

TABLE I

| Substrate | Oxidant | Temp., °C. | Atm. | Rate of Addn. | Total Time, Hr. | Diphenoquinone | Benzoquinone | Remarks |
|----------------|--------------------------------------|----------------|----------------|---------------|------------------|------------------|--------------------|--------------------------|
| Phenol | Ferricyanide | 25 ± 2 | N ₂ | Immediate | 4 | 98% ^a | Trace ^c | |
| | | | | Dropwise | 43 | 71% ^b | 7% | |
| Aldehyde | Lead dioxide | 25 ± 2 | O ₂ | Immediate | 12 | 96% ^a | None | |
| | Ferricyanide | | N ₂ | Dropwise | 48 | 95% ^a | Trace ^c | |
| | | | O ₂ | Immediate | 17 | 98% ^a | Trace ^c | |
| | | | O ₂ | Dropwise | 36 | | 30% | Remainder diphenoquinone |
| | | | O ₂ | 48 | | 58% | | |
| | Lead dioxide | O ₂ | Immediate | 96 | None | 39% | Remainder tar | |
| Acid | Moist lead dioxide | 25 ± 2 | O ₂ | Dropwise | 72 | None | 26% | |
| | | | N ₂ | | 24 | 97% ^a | None | |
| | Ferricyanide | | O ₂ | 44 | 96% | None | | |
| | | | O ₂ | 2 | 95% ^a | None | | |
| Diphenoquinone | Lead dioxide | | Immediate | 2 | 95% ^a | None | | |
| Aldehyde | Ferricyanide | | O ₂ | Immediate | 48 | | | No reaction |
| | H ₂ O + O ₂ | | O ₂ | | 75 | | | |
| | H ₂ O/NaOH/O ₂ | | O ₂ | | 72 | | | |

^a Before recrystallization. ^b Loss in handling during recrystallization. ^c Less than 0.1%.

to the reaction mix containing the remainder of the solvent by means of a capillary dropping funnel. After reaction, the benzene layer was evaporated to dryness and the 2,6-di-*t*-butylbenzoquinone separated by sublimation at 65°. The diphenoquinone was recrystallized from ethanol, m.p. 240–241° (uncorr.), reported 245–247°,^{5,6} and identified by the identity of the mixture melting point and ultraviolet spectrum with those of an authentic sample.⁷

In those cases where no benzoquinone was formed, the crude diphenoquinone melted within two degrees of the purified product.

Anal. Calcd. for C₂₈H₄₀O₂: C, 82.30; H, 9.87. Found: C, 82.35; H, 9.77.

The 2,6-di-*t*-butylbenzoquinone was recrystallized from methanol-water, m.p. 65–66°, reported 65–66°,⁸ 67.5–68.5°.⁹ A mixture melting point with a known sample showed no depression.

In several runs using the aldehyde, the aqueous layer was strongly acidified. Essentially quantitative (98%) evolution of carbon dioxide resulted.

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(5) H. Hart and F. A. Cassis, Jr., *J. Am. Chem. Soc.*, **73**, 3179 (1951).

(6) W. J. Detroit and H. Hart, *J. Am. Chem. Soc.*, **74**, 5215 (1952).

(7) Kindly provided by Dr. Harold Hart.

(8) S. J. Metro, *J. Am. Chem. Soc.*, **77**, 2901 (1955).

(9) C. D. Cook, R. C. Woodworth, and P. Fianu, *J. Am. Chem. Soc.*, **78**, 4159 (1956).

Application of a Silicic Acid Chromatostrip Technique for Observing the Sequential Methylation of β -Resorcylic Acid and Related Reactions

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During recent work on the characterization of the naturally occurring estrogen, coumestrol,² it became necessary to prepare the 2,4-dimethoxy and 2-hydroxy-4-methoxybenzoic acids. We found that these acids, together with their methyl esters, form sequentially as discrete products during the methylation of β -resorcylic acid in alkaline solution. Utilization of a fluorescent silicic acid chromatostrip technique³ permitted us to observe under ultraviolet light the sequence of formation of each intermediate compound during methylation. As a result, it was possible to stop the reaction at any time in order to obtain the desired derivative. Thus, from one methylation reaction mixture all the methylated derivatives of β -resorcylic acid could be readily isolated.

The chromatostrip technique used to follow the chemical reactions was originally developed by Kirchner *et al.*³ and Miller and Kirchner⁴ and has

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(2) E. M. Bickoff, A. N. Booth, R. L. Lyman, A. L. Livingston, C. R. Thompson, and F. DeEds, *Science*, **126**, 969 (1957).

(3) J. G. Kirchner, J. M. Miller, and G. J. Keller, *Anal. Chem.*, **23**, 420 (1951).

been successfully applied to a variety of isolations from natural materials.⁵⁻⁷

This communication describes a further application of the chromatostrip for following and maintaining control of the methylation of a polyhydroxybenzoic acid. The simplicity of its use suggests that the method may be advantageously employed to follow a variety of reactions involving other types of aromatic compounds.

Table I summarizes the R_f values of a variety of hydroxy aromatic compounds and related derivatives. All could be identified under ultraviolet light either as a fluorescing spot or as a dark spot resulting from absorption of the ultraviolet rays.

From the R_f values in Table I, it may be seen that a mixture of nearly all of the compounds shown could be resolved using only the 3 solvent systems described. In addition to its high resolution, the chromatostrip is suitable for following slow chemical reactions because a separation may be completed in 15 min. or less, permitting reasonably close control of the reaction. Also detection of compounds which fluoresce or absorb ultraviolet light may be made instantaneously without resorting to further manipulations. If necessary for detection, however, it is possible to carry out a number of color tests or other chemical reactions directly on the glass strip.

In the procedure described the termination of a reaction was made at a time when only a single component of the system was present. This made it very easy to isolate the compound in high yield. However, it may not always be possible to obtain single intermediate compounds. Thus, when phloroglucinol was methylated with methyl sulfate and potassium hydroxide, the monomethoxyl, dimethoxyl, and trimethoxyl derivatives formed in an overlapping manner with all 3 being present after 1 hr. Further methylation resulted in the gradual disappearance of first the monomethoxyl, then the dimethoxyl derivatives, leaving finally the trimethoxylated compound. Whether the conditions of the reaction might have been altered to produce any desired intermediate was not investigated.

EXPERIMENTAL

Chromatographic procedure. The chromatostrips, having the dimensions of about 0.5 inch by 5.5 inches were prepared from $\frac{1}{16}$ -inch window glass. The strips were coated on one side with a paste made up of silicic acid, starch, and inorganic fluorescing materials as described by Kirchner *et al.*³ and Miller and Kirchner.⁴ The inorganic fluorescing agent⁸ allows detection of compounds that do not fluoresce,

(4) J. M. Miller, and J. G. Kirchner, *Anal. Chem.*, **26**, 2002 (1954).

(5) J. M. Miller, and J. G. Kirchner, *Anal. Chem.*, **24**, 1480 (1952).

(6) J. M. Miller, and J. G. Kirchner, *Anal. Chem.*, **25**, 1107 (1953).

(7) W. L. Stanley, and S. H. Vannier, *J. Assoc. Offic. Agr. Chem.*, **40**, 582 (1957).

(8) J. W. Sease, *J. Am. Chem. Soc.*, **70**, 3630 (1948).

TABLE I

R_f VALUES OF A VARIETY OF POLYHYDROXY AROMATIC COMPOUNDS WHEN DEVELOPED ON SILICIC ACID CHROMATOSTRIPS

| | Ether: Skelly- solve B (7:3) | Ethyl Acetate: Skelly- solve B (3:1) | Acetone: Skelly- solve B (1:3) | Appearance of Spot under Ultraviolet Wave Length |
|--|---------------------------------------|--|---|--|
| | R_f^a | R_f^a | R_f^a | 2540 Å |
| Resorcinol | 0.49 | 0.78 | 0.19 | Dark purple |
| Hydroquinone | 0.45 | 0.78 | 0.17 | Purple fluor. |
| Catechol | 0.65 | 0.89 | 0.41 | Black |
| Phloroglucinol | 0.18 | 0.52 | 0.0 | Black (faint) |
| Pyrogallol | 0.39 | 0.72 | 0.17 | Black (faint) |
| α -Resorcylic acid | 0.21 | 0.61 | 0.73 | Purple |
| β -Resorcylic acid | 0.57 | 0.85 | 0.19 | Blue fluor. |
| γ -Resorcylic acid | 0.10 | 0.15 | 0.10 | Purple |
| Salicylic acid | 0.72 | 0.88 | 0.48 | Blue fluor. |
| Protocatechuic acid | 0.32 | 0.55 | 0.10 | Purple |
| Gentisic acid | 0.35 | 0.65 | 0.17 | Blue fluor. |
| 2,4,6-Trihy- droxybenzoic acid | 0.15 | 0.50 | 0.10 | Black (faint) |
| <i>p</i> -Hydroxy- phenylacetic acid | 0.35 | 0.72 | 0.14 | Purple |
| 2,5-Dihydroxy- phenylacetic acid | 0.67 | 0.85 | 0.43 | Black |
| <i>m</i> -Hydroxy- phenylacetic acid | 0.32 | 0.65 | 0.19 | Purple |
| <i>p</i> -Hydroxy- aceto- phenone | 0.48 | 0.83 | 0.40 | Purple |
| <i>o</i> -Hydroxy- aceto- phenone | 0.85 | 1.0 | 0.72 | Purple |
| 2,6-Dihydroxy- aceto- phenone | 0.71 | 1.0 | 0.54 | Purple |
| Orcinol | 0.49 | 0.81 | 0.27 | Black (faint) |
| 3,5-Dimethoxy- benzoic acid | 0.64 | 0.86 | 0.36 | Blue-purple |
| <i>p</i> -Methoxy- benzoic acid | 0.63 | 0.92 | 0.39 | Blue-purple |
| 4-Hydroxy-3- methoxy- benzoic acid | 0.46 | 0.76 | 0.29 | Blue-purple |
| 3,4,5-Trimeth- oxybenzoic acid | 0.43 | 0.75 | 0.31 | Purple |
| Ferulic acid | 0.42 | 0.76 | 0.16 | Blue |
| Syringic acid | 0.33 | 0.57 | 0.16 | Blue-purple |
| 2,4-Dimethoxy- phenylaceto- nitrile | 0.77 | 1.0 | 0.56 | Purple |

^a The R_f 's are only relative and will vary somewhat between different batches of chromatostrips. Each strip was spotted with 10-20 gamma of material and the solvent front allowed to travel 10-11 cm.

but do absorb under ultraviolet light in the wavelength range of 230-390 m μ . The silicic acid-starch paste was applied by spreading it on a glass strip and drawing the strip

under a straight-edged object adjusted to give a coating about 0.5 mm. thick. It was found advantageous to dry the chromatostrips at 105° for 30 min. and to store them over potassium hydroxide in a desiccator until used.

For chromatography, a small spot of the solution to be developed was placed one centimeter above one end of a strip. The strip was immersed in the appropriate developing solution contained in a test tube. About 1.5 ml. of solvent in the tube is sufficient for development. Three solvent mixtures that we have found to be particularly useful are ether-Skellysolve B (7:3); ethyl acetate-Skellysolve B (3:1); and acetone-Skellysolve B (1:3).^{9a}

For following the methylation of β -resorcylic acid, the ether: Skellysolve B (7:3) mixture was employed as the developer. The methoxy derivatives were observed on the strip as dark absorption spots when viewed under an ultraviolet lamp having a peak emission wave length at 2540 Å. At 15-min. intervals, samples were removed from the reaction mixture by means of a micropipet, acidified, and chromatographed. In this way, formation of the new methoxy compound and the disappearance of the reactant were simultaneously observed.

Methyl 2,4-dihydroxybenzoate. To 10 g. of β -resorcylic acid dissolved in acetone was added 10 g. of sodium carbonate, and while the mixture was boiling on a steam bath dimethyl sulfate and 10% methanolic potassium hydroxide were alternately added dropwise to maintain the pH at 7-8. The original spot representing β -resorcylic acid ($R_f = 0.57$) gradually changed during 1.5 hr. to a faster-moving spot ($R_f = 0.73$). After acidification of the reaction mixture and removal of acetone, the compound was extracted from the mixture with ether and crystallized from methanol to give colorless needles, producing a positive ferric chloride reaction, m.p. 117°. Hydrolysis of this compound with 10% methanolic potassium hydroxide for 0.5 hr. on the steam bath gave the original β -resorcylic acid. Therefore, the first product formed was the methyl 2,4-dihydroxybenzoate.

Anal. Calcd. for $C_8H_8O_4$: C, 57.1; H, 4.77; OMe, 18.4. Found: C, 57.1; H, 4.85; OMe, 18.4.

Rangaswami⁹ described the preparation of the methyl- β -resorcylicate by means of methanol and hydrochloric acid. The completely dried compound melted at 119°.

Methyl 2-hydroxy-4-methoxybenzoate. Continued heating of the solution of methyl ester and maintenance of the pH at 7-8 for 2.5 additional hours produced another compound which gave a single spot ($R_f = 0.86$). This material crystallized in the refrigerator from methanol and water as white needles, m.p. 46°. Mauthner¹⁰ gave 48° as the melting point for this compound. The compound was the methyl 2-hydroxy-4-methoxybenzoate.

Anal. Calcd. for $C_9H_{10}O_4$: C, 59.3; H, 5.49; OMe, 34.1. Found: C, 59.4; H, 5.62; OMe, 33.8.

2-Hydroxy-4-methoxybenzoic acid. Saponification of the acetone solution of methyl 2-hydroxy-4-methoxybenzoate for 4 hr. on the steam bath with 10% methanolic potassium hydroxide resulted in another pure compound as shown by a single blue fluorescent spot ($R_f = 0.67$). The isolated material, after removal of acetone and extraction into ether, was crystallized from methanol as colorless needles, m.p. 158°. The compound gave a positive ferric chloride test and was soluble in 5% sodium carbonate solution.

Calcd. for $C_9H_8O_4$: C, 57.1; H, 4.77; OMe, 18.4. Found: C, 57.1; H, 4.77; OMe, 18.8.

Mauthner¹⁰ indicated 158° as the melting point for 2-hydroxy-4-methoxybenzoic acid.

Methyl 2,4-dimethoxybenzoate. The methyl dimethoxybenzoate was conveniently formed by the alternate addi-

(9) M. S. Rangaswami, *J. Ind. Chem. Soc.*, **16**, 160 (1939). (a) Mention of specific products does not constitute endorsement by the Department of Agriculture over others of a similar nature not mentioned. (b) All melting points were obtained on the Kofler block.

(10) N. Mauthner, *J. prakt. Chem.*, **159**, 36 (1941).

tions of dimethyl sulfate and 10% potassium hydroxide to a boiling acetone solution of β -resorcylic acid so as to maintain the pH at 11-12. The reaction went *via* the same methyl ester intermediates described above, but at the higher pH, the reaction was completed within 2 hr. and the chromatostrip showed only a single spot ($R_f = 0.63$). The material isolated was an amber-colored liquid at room temperature. It was further purified to a colorless liquid by distillation at 120° and 0.75 mm. It boiled at 293-296° under atmospheric pressure. No ferric chloride reaction was observed and the compound was not extracted from ether by 5% sodium carbonate.

Anal. Calcd. for $C_{10}H_{12}O_4$: C, 61.2; H, 6.12; OMe, 47.5. Found: C, 60.9; H, 6.44; OMe, 45.0.

Perkin and Schiess¹¹ had previously described the preparation of this compound with a boiling point of 294-296° at atmospheric pressure.

2,4-Dimethoxybenzoic acid. Saponification of the methyl 2,4-dimethoxybenzoate with 10% methanolic potassium hydroxide for 2 hr. on the steam bath resulted in formation of a compound giving a new dark spot on the chromatostrip ($R_f = 0.27$), which when isolated crystallized from water as colorless needles, m.p. 108°. The material gave no ferric chloride reaction and was soluble in sodium carbonate.

Anal. Calcd. for $C_9H_{10}O_4$: C, 59.1; H, 5.49; OMe, 34.1. Found: C, 59.4; H, 5.54; OMe, 34.4.

Karrer *et al.*¹² have described the dimethoxybenzoic acid as having a melting point of 107°.

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(11) H. P. Perkin and E. Schiess, *J. Chem. Soc.*, 159 (1904).

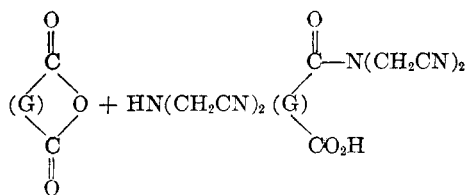
(12) G. Karrer, A. Rebmann, and E. Zeller, *Helv. Chim. Acta*, **3**, 261 (1920).

N,N-Bis(cyanomethyl)carboxamic Acids

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In a previous note¹ the synthesis of a series of *N,N*-bis(2-cyanoethyl)carboxamic acids was reported. This note describes the synthesis and properties of several analogous compounds, *N,N*-bis(cyanomethyl)carboxamic acids, which were prepared by the reaction of iminodiacetonitrile with cyclic anhydrides in an inert solvent.



(1) J. W. Lynn, *J. Am. Chem. Soc.*, **78**, 5829 (1956).