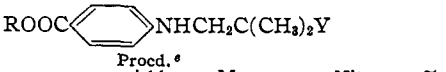
TABLE I

SUBSTITUTED ALKYL *p*-(N-AMINO)-BENZOATES



R	Y	Procd.[e] yield, %	M. p., °C.	Nitrogen, % Calcd.	Found
CH_3	NO_2	A90	132.5–134	11.11	11.44
C_2H_5	NO_2	A56	137–138	10.50	10.15
$CH_3CH_2CH_2$	NO_2	A51	103–104	10.00	10.17
$(C_2H_5)_2N(CH_2)_3$	NO_2	B94	110–110.5	11.97	12.03
CH_3	NH_2	D82	79–80	12.61	12.60
CH_3	NH_2	C92[a]			
C_2H_5	NH_2	C74	65.5–66.5	11.87	12.09
$CH_3CH_2CH_2$	NH_2	C81	Oil	11.20	11.51
$(C_2H_5)_2N(CH_2)_3$	NH_2	B58	20–22	13.09	12.97
$(C_2H_5)_2N(CH_2)_3$	NH_2	D84	18–20		
H	$(CH_3)_2N$	E68	238 (dec.)[b]	11.87	11.73
CH_3	$(CH_3)_2N$	C60	90.5–91.5	11.20	10.74[c]
CH_3	$(CH_3)_2N$	E68	90.5–91.5		
C_2H_5	$(CH_3)_2N$	E66	70–71	10.60	10.57
C_2H_5	$(CH_3)_2N$	C12	72–73		
$CH_3CH_2CH_2$	$(CH_3)_2N$	C35[d]	65.5–66.5	10.08	10.33
$(C_2H_5)_2N(CH_2)_3$	$(CH_3)_2N$	B92	38–39	12.03	12.56

[a] This is the hydrochloride, m. p., 166–168° (dec.). [b] The hydrochloride melts at 256° (dec.). [c] Analysis after further drying: Calcd. for C, 67.20; H, 8.70. Found: C, 67.40; H, 8.39. [d] Yields for other runs of this compound were much lower. [e] Letter preceding percentage yield represents procedure followed.

to one-fourth volume, adding water and cooling gave an additional 6.4 g. (94% yield).

The other γ-diethylaminopropyl esters (V and VI, R = $(C_2H_5)_2N(CH_2)_3$) were prepared in a similar way with the following exceptions. After refluxing, the solvent was removed under reduced pressure, water was added and the product was extracted with ether. After washing the ether solution with dilute sodium carbonate solution, the ether was evaporated. Solid esters were recrystallized from petroleum ether.

Procedure C. Methyl *p*-(2-Amino-2-methylpropylamino)-benzoate.—A suspension of 9 g. of finely divided *p*-(2-amino-2-methylpropylamino)-benzoic acid[2] in 100 ml. of methanol was saturated with dry hydrogen chloride, finally with cooling in ice. After standing at room temperature overnight, the reaction mixture was refluxed for twenty minutes. After cooling in the refrigerator, the mixture was filtered and washed with methanol, ylelding 11.7 g. of the hydrochloride of the ester, V (R = CH_3).

A portion of the hydrochloride was dissolved in water. Upon adding sodium hydroxide, the ester, m. p. 79–80.5°, precipitated.

Other esters required refluxing for a longer time (up to twenty-four hours) in order to get satisfactory yields. In most cases the product was isolated as described in the last paragraph under Procedure B.

Procedure D. Methyl *p*-(2-Amino-2-methylpropylamino)-benzoate.—A solution of 130 g. of methyl *p*-(2-nitro-2-methylpropylamino)-benzoate in 1000 ml. of methanol, to which was added 10 g. of Raney nickel, was hydrogenated at 100° and 1000 p.s.i. After filtering off the catalyst, the methanol was distilled and the remaining liquid distilled under reduced pressure. There was obtained 94.5 g. of the ester, b. p. 160–161° at 1 mm., which solidified upon standing, m. p. 79–81°.

γ-Diethylaminopropyl *p*-(2-nitro-2-methylpropylamino)-benzoate was hydrogenated in benzene. The amine was purified by washing with aqueous sodium hydroxide, drying over anhyd. sodium carbonate and removing the solvent under reduced pressure.

Procedure E. *p*-(2-Dimethylamino-2-methylpropylamino)-benzoic Acid.—To 41.6 g. (0.2 mole) of *p*-(2-amino-2-methylpropylamino)-benzoic acid was added 50 ml. of formalin and 52 ml. of 98% formic acid. In about fifteen minutes the flask was placed in an oil-bath heated to 140° and the mixture was allowed to reflux for four hours. After adding 24 ml. of concd. hydrochloric acid to the hot reaction mixture, it was evaporated to dryness under reduced pressure with warming on the steam-bath. After adding 50 ml. of warm water and cooling in the refrigerator, the mixture was filtered and the solid washed with water, yielding 37.1 g. of the hydrochloride of the acid, m. p. 256° (dec.).

A sample of the hydrochloride was dissolved in boiling water and sufficient 20% aqueous sodium hydroxide was added to make the solution faintly basic. Filtering and washing with hot water gave the free acid, m. p. 238° (dec.).

Esters prepared in this manner were purified as in Procedure C.

Summary

A number of esters of *p*-(2-nitro-2-methylpropylamino)-, *p*-(2-amino-2-methylpropylamino)- and *p*-(2-dimethylamino-2-methylpropylamino)-benzoic acid have been prepared.

GREENCASTLE, INDIANA RECEIVED NOVEMBER 15, 1948

[CONTRIBUTION FROM THE STERLING-WINTHROP RESEARCH INSTITUTE]

Ring Substituted Benzoylacrylic Acids as Antibacterial Agents

BY FRED. K. KIRCHNER, JOHN HAYS BAILEY AND CHESTER J. CAVALLITO

In an investigation of model antimicrobial compounds containing unsaturated lactone or carbonyl structures, one of the types which appeared to be of interest because of its solubility and activity against both gram positive and gram negative bacteria was benzoylacrylic acid. Worrall[1] has reported antibacterial activity of methyl and propyl esters of the acid and Rinderknecht, *et al.*,[2] have observed activity of the acid as well as the *p*-chloro-, 2,4-dichloro- and *p*-acetamido derivatives.[2,3]

Observations have been made relative to some non-ionic antibiotics that the degree of activity against gram positive bacteria was associated with the lipophilic properties of the compound.[4,5] It was of interest to determine whether this relationship could be shown with the anionic benzoylacrylic acids.

Para alkyl-substituted β-benzoylacrylic acids

(1) Worrall, "Med. World" (London), 1946, 2 pp., *C. A.*, **41**, 6598 (1947).

(2) Rinderknecht, Ward, Bergel and Morrison, *Biochem. J.*, **41**, 463 (1947).

(3) British Patent 588,108; *C. A.*, **42**, 329 (1948).

(4) Small, Bailey and Cavallito, THIS JOURNAL, **69**, 1710 (1947).

(5) Cavallito and Bailey, *J. Bact.*, in press.

have been prepared with substituent groups ranging from methyl to dodecyl. A marked increase in activity against gram positive bacteria resulted up to approximately the nonyl derivative, beyond which there was a decrease. The *p*-methoxy and ethoxy derivatives were of the same order of activity as the methyl and ethyl compounds. The 2,4-dihydroxy derivative was prepared with some difficulty and was found to be less active than benzoylacrylic acid. The *m*-nitro-, *p*-chloro- and *p*-bromobenzoylacrylic acids were only moderately active.

The benzoylacrylic acids prepared were presumably all of the *trans* form.[6] Orientation in the benzene ring was determined by hypobromite oxidation of the benzoylacrylic acids to the corresponding alkylbenzoic acids.

Unsaturated lactones and other carbonyl derivatives have been indicated to inhibit growth by reaction with biologically essential —SH and possibly amino groups.[7,8] It was thus of interest to determine the nature of the reaction of cysteine with benzoylacrylic acid. An amphoteric reaction product was obtained and it appeared to be a simple addition product of cysteine to the —CO—CH=CH—COOH system to yield the —CO—CH_2—CH(SR)—COOH type of derivative. The cysteine derivative retained considerable antibacterial activity which is believed to result from the ease of dissociation of the addition product to regenerate benzoylacrylic acid.

The low activity of 2,4-dihydroxybenzoylacrylic acid is believed to result from interruption of the conjugated system by chelation involving the carbonyl and the 2-hydroxy group. It would be of interest to determine whether the alkyl substituted phenylglyoxylic acids of Hunsberger and Amstutz[9] would be more active if devoid of the orthohydroxy group.

With the lichesterinic acid type of antibiotic[10] it was found that the unsaturated structure was subordinate to the surface-active properties in contributing to antibacterial activity. The higher alkyl-substituted benzoylacrylic acids, on the other hand, have been found to be relatively poor surface tension depressants. The importance of the double bond in contributing to antibacterial activity was demonstrated by *p*-hexylbenzoylacrylic acid which was approximately one-hundred fold as active as *p*-hexylbenzoylpropionic acid.

Experimental[11]

Phenyl Alkyl Ketones.—Except for propiophenone, which was purchased, the ketones were prepared through the Friedel–Crafts reaction. The acid chlorides were prepared according to Bauer's recommendation.[12]

Alkyl Benzenes.—Toluene, and ethyl and isopropyl benzene, were purchased and redistilled before use. The *t*-butylbenzene was prepared by the method of Norris and Sturgis.[13] The remaining *n*-alkyl benzenes were obtained by the Wolff–Kishner reduction of the phenyl *n*-alkyl ketones, using the Huang-Minlon modification.[14]

The constants previously reported[15] for *n*-decylbenzene were determined on a substance whose structure was questioned. Reduction of the *n*-nonyl phenyl ketone gave *n*-decylbenzene of b. p. 115–116° (2 mm.), and $n^{25}D$ 1.4816.

Anal. Calcd. for $C_{16}H_{26}$: C, 88.00; H, 12.00. Found: C, 88.17; H, 11.76.

β-(4-Alkylbenzoyl)-acrylic Acids.—The methyl derivative was prepared using an excess of toluene as a solvent.[16] The other alkyl derivatives were prepared by the following general procedure. In a suitably equipped flask, cooled with an ice-bath, were placed 0.20 mole of the alkyl benzene, 0.21 mole of powdered maleic anhydride, and 300 ml. of dry carbon disulfide. There was added in small portions with stirring a total of 0.42 mole of anhydrous aluminum chloride. When the addition was complete the ice-bath temperature was maintained for twenty minutes longer, whereupon the ice-bath was removed and stirring was continued at room temperature for four hours. The flask then was warmed gently for two hours, with continued stirring.

At the end of this period the cooled reaction mixture was decomposed with 300 g. of ice and 10 ml. of concentrated hydrochloric acid. The resulting mixture was steam-distilled and the yellow residue was taken up in ether and the ether solution dried with Drierite.

After removing the Drierite and ether, the yellow residue was recrystallized from a hot benzene–Skellysolve B mixture, or from hot Skellysolve B alone. In this manner there was obtained the compounds listed in Table I.

With the isopropyl and the *t*-butyl derivatives the above procedure was modified at one point. When the aluminum chloride addition was completed, the stirring was continued for four hours at ice-bath temperature, then two hours at room temperature.

The orientation of the substituents in the benzene ring was determined by the sodium hypobromite oxidation[17] of the acrylic acids to known alkylbenzoic acids, and comparison of their constants. The melting point of 4-*n*-octylbenzoic acid was found to be 98–99°, in close agreement with Zaki and Fahim. Beran[18] had reported a melting point of 139°. Melting points for the 4-*n*-nonyl-, 4-*n*-decyl- and 4-*n*-dodecylbenzoic[19] acids were not found in the literature; therefore, we are reporting the following values based on analytically pure samples.

4-*n*-Nonylbenzoic acid, m. p. 96–97° (cor.)

Anal. Calcd. for $C_{16}H_{24}O_2$: C, 77.37; H, 9.74. Found: C, 77.38; H, 9.56.

4-*n*-Decylbenzoic acid, m. p. 95–96° (cor.)

Anal. Calcd. for $C_{17}H_{26}O_2$: C, 77.81; H, 9.99. Found: C, 77.53; H, 9.77.

4-*n*-Dodecylbenzoic acid, m. p. 93–95° (cor.)

Anal. Calcd. for $C_{19}H_{30}O_2$: C, 78.60; H, 10.42. Found: C, 78.44; H, 10.20.

The above acids, oxidized with potassium permanganate, gave terephthalic acid, identified through its methyl ester, m. p. 140°.

(6) Lutz and Scott, *J. Org. Chem.*, **13**, 284 (1948).
(7) Cavallito and Haskell, THIS JOURNAL, **67**, 1991 (1945).
(8) Geiger, *Arch. Biochem.*, **16**, 423 (1948).
(9) Hunsberger and Amstutz, THIS JOURNAL, **70**, 671 (1948).
(10) Cavallito, Fruehauf and Bailey, *ibid.*, **70**, 3724 (1948).
(11) In the experimental section physical constants and analyses are reported only for those compounds which have not been reported previously, and for those whose preparation or properties differ from that found in the literature.
(12) Bauer, *Oil and Soap*, **23**, 1 (1946).
(13) Norris and Sturgis, THIS JOURNAL, **61**, 1413 (1939).
(14) Huang-Minlon, *ibid.*, **68**, 2487 (1946).
(15) Francis, *Chem. Rev.*, **42**, 144 (1948).
(16) von Pechmann, *Ber.*, **15**, 881 (1882).
(17) Zaki and Fahim, *J. Chem. Soc.*, 307 (1942).
(18) Beran, *Ber.*, **18**, 139 (1885).
(19) Balle, Wagner and Nold, U. S. Patent 2,195,198, report the preparation of a *n*-dodecylbenzoic acid, but give no melting point.

TABLE I

ANALYTICAL, PHYSICAL AND ANTIBACTERIAL PROPERTIES

$RC_6H_4COCH=CHCOOH$ R	Analyses, % Carbon Calcd.	Carbon Found	Hydrogen Calcd.	Hydrogen Found	M. p., °C.[a]	Surface tension,[b] dynes per cm.	Minimum bacteriostatic[c] concn. as moles per ml. × 10^4 *Staphylococcus aureus 209*	*Cl. perfringens*	*M. tuberculosis H37Rv*	*E. coli*	*K. pneumoniae*
Hydrogen[i]	68.18	68.00	4.58	4.53	93–94	72.6	7.5	12.5	25.0	25	100
4-Methyl	69.46	69.52	5.30	5.34	139–140	70.4	5	12.5	7.5	25	75
4-Ethyl	70.57	70.72	5.93	5.77	105–106	72.1	1	3.75	5.0	25	75
4-*n*-Propyl	71.54	71.54	6.46	6.67	87–89	70.2	0.5	3.75	1.0	25	75
4-*n*-Butyl	72.39	72.39	6.94	6.67	90–91	72.1	0.1	1.25	0.5	25	75
4-*n*-Amyl	73.15	72.91	7.37	7.56	81–82	66.0	0.075	1.25	0.1	50	50
4-*n*-Hexyl	73.82	73.83	7.74	7.72	86–87	68.0	0.05	3.75	0.05	25	75
4-*n*-Heptyl	74.42	74.64	8.08	8.26	79–80	60.0	0.025	0.05	0.01	10	>10
4-*n*-Octyl	74.97	75.07	8.39	8.26	77–78	50.1	0.0075	0.0375	0.005	10	>10
4-*n*-Nonyl	75.46	75.78	8.67	8.55	82–83	33.8	0.025	0.025	0.01	...	...
4-*n*-Decyl	75.91	75.81	8.92	8.78	82–83	34.6	0.025	0.0025	0.0075	...	...
4-*n*-Dodecyl	76.70	76.76	9.37	9.13	81–82	53.6	<0.01[h]	<0.05[h]	<0.01[h]	...	...
4-*i*-Propyl	71.54	71.58	6.46	6.57	95–98	72.6	0.5	0.25	5.0	10	50
4-*t*-Butyl[j]	72.39	72.17	6.94	7.06	123–125	72.0	0.25	0.125	5.0	25	50
4-Methoxy	64.07	64.20	4.89	5.01	129–131	..	10.0	2.5	10	25	75
4-Ethoxy	65.44	65.31	5.49	5.20	141–145	..	2.5	2.5	10	25	75
4-Hexyloxy	69.54	69.55	7.30	7.41	118–119	..	0.25	0.5	0.5	>10	>10
4-Chloro[d]	57.02	56.99	3.35	3.34	156–157	..	2.5	3.75	10	25	50
4-Bromo[e]	47.09	47.15	2.77	2.77	159–160	..	2.5	3.75	7.5	25	50
3-Nitro[f]	54.30	54.58	3.19	3.19	190–192	..	50	25.0	50.0	100	>100
2,4-Dihydroxy	57.69	57.43	3.87	4.17	195	..	>100	>100	>100	>100	>100
Cysteine deriv. of benzoylacrylic acid[g]	52.51	52.62	5.08	5.14	160–161 (dec.)		25	12.5	50	25	>100
β-4-*n*-Hexylbenzoyl propionic acid	73.25	73.42	8.46	8.51	97–98	67.8	5	12.5	0.25	>100	100

[a] Corrected. [b] Solvent is 0.1 molar potassium phosphate buffer of *p*H 7; solvent surface tension = 72.0 dynes per cm. Measurements made with a Du Nouy interfacial tensiometer at 27°. Concentration of acids = 7×10^{-3} moles per ml. [c] The bactericidal activity is approximately one-tenth the bacteriostatic power. [d] Calcd.: Cl, 16.84. Found: Cl, 16.90. [e] Calcd.: Br, 31.33. Found: Br, 30.90. [f] Calcd.: N, 6.33. Found: N (Kjeldahl), 6.01. [g] Calcd.: N, 4.71. Found: N (Kjeldahl), 4.70. [h] Lack of solubility prevented attaining a bacteriostatic concentration. [i] Fieser and Fieser, THIS JOURNAL, **57**, 1679 (1935). [j] Price, *et al.*, *J. Org. Chem.*, **7**, 517 (1942).

β-(2,4-Dihydroxybenzoyl)-acrylic Acid.[20]—In a round-bottom flask were placed 40 g. (0.4 mole) of resorcinol, 40 g. (0.41 mole) of powdered maleic anhydride, and 600 ml. of freshly-distilled nitrobenzene. A solution of 108 g. (0.81 mole) of anhydrous aluminum chloride in 300 ml. of nitrobenzene was added dropwise to the stirred solution over a period of ninety minutes, keeping the temperature of the reaction mixture between 20–30°. When the addition was complete stirring was continued for three hours longer, maintaining the temperature at 50–60°. At the end of this period the stirring was discontinued and the reaction allowed to cool to room temperature and stand overnight.

To the reaction mixture was added 300 ml. of ice-cold 12% hydrochloric acid solution and the mixture was stirred for three hours while keeping the temperature at 0–10°. At the end of this period the supernatant liquors were decanted from the brown, viscous mass which had formed. The latter was washed several times with benzene, then crystallized from 60% ethanol. A second smaller crop of the product was obtained by concentrating and cooling the nitrobenzene liquor. The orange-colored product was dried at 100° to give a lemon-yellow compound of m. p. 195°, yield 20–25%.

Catalytic hydrogenation of the unsaturated acid with platinum catalyst gave β-(2,4-dihydroxybenzoyl)-propionic acid, m. p. 199–200°, previously reported by Desai and Shroff.[21]

Other β-Substituted Benzoylacrylic Acids.—The 4-methoxy,[22] 4-ethoxy,[23] 4-chloro,[24] 4-bromo,[25] and 3-nitro[26] compounds were prepared according to procedures in the literature.

The hexyloxy compound was prepared according to the method used for the *n*-alkyl derivatives with a slight modification. After the addition of the aluminum chloride was complete, the reaction mixture became so viscous that it was allowed to stand at room temperature for five hours. At the end of this period the solvent was decanted and the viscous mass decomposed with ice and concentrated hydrochloric acid. The required hexyloxybenzene[27] was prepared according to a procedure given by Vogel.[28]

(20) Bogert and Ritter, *Proc. Nat. Acad. Sci. Washington*, **10**, 363 (1924); *cf.* Thomas "Anhydrous Aluminum Chloride in Organic Chemistry," Reinhold Pub. Corp., N. Y., 1941, p. 581. In the original article the authors make no mention of an attempt to prepare this acid. However, the article in Thomas states that Bogert and Ritter had obtained only fumaric acid in attempting to condense resorcinol with maleic anhydride in the presence of aluminum chloride and carbon disulfide.

(21) Desai and Shroff, *J. Univ. Bombay*, **10**, Pt. 3, 97 (1941).

(22) Dave and Nargund, *ibid.*, **7**, Pt. 3, 191 (1938).

(23) Rice, THIS JOURNAL, **46**, 2319 (1924).

(24) Rinderknecht, *et al.*, *Biochem. J.*, **41**, 463 (1947).

(25) Kohler and Woodward, THIS JOURNAL, **58**, 1933 (1936).

(26) Bogert and Ritter, *ibid.*, **47**, 526 (1925).

(27) Tronow and Ladigina, *Ber.*, **62**, 2844 (1929).

(28) Vogel, "A Textbook of Practical Organic Chemistry," Longmans, Green and Co., N. Y., 1948, p. 641.

The hexyloxy compound was oxidized[17] to the known 4-*n*-hexyloxybenzoic acid.[29]

β-(4-*n*-Hexylbenzoyl)-propionic Acid.—This compound was prepared in the same manner as the majority of the aroylacrylic acids. The compound was oxidized[17] to the known 4-*n*-hexylbenzoic acid.

Reaction Product of β-Benzoylacrylic Acid with Cysteine.—Three grams (0.017 mole) of β-benzoylacrylic acid was dissolved in 12 ml. of water and the solution adjusted to *p*H 7 with aqueous sodium hydroxide solution. A solution of 2.68 g. (0.017 mole) of cysteine hydrochloride in 12 ml. of water was treated with aqueous sodium hydroxide until the *p*H was 7. The solutions were mixed and allowed to stand overnight.

The mixture had become slightly yellow and the *p*H was 7.3. Dilute hydrochloric acid was added dropwise until *p*H 2 was reached. The white precipitate was filtered off, washed several times with water, then recrystallized from 60% ethanol solution.

Antibacterial Tests.—The compounds were tested as growth inhibitors of *Staphylococcus aureus*, *Escherichia coli* and *Klebsiella pneumoniae* in nutrient broth by the serial dilution method using as an inoculum, 1 ml. of a 1:1000 dilution of a twenty-four hour culture of the test organism and a final volume of 5 ml. in each tube. Incubation was for eighteen hours at 37°. Tests with *Clostridium perfringens* and *Mycobacterium tuberculosis* H37Rv were conducted as previously described.[10] To avoid fluctuations in susceptibility of the test organisms, all compounds of a series were tested against a single organism at the same time and benzoylacrylic acid was used as a reference test compound for series of tests conducted on different days.

Acknowledgment.—The authors wish to express their appreciation for the analytical determinations and the corrected melting points which were carried out under the direction of Mr. M. E. Auerbach in the Analytical Laboratories of this Institute. We are indebted to Dr. F. C. Nachod and his staff for the surface-tension measurements. We are appreciative also of the technical assistance given by Mrs. G. W. Schraver during the course of this investigation.

(29) Jones, *J. Chem. Soc.*, 1874 (1935).

Summary

1. A number of substituted β-aroylacrylic acids were prepared and their antibacterial activity determined *in vitro*.

2. With the *n*-alkyl series there was a noticeable increase in activity against gram positive bacteria as the length of the alkyl chain was increased, the C_9 chain being approximately the optimum chain length. The antibacterial activity appears to result from the presence of the highly conjugated benzoylacryl- system which may react with biologically essential —SH groups; the lipophilic ring substituents quantitatively increase the activity against gram positive bacteria.

3. The following 4-*n*-alkylbenzoic acids are considered to be reported for the first time: *viz.*, *n*-nonyl, *n*-decyl and *n*-dodecyl.

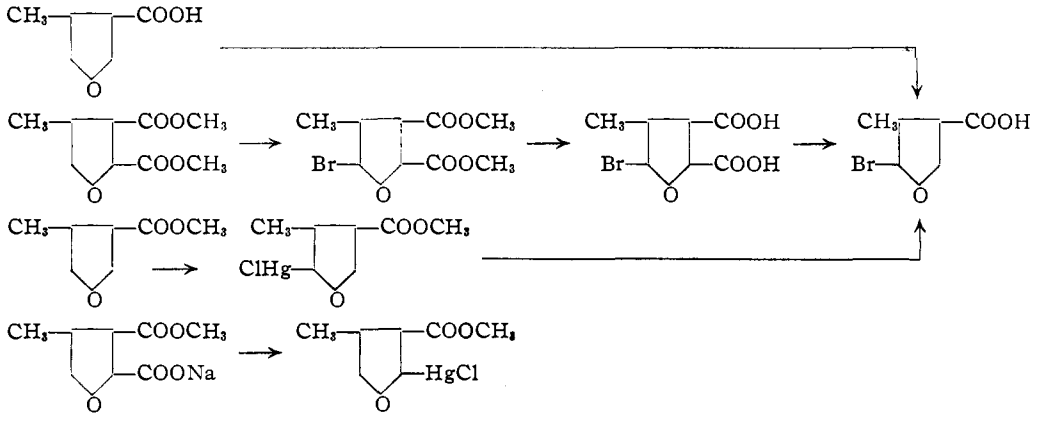
RENSSELAER, NEW YORK RECEIVED NOVEMBER 12, 1948

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF IOWA STATE COLLEGE]

Orientation in the Furan Nucleus. XII. 3-Methyl-4-furoic Acid and Some of its Derivatives

BY HENRY GILMAN AND ROBERT R. BURTNER[1]

In a previous paper dealing with orientation of β-substituted furans[2] the prediction was made on the basis of evidence then available that nuclear substitution of 3-methyl-4-furoic acid would involve replacement of hydrogen on the 2-position. The validity of this assumption as regards bromination of the acid, mercuration of the methyl ester, and nitration of the ethyl ester has now been established by a sequence of reactions as shown.



(1) Present address: G. D. Searle & Co., Chicago, Ill.

(2) Gilman and Burtner, THIS JOURNAL, **55**, 2903 (1933).