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Some pyrroline derivatives as potential anti-inflammatory agents

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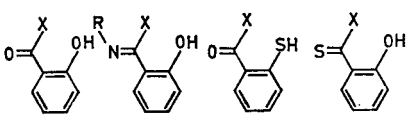
Eight *N*-substituted-4-carbethoxy-2-oxo-3-hydroxypyrrolines and six *N*-β-phenethyl-2-oxo-3-arylaminopyrrolines were synthesised and tested for possible anti-inflammatory action. Infrared and nuclear magnetic resonance spectral evidence is presented to show that in the solid state and in solution ($CDCl_3$) these compounds exist solely as the tautomer in which the double bond is within the heterocyclic ring. The compounds showed no important pharmacological activity.

THERE are four main types of anti-inflammatory agent in clinical use viz. salicylates, phenylbutazone derivatives, corticosteroids, and quinoline type compounds. None of these are totally satisfactory because of their side-effects. Whitehouse & Bostrom (1962) proposed two prerequisites for anti-inflammatory activity in salicylates; the molecule must chelate with metal ions and it must be lipid soluble. On this basis it was suggested that the structures (Ia-Id, where R = alkyl, X = H, OH, OR) may have anti-inflammatory activity.



Ia Ib Ic Id

The purpose of the present work is to replace the six-membered aromatic structure with a planar five-membered heterocyclic ring suitable for simple modification to improve lipid solubility and chelating ability.

Experimental

PREPARATION OF THE COMPOUNDS

General method of preparation of 1-*substituted*-4-*carbethoxy*-2-*oxo*-3-*hydroxypyrrolines* (*II*). Compounds of type III were prepared from appropriately substituted amines and ethyl acrylate in equimolar amounts (0·1–0·2 mol) by heating under reflux in absolute ethanol (100 ml) for 30 min, and the solution allowed to stand for 24 hr before adding diethyl oxalate. This solution was added portionwise to a solution prepared by dissolving sodium (0·1–0·2 mol) in absolute ethanol (100 ml); the mixture was heated under reflux on a steam-bath for 1 hr and the solvent evaporated off. The solid residue was dissolved in the minimum amount of water and the solution acidified with concentrated hydrochloric acid to yield a precipitate which was recrystallised from ethanol.

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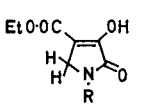
The following new compounds were prepared: II, R = *p*-chlorophenethyl, yield 52%, m.p. 172–172·5°. Found: C, 58·6; H, 5·3; N, 4·4%; $C_{15}H_{16}NO_4$ requires C, 58·15; H, 5·2; N, 4·5%; ν 3150, 1670 cm^{-1}. II, R = *p*-methoxyphenethyl, yield 61%, m.p. 152–152·5°. Found: C, 62·8; H, 6·3; N, 4·4%; $C_{16}H_{19}NO_5$ requires C, 62·9; H, 6·2; N, 4·6%; ν 3150, 1660 cm^{-1}. II, R = β-phenylisopropyl, yield 57%, m.p.

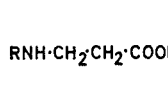
TABLE 1. ULTRAVIOLET SPECTRAL DATA OF COMPOUNDS OF TYPE II IN ETHANOL

No.	R =	λ_{max} in mμ	ε
1	Methyl	249	13,500
2	*n*-Butyl	247	11,400
3	Cyclohexyl	247	10,500
4	Benzyl	249	15,100
5	Phenethyl	248	11,300
6	β-Phenylisopropyl	248	14,200
7	*p*-Methoxyphenethyl	226, 249	16,300; 12,600
8	*p*-Chlorophenethyl	221, 247	15,200; 12,300

114–115°. Found: C, 66·4; H, 6·7; N, 4·7%; $C_{16}H_{19}NO_4$ requires C, 66·4; H, 6·6; N, 4·8%; ν 3120, 1660 cm^{-1}. The other compounds recorded in Table 1 were prepared as described by Southwick, Previc, Casanova & Carlson (1956) and Southwick & Crouch (1953); the m.p. values of these materials were in agreement with reported values.



II



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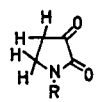

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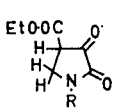

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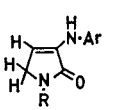
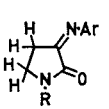


VI



VII

General method of preparation of 1-*β-phenethyl*-2-*oxo*-3-*arylamino-pyrrolines* (VI, R = phenethyl). 1-β-Phenethyl-2,3-dioxopyrrolidine (IV, R = phenethyl) (0·01 mol) (Southwick & others, 1956) and the appropriate aromatic amine (0·01 mol) were dissolved in absolute ethanol (40 ml) and heated under reflux for 1·0 hr. The solid which separated upon cooling was recrystallised from ethanol. The following new compounds were prepared: VI, Ar = phenyl, yield 1·0 g (36%), m.p. 164–165°. Found: C, 77·4; H, 6·65; N, 10·35%; $C_{18}H_{18}N_2O$ requires C, 77·7; H, 6·5; N, 10·1%. VI, Ar = *p*-nitrophenyl, yield 1·0 g (30%), m.p. 215–216°. Found: C, 67·2; H, 5·2; N, 12·6%; $C_{18}N_{17}N_3O_3$ requires C, 66·9; H, 5·3; N, 13·0%. VI, Ar = *p*-tolyl, yield 1·3 g (34%), m.p. 182–183°. Found: 78·5; H, 7·2; N, 9·8%; $C_{19}H_{20}N_2O$ requires C, 78·1; H, 6·9; N, 9·6%. VI, Ar = *p*-methoxyphenyl, yield 1·6 g (50%) m.p. 177·5–179°. Found: C, 74·1; H, 6·35; N, 9·35%; $C_{19}H_{20}N_2O_2$ requires C, 74·0; H, 6·5; N, 9·1%. VI, Ar = *p*-chlorophenyl, yield 1·3 g (41%) m.p. 206–207°. Found: C, 69·4; H, 5·6; N, 9·35%; $C_{18}H_{17}N_2OCl$ requires C, 69·1; H, 5·5; N, 9·0%. VI, Ar = β-naphthyl, yield 1·5 g (45%),

m.p. 199·5–200·5°. Found: C, 81·0; H, 6·1; N, 8·8%; $C_{22}H_{20}N_2O$ requires C, 80·5; H, 6·1; N, 8·5%.

PHYSICAL MEASUREMENTS

The infrared spectra of the compounds as Nujol mulls were obtained on a Unicam SP200, ultraviolet spectra of the compounds dissolved in absolute ethanol were obtained on a Unicam SP 800, and NMR spectra of the compounds in deuterochloroform using tetramethylsilane as reference were obtained using a Varian A60 instrument.

TABLE 2. NMR SPECTRAL DATA OF COMPOUNDS OF TYPE II IN $CDCl_3$ AT 60 MC. (τ values from tetramethylsilane)

R =	Me	Bu	Benzyl	Phenethyl
Protons:				
C–Me (ester) ..	8·64 (3H, T)	8·68 (3H, T)	8·72 (3H, T)	8·67 (3H, T)
N–Me	6·88 (3H, S)	—	—	—
Ph–CH_2– ..	—	—	—	7·10 (2H, M)
N–CH_2–	—	6·50 (2H, M)	5·32 (2H, S)	6·30 (2H, M)
–CH_2– (ring) ..	6·00 (2H, S)	6·00 (2H, S)	6·13 (2H, S)	6·15 (2H, S)
CH_2–C (ester) ..	5·70 (2H, Q)	5·68 (2H, Q)	5·70 (2H, Q)	5·70 (2H, Q)
Aromatic ..	—	—	2·67 (5H, S)	2·75 (5H, S)
OH*	0·50 (1H, S)	−1·10 (1H, S)	0·75 (1H, S)	0·84 (1H, S)

* Disappears on deuteration.
(S = singlet, D = doublet, T = triplet, Q = quartet, M = multiplet.)

PHARMACOLOGICAL TESTING

Compounds of structure II (Nos. 1, 4, 5, 6, 7, 8 Table 1) were tested for anti-inflammatory activity by measuring the delay in onset of skin erythema following exposure of the depilated backs of guinea-pigs to ultraviolet light (Hardy, Wolff & Goodell, 1952). Only compound 8 showed any activity (at 60 mg/kg); other compounds were inactive at 200 mg/kg. Compound 8, tested by the method of Randall & Selitto (1957), gave an ED50 of 83 mg/kg as compared with 2 mg/kg for phenylbutazone. Compound 1 showed some antipyretic and analgesic activity after 4 hr at a dose of 200 mg/kg.

Compounds of structure VI (Nos. 9, 10, 11 and 14 Table 3) were also tested by the ultraviolet light induced erythema test. Only compound 11 showed any activity at a dose level of 200 mg/kg. Compounds 10, 11 and 14 showed slight sedative effects at doses above 1 g/kg.

Results and discussion

PREPARATION OF THE COMPOUNDS

Attempts to hydrolyze the ester group in compounds of type II failed. Refluxing in 6N hydrochloric acid resulted in decarboxylation and rearrangement to the corresponding 2,3-dioxopyrrolidine (IV), except in (II, R = Me) where no product could be isolated. The reaction of 2,3-dioxopyrrolidine (IV, R = phenethyl) with aromatic amines was used to prepare 1-substituted-2-oxo-3-arylaminopyrrolines (VI, R = phenethyl) [cf. the reaction of 1-benzyl-2,3-dioxopyrrolidine with aniline to form VI, R = benzyl, Ar = phenyl (Southwick & others, 1956)]. Compounds of structure II and VI gave coloured complexes with ferric or cupric ions.

SPECTRAL DATA AND STRUCTURAL CONSIDERATIONS

On the basis of pK_a and infrared spectral data, it was suggested by Southwick & others (1956) that compounds of type II existed mainly in the enolic form rather than the ketonic form V. Present evidence based on infra-red and NMR data (Table 2) shows that compounds of type II exist in the solid state and in solution ($CDCl_3$) solely in the enolic form; the evidence is as follows.

TABLE 3. ULTRAVIOLET AND INFRARED SPECTRA OF COMPOUNDS OF TYPE VI

No.	Ar =	Ultraviolet (ethanol) mμ/extinction		Infrared (Nujol) cm^{-1}
9	Phenyl	243 15,000	291 9,600	3250, 1660, 1630
10	*p*-Nitrophenyl	250sh 7,900	374 12,100	3300, 1660, 1640
11	*p*-Tolyl	243 17,000	293 13,000	3300, 1670, 1640
12	*p*-Methoxy	241 16,500	296 10,400	3300, 1670, 1640
13	*p*-Chlorophenyl	247 14,000	292 14,600	3300, 1670, 1640
14	β-Naphthyl	265sh 17,000 309 13,600	275sh 12,300 350 2,600	3300, 1680, 1640, 1620

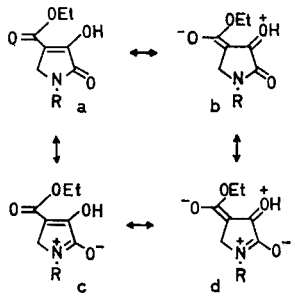
sh = shoulder.

The hydrogen-bonded hydroxyl group of II is indicated in the solid state by the medium strength band around 3100 cm^{-1} in the infrared spectrum (Nujol mull) of these compounds; the carbonyl group is shown by the broad band at 1650 ± 20 cm^{-1}. The hydroxyl proton in structure II is also shown by the broad band (1 proton) in the NMR spectra of the four 1-substituted 4-carbethoxy-2-oxo-3-hydroxypyrrolines examined (Table 2); this signal disappears on deuteration, under neutral conditions. Supporting evidence is the two-proton *singlet* (*ca* 6·0 τ) which is assigned to the C_5 protons; this signal is again consistent only with structure II. [The predicted NMR spectrum of V would show no active hydrogen and have the signal of the aliphatic proton on C_4 split into a multiplet by the two adjacent C_5 protons; the signal of these latter protons would also be split by the adjacent C_4 proton.] The NMR spectra and their integrals show no detectable amount of tautomer V in $CDCl_3$ solutions of compounds of type II.

Other aspects of the NMR spectra include an unsplit two-proton singlet in the spectrum of the *N*-benzyl analogues which is assigned to the methylene group of the benzyl side-chain. In the phenethyl analogues, multiplets appear at 7·10 and 6·30 τ each representing 2 protons. The 7·10 τ peak is assigned to the methylene group next to the benzene ring.

The tautomer present in ethanol solution cannot be deduced unequivocally from the ultraviolet spectra of solutions of compounds (II) due to the lack of suitable model compounds. However, since from the infra-red and NMR evidence the enolic structure II is present in the solid state and in deuterochloroform solution, the high ultraviolet absorption at about 248 mμ (ϵ ca. 13,000, Table 1) may be assigned with reasonable

certainty to the enolic form and to resonance between the hybrids (VIIIa←→VIIId; R = benzyl).



VIII

The preference of 1-substituted-4-carbethoxy-2-oxo-3-hydroxypyrrolines for the enolic form II may be due to the additional resonance stabilisation (VIIIa←→VIIIb←→VIIId) which is not possible with the ketonic form V. [The NMR spectrum of the compound without the ester group (IV, R = phenethyl) shows only a complicated set of peaks for eight protons indicating that the ketonic form of this compound is present in deuterochloroform solution.]

On the basis of infra-red evidence (Southwick & others, 1956), structure VI and not VII (R = benzyl, Ar = phenyl) was assigned to 1-benzyl 2-oxo-3-phenylaminopyrroline. Our infra-red and NMR evidence (Tables 3 and 4) indicates that the six new phenethyl analogues we report exist in the solid state and in solution ($CDCl_3$) as structure VI. The evidence is as follows.

TABLE 4. NMR SPECTRA OF N-SUBSTITUTED 2-OXO-3-PHENYLAMINOPYRROLINES (VI, Ar = PHENYL) IN $CDCl_3$ AT 60 MC (τ values from tetramethylsilane)

	R = Benzyl	R = Phenethyl
Protons:		
$-CH_2$ (adjacent to Ph)	—	7·08 (2H, multiplet)
$-CH_2$ (ring)	6·18 (2H, doublet)	6·20 (4H, multiplet)
$-CH_2$ (adjacent to N) ..	5·30 (2H, singlet)	
Olefinic	4·08 (1H, triplet)	4·17 (1H, triplet)
N-H*	3·34 (1H, broad singlet)	3·40 (1H, broad singlet)
Aromatic	2·84 (10H, multiplet)	2·90 (10H, multiplet)

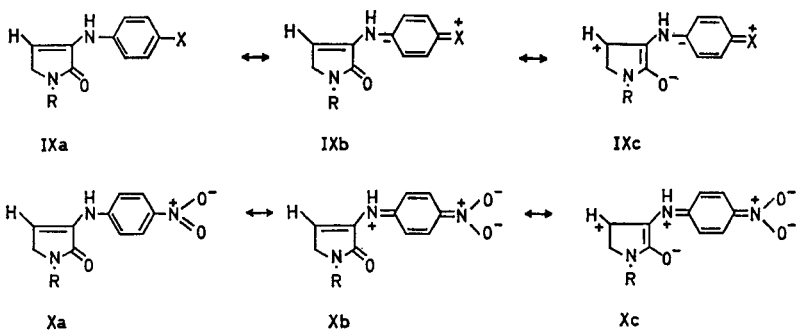
* Disappears on deuteration.

The presence of the imino-group in structure VI is shown in the solid state by the N-H bond 3300 cm^{-1} (Table 2) in the infra-red spectrum (Nujol mull); the lactam carbonyl and the carbon-carbon double bond bands are at 1670 and 1640 cm^{-1} respectively. For NMR measurements, 1-benzyl-2-oxo-3-phenylaminopyrroline was used as a prototype of the series since the phenethyl group resonance partly obliterates the ring proton signals in the corresponding 1-phenethyl analogues (see Table 4). The N-H proton is indicated by a low broad one-proton signal at 2·34 τ which disappears upon deuteration. A one-proton triplet at 4·08 τ (J = 2·5 cps) can only be the signal of the olefinic proton at C_4 split by the two ring protons at C_5; these latter appear as a doublet at 6·18 τ

(identical J). [The predicted NMR spectrum for structure VII would have no active proton, no olefinic proton, and a complicated pattern for four ring protons.] The position, multiplicity, and integral of the NMR signals are consistent only with Structure VI; there is no detectable amount of structure VII.

The ultraviolet spectra of these compounds in ethanol cannot distinguish unequivocally between the two tautomers because both VI and VII are equally conjugated. The ultraviolet data in Table 3 show that substitution in the *para* position of the benzene ring of compounds of type VI leads in some instances, e.g. compounds 12 and 13, Table 3, to only small changes in wavelength, while the substitution of a *p*-NO_2 group leads to a bathochromic shift of 80 mμ. Since the infra-red and NMR data indicate structure VI in the solid state and $CDCl_3$ solution, it is concluded that the *p*-OMe and *p*-Cl may exert a mesomeric effect on the benzene ring which is not transmitted to the pyrroline double bond (i.e. IXa$\leftrightarrow$IXc; R = phenethyl, X = Cl or OMe); thus only small wavelength shifts are produced by these substitutions.

On the other hand, the *p*-NO_2 group (No. 10 Table 2) can interact with the double bond in the pyrroline ring forming a system encompassing four conjugated double bonds (Xa$\leftrightarrow$Xb$\leftrightarrow$Xc; R = phenethyl) and this causes the large bathochromic shift.



IXa IXb IXc

Xa Xb Xc

PHARMACOLOGICAL RESULTS

The lack of significant pharmacological activity in compounds of type II and VI precludes any structure-activity correlations.

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