lead from **4** to **6** and from **6** to **7**.

Pharmacology

Results. The antiinflammatory activity of AOPAs **7a–t** (see Table I) was evaluated by the method of carrageenan-induced paw edema described in the Experimental Section. Diclofenac was used as the reference drug. All compounds were administered intraperitoneally (ip) at a dose of 50 mg/kg. The compounds showing 50% or more inhibition were tested at 25 mg/kg. The compounds showing 20–50% inhibition were further tested at a dose of 100 mg/kg; for the compounds that showed no increase in activity at 100 mg/kg, a full dose–response curve starting from 25 mg/kg was also made.

As shown in Table I, some of the compounds tested revealed significant antiinflammatory activity at the dose of 50 mg/kg ip. In particular, the highest antiinflammatory activity was exhibited by the benzylidene derivative **7q**, which prevented the formation of paw edema by 15% at 25 mg/kg and by 66% at 50 mg/kg; the approximate ID_{50} was in this case 40 mg/kg. In the same experimental conditions, the reference compound diclofenac (**3**) inhibited intraplantar edema with an approximate ID_{50} (35 mg/kg) close to that of the benzylidene derivative **7q**. The aliphatic compounds **7a** and **7e** exerted a dose-dependent antiinflammatory activity at both 50 (21% and 24%, respectively) and 100 mg/kg (35% and 53%). Compounds **7d**, **7m**, and **7n** showed dose-dependent antiinflammatory effects at the doses of 25 (21%, 23, and 19% respectively) and 50 mg/kg (35%, 40%, and 28%), while the dose of 100 mg/kg did not lead to any enhancement of the activity. At 50 mg/kg compounds **7b**, **7c**, **7g**, **7h**, **7j**, **7o**, and **7s** showed insignificant antiinflammatory effects that ranged between 10% and 20%, whereas compounds **7f**, **7i**, **7k**, **7l**, **7p**, **7r**, and **7t** were completely inactive.

Discussion. The newly synthesized β-aminoxypropionic acids **7** can be considered, from the structural point of view, as oxime ethers and can be subdivided into different groups, depending on whether they derive from oximes of aliphatic (**7a–j**) or aromatic (**7m–p**) ketones or aliphatic (**7k,l**), aromatic (**7q,r**), or heteroaromatic (**7s,t**) aldehydes.

An analysis of the pharmacological data shown in Table I reveals that all the groups contain one or more compounds with a significant antiinflammatory activity, except those of the oxime ethers of aliphatic (**7k,l**) and heteroaromatic (**7s,t**) aldehydes. The highest degree of activity is found in acid **7q** (a benzaldehyde derivative), in **7m** and **7n** (which derive from acetophenone and propiophenone), and in **7a** and **7d** (acetone and ethyl isopropyl ketone derivatives, respectively).

The fact that several AOPAs present an appreciable antiinflammatory activity, in spite of the considerable structural differences at the level of the R and R^1 substituents, makes it difficult to determine a precise structure–activity relationship for these compounds. However, among those derivatives that are structurally comparable, some significant, characteristic trend may be detected by a comparative examination of the values of their pharmacological activity with the type of substituent linked to the amino carbon.

Compounds **7m–o** and **7q** represent a homogeneous series in which R is a phenyl and R^1 varies from an atom of hydrogen (**7q**) to a phenyl group (**7o**), passing through a methyl (**7m**) and an ethyl (**7n**) group. In these compounds,

there is a gradual decrease in the activity in proportion to the increase in the dimensions of the R^1 substituent. An analogous behavior is found in the oxime ethers of aliphatic methyl ketones **7a**, **7b**, and **7i**, in which R^1 is a methyl and R is a methyl (**7a**), a cyclopropyl (**7b**), or a cyclohexyl (**7i**), and the activity decreases until it finally disappears completely, passing from **7a** to **7b** and then to the cyclohexyl derivative **7i** (the cyclohexyl group is also present in the derivative **7l**, which is likewise inactive). In the case of the two AOPAs **7d** and **7g**, where R is an isopropyl group and R^1 is an ethyl or an isopropyl, respectively, the more active derivative is the one with the less bulky substituent, i.e. **7d**.

As regards the cyclic ketone derivatives **7c**, **7e**, **7h**, **7j**, and **7p**, the most active compound proved to be the cyclohexane derivative **7e**; the activity decreases or disappears both in the inferior homologue **7c** and in the other compounds **7h**, **7j**, and **7p**, which are structurally more complex.

A comparison of the activity of **7q** with that of the heteroaromatic aldehyde derivatives **7s** and **7t** indicates that the substitution of the phenyl with an aromatic heterocycle, such as furane or thiophene, leads to a drastic reduction or to the disappearance of the activity.

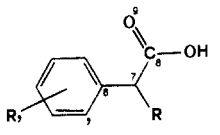
Furthermore, it is interesting to note that the inversion of the configuration around the methyleneaminoxy double bond of the (*E*)-benzaldoxime derivative **7q** leads to the *Z* isomer (**7r**), which is totally devoid of any antiinflammatory activity.

Taken together, these pharmacological results indicate that ArAAs also generally exhibit an antiinflammatory activity after the substitution of the Ar with the MAOMM. It would thus appear to also be possible to speak of the existence of a bioisosterism between these two molecular portions in the class of arylacetic nonsteroidal antiinflammatory drugs (ArAAs).

Structural and Theoretical Studies

Today it is commonly accepted that the antiinflammatory action of NSAIDs involves the inhibition of the conversion of arachidonic acid to prostaglandins, which are the mediators of the inflammatory processes.[2a] Several hypotheses have been advanced in order to describe the mechanistic action of NSAIDs at a molecular level and several steric models have been proposed in order to describe and rationalize their interaction at the receptor.[8]

Starting from the results of experimental and theoretical studies[8] carried out on series of arylalkanoic NSAIDs (C),[9]



C

which showed that all active compounds possess low-energy conformations with values of C(1)–C(6)–C(7)–C(8) (τ_1) and C(6)–C(7)–C(8)–O(2) (τ_2) torsion angles near to 90°,[10] the importance of a suitable spatial relationship between the carboxylic group and the phenyl ring for antiinflammatory activity[8] was established.

In the light of these hypotheses, it appeared to be of interest to investigate, by means of structural and theoretical studies, the possible existence of analogies at the molecular level between the new antiinflammatory AOPAs (B) and the antiinflammatory arylacetic drugs of type A, in order to see whether support could be found for the hypothesis that the MAOMM may be a bioisostere of an Ar also in type A drugs. X-ray crystallographic analysis was performed on one of the new antiinflammatory acids **7**. Acid **7c** was selected because it gave crystals suitable for X-ray structural studies. An ORTEP plot of the X-ray structure of **7c**, showing the atom-numbering scheme, is given in Figure 2. Relevant structural parameters are shown in Table III.

As far as distances and angles are concerned, the values for compound **7c** (see Table III) are in the range of those found in the literature for similar compounds.[11] The

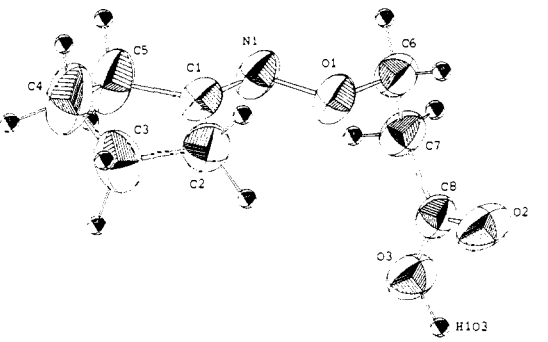


Figure 2. ORTEP plot (50% probability level for thermal ellipsoids) of the molecule showing the atom-numbering scheme.

Table III. Distances (Å), Angles (deg), and Torsion Angles (deg) for Non-Hydrogen Atoms of Compound **7c**[a]

C(1)–C(2)	1.4849 (31)
C(1)–C(5)	1.5072 (31)
C(1)–N(1)	1.2709 (32)
C(2)–C(3)	1.5158 (41)
C(3)–C(4)	1.4776 (42)
C(4)–C(5)	1.4870 (59)
N(1)–O(1)	1.4297 (22)
O(1)–C(6)	1.4244 (34)
C(6)–C(7)	1.5142 (36)
C(7)–C(8)	1.4969 (26)
C(8)–O(2)	1.2297 (26)
C(8)–O(3)	1.3042 (31)
C(5)–C(1)–N(1)	121.39 (21)
C(2)–C(1)–N(1)	128.81 (21)
C(2)–C(1)–C(5)	109.80 (21)
C(1)–C(2)–C(3)	104.93 (19)
C(2)–C(3)–C(4)	106.18 (26)
C(3)–C(4)–C(5)	107.24 (33)
C(1)–C(5)–C(4)	103.69 (27)
C(1)–N(1)–O(1)	109.94 (20)
N(1)–O(1)–C(6)	108.61 (20)
O(1)–C(6)–C(7)	111.70 (20)
C(6)–C(7)–C(8)	113.38 (20)
C(7)–C(8)–O(3)	114.79 (19)
C(7)–C(8)–O(2)	122.73 (21)
O(2)–C(8)–O(3)	122.47 (21)
C(5)–C(1)–N(1)–O(1)	178.94 (22)
C(2)–C(1)–N(1)–O(1)	-1.21 (34)
N(1)–C(1)–C(5)–C(4)	-164.64 (27)
C(2)–C(1)–C(5)–C(4)	15.49 (33)
C(5)–C(1)–C(2)–C(3)	1.57 (29)
N(1)–C(1)–C(2)–C(3)	-178.29 (26)
C(1)–C(2)–C(3)–C(4)	-18.34 (33)
C(2)–C(3)–C(4)–C(5)	28.82 (39)
C(3)–C(4)–C(5)–C(1)	-27.07 (38)
C(1)–N(1)–O(1)–C(6)	176.67 (20)
N(1)–O(1)–C(6)–C(7)	-78.62 (24)
O(1)–C(6)–C(7)–C(8)	-70.06 (26)
C(6)–C(7)–C(8)–O(2)	-27.74 (32)
C(6)–C(7)–C(8)–O(3)	152.92 (21)

[a] Esd's are in parentheses.

(8) Gund, P.; Jensen, N. P. In *Quantitative Structure–Activity Relationships of Drugs*; Academic Press: New York, 1983; Chapter 7.

(9) The numbering scheme of C was chosen in analogy with that used in the crystallographic study of **7c**.

(10) Dive, G.; Lapiere, C. L.; Leroy, G. *Bull. Soc. Chim. Belg.* **1977**, *86*, 73.

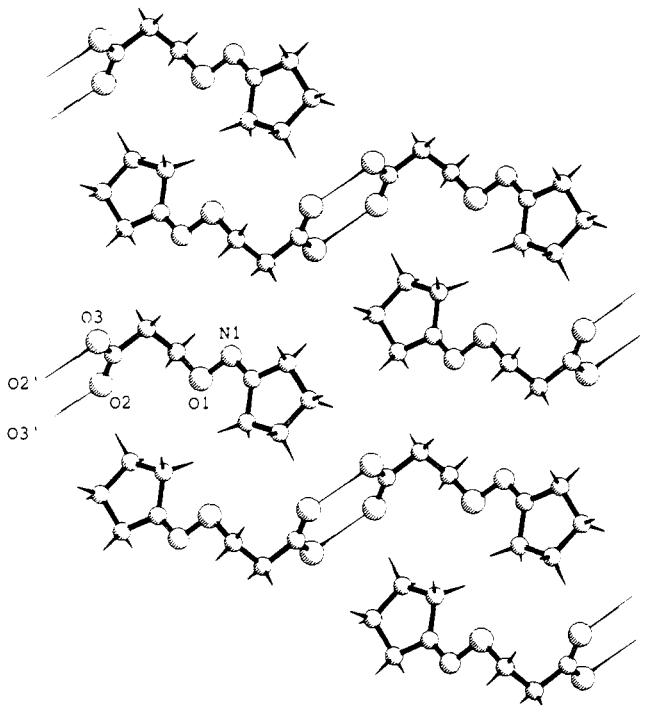


Figure 3. Perspective view of the unit cell showing the hydrogen bonding.

cyclopentane ring adopts an envelope form in which the C(4) atom is displaced by 0.415 Å from the plane through the other four atoms, with a mean C–C bond length of 1.494 (36) Å and a mean C–C–C angle of 106.37 (26)°. The double bond C(1)–N(1) = 1.271 (3) Å compares well with the mean value of 1.274 (8) Å reported by Jones et al.[12] for 20 oximes recorded in the Cambridge Structural Database. The two bonds involving the O(1) atom are comparable in length, whereas the C(7)–C(8) bond is significantly shorter than the equivalent C(6)–C(7) one, in agreement with the values reported[11] for analogous types of bonds. Two molecules form centrosymmetric dimers through the hydrogens of the carboxylic groups, as can be seen from the PLUTO drawing of the cell (see Figure 3) and the bond distances $O(2)–O(3)^i$ and $O(3)–O(2)^i$ equal to 2.672 (3) Å, where $i = (-x, 1 - y, 1 - z)$.

The planarity analysis for compound **7c** shows that this molecule can be considered to be composed of two planar fragments C(2)–C(1)–N(1)–O(1)–C(6) and C(6)–C(7)–C(8), almost at right angles to each other; the angle between the two planes C(2)–C(1)–N(1)–O(1)–C(6) and C(6)–C(7)–C(8) is 75.84° and the torsion angle N(1)–O(1)–C(6)–C(7) is −78.2(2)°.[13] There are no unusual short contacts.

The results of this structural analysis confirm the structures assigned to acids **7** and consequently also those of esters **6**.

The conformational data obtained for **7c** by the X-ray analysis were to be compared with the analogous conformational data of NSAIDs possessing an arylacetic structure A (i.e. C, in which R = H). Cambridge Structural Database

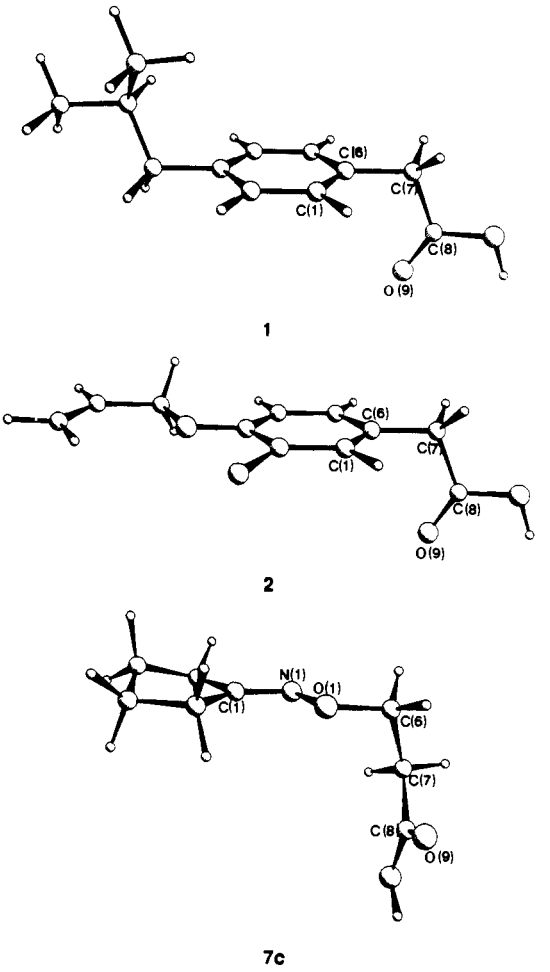


Figure 4. PLUTO plot of the preferred conformation of ibufenac (1) and aclofenac (2) and of the solid-state conformation of the compound **7c**.

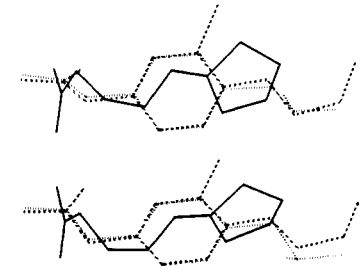
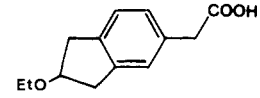


Figure 5. Stereo overlap mapping of the structures of ibufenac (dotted line), aclofenac (dashed line), and **7c** (solid line).

reports only a few X-ray data of compounds possessing such a structure; none of these compounds, however, is of any interest from the therapeutic point of view; an example is offered by (2-ethoxy-5-indanyl)acetic acid (8), which possesses a slight antiinflammatory activity.[15]



8

On the other hand, complete sets of geometrical parameters were not available in the literature for the ArAAs studied by means of theoretical calculations. Consequently, the preferred conformations of two antiinflammatory ArAAs largely used in therapy, ibufenac (1) and

(11) Allen, F. H.; Kennard, O.; Watson, D.; Brammer, L.; Orpen, G.; Taylor, R. *J. Chem. Soc., Perkin Trans. 2* **1987**, S1.
(12) Jones, P. G.; Edwards, M. R.; Kirby, A. J. *Acta Crystallogr. Sect. C* **1986**, *C42*, 1222.
(13) The planar arrangement found for the C(1)–N(1)–O(1)–C(6) portion of **7c** is in agreement with findings of X-ray studies[14] and theoretical calculations,[3] for the same group in β-adrenergic drugs in which the MAOMM is present.
(14) (a) Carpy, A.; Gadret, M.; Leger, J. M.; Wermuth, C. G.; Leclerc, G. *Acta Crystallogr. Sect. B* **1979**, *B35*, 1144. (b) *Ibid.* **1980**, *B36*, 1715.
(15) Hata, T.; Sato, S.; Tamura, C. *Acta Crystallogr. Sect. C* **1986**, *C42*, 449.

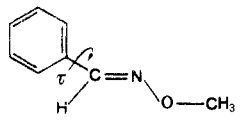
aclofenac (2), were determined by using the MMPMI program, which is a version of the Allinger's molecular mechanics method.[16] The conformations obtained for 1 and 2 are shown in Figure 4, together with the solid-state conformation of **7c**.

Drugs 1 and 2 possess very similar preferred conformations: the values of the C(1)–C(6)–C(7)–C(8) (τ_1) and C(6)–C(7)–C(8)–O(2) (τ_2) torsion angles are −83.8° and −11.0° for 1 and −79.0° and −11.2° for 2, respectively. Values of τ_1 show that in these conformations the carboxylic group of 1 and 2 lies outside the aromatic ring plane in a manner very similar to that that had been found for low-energy conformations of type C antiinflammatory arylalkanoic acids (see above). Furthermore these conformations almost correspond to the one found for compound 8 in the solid state (τ_1 = −81.2° and τ_2 = 25.7°).

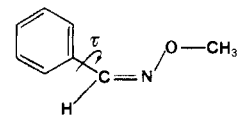
The conformational study of compound **7c** showed that the MAOMM is planar like the Ar of the ArAAs, which is the molecular moiety that the MAOMM would simulate. A suitable superimposition of the structure of **7c**, obtained by X-ray crystallographic analysis, with those of 1 and 2, obtained by molecular mechanical calculations, also shows that the spatial relationship between the C(8) carbon of the carboxylic group and the planar C=NOC portion of compound **7c** is not substantially different from that found for the same carbon atom and the aryl plane in 1 and 2, as can be observed in Figure 5.[17]

The results of this conformational comparison between AOPA **7c** and the two ArAAs 1 and 2 would appear to support the existence of a bioisosterism between the MAOMM of AOPAs and the Ar of the ArAAs, at least from a steric point of view.

In order to verify whether the different antiinflammatory activity of (*E*)-benzaldoxime derivative **7q** (the most active compound of the series) and of its *Z* isomer **7r** (completely inactive), could be explained in conformational terms, the two model compounds of **7q** (**9a**) and of **7r** (**9b**)



9a



9b

were studied. In these two model compounds, a hydrogen atom replaces the CH_2COOH portion of **7q** and **7r**; as in previous studies,[3] this simplification was made in order to reduce the computation time. The torsion angle τ was optimized at the SCF-STO3G "ab initio" level, by fixing the geometry of the MAOMM as in **7c** and using standard values for the phenyl-ring geometry.

E compound **9a** shows the lowest energy conformation when τ = 0° (i.e. when the phenyl ring is coplanar with the MAOMM), while for *Z* compound **9b**, this occurs when τ = 90° (i.e. when the phenyl ring is perpendicular to the MAOMM).[18]

In order to compare the reactivity of the MAOMM of AOPAs (B) with that of the Ar of the ArAAs (A) at the molecular level, the electrostatic molecular potential (EMP) of **7c**, 1, and 2 was considered. The EMP maps, shown in Figure 6, were calculated in a plane parallel to that of the MAOMM of **7c** or by the Ar of 1 and 2, at a distance of 1.7 Å from these planes,[3] on the other side with respect to the carboxylic group; the wave functions of 1,

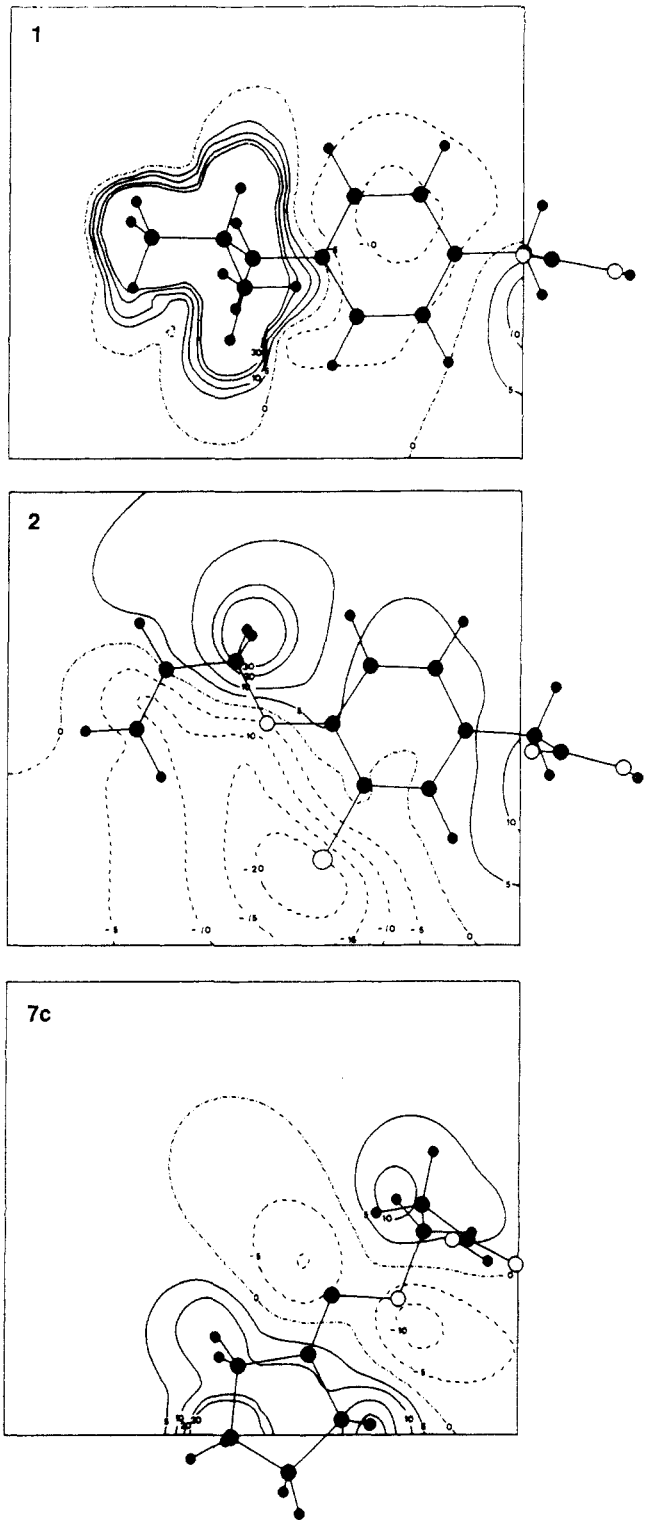


Figure 6. EMP contours maps of ibufenac (1), aclofenac (2), and **7c** on the plane parallel to those of the phenyl ring of ibufenac and aclofenac and to that of the MAOMM of **7c**, at a distance of 1.7 Å. Isopotential levels are in kcal mol^{-1}. The dashed lines represent negative potentials.

(16) Standard (see, for example: Allinger, N. L. In *Advances in Physical Organic Chemistry*; Academic Press: New York, 1976; Vol. 13, Chapter 1.) or MMPMI-generalized force field parameters were used except for the chlorine atom (compound 2) for which previously reported parameters were used (Squier, G. J.; van der Schyf, C. J.; Venter, D. P.; Oliver, D. W.; van Rooyen, P. H.; Dillen, J. L. M. *Eur. J. Med. Chem.* **1986**, *21*, 213.).

(17) It should be noted, however, that the C(7) atom of **7c** does not coincide with the C(7) atoms of 1 and 2, because in 1 and 2 this atom lies on the ring plane, while in **7c** it lies outside the MAOMM plane.

(18) The X-ray geometries of the *E* and *Z* isomers of *p*-Chlorobenzaldoxime (in which, however, there is an oximic hydroxyl group with a reactivity very different from that of the methoxy group of **9a** and **9b**) do not display a marked difference in their τ values (9.4° and 21.2° for *E* and *Z* isomers, respectively).[19]

2, and 7c were calculated at the SCF-STO3G "ab initio" level, considering the geometries of the preferred conformations obtained by X-ray analysis for 7c and MMPMI calculations for 1 and 2.

An analysis of the general trend of the EMP shows that over the planar C=NOC moiety of 7c there is a negative region with two close minima in which the EMP value is less than −10 kcal mol^{-1}, approximately situated above that of the heteroatoms of the MAOMM; this negative region decidedly separates the positive EMP regions generated by the cyclopentanic portion and by the C(7) atom, respectively. The EMP trend of compound 1 is not very different on the phenyl ring there is a negative region with an extension similar to that of the negative region of 7c and a minimum value less than −10 kcal mol^{-1}; this EMP region separates the positive regions generated by the isobutyl portion and by the C(7) atom, respectively. Thus, results show that the EMP trend on the MAOMM of 7c and on the Ar of 1 are similar; this fact tends to indicate an analogous reactivity of these spatially corresponding molecular portions.[20]

The EMP trend of compound 2 is very different from that of both 1 and 7c; the electronic characteristics of the two substituents on the phenyl ring of 2 make the EMP positive over the ring, while a region with a very negative EMP is generated outside the ring.

The fact that the antiinflammatory ArAA 2 presents an EMP trend over the planar region rather different from those of the other two antiinflammatory agents 1 and 7c seems to indicate that EMP analysis of this region is not able to account for the common antiinflammatory activity of these drugs.

Conclusions

On the basis of hypotheses previously advanced about the possibility of the existence of a bioisosterism between an aryl group (Ar) and an (metyleneaminoxy)methyl moiety (MAOMM), a series of β-aminoxypropionic acids (AOPAs) were synthesized as analogues of antiinflammatory arylacetic acids (ArAAs) in which the Ar is substituted by the MAOMM. The pharmacological data showed that most of the AOPAs examined possess a significant antiinflammatory activity which in the case of the drug 7q is very close to that of the reference drug diclofenac (3).

The fact that these AOPAs exhibit an antiinflammatory activity supports the working hypothesis about the bioisosterism between the Ar and the MAOMM, showing that this bioisosterism also exists in this case.

The results of conformational studies would appear to confirm the existence of this bioisosterism, at least from a steric point of view.

An analysis of the EMP trend calculated for one of the AOPAs synthesized (7c) and for ibufenac (1), one of the most widely used ArAAs, shows that the MAOMM of 7c and the Ar of 1 possess a similar reactivity. However, the fact that another widely used antiinflammatory ArAA, aclofenac (2), presents an EMP trend rather different from those of 1 and 7c seems to indicate that the EMP analysis is not able to account for the common antiinflammatory activity of these drugs.

Experimental Section

Melting points were determined on a Kofler hot-stage apparatus and are uncorrected. IR spectra for comparison of compounds were taken as paraffin oil mulls or as liquid films, on a Perkin-Elmer Model 1310 instrument. ^{1}H NMR spectra were obtained with a Varian EM 360 A instrument in a ca. 10% $CDCl_3$ solution using Me_4Si as the internal standard. The proton magnetic resonance assignments were established on the basis of the expected chemical shifts and the multiplicity of the signals. *E* oximes of symmetric and asymmetric carbonyl compounds (**4a**,**c**,**e**,**f**,**g**,**j**,**o**,**p** and **4b**,**d**,**h**,**i**,**k**,**l**,**m**,**n**,**q**,**s**,t, respectively) and (*Z*)-benzaldoxime (**4r**) were prepared by the usual methods and their physical constants were in accordance with those reported in literature.

Analytical TLCs were carried out on 0.25-mm layer silica gel plates (Merck F_{254}) containing a fluorescent indicator; spots were detected under UV light (254 nm), in the case of completely or partially aromatic compounds, or by spraying with 0.2 M $K_2Cr_2O_7$ in 40% aqueous sulfuric acid followed by gentle heating, in the case of completely aliphatic compounds. Column chromatographies were performed by the flash technique, using 230–400 mesh silica gel. Petroleum ether refers to the fraction boiling at 40–60 °C. Magnesium sulfate was always used as the drying agent. Evaporations were made in vacuo (rotating evaporator). Elemental analyses were performed by our analytical laboratory and agreed with the theoretical values to within ±0.4%.

Ethyl β-Aminoxypropionate Derivatives 6a–t. A solution of the appropriate oxime (**4a**–t, 0.5 mol) in anhydrous EtOH (250 mL) was treated with ethyl acrylate (**5**, 0.42 mol) and then with a solution of 2 N KOH in anhydrous EtOH (42 mL). The resulting mixture was stirred at 35 °C for 48 h (60 °C for 72 h in the case of **4r**) and then evaporated. The residue was dissolved in Et_2O, washed (aqueous 10% NaOH and H_2O), dried, filtered, and evaporated to dryness to yield crude ester **6**, which was purified by the appropriate procedure (see Table II).

The ^{1}H NMR spectra of these esters exhibit two triplets whose middle points range from 2.56 to 2.98 ppm for the CH_2COO and from 4.16 to 4.67 ppm for the $NOCH_2$ protons. For other physical and microanalytical data (see Table II).

The configurational stability of the oximes of asymmetric carbonyl compounds was tested by treating the appropriate oxime 4 with KOH in anhydrous EtOH under the above-described conditions. After 48 h at 35 °C (70 h at 60 °C in the case of **4r**), the solution was evaporated and the residue was dissolved in H_2O, acidified to pH 4–5 with 20% aqueous H_3PO_4, and then extracted with Et_2O. Evaporation of the washed (H_2O), dried, and filtered extracts yieled the practically pure starting oxime 4.

β-Aminoxypropionic Acid Derivatives 7a–t. Each of the esters **6** (0.05 mol) was added to a solution of KOH (0.06 mol) in EtOH (30 mL). After 48 h at room temperature, the solvent was evaporated and the residue was taken up with H_2O. The aqueous phase was washed with Et_2O, acidified to pH 4–5 with 20% aqueous H_3PO_4, and extracted with Et_2O. Evaporation of the washed (H_2O), dried, and filtered extracts gave crude acid **7**, which was purified by the appropriate procedure (see Table I).

In the ^{1}H NMR spectra of **7a–t** there are two triplets attributed to the ethylenic portion of the propanoyl moiety [middle points of the signals: δ 2.65–2.93 (CH_2COO) and 4.21–4.46 ($NOCH_2$)]. For other physical and microanalytical data, see Table I.

For the stability tests, the oximes of asymmetric carbonyl compounds 4 were treated, under the conditions described above for the preparation of 7, with KOH in 95% aqueous EtOH. The usual work-up made it possible to recover the unaltered starting oximes 4.

Crystallography. Crystals of **7c**, mp 62–63 °C, were obtained by slow evaporation of a CH_2Cl_2 solution containing hexane.

$C_{13}H_9NO_3$, M = 171.96, triclinic, space group $P\bar{1}$, a = 10.151 (2) Å, b 10.191 (2) Å, c = 4.929 (1) Å, α = 97.41 (2)°, β = 101.54 (2)°, γ = 111.57 (2)°, Z = 2, U = 452.98 (18) Å^3, D_c = 1.255 g·cm^{-3}, $F(000)$ = 184, λ(Cu-Kα) = 1.54178 Å, μ(Cu-Kα) = 7.63 cm^{-1}.

A crystal of the approximate size 0.10 × 0.10 × 0.24 mm was used to collect data with a Siemens AED diffractometer on line to a General Automation Jumbo 220 microcomputer.[21] The cell parameters and orientation matrix were obtained by least-squares refinement on 22 high-angle reflections. A total of 1880 independent reflections were measured with the ϑ – 2ϑ step scan (4°

(19) (a) Folting, K.; Lipscomb, W. N.; Jerslev, B. *Acta Crystallogr.* **1964**, *17*, 1263. (b) Jensen, K. G. *Acta Chem. Scand.* **1970**, *24*, 3293.

(20) A comparable reactivity of an MAOMM and a suitable substituted aromatic portion has also been found in the field of the β-adrenergic drugs.[3]

(21) Belletti, D.; Ugozzoli, F.; Cantoni, A.; Pasquinelli, G. *Gestione on line di diffrattometro a cristallo singolo SIEMENS AED con sistema General Automation Jumbo 220*, Internal Reports 1, 2, 3, 1979.

$\leq 2\vartheta \leq 140°$) and the five-point method, monitoring one standard reflection every 50. Only 1430 reflections with $I > 2\sigma(I)$ [$\sigma(I)$ based on counting statistics] were considered as observed and used in the analysis. Seven low order reflections (0,2,0/-1,2,0/1,0,1/-2,0,1/-1,1,1/1,-1,1/-1,-1,1) were discarded in the last cycle of refinement. Lorentz and polarization were applied but not absorption.

The structure was solved via direct methods and refined by full-matrix least squares[22] with anisotropic and isotropic thermal parameters for non-hydrogen and hydrogen atoms respectively. All the hydrogens were located from difference Fourier map; H(41), H(42), H(51), and H(52) were constrained to ride on C(4) and C(5), respectively, during the last cycle of refinement. Final $R = 0.0853$, $R_w = 0.0869$, $w = [(F)^2 + 0.062F^2]^{-1}$ for 1430 reflections. The figures were obtained by using ORTEP[23] and PLUTO[24] whereas geometric calculations were performed with the PARST program.[25] All the calculations were performed using CRYSRULER.[26]

Pharmacological Methods. Carrageenan-Induced Paw Edema. Ten or 20 female Wistar rats (200–250-g body weight) were used for each group. Hind paw volumes were measured using a water pletysmometer (Basile, Varese, Italy) according to the method described by Winter et al.[27] Then the compounds to be tested were administered intraperitoneally (ip) at a constant dose of 50 mg/kg, by using a solution in aqueous $NaHCO_3$ (pH 7.4) at a concentration of 10 mg/mL. Control rats received the same volume of the vehicle. Thirty minutes later, 0.05 mL of 1% solution of carrageenan (Sigma, St. Louis, MO) was subcutaneously injected into the plantar surface of the right hind paw. The increase in paw volume 4 h after the injection of the phlogistic agent was adopted as a measure of the effect, and results were expressed as a precent reduction of control edema. Compounds showing 50% or more of inhibition (**7q** and **3**) were further tested at 25 mg/kg following the procedure described above. Compounds showing 20–50% of inhibition (**7a**, **7d**, **7e**, **7m**, and **7n**) were tested at 100 mg/kg; for compounds (**7d**, **7m**, and **7n**) that showed no increase in activity at 100 mg/kg, a full dose–response curve starting from 25 mg/kg was also carried out. Approximate ID_{50}s were calculated by plotting the log of the dose of the compound versus inhibition of paw edema. Student's t test for grouped data was used for statistical evaluations. Differences were considered to be statistically significant when p was 0.05 or lower.

Acknowledgment. This work was supported in part by a grant from the Consiglio Nazionale delle Ricerche and the Ministero della Pubblica Istruzione.

Registry No. 1, 1553-60-2; **2**, 22131-79-9; **4a**, 127-06-0; **4b**, 80606-74-2; **4c**, 1192-28-5; **4d**, 49805-38-1; **4e**, 100-64-1; **4f**, 1188-63-2; **4g**, 1113-74-2; **4h**, 4576-48-1; **4l**, 80606-77-5; **4j**, 4500-12-3; **4k**, 49805-57-4; 4l, 30950-35-7; **4m**, 10341-75-0; **4n**, 23517-42-2; **4o**, 574-66-3; **4p**, 2157-52-0; **4q**, 622-31-1; **4r**, 622-32-2; **4s**, 620-03-1; 4t, 38266-87-4; **5**, 140-88-5; **6a**, 119881-14-0; **6b**, 125803-04-5; **6c**, 119881-16-2; **6d**, 125803-05-6; **6e**, 119881-17-3; **6f**, 125803-06-7; **6g**, 103586-50-1; **6h**, 125803-07-8; **6l**, 125803-08-9; **6j**, 125803-09-0; **6k**, 125803-10-3; **6**, 125803-11-4; **6m**, 103586-51-2; **6n**, 125803-12-5; **6o**, 125803-13-6; **6p**, 119881-19-5; **6q**, 103586-48-7; **6r**, 125803-14-7; **6s**, 125803-15-8; **6t**, 125803-16-9; **7a**, 103586-55-6; **7b**, 125803-17-0; **7c**, 103586-53-4; **7d**, 125803-18-1; **7e**, 103586-54-5; **7f**, 103586-56-7; **7g**, 103604-67-7; **7h**, 103586-57-8; **7i**, 125803-19-2; **7j**, 125803-20-5; **7k**, 125803-21-6; **7l**, 125803-22-7; **7m**, 103586-52-3; **7n**, 125803-23-8; **7o**, 15985-45-2; **7p**, 103586-60-3; **7q**, 103586-49-8; **7r**, 103586-70-5; **7s**, 103586-62-5; **7t**, 103586-69-2.

Supplementary Material Available: Table IV giving final atomic coordinates and isotropic B equivalent, Table V giving anisotropic and isotropic thermal parameters, and Table VII giving cartesian coordinates of compounds 1, **2**, and **7c** (3 pages); Table VI giving observed and calculated structure factor amplitudes (8 pages). Ordering information is given on any current masthead page.

(22) Sheldrick, G. M. *SHELX-76, Program for Crystal Structure Determination*; University of Cambridge: Cambridge, England, 1976.
(23) Johnson, C. K. *ORTEP II, Fortran Thermal Ellipsoids Plot Program for Crystal Structure Illustration*, Report ORNL-5138, Oak Ridge, 1976.
(24) Motherwell, W. D *PLUTO, A Program for Plotting Molecular and Crystal Structures*, University of Cambridge: Cambridge, England, 1978.
(25) Nardelli, M., PARST, A System of Fortran Routines for Calculating Molecular Structure Parameters from Results of Crystal Structure Analyses. *Comput. Chem.* **1983**, *7*, 95.
(26) Rizzoli, C.; Sangermano, V.; Calestani, G.; Andreetti, G. D. CRYSRULER: An Integrated System of Computer Programs for Crystal Structure Analysis on Personal Computer. *J. Appl. Crystallogr.* **1987**, *20*, 436.
(27) Winter, C. A.; Risley, E. A.; Nuss, G. W. *Proc. Soc. Exp. Biol. Med.* **1962**, *111*, 544.

Retinobenzoic Acids. 5. Retinoidal Activities of Compounds Having a Trimethylsilyl or Trimethylgermyl Group(s) in Human Promyelocytic Leukemia Cells HL-60

Takeru Yamakawa, Hiroyuki Kagechika, Emiko Kawachi, Yuichi Hashimoto, and Koichi Shudo*

Faculty of Pharmaceutical Sciences, University of Tokyo, 7-3-1 Hongo, Bunkyo-ku, Tokyo 113, Japan.
Received August 4, 1989; Revised Manuscript received November 15, 1989

The retinoidal activities of trimethylsilyl or trimethylgermyl-containing retinobenzoic acids are discussed on the basis of differentiation-inducing activity on human promyelocytic leukemia cells HL-60. Compounds with a trimethylsilyl or trimethylgermyl group at the meta position of the generic formula **2** have more potent activities than the corresponding retinobenzoic acids with a *m-tert*-butyl group. Compounds having two *m*-trimethylsilyl or -trimethylgermyl groups also have strong activities, and (*E*)-4-[3-[3,5-bis(trimethylsilyl)phenyl]-3-oxo-1-propenyl]benzoic acid (**22**, Ch55S) and (*E*)-4-[3-[3,5-bis(trimethylgermyl)phenyl]-3-oxo-1-propenyl]benzoic acid (**35**, Ch55G) are more active than retinoic acid by 1 order of magnitude. However, in the para-substituted chalcone derivatives, the replacement of a *tert*-butyl group (**49**, Ch40) with a trimethylsilyl (**27**, Ch40S) or a trimethylgermyl (**30**, Ch40G) group caused the disappearance of the activity.

Retinoids are defined as "substances that elicit specific biological responses (that is, the specific activities of retinoic acid) through binding to the specific receptor(s)".[1,2] Retinobenzoic acids are "a series of benzoic acid derivatives with potent retinoidal activities".[3] They modulate the cellular differentiation and proliferation in many types of cells in cases where retinoic acid (**1**, Chart I) acts as a modulator.[2,4] Their mechanism of action seems to be the same as that of retinoic acid,[5,6] and they also bind to the

(1) Sporn, M. B., Roberts, A. B., Goodman, D. S., Eds. *The Retinoids*; Academic Press, Inc.: Orlando, FL, 1984.
(2) Sporn, M. B.; Roberts, A. B.; Roche, N. S.; Kagechika, H.; Shudo, K. *J. Am. Acad. Dermatol.* **1986**, *15*, 756.
(3) Shudo, K.; Kagechika, H. *Chemistry and Biology of Synthetic Retinoids*; Dawson, M. I., Okamura, W. H., Eds.; CRC Press: Boca Raton, FL, in press.
(4) Jetten, A. M.; Anderson, K.; Deas, M. A.; Kagechika, H.; Lotan, R.; Rearick, J. I.; Shudo, K. *Cancer Res.* **1987**, *47*, 3523.

0022-2623/90/1833-1430$02.50/0