REACTION OF 4-AROYL-2(3*H*)-DIHYDROFURANONES WITH HYDROXYLAMINE HYDROCHLORIDE. SYNTHESIS AND PHARMACOLOGICAL STUDY OF A SERIES OF 3-ARYL-4,5--DIHYDRO-4-ISOXAZOLEACETIC ACIDS

Maria M. CURZU[a], Gérard A. PINNA[a], Giorgio CIGNARELLA[b], Daniela BARLOCCO[b] and Maria Piera DEMONTIS[c]

[a] *Istituto Chimica Farmaceutica, Università di Sassari, Via Muroni 23, 07100 Sassari, Italy*
[b] *Istituto Chimica Farmaceutica e Tossicologica, Università di Milano, Viale Abruzzi 42, 20131 Milano, Italy*
[c] *Istituto Farmacologia, Facoltà di Medicina e Chirurgia, Università di Sassari, 07100 Sassari, Italy*

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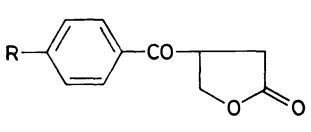
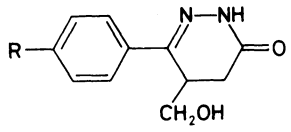
Dedicated to Dr Miroslav Protiva on the occasion of his 70th birthday.

We have synthesized and tested for their antiinflammatory activity a new series of 3-aryl-4,5-dihydro-4-isoxazoleacetic acids (*IVa—IVg*). Preliminary pharmacological results seem to indicate the 3-phenyl derivative *IVa* as the most interesting compound. In fact, when tested against carrageenin edema in Wistar rats, it shows antiinflammatory activity comparable to that of naproxene, taken as reference drug, though of shorter duration. All the substituted phenyl derivatives were less active (*IVd—IVf*) or inactive (*IVb, IVc, IVg*).

In a previous paper[1] we reported that the reaction between β-benzoyl-γ-butyrolactones (*I*) and hydrazine hydrate in ethanol led to 5-hydroxymethyl-6-substituted phenyl-4,5-dihydro-3(2*H*)pyridazinones (*II*) and to the corresponding dehydrated 5-methyl-6-substituted phenyl-3(2*H*)pyridazinones (*III*) when acetic acid was used as a solvent.



I



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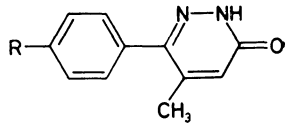


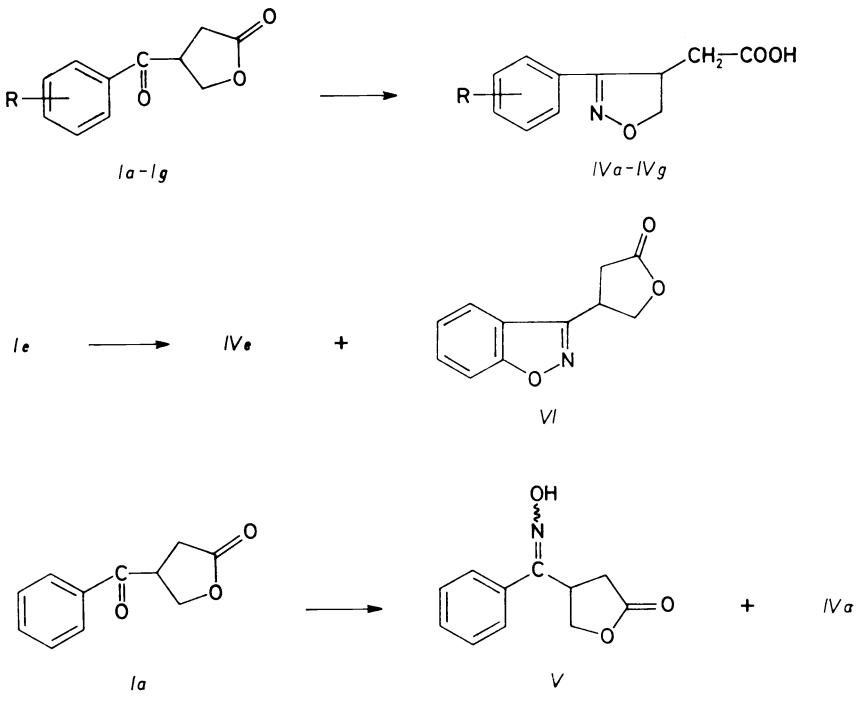
III

Continuing our interest in the chemistry of β-benzoyl-γ-butyrolactones (*I*) we have now extended our studies to the reaction between *I* and hydroxylamine hydro-

chloride. A preliminary experiment carried out by reacting *Ia* with excess hydroxylamine hydrochloride and ammonium acetate in 98% ethanol at reflux for 3·5 h led to the isolation of a $NaHCO_3$-soluble compound, identified as the unknown 3-phenyl-4,5-dihydro-4-isoxazoleacetic acid (*IVa*, 32% yield) and to an alkali-insoluble fraction, mainly constituted by a *syn* : *anti* mixture of oximes *V* (37% yield).

Unexpectedly, by carrying out the reaction in 60% aqueous ethanol the only product isolated (46% yield) was *Ia*. The extension of this procedure to a series of β-substituted benzoyl-γ-butyrolactones (*Ib*–*Ig*) led to the corresponding *IVb*–*IVg* in satisfactory yields.

Interesting to note, when starting from the *ortho*-hydroxy substituted lactone (*Ie*), besides *IVe* a second compound was obtained, which by analytical and spectral properties was identified as the unknown 3-[2-oxo-tetrahydrofuran-4-yl]benzoisoxazole (*VI*). (See Scheme 1). Compounds *IVa*–*IVg* have been tested for their antiinflammatory activity.



In formulae *I* and *IV*: *a*, R = H ; *b*, R = *p*-Cl ; *c*, R = *p*-CH_3 ; *d*, R = *p*-OCH_3 ; *e*, R = *o*-OH ; *f*, R = *p*-$NHCOCH_3$; *g*, R = *p*-OH

SCHEME 1

EXPERIMENTAL

Chemistry

Melting points were determined with a Büchi 510 capillary apparatus and are uncorrected. All elemental analyses (C, H, Cl, N) for the new substances were within ±0·4 of the theoretical value. IR spectra (Nujol mull) were recorded on a Perkin Elmer 297 infrared spectrophotometer. ^{1}H NMR spectra were recorded on a Varian XL 200 in $CDCl_3$ with tetramethylsilane as the internal standard. Chemical shifts are expressed in δ units. Yields refer to crystallized compounds.

4-(Substituted Benzoyl)-2(3*H*)-dihydrofuranones (*Ia*—*Ig*), General Method

According to a previously reported method[2], 37% formaldehyde solution (0·3 mol) is added to a stirred solution of the required 3-benzoylpropionic acid (0·27 mol) in 0·5M sodium hydroxide (0·3 mol). After 1 h at room temperaure, the mixture is acidified with hydrochloric acid (~30 ml), stirred for additional 12 h and then extracted with chloroform. After evaporation of the solvent, the residue is chromatographed on silica gel (Merck Silica gel 60, 70—230 mesh, eluent cyclohexane–ethylacetate 9 : 1) to give in the order the desired *I* and low amounts (<10%) of the corresponding 3-aroyl-3-butenoic acid. (See Table I).

TABLE I

Physical properties and spectral data of compounds *Ia*—*Ig*

Compound	Yield, %	M.p., °C (solvent)	IR, cm^{-1}	^{1}H NMR
Ia	80	63—65 (ether)	1 761 1 679	2·7—3·1 m, 2 H; 4·0—4·8 m, 3 H; 7·3—8·2 m, 5 H
Ib	61	87—89 (ether)	1 770 1 685	2·2—2·9 m, 2 H; 4·0—4·6 m, 3 H; 7·4 d, 2 H (J = 10); 7·5 d, 2 H (J = 10)
Ic	70	85—87 ethanol)	1 765 1 672	2·4 s, 3 H; 2·7—3·2 m, 2 H; 3·9—4·8 m, 3 H; 7·3—7·6 m, 4 H
Id	80	55—56 (ethanol)	1 720 1 660	2·9 m, 2 H; 3·9 s, 3 H; 4·3—4·7 m, 3 H; 6·9—7·3 m, 2 H; 7·8—8·1 m, 2 H
Ie	68	169—171 (ethanol)	3 250 1 740 1 670	2·6—2·9 m, 2 H; 4·0—4·6 m, 3 H; 6·7 d, 2 H (J = 9); 7·5 d, 1 H (J = 9); 9·5 s, 1 H
If	85	140—142 (ethanol)	3 300 1 770 1 670	2·6 s, 3 H; 3·3—3·7 m, 2 H; 4·7—5·2 m, 3 H; 8·0—8·3 m, 4 H; 10·5 br s, 1 H
Ig	65	50—52 (ethanol)	1 780 1 640	2·7—3·1 m, 2 H; 4·4—4·7 m, 3 H; 6·9—7·1 m, 2 H; 7·5—7·6 m, 2 H; 11·9 s, 1 H

3-Substituted Phenyl-4,5-dihydro-4-isoxazoleacetic Acids (*IVa—IVg*), General Method

A solution of the required *I* (0·01 mol), hydroxylamine hydrochloride (0·02 mol) and ammonium acetate (0·02 mol) in water (7 ml) and ethanol (10 ml) is refluxed for 3·5 h. After cooling, the ethanol is removed under vacuum, the solution acidified and extracted with chloroform. After evaporation of the chloroform, the residue is treated with 5% sodium bicarbonate and re-extracted with ether. The alcaline solution is then acidified and the solid which separates filtered and crystallized from a proper solvent. (See Table II).

Reaction of *Ia* with Hydroxylamine Hydrochloride

A suspension of *Ia* (2 g, 0·01 mol), hydroxylamine hydrochloride (1·46 g, 0·02 mol) and ammonium acetate (1·62 g, 0·02 mol) in 98% ethanol (10 ml) was refluxed for 3·5 h. After the above

TABLE II

Physical properties and spectral data of compounds *IVa—IVg*

Compound	Yield, %	M.p., °C (solvent)	IR, cm^{-1}	^1H NMR
IVa	46	115—117 (ethanol)	1 690	2·3—2·8 m, 2 H; 3·9—4·2 m, 1 H; 4·3—4·8 m, 2 H; 7·0—7·3 br s, 1 H; 7·3—7·8 m, 5 H
IVb	71	163—166 (ethanol)	1 690	2·4 dd, 1 H (J = 10); 2·6 dd, 1 H (J = 3); 4·2—4·6 m, 3 H; 7·4 d, 2 H (J = 8); 7·6 d, 2 H (J = 8)
IVc	65	123—124 (ethanol)	1 690	2·4 s, 3 H; 2·5 dd, 1 H (J = 10·8); 2·8 dd, 1 H (J = 3); 3·9—4·6 m, 3 H; 7·2 d, 2 H (J = 8); 7·5 d, 2 H (J = 8)
IVd	69	155—158 (ethanol)	1 680	2·4 dd, 1 H (J = 11); 2·7 dd, 1 H (J = 3); 3·8 s, 3 H; 3·9—4·1 m, 1 H; 4·3—4·5 m, 2 H; 6·9 d, 2 H (J = 8); 7·6 d, 2 H (J = 8)
IVe	32	130—134 (ethanol)	3 380 1 700	2·4—2·5 m, 1 H; 2·6—2·8 m, 1 H; 4·1—4·2 m, 1 H; 4·3—4·4 m, 2 H; 7·1 br s, 1 H; 7·2—7·4 m, 4 H; 9·7 br s, 1 H
IVf	53	209—212	3 320	2·1 s, 3 H; 2·4 dd, 1 H (J = 10); 2·6 dd, 1 H (J = 3); 3·9—4·5 m, 3 H; 7·5—7·7 m, 4 H; 9·7 s, 1 H
IVg	43	168—170 (ethanol)	3 220	2·4—2·7 m, 2 H; 4·0—4·5 m, 3 H; 6·8 d, 2 H (J = 7); 7·5 d, 2 H (J = 7); 9·4 br s, 1 H

reported work-up, the organic layer was separated to give, after evaporation of the solvent, 0·8 g (37%) of a 1 : 1 *syn* : *anti* mixture (ratio determined by ^{1}H NMR) of the lactone oxime *V*, as an oil which spontaneoeusly solidifies by air exposure. By acidification of the alkaline solution, 0·7 g (32%) of *IVa* were recovered.

3-[2-Oxotetrahydrofuran-4-yl]benzoisoxazole (*VI*)

A solution of *Ie* (3 g, 0·014 mol), hydroxylamine hydrochloride (2 g, 0·029 mol) and ammonium acetate (2·2 g, 0·029 mol) in water (10 ml) and ethanol (15 ml) is refluxed for 3·5 h. After cooling, the solution is acidified with 4M hydrochloric acid and the solvent evaporated. The residue is treated with 5% sodium bicarbonate and extracted with ether. The organic layer is dried over sodium sulfate, the solvent evaporated and the semisolid residue triturated with ether to give 0·5 g (17%) of *VI*; m.p. = 71—73°C (ethanol). IR: 1 775 cm^{-1} (lactone C=O). ^{1}H NMR: 2·91—3·25 m, 2 H; 4·18—4·39 m, 1 H; 4·69—4·75 m, 1 H; 4·78—4·98 m, 1 H; 7·09—7·78 m, 4 H.

Pharmacology

The antiinflammatory activity was determined in the carrageenin-induced edema of rat paw by a modification of the method of Winter et al.[3] Male Wistar rats weighing 160—180 g, fasted for 18 h, were randomly divided into groups of six. Each test compound was suspended in arabic gum (20 mg/5 ml) and administered orally at a dose of 20 mg/kg, with controls receiving the vehicle only. One hour after medication 0·1 ml of 1% carrageenin in normal sterile saline was injected into the plantar tissue of the right hind paw. Paw volume was measured by a plethysmometer at time intervals of 3, 5 and 7 h after induction of inflammation. Mean percentages of edema inhibition were calculated according to the formula: inhibition (in%) = $(\Delta V_c - \Delta V_t / \Delta V_c) \cdot 100$, where ΔV_c and ΔV_t were the increase in paw volume for control and treated animals, respectively. Results for compounds *IVa*—*IVg* are reported in Table III.

TABLE III
Antiinflammatory activity of compounds *IVa*—*IVg*

Compound	Antiinflammatory activity % inhibition vs controls		
	3 h	5 h	7 h
IVa	37·5[a]	33·1[a]	22·7
IVb	N.A.	N.A.	N.A.
IVc	N.A.	N.A.	N.A.
IVd	10·3	1·0	5·8
IVe	12·5	14·7	15·1
IVf	11·1	6·1	6·8
IVg	N.A.	N.A.	N.A.
Naproxene	37·5[a]	42·6[a]	36·9[a]

[a] $P < 0{\cdot}01$.

RESULTS AND DISCUSSION

The data reported in Table III indicate that only the unsubstituted compound *IVa* was provided with an antiinflammatory activity comparable to that of naproxene, taken as reference drug, though of shorter duration. Introduction of Cl (*IVb*), CH_3 (*IVc*) or OH (*IVg*) substituents in the *para* position of the phenyl ring led to inactive compounds, while the presence in the same position of a methoxy (*IVd*) or an acetylamino group (*IVf*) dramatically reduced the antiinflammatory properties. The only *o*-substituted term tested (*IVe*) was also found scarcely active. Since the pharmacological data presently available seem to indicate that *para* substitution of the phenyl ring brings about loss of activity independently of the electronic features of the substituent, we are planning the synthesis of a series of *ortho*- or *meta*-substituted derivatives, whose chemical and pharmacological properties will be the subject of a next paper.

REFERENCES

1. Barlocco D., Martini A., Pinna G. A., Curzu M. M., Sala F., Germini M.: Il Farmaco Ed. Sc. *42*, 585 (1987).
2. Cignarella G., Grella G., Curzu M. M.: Synthesis *1980*, 825.
3. Winter C. A., Risley E. A., Nuss G. W.: Proc. Soc. Exp. Biol. Med. *111*, 544 (1962).