Synthesis of Hydantoins Transformable to 3-Alkylorotic Acids from Aminofumarate and Isocyanates

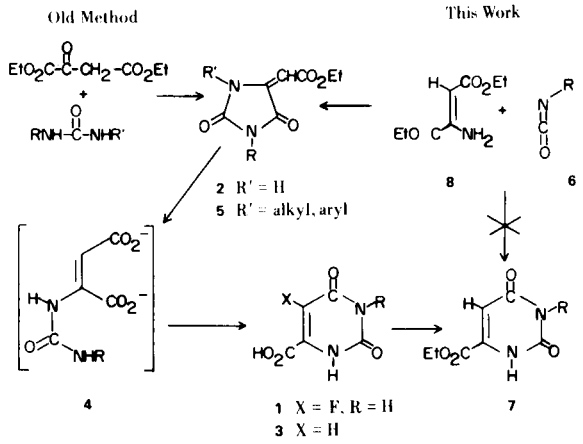
Stephen S. Washburne and Kwanghee K. Park

Department of Chemistry, Temple University, Philadelphia, Pennsylvania 19122

Received September 4, 1974

The utility of orotic acids and their glycosides as antitumor agents has been vigorously pursued since the seminal synthesis of fluoroorotic acid **1** by Heidelberger (1). Other uses, ranging from analytical reagents for metals (2) to insect chemosterilants (3) have since evolved for these pyrimidine derivatives. 1,3-Dimethylorotic acid has been utilized as a probe for study of the metabolically central, enzymatic decarboxylation of orotidine-5′-phosphate (4). The classical synthesis of alkylorotic acids, first realized by Bachstez (5), and since elaborated by numerous groups (6), involves condensation of a urea with a dialkyl oxaloacetate to form a carboxymethylidene hydantoin **2**, which can be approached by alternative routes (7). Hydantoin **2** suffers rearrangement to orotic acid **3** upon saponification (6), a rearrangement which has been reported to proceed *via* a uraminomaleate (8) or uraminofurmarate **4**. Hydantoins substituted in the 1-position, *e.g.*, **5**, saponify without rearrangement (9), possibly due a higher rotational barrier in the disubstituted amide bond.

In view of our interest in uracil synthesis (10), and the ready availability of alkyl isocyanates (11), we hoped that the condensation of diethyl aminofurmarate with an isocyanate would afford a 3-alkylorotic acid ester directly, *i.e.*, **8** + **6** → **7**.



This goal could not be realized, and a hydantoin **2** (R′ = H) was inevitably the product when diethyl aminofumarate and isocyanates were combined at 120° in the presence of aluminum chloride catalyst (See Table I). In the reaction of *t*-butyl isocyanate with aminofumarate it was necessary to add sodium ethoxide to effect cyclization of initially formed *N*-*t*-butyl-*N*′-diethylfumarylurea, but in other cases shown in Table I, pure hydantoin was isolated by recrystallization of the solidified reaction mixture. Following the method of Lemieux (6c), the hydantoins were converted to the corresponding orotic acid **3** by treatment with aqueous alcoholic potassium hydroxide. Spectroscopic and analytical parameters were in accord with the orotic acid structure, and except in the R = *t*-butyl case, the yields were good.

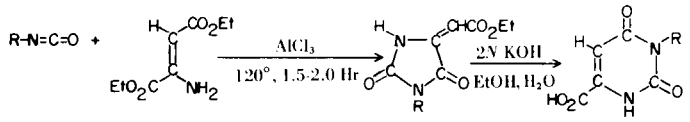
This procedure constitutes a new route to 3-substituted orotic acids. By passage of dry hydrogen chloride into an ethanol solution of **3a**, the ethyl ester **7a** isomeric with **2a** was obtained in good yield. Spectroscopically it was easy to distinguish these isomers (Table II). The hydantoin structure exhibits a complex carbonyl stretching region with three or four bands between 1790 and 1660 cm^{-1} while the orotic acid exhibits a similar complex between 1730 and 1620 cm^{-1}. Even more specific is the position of the single olefinic resonance in the pmr spectrum; hydantoins were in the δ 5.68 to 5.87 region, while orotic acids and the ester **7a** were between δ 6.10 and 6.28 ppm.

This orotic acid synthesis will be of especial utility where the mono-substituted urea required for the classical preparation is difficultly accessible. An example *au point* is the 3,3′-trimethylenebisurea $(H_2NCONHCH_2)_2CH_2$ required for preparation of **2e**. Contrariwise, 3,3′-trimethylene diisocyanate **6e** is preparable in 85% yield from glutaryl dichloride and trimethylsilyl azide (11).

Of further interest is the stereochemistry of the olefinic bond of **2**. In previous work (**6b**,**6c**) this was not rigorously determined, as **2** was synthesized from a non-rigid precursor, *i.e.*, a ketosuccinate. In the present case, use of aminofumarate assures that **2** will have the *Z* configuration required for closure to orotate to be effected.

TABLE I

3-Alkylorotic Acids from Isocyanates and Diethyl Aminofumarate



6a-e 8 2a-e 3a-e

Run	R	mmoles 6	mmoles 8	Yield 2	Yield 3 (a)
a	C_6H_5–	30	15	49%	84%
b	nC_4H_9–	20	10	67%	87%
		20	20	38%	
c	tC_4H_9–	36	30	67% (b)	9.5% (c)
d	½-$(CH_2)_6$– (d)	10	20	18%	76%
e	½-$(CH_2)_3$– (d)	50	100	16%	91%

(a) Based on **2**. (b) Reaction at 75°, 2 hours, sodium ethoxide required to effect cyclization. (c) *t*-Butyl urea (31%) was major product. (d) **6d** = $OCN(CH_2)_6NCO$; **6e** = $OCN(CH_2)_3NCO$.

TABLE II

Spectroscopic and Analytical Data on 3-Alkyl-5-carboethoxymethylidenehydantoins **2** and 3-Alkylorotic Acids **3**

Compound	ν c=o (cm^{-1})	δ HC=C (ppm)		Analysis Carbon	Hydrogen	Nitrogen	M.p.
2a	1780, 1740, 1700, 1680	5.68	Calcd.	---	---	---	191°
			Found	---	---	---	
2b	1780, 1730, 1700, 1660	5.87	Calcd.	54.99	6.71	11.66	78-79°
			Found	55.21	6.72	11.67	
2c	1790, 1730, 1690	5.76	Calcd.	54.99	6.71	11.66	54-55°
			Found	55.08	6.63	11.67	
2d	1790, 1740, 1700	5.87	Calcd.	53.33	5.82	12.44	177°
			Found	53.41	5.88	12.31	
2e	1790, 1745, 1690	5.88	Calcd.	50.00	4.94	13.72	203°
			Found	49.98	5.00	13.67	
3a	1710, 1660, 1640	6.26	Calcd.	---	---	---	280° dec.
			Found	---	---	---	
3b	1730, 1710, 1660, 1640	6.14	Calcd. (a)	46.96	6.13	12.17	197-198°
			Found	47.09	6.16	12.18	
3c	1720, 1710, 1650, 1620	6.10	Calcd. (a)	46.96	6.13	12.17	>320° dec.
			Found	46.78	6.02	12.34	
3d	1710, 1650, 1650	6.10	Calcd. (b)	42.86	5.40	12.50	294° dec.
			Found	42.69	5.39	12.46	
3e	1735, 1645	6.13	Calcd. (a)	42.17	3.81	15.13	>310° dec.
			Found	42.41	3.62	15.23	
7a	1740, 1730, 1710, 1660	6.28	Calcd.	60.00	4.65	10.76	204°
			Found	60.10	4.55	10.75	

(a) Assuming one mole of water of crystallization. (b) Assuming three moles of water of crystallization.

That *t*-butyl hydantoin **2c** did not form upon admixture of **6c** and **8**, and required ethoxide, to induce cyclization of *t*-BuNHCONHC(CO_2Et)=$CHCO_2Et$, is a consequence of lessened nucleophilicity of the *t*-BuNH- moiety caused by steric hindrance. This factor of steric hindrance also explains the reluctance of **2c** to isomerize to **3c**, *t*-butyl urea formed by cleavage of a species like **4e** being the major product in this reaction.

This bis-orotic acids **3d** and **3e** were synthesized in order to investigate possible photodimerizations (12), results of which study will be reported later.

EXPERIMENTAL

General Comments.

All reactions with isocyanates, or with trimethylsilyl azide were carried out under anhydrous conditions; glassware was twice flame-dried and dry nitrogen was passed through the reaction systems. Ir spectra were determined on Perkin-Elmer Model 727 spectrophotometer and nmr spectra on Varian Model XL-100 spectrometer. Chemical shifts are given in δ units (parts per million) down-field from internal TMS. Micro-analyses were performed by Galbraith Laboratories, Knoxville, Tennessee.

Preparation of Diethyl Aminofumarate (**8**).

This was made by a slight modification of the procedure of Huisgen (13). Anhydrous ether (Mallinckrodt), 500 ml., was saturated with ammonia (Matheson gas products) at 0°; and at -10°, 34.0 g. (0.2 mole) of diethyl acetylenedicarboxylate (Aldrich) in 100 ml. of anhydrous ether was added slowly with swirling. The solution was stirred for 3 hours at 0°. Evaporation of ether and distillation of the residue gave 35.4 g. (0.19 mole, 95%) of diethyl aminofumarate, b.p. 70-72° (0.35-0.40 mm).

3-Alkyl-5-carboethoxymethylidenehydantoins (**2**). General Procedure.

A mixture of isocyanate **6** and **8** (quantities specified in Table I) were heated with *ca.* 0.1 g. of anhydrous aluminum chloride for 1.5 to 2 hours at 120°. The reaction mixture solidified on cooling, and was recrystallized from ethanol/water to afford pure hydantoin **2**: yields, spectra, and analyses given in Tables I and II. The product **2a** was identical in m.p., ir, and pmr with phenylhydantoin prepared by Clerk-Bory's procedure (6b).

3-*t*-Butyl-5-carboethoxymethylidenehydantoin (**2c**).

The cooled reaction mixture from the general procedure contained mostly *N*-*t*-butyl-*N'*-(diethylfumaryl)urea. After removal of unreacted starting material (90°, 0.15 mm) by Kugelrohr distillation, the residue was refluxed with 15 ml. of ethanol and *ca.* 0.1 g. of sodium ethoxide for 1.5 hours. Removal of solvent and distillation (Kugelrohr, 100°, 0.15 mm) afforded 67% of **2c**, m.p. 54-55°.

3-Alkylorotic Acids (**3**). General Procedure.

A mixture of x mmole of hydantoin **2**, x ml. of 2*N* potassium hydroxide solution, and 0.5 x ml. of ethanol was heated on a steam bath for 2.5-16 hours, then collected and acidified with 2*N* aqueous hydrochloric acid. The precipitate was filtered, then recrystallized from ethanol/water to afford **3**: yields, spectra and analyses given in Table I and II.

3-*t*-Butylorotic Acid (**3**).

A mixture of 4.8 g. (20 mmoles) of **2c** and 20 ml. of 5*N* aqueous potassium hydroxide was heated at 65° for 16 hours. The cooled solution precipitated 0.73 g. of *t*-butylurea, m.p. 186°, identified by ir (14). The filtrate was acidified with 2*N* hydrochloric acid. On standing overnight a precipitate formed, which after recrystallization from ethanol/water, gave 0.43 g. (9.5%) of 3-*t*-butylorotic acid (monohydrate), m.p. $>$ 320° dec.

Ethyl 3-Phenylorotate (**7a**).

A slow stream of gaseous hydrogen chloride was passed through a boiling solution of 1.25 g. (5 mmoles) of **3a** in 20 ml. of absolute ethanol, affording a precipitate which was recrystallized from 95% ethanol to give 1.1 g. (84%) of **7a**, m.p. 203-204°.

Acknowledgement.

This investigation was sponsored by grant No. CA-13120-03 from the National Cancer Institute.

REFERENCES

(1) R. Duschinsky, E. Pleven, and C. Heidelberger, *J. Am. Chem. Soc.*, 79, 4559 (1957).

(2) B. C. Lewis and W. I. Stephen, *Anal. Chim. Acta*, 36, 234 (1966).

(3) M. D. Boston, R. S. Patterson, and C. S. Lofgren, *Fla, Entomol.*, 54, 215 (1970); *Chem. Abstr.*, 74, 110856u (1971).

(4) P. Beak and B. Siegel, *J. Am. Chem. Soc.*, 95, 7919 (1973).

(5) M. Bachstez, *Chem. Ber.*, 64, 2683 (1931).

(6a) H. K. Mitchell and J. F. Nye, *J. Am. Chem. Soc.*, 69, 674 (1947); (b) C. Clerc-Bory and C. Mentzer, *Bull. Soc. Chim. France*, 436 (1958); (c) R. U. Lemieux and I. Puskas, *Can. J. Chem.*, 42, 2909 (1964).

(7a) M. R. Atkinson, M. H. Maguire, R. K. Ralph, G. Shaw and R. N. Warrener, *J. Chem. Soc.*, 2363 (1957); (b) G. V. Rutkovskii, B. A. Ivin, E. G. Sochilin, and I. Y. Tsereteli, *J. Gen. Chem.* USSR, 40, 357 (1970).

(8) B. A. Ivin, G. V. Rutkovskii, E. G. Sochilin, and I. Y. Tsereteli, *Zhur. Org. Khim.*, 8, 640 (1972).

(9) R. K. Ralph, G. Shaw, and R. N. Naylor, *J. Chem. Soc.*, 1169 (1959).

(10) S. S. Washburne, W. R. Peterson, Jr., and D. A. Berman, *J. Org. Chem.*, 37, 1738 (1972).

(11) S. S. Washburne and W. R. Peterson, Jr., *Syn. Commun.*, 2, 227 (1972).

(12) M. W. Logue and N. J. Leonard, *J. Am. Chem. Soc.*, 94, 2842 (1972), and references therein.

(13) R. Huisgen, K. Herbig, A. Siegl and H. Huber, *Chem. Ber.*, 99, 2526 (1966).

(14) Sdatler Laboratories, Philadelphia, Pa; Spectrum #9751.