seem to suggest polymerization rather than a shift of tautomeric forms. Ready polymerization of N-methylmyosmine was encountered in the work previously cited.[13] This phenomenon was not studied further. The values of k' as reported were calculated from the major peak of these curves.

Values much greater than 10 or less than 0.1 cannot be determined accurately with an eight-tube transfer. For this reason, the values of the observed partition coefficients as reported in Table I for 4-methylamino-1-(3-pyridyl)-1-butanol, nornicotine, anabasine and metanicotine deviate from a straight line function sufficiently to make it impossible to calculate good values for the true partition coefficient or hydrolysis constant of these compounds. The observed values are in the correct order of magnitude and are reported here for use in separations of these compounds by countercurrent techniques.

PHILADELPHIA 18, PENNA.

[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, COLLEGE OF AGRICULTURE, UNIVERSITY OF WISCONSIN]

Relation between Precursor Structure and Biosynthesis of Penicillins[1]

BY DONALD C. MORTIMER[2] AND MARVIN J. JOHNSON

RECEIVED APRIL 19, 1951

The addition of various carboxylic acids to synthetic-medium fermentation of *Penicillium chrysogenum* Q176 brought about the synthesis of the corresponding penicillin if the acid added was not substituted in the α-position. The paper chromatographic assay for penicillin types showed that in the presence of sorbic, cyclohexaneacetic and phenoxyacetic acids, the percentage of precursor-produced penicillin was as high as in the presence of phenylacetic acid. The over-all precursor efficiency of these acids was, however, much lower than that for phenylacetic acid. The rate of metabolism of certain precursor acids has been found to bear an inverse relation to precursor efficiency. Acids substituted in the α- or β- positions were not readily metabolized by the mold. If a rapidly metabolized acid such as sorbic was added to the fermentation at frequent intervals, its precursor efficiency became comparable with that for the slowly metabolized phenylacetic acid.

During the early coöperative investigations on penicillin[3] work was done in many laboratories on biosynthesis of penicillins. A large number of compounds were tested as possible precursors. It was found that when any one of a number of carboxylic acids was added to the fermentation, as such or as a suitable derivative, a penicillin was produced which was a substituted amide of the acid added, just as benzyl penicillin is a substituted amide of phenylacetic acid. Eleven new penicillins were crystallized. Later, Behrens and coworkers[4,5] reported tests on further possible precursors, and isolated 18 additional new penicillins. An additional five new penicillins were reported by Philip and others.[6] At the time most of this work was done, convenient criteria for production of new penicillins were not available. Stimulation of yield and variation of the penicillin activity ratio as measured on two test organisms were the methods used. After the advent of convenient paper chromatographic techniques, Thorn and Johnson[7] were able to show that the lower saturated fatty acids regularly produced biosynthetic penicillins, although such compounds had given no evidence of precursor activity in previous work.

The tentative conclusions regarding the relation of structure to precursor activity drawn from earlier work were[1] (a) that ring-substituted phenylacetic acids were excellent precursors, (b) that certain other ring systems could be substituted for the benzene ring, (c) that an "interrupting group" in the carbon chain, apparently to prevent β-oxidation, appeared advantageous, and (d) that substitution in α-position led to structures with no precursor activity.

The fact that acids which should be resistant to β-oxidation tended to be good precursors, together with the finding[5] that straight-chain aliphatic acids often acted as precursors for penicillins corresponding to acids containing two fewer carbon atoms, made it appear likely that precursor activity was inversely related to oxidizability. In the present investigation the chromatographic method for detection and determination of new penicillins has been applied to a number of possible precursors, many of which have been tested by other workers. A more accurate evaluation of their precursor activity has thus been gained, and the relation of precursor activity to oxidizability by the mold has been studied.

Results and Discussion

Table I lists the compounds tested as precursors, and the total penicillin yields obtained when they were used. The last column gives the percentage of "new" (*i.e.*, biosynthetic) penicillin formed, as determined by paper chromatography. Diagrams of representative paper chromatograms are given in Fig. 1. The data of Table I show that substitution of any one of a variety of groups at the α-carbon of the precursor acid made the resulting compound unavailable as precursor. An exception was α-ethylphenylacetic acid, which, in one of

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(2) Division of Applied Biology, National Research Council, Ottawa, Canada.

(3) O. K. Behrens, in H. T. Clarke, J. R. Johnson and R. Robinson, "The Chemistry of Penicillin," Princeton University Press, Princeton, N. J., 1949, p. 657.

(4) O. K. Behrens, J. Corse, D. E. Huff, R. G. Jones, Q. F. Soper and C. W. Whitehead, *J. Biol. Chem.*, **175**, 771 (1948).

(5) O. K. Behrens, J. Corse, J. P. Edwards, L. Garrison, R. G. Jones, Q. F. Soper, F. R. van Abeele and C. W. Whitehead, *ibid.*, **175**, 793 (1948).

(6) J. E. Philip, A. P. Saunders, A. F. DeRose, D. W. MacCorquodale, J. C. Sylvester and A. W. Weston, *ibid.*, **189**, 479 (1951).

(7) J. A. Thorn and M. J. Johnson, THIS JOURNAL, **72**, 2052 (1950).

the experiments yielded a biosynthetic penicillin, but in the other experiment failed to do so.

TABLE I

THE EFFECT OF CARBOXYLIC ACIDS USED AS PRECURSORS UPON THE TOTAL PENICILLIN YIELD

Acid added[a]	Penicillin at 120 hr.,[b] units/ml. Test flask	Control flask[c]	Stimulation ratio test/control	New penicillin, % of total units[d]
Crotonic	200	215	0.93	0
β-Chlorocrotonic	185	215	0.86	0
Sorbic	125	113	1.10	91.8
	175	152	1.15	100
α,β-Hexenoic	168	167	1.0	86.0
Caproic	133	167	0.80	69.2
α-Ethylbutyric	148	167	0.89	0
	75	*85*	*0.88*	0
α-Ethylcaproic	*63*	*85*	*0.74*	0
Benzoic	162	147	1.10	0
	178	171	1.04	0
p-Nitrobenzoic	153	171	0.89	0
Phenylacetic	285	171	1.67	89.7
	229	152	1.51	
α-Hydroxyphenylacetic	*95*	*120*	*0.79*	0
α-Aminophenylacetic	*90*	*120*	*0.75*	0
α-Ethylphenylacetic	180	195	0.92	14.6
	95	*120*	*0.79*	0
α-Phenylphenylacetic	*80*	*120*	*0.67*	0
Cinnamic	16	113	0.14	0
	12	106	0.11	0
Hydrocinnamic	112	171	0.65	50
	114	152	0.75	
Cyclohexaneacetic	59	106	0.56	83.5
	95	*120*	0.79	
Cyclohexanepropionic	45	215	0.21	20–40
	27	*85*	*0.32*	
Cyclohexanebutyric	*23*	*85*	*0.27*	0
Phenoxyacetic	160	106	1.51	90
	205	152	1.35	
p-Chlorophenoxyacetic	245	171	1.43	87

[a] 100 mg. added every 24 hours, beginning at 48 hours. [b] *Staphylococcus aureus* units except for numbers in italics, *Bacillus subtilis* units for numbers in italics. The two units are identical only for benzylpenicillin. [c] Control flask: flasks to which no precursor was added. The penicillin activity shown is the average of two or more flasks from the same run as the test flasks, of which two were usually run. [d] From paper chromatographic analysis. See Methods section.

α,β-Unsaturated acids differed in their behavior. Crotonic, β-chlorocrotonic and cinnamic acids had no precursor activity. α,β-Hexenoic acid was active, but it was found previously[5] that the penicillin produced had the chromatographic position of *n*-propylpenicillin. Therefore, β-oxidation of the acid probably accounted for its activity. The high precursor activity of sorbic acid is more difficult to explain. It does not appear likely that this compound was active as added, and β-oxidation would result in the production of crotonic acid, which is inactive. The penicillin produced (Fig. 1) has a chromatographic position close to that of penicillin F.

The β-substituted acids used were, as expected, good precursors, unless they were also α-substituted.

In order to obtain a clearer picture of the possible

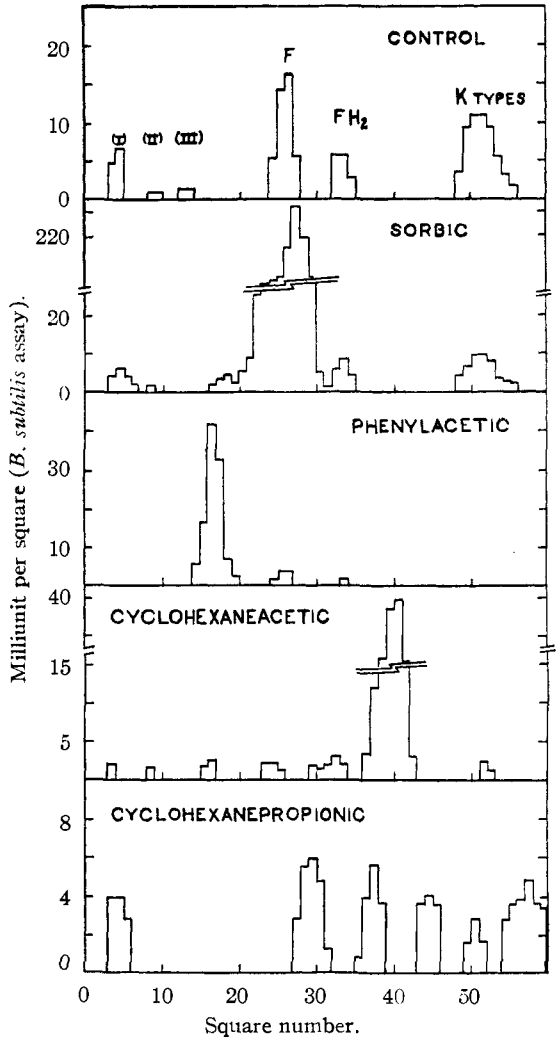

Fig. 1.—Position on the paper chromatogram of some of the penicillins formed by the addition of carboxylic acids to fermentations of *Penicillium chrysogenum* Q176. Precursors were added at a level of 100 mg. every 24 hours beginning 48 hours after inoculation. The sample (about 8 μl. of broth) was applied at square No. 4.

reasons for the differences in behavior of the active precursors, the rates of catabolism of a number of them were studied. Preliminary experiments on butyric, caproic and β,γ-hexenoic acids showed that the rate of disappearance of these acids was approximately constant throughout the fermentation period. The amount of an acid remaining 12 hours after addition was taken as a measure of its rate of disappearance. The figures in Table II show that, in general, α- or β-substituted acids were more slowly metabolized than unsubstituted acids. Since the α-substituted compounds are apparently unsuited as precursors for steric reasons, the reason for the superiority of β-substituted compounds as precursors appears to be, at least in part, that they are more stable to enzymatic oxidation. In Table III data are given on the amounts of various acids present at the end of fermentations. In these cases the acids were added at intervals in order to give the mold a continuing supply of precursor. It will again be noted

that the α- and β-substituted compounds were most resistant to oxidation.

The neutral and acid steam distillates of each of the broths were examined for breakdown products of the metabolized acids. Countercurrent distribution analysis and Duclaux-type distillation of the acid fractions failed to show significant quantities of acids other than the one added to the fermentation. A small proportion of acetic acid was usually present. The only product detected in the neutral volatile fractions following phenylhydrazine, iodoform and dichromate oxidation tests was methyl *n*-propyl ketone. This ketone appeared in the flasks to which caproic acid had been added. As much as 35% of the added acid appeared as ketone within 12 hours. The presence of this ketone indicates β-oxidation of the acid followed by decarboxylation of the β-ketocaproic acid.

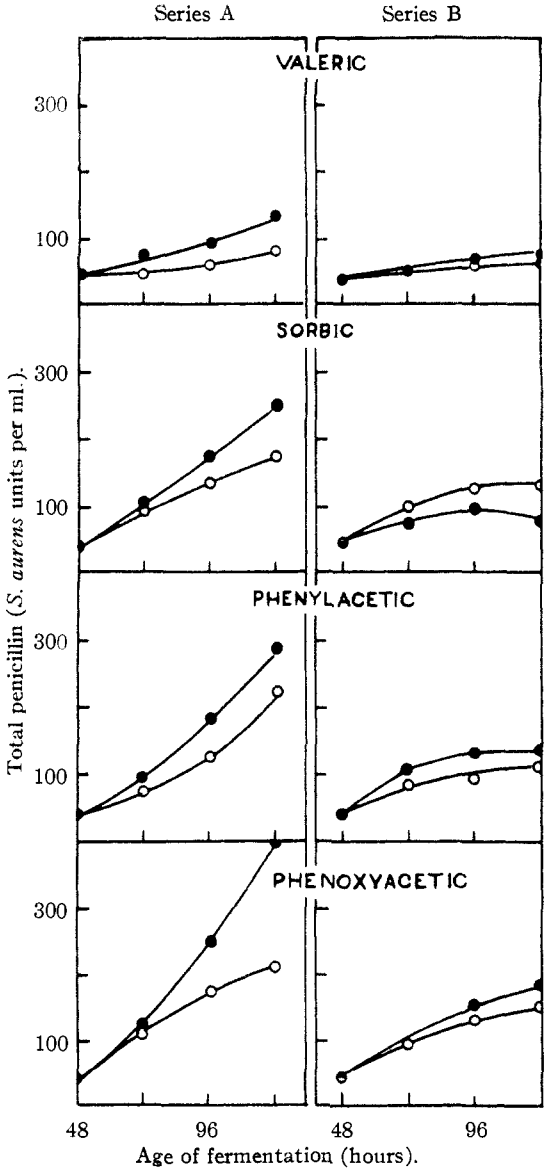

Fig. 2.—Effect upon yield of adding 100-mg. aliquots of precursor acids to synthetic medium fermentations of *Penicillium chrysogenum* Q176: added at 12 hour intervals beginning at 48 hours —●—, added at 24 hour intervals beginning at 48 hours —○—. Series A flasks grew at the normal *p*H of 7.7 to 7.8, yielding an average of 152 units per ml. at 120 hours in the absence of precursor. Series B flasks grew at a *p*H of 7.2 to 7.4, yielding an average of 106 units per ml. at 120 hours in the absence of precursor.

TABLE II

DISAPPEARANCE OF ACIDS IN FERMENTATIONS

Acid added	Amount added,[a] mM.	Recovery of added acid,[b] %
Propionic	1.01	12
Butyric	1.05	54
Valeric	0.95	52
α-Methylbutyric	1.05	82
Sorbic	0.68	10
Hydrosorbic	.62	10
β,γ-Hexenoic	.95	22
Caproic	.90	14
α-Ethylbutyric	.83	81
Phenylacetic	.78	78
Phenoxyacetic	.61	92
Cyclohexaneacetic	1.10	54
Caprylic	0.88	21

[a] Fermentations were about 48 hours old and at a *p*H of 7.45 to 7.55 when the acid was added. [b] Twelve hours after addition.

TABLE III

AMOUNT OF PRECURSOR IN THE FERMENTATION MEDIUM AT THE END OF THE FERMENTATION

Acid added	Total amount added,[a] mM.	Recovery of added acid,[b] %
Valeric	2.84	9.9
Sorbic	2.82	6.8
Caproic	3.30	13.0
α-Ethylbutyric	3.48	87.5
Phenylacetic	2.34	43.5
Phenoxyacetic	1.82	83.2
Cinnamic	2.46	4.9

[a] Acid was added in 3 equal portions at 48, 72 and 96 hours. [b] At 120 hours.

Figure 2 summarizes data obtained in a series of experiments in which precursor was added to the medium at intervals during the fermentation. It will be seen that more frequent precursor addition resulted in higher yields, and that fermentations carried out at higher *p*H values also gave higher yields. It is interesting to note that even in the case of phenylacetic and phenoxyacetic acids, when much of the added precursor was still present at the end of the fermentation (Table III), addition of more precursor resulted in higher yields.

Many of the precursors used were toxic to the fermentation. There was no consistent relation between the structure of an acid and its toxicity. The amount of acid which could be added before abnormalities in the fermentation occurred also varied. For example, cyclohexanepropionic acid caused a sharp decrease in yield whereas the same amount (100 mg. per flask) of phenylpropionic acid depressed the yield much less. One-tenth this amount of cinnamic acid caused abnormal pigmentation and a greatly reduced penicillin yield. The fermentation would tolerate the addition of at least three times as much α-ethylbutyric acid as caproic acid without affecting the yield. With the

present information it is not possible to assess the extent to which toxicity of the precursor (or its metabolic products) influences the final yield of penicillin in the fermentation.

Experimental

Fermentation Methods.—*Penicillium chrysogenum* Q176 was employed throughout as the fermentation organism. Fermentations, 100 ml. in volume, were conducted in 500-ml. erlenmeyer flasks at 25°, on a shaker. Except for the conditions noted below, the fermentation methods and media were essentially those used by Thorn and Johnson.[7]

The inoculation medium contained in grams per liter: glucose 40.0, ammonium lactate 21.0, potassium dihydrogen phosphate 3.0, calcium carbonate 15.0, and the standard salt mixture of Jarvis and Johnson,[8] omitting the copper sulfate. The 42-hour inoculum was diluted with an equal volume of sterile water to permit easier pipetting. Five ml. of diluted inoculum was transferred to each of the fermentation flasks. In some of the later work, the level of phosphate in the fermentation medium was raised to 5.0 g. per liter to aid in stabilizing the *p*H of the fermentation.

Chromatographic Methods.—The techniques described by Karnovsky and Johnson[9] were modified in certain respects. The room temperature humidification stage was omitted, and the ether used was not completely water-saturated.

Acids Used as Precursors.—Except as noted below, acids were obtained from the Eastman Kodak Company. Recrystallization or distillation, where necessary, were the only methods of purification. The cyclohexaneacetic, cyclohexanepropionic and cyclohexanebutyric acids were from experimental lots provided by the Dow Chemical Company, and were distilled before use. Boiling points obtained were: cyclohexaneacetic acid 242–244° (uncorrected), cyclohexanepropionic acid 166–168° at 30 mm., cyclohexanebutyric acid 187–189° at 30 mm.

α-Ethylbutyric acid was prepared from the corresponding alcohol, 2-ethylbutanol, obtained from Union Carbide and Carbon Corporation. The method used was a room temperature oxidation with 5% alkaline potassium permanganate.[10] The final yield of redistilled product was about 55% of theory, b.p. 188–190° (uncorrected).

α-Ethylcaproic acid was prepared in a like manner from 2-ethyl-*n*-hexanol, a Union Carbide and Carbon product. The final yield was about 15% of theory, b.p. 140–142° at 30 mm.

β-Chlorocrotonic acid was obtained through the courtesy of Dr. H. A. Lardy of the University of Wisconsin, and was prepared in his laboratory.

α,β- and β,γ-Hexenoic acids were prepared by the malonic acid–butyraldehyde condensation methods of Boxer and Linstead.[11]

All acids were added to the fermentations as their potassium salts in aqueous solution.

Analytical Methods.—Penicillin assays were by the Oxford cup assay procedure with *Staphylococcus aureus* H. as the test organism. The blotter disc assay with *Bacillus subtilis* spore inoculum, as described by Karnovsky and Johnson,[7] was used for some of the assays.

Volatile acids were determined as follows: An 80-ml. aliquot of filtered broth was distilled at *p*H 7. The first 50 ml. of distillate was collected as the neutral volatile fraction. The solution remaining was adjusted to *p*H 1.5 with phosphoric acid and was distilled with slow addition of water. The volume in the flask was maintained at 20 ml. while 100 ml. was collected in the volatile acid fraction. This fraction was titrated to the phenolphthalein end-point with 0.10 *N* potassium hydroxide. The resultant salt solution was set aside for further analysis, either by countercurrent distribution or Duclaux distillation.

The sample for countercurrent distribution was prepared by evaporating the above-mentioned salt solution to a volume of 2 ml., adding 0.2 g. of ammonium sulfate, acidifying and steam distilling the resultant solution. Ten ml. of distillate was collected. An aliquot of this distillate was used as sample for a ten-plate countercurrent distribution. The solvent system (distilled water and redistilled chloroform) served to separate six-carbon and higher acids from butyric and lower acids.

Duclaux distillation data were obtained by distilling the 100-ml. volume of the volatile acid fraction after adding 2.0 ml. of 1.0 *N* sulfuric acid. The first 10 ml. and three subsequent 25-ml. fractions of the distillate were titrated with 0.10 *N* potassium hydroxide. The fraction of the total titration appearing in each of these aliquots was then calculated. These values were compared with similarly determined constants for known amounts of the pure acid.

In most cases, the only acid which appeared to be present in addition to the one added to the fermentation was acetic acid.

Methyl ketone determinations were made on a 15-ml. aliquot of the neutral volatile fraction described above. This aliquot was treated with 5.0 ml. of 1.0 *N* sodium hydroxide, followed by 5.0 ml. of 0.1 *N* iodine potassium iodide solution. After 15 minutes, the contents of the flask were made acid with 6.0 ml. of 1.0 *N* sodium thiosulfate to the starch end-point. From the difference in titration between the sample and the water blank treated in a like manner, the number of millimoles of methyl ketone present was calculated.

Phenylacetic acid was determined by direct application of the Kappeler–Adler reaction previously utilized for the determination of benzylpenicillin.[12] An aliquot of broth containing 0.2 to 0.8 mg. of free phenylacetic acid was diluted to 10 ml. and, after acidification with 0.5 ml. of 40% phosphoric acid, extracted by shaking with 25 ml. of chloroform. The chloroform phase was drained into a 50-ml. erlenmeyer flask containing 0.5 g. of anhydrous sodium sulfate. A brief shaking served to dry the chloroform. A 20-ml. aliquot of the dried chloroform was pipetted into a 25 × 200 mm. Pyrex test-tube. Two glass beads were added and the chloroform was evaporated by heating the tube in a boiling water-bath. Careful agitation of the tube during the initial stages of evaporation was essential to avoid superheating.

Nitration and color development procedures were carried out in the tubes as described in the original publication.[10] Light absorption was measured with an Evelyn colorimeter at 580 mμ, with an 18-mm. Pyrex test-tube as the cell.

In estimating the amount of *phenoxyacetic, sorbic* and *cinnamic acids* present in broths, advantage was taken of the characteristic high ultraviolet absorption of these acids. A Beckman model DU spectrophotometer was used throughout. The background absorption due to metabolic products in the broth was taken into account in calculating the extinction due to the acid. In distilled water the molecular extinction and absorption maxima of these acids were: sorbic acid, E_M 25,000 at 257 mμ; cinnamic acid, E_M 22,000 at 259 mμ; phenoxyacetic acid, E_M 1,580 at 269 mμ. Broth from control flasks gave a spectrum with a broad peak between 280 and 295 mμ, and a minimum plateau between 250 and 260 mμ. At a dilution of one to a hundred, the respective extinctions were 0.275 and 0.238.

Precursor acids were added as the potassium salts when the fermentations reached the ages of 48, 72 and 96 hours. The samples for assay were taken at 120 hours and adjusted to *p*H 6.0 to 6.5 with strong phosphoric acid. If the chromatographic assay was not set up immediately, the samples were frozen. The amount of precursor acid added daily was normally 100 mg.

In studying the rates of disappearance of the acids, various procedures were employed. It was necessary to have the fermentations in a similar state if comparable results were to be obtained. At the age of 44 to 48 hours, the fermentations usually had a *p*H value between 7.45 and 7.55. Therefore rate studies were made only in flasks meeting these conditions. The usual amount of precursor salt (100 mg. of the acid neutralized with potassium hydroxide) was added to the flask. Twelve hours later the flask was removed from the shaker for analysis by the methods described above. Flasks were usually analyzed in duplicate.

To determine how much precursor was left in the fermentation at 120 hours, after the usual 3 daily additions had been made, the flasks were analyzed for the appropriate acid at the age of 120 hours.

(8) F. G. Jarvis and M. J. Johnson, THIS JOURNAL, **69**, 3010 (1947).

(9) M. L. Karnovsky and M. J. Johnson, *Anal. Chem.*, **21**, 1125 (1949).

(10) W. J. Hickinbottom, "Reactions of Organic Compounds," 2nd Edition, Longmans, Green and Company, New York, N. Y., 1948, p. 108.

(11) S. E. Boxer and R. P. Linstead, *J. Chem. Soc.*, 740 (1931).

(12) G. E. Boxer and P. M. Everett, *Anal. Chem.*, **21**, 670 (1949).

Each of the broths to which precursor had been added was tested qualitatively for the presence of ketone by the addition of an acidic solution of 2,4-dinitrophenylhydrazine. An aliquot of the neutral volatile fraction was analyzed quantitatively for methyl ketone as described above. Only when caproic acid was added to the flask, was an appreciable amount of ketone obtained. The structure assigned to this ketone was methyl *n*-propyl ketone. The quantitative iodoform method was mild enough that alcohols such as isopropyl alcohol reacted very slowly. The compound must then be a methyl ketone. The recrystallized 2,4-dinitrophenylhydrazone melted at 140–141°, which corresponded with the value given[13] for the hydrazone of methyl *n*-propyl ketone.

(13) R. L. Shriner and R. C. Fuson, "Identification of Organic Compounds," 2nd Edition, John Wiley and Sons, Inc., New York, N. Y., 1940, p. 221.

MADISON, WISCONSIN

[CONTRIBUTION FROM THE STERLING-WINTHROP RESEARCH INSTITUTE]

New Anthelmintics. The Synthesis of Some 9-Hydroxyalkyl- and Dihydroxylalkyl-aminoalkylaminoacridines

BY A. R. SURREY, C. M. SUTER AND J. S. BUCK

RECEIVED MARCH 14, 1952

A series of substituted 9-aminoacridines containing a primary or/and a secondary or tertiary hydroxyl group in the basic side chain has been prepared. The preparation of N-(2-hydroxypropyl)- and N-(3-hydroxybutyl)-1,3-propanediamine and N-(2,3-dihydroxypropyl)-ethylenediamine is also described. Some of the acridines have been found to possess marked anthelmintic activity.

The present communication reports the synthesis of 9-aminoacridine derivatives (Table I) containing a primary, or/and a secondary or tertiary hydroxyl group in the basic side chain. In addition, one compound having a morpholino group in the side chain is also included. Some of the compounds have been found to be unexpectedly effective as anthelmintic agents.

The acridines described in Table I were prepared from the corresponding 9-chloroacridine by the reaction with phenol to give the 9-phenoxyacridine (not isolated) followed by treatment with the appropriate primary–secondary diamine. The products were isolated as yellow crystalline dihydrochlorides containing varying amounts of water. It was found that in most instances complete removal of water from these salts is very difficult.

Two of the basic side chains, N-(2-hydroxyethyl)-ethylenediamine and 2-morpholinoethylamine employed in the present work are commercially available.[1] The preparation of N-(2-hydroxypropyl)- and N-(2-hydroxyisobutyl)-ethylenediamine have been reported by Kitchen and Pollard.[2] The 2-hydroxyethyl-,[3] 2-hydroxypropyl and 3-hydroxybutyl-1,3-propanediamines were prepared by condensing ethanolamine, 2-hydroxypropylamine and 3-hydroxybutylamine with acrylonitrile to give the substituted aminopropionitriles which were then reduced catalytically with Raney nickel. The reaction of glycidol with ethylenediamine gave N-2,3-dihydroxypropylethylenediamine which was used without purification. The same diamine was also obtained directly from glycerol α-monochlorohydrin by reaction with ethylenediamine and potassium hydroxide.

Of the compounds listed in Table I, 9-(2-hydroxyethylaminoethylamino)-2-methoxyacridine,[4] 6-chloro-9-(3-(2-hydroxyethylamino)-propylamino)-2-methoxyacridine and 9-(2-(2,3-dihydroxypropylamino)-ethylamino)-2-methoxyacridine appear to be the best anthelmintic agents[5] when tested in Swiss mice against the oxyurid worms, *Aspicularis tetraptera* and *Syphacea obvelata*.

Experimental

2-Hydroxypropylaminopropionitrile.—Acrylonitrile (66.5 g.) was added dropwise with stirring over a period of 90 minutes to 141.2 g. of monoisopropanolamine (temperature below 30°). After stirring for five additional hours the reaction mixture was heated on the steam-bath for 30 minutes and then allowed to stand overnight at room temperature. The product was fractionally distilled, 40 g. (25%); 111–113° at 0.6 mm. The product solidified on standing, m.p. 47–51°.

Anal. Calcd. for $C_6H_{12}N_2O$: N (basic), 10.93. Found: N (basic), 11.01.

3-Hydroxybutylaminopropionitrile.—Prepared as above in 74% yield, b.p. 132–135° at 0.8 mm., $n^{25}D$ 1.4615.

Anal. Calcd. for $C_7H_{14}N_2O$: N (basic), 9.85. Found: N (basic), 9.98.

N-2-Hydroxypropyl-1,3-propanediamine.—2-Hydroxypropylaminopropionitrile (38 g.) in 200 ml. of ammoniacal ethanol (approx. 12%) was reduced catalytically with Raney nickel at 120° and an initial hydrogen pressure of 1180 pounds. The product 19 g. (48%) distilled at 105–110° at 1.5 mm., $n^{25}D$ 1.4747.

Anal. Calcd. for $C_6H_{16}N_2O$: N, 21.30. Found: N, 21.50.

N-3-Hydroxybutyl-1,3-propanediamine.—Prepared as above in 58% yield, b.p. 95–100° at 0.25 mm., $n^{25}D$ 1.4738.

Anal. Calcd. for $C_7H_{18}N_2O$: N, 19.00. Found: N, 18.98.

N-(2,3-Dihydroxypropyl)-ethylenediamine.—Glycidol (17.5 g.) was added dropwise with stirring at 70–80° to 91 g. of ethylenediamine over a period of 90 minutes. Fractional distillation gave a crude product, b.p. 171° at 0.4 mm., which solidified on standing.

Glycerol α-monochlorohydrin (110 g.) was added dropwise with stirring over a period of 90 minutes to a mixture of 54 g. of potassium hydroxide in 400 g. of ethylenediamine at 70–80°. After the addition was complete the mixture was allowed to stand overnight at room temperature, filtered, and the filtrate was distilled under reduced pressure. After

(1) Samples of these amines were obtained through the courtesy of the Carbide and Carbon Chemicals Corporation.

(2) L. J. Kitchen and C. B. Pollard, *J. Org. Chem.*, **8**, 342 (1943).

(3) A. R. Surrey and H. F. Hammer, THIS JOURNAL, **72**, 1814 (1950).

(4) The compound can be regarded as an ethylenediamine derivative, N¹-(2-hydroxyethyl)-N²-(2-methoxy-9-acridyl)-ethylenediamine.

(5) The authors are indebted to Dr. E. W. Dennis and Dr. D. A. Berberian of this Institute for the testing of these compounds, the details of which will be published elsewhere.