

confirming the previous finding that the *cis* is converted to *trans*-form on treatment with thionyl chloride.

*Anal.* Calcd. for  $C_{10}H_{19}ON$ : N, 8.28. Found: N, 8.36.

The anilide prepared in a similar manner melted at  $192^\circ$  and showed no depression in melting point when mixed with anilide prepared from *cis*-synthetic acid.

*Anal.* Calcd. for  $C_{16}H_{23}ON$ : N, 5.71. Found: N, 5.87.

### Summary

Through the use of a number of fractionation procedures in a continuation of the study of acids from California petroleum, the following naph-

thenic acids have been isolated and identified for the first time from petroleum: 2-methylcyclopentanecarboxylic, 3-methylcyclopentanecarboxylic, 2,3-dimethylcyclopentanecarboxylic, cyclohexanecarboxylic, and *cis*-2,2,6-trimethylcyclohexanecarboxylic acids.

The following acids previously reported by Nenitzescu from European petroleum were isolated also from California acids: cyclopentanecarboxylic, cyclopentanecarboxylic and 3-methylcyclopentanecarboxylic acids.

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[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY AND THE PURDUE RESEARCH FOUNDATION, PURDUE UNIVERSITY]

## New Methods for the Resolution of Enantiomorphs. II. Liquid-Liquid Extraction<sup>1</sup>

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In the first paper of this series Bailey and Hass<sup>3</sup> showed that resolution of enantiomorphs could be brought about by their conversion to volatile diastereoisomers followed by rectification and re-conversion to the original compounds. This report deals with resolution by means of distribution between two liquids.

The use of liquid-liquid extraction to separate diastereoisomers depends upon the fact that these compounds have different distribution ratios between two incompletely miscible solvents. The concentrations of one of the diastereoisomers, A, in the solvents S and S' may be expressed as  $C_A$  and  $C'_A$ , respectively, and those of the other, B, as  $C_B$  and  $C'_B$ . The relative ease with which the diastereoisomers may be separated is then determined by the quantity  $C'_A C_B / C_A C'_B$ , which is the ratio of the two distribution ratios  $C'_A / C_A$  and  $C'_B / C_B$ . If this quantity is unity, no separation is possible, and the greater its deviation from unity, the more easily can separation be made.

Fractional distribution of two solutes between two solvents may be carried out either by a multiple contact method, for which separatory funnels were used, or by a countercurrent contact method, using a column. Both methods were successfully applied in this investigation.

(1) Based on a doctoral thesis, Purdue University. Presented before the Division of Physical and Inorganic Chemistry at the Buffalo meeting of the American Chemical Society, September, 1942.

(2) Eli Lilly Research Fellow, 1941-1942.

(3) Bailey and Hass, *THIS JOURNAL*, **63**, 1969 (1941).

### Experimental

**Method and Apparatus.**—The scheme used for fractional distribution by the multiple contact method was that described by Hunter and Nash.<sup>4</sup> The solutions in each case were shaken vigorously in separatory funnels for one-hour periods to ensure the attainment of equilibrium.

Separations by the countercurrent contact method were made in an extraction apparatus represented schematically in Fig. 1. Brucine *d*-mandelate and brucine *l*-mandelate were the compounds chosen for trial. The column was designed for operation in a manner entirely analogous to that of a rectification apparatus. Solute in water and in chloroform correspond, respectively, to the vapor and liquid phases. Removal of water from the solute and its replacement by chloroform corresponds to condensation, and removal of chloroform and its replacement by water corresponds to evaporation. The water solution after leaving the top of the column is raised by a water vapor lift D, to a boiler E, provided with an overflow tube H. The solution is concentrated in the boiler and the concentrated solution flows through the overflow tube back to the column. On its way to the column it passes through a section of 10-mm. Pyrex tubing, M, packed with glass helices. At the same time chloroform, which has distilled from the chloroform boiler, C, passes over the helices so that some transfer of brucine mandelate from the aqueous phase to the chloroform occurs. Similarly in the packed section G, concentrated chloroform solution from C is put into contact with distilled water from E. The column A was packed with a nickel screen spiral as described by Lecky and Ewell.<sup>5</sup> The diameter of the column was 28 mm., the packed height was 95 cm., and there was one turn of 60-mesh screen per cm.

Analyses of samples taken from the water boiler showed a large excess of mandelic acid over brucine. Evidently,

(4) Hunter and Nash, *Ind. Eng. Chem.*, **27**, 836 (1935).

(5) Lecky and Ewell, *Ind. Eng. Chem., Anal. Ed.*, **12**, 544 (1940).

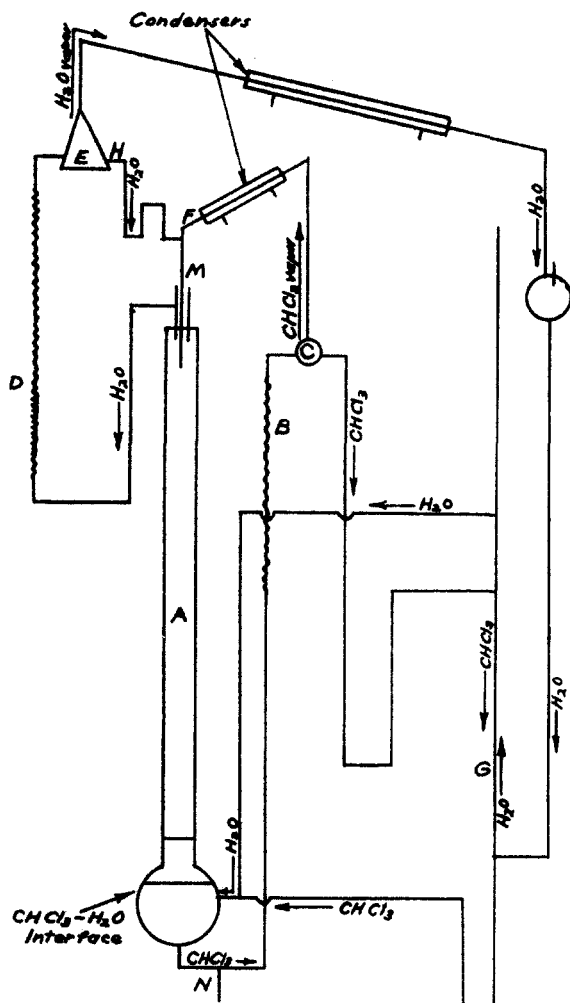


Fig. 1.—Countercurrent extraction apparatus.

the chloroform in passing through the column extracted from the aqueous phase some of the brucine which had resulted from hydrolysis. The device used to diminish the extent of the extraction is shown in Fig. 2, which is mostly self-explanatory. Water under a constant head is delivered through a partially opened stopcock at a uniform rate to the bucket C. As the float F rises, mercury flows from A into R, forcing chloroform solution of brucine into the column, and simultaneously the flow of mercury from S into B withdraws chloroform solution from the bottom of the column. A 75–100 ml. sample was collected in S every twenty-four hours. The presence of excess brucine in the chloroform which passed through the column reduced the excess mandelic acid in the aqueous phase from 400 to 25%.

**Experiment 1—Resolution of *dl*-Mandelic Acid by Multiple Contact Method of Fractional Distribution.**—An aqueous solution (105 ml.) of brucine mandelate (6 g.) was prepared by dissolving equivalent weights of mandelic acid and brucine in hot water. This aqueous solution was shaken for one hour with chloroform (15 ml.). After the two phases had separated, the chloroform phase was transferred to a separatory funnel containing water (105

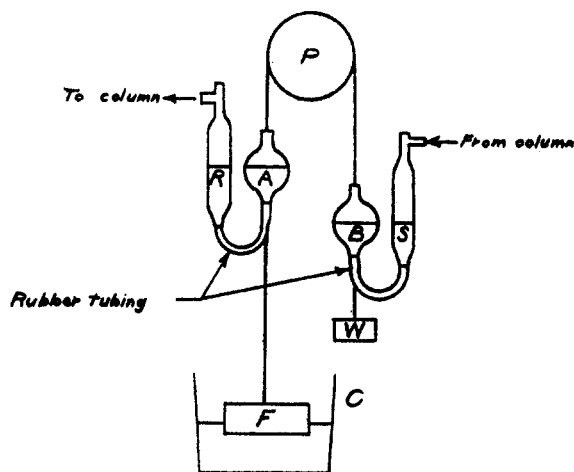


Fig. 2.

ml.), and chloroform (15 ml.) was added to the aqueous solution remaining in the first funnel. Both funnels were shaken for one hour.

The aqueous phase in the first separatory funnel was made ammoniacal and the brucine which precipitated removed by filtration. The solution of ammonium mandelate was concentrated under vacuum (18 mm.) to a volume of about 40 ml. The concentrate was acidified with sulfuric acid and extracted three times with 40-ml. portions of ether. The ether extracts were combined and extracted with 5 ml. of water. The ether was removed from the ether phase and the residue taken up with water. The aqueous solution was clarified by treatment with charcoal followed by filtration. After the optical activity of the solution had been measured, the concentration of mandelic acid was determined by titration with standard sodium hydroxide (0.2 *N*) using phenolphthalein indicator  $\alpha = -0.11$ ,  $c = 1.29$ ,  $l = 2$ ,  $[\alpha]^{20}_D -4.3^\circ$ .

The chloroform phase was extracted with 50 ml. of dilute sulfuric acid (1 *N*) and with 50 ml. of water. The aqueous portions were combined and treated in the manner described in the preceding paragraph to give an aqueous solution of mandelic acid  $\alpha = 0.09^\circ$ ,  $c = 1.17$ ,  $l = 2$ ,  $[\alpha]^{20}_D 3.9^\circ$ .

**Experiment 2—Partial Resolution of *dl*-Mandelic Acid by Countercurrent Method of Fractional Distribution.**—Brucine (25 g.) and mandelic acid (9.6 g.) were dissolved in chloroform (150 ml.). This solution was shaken for several minutes with water (1000 ml.) to distribute the salt between both phases. The chloroform solution was diluted to 250 ml. and put into the column. The aqueous solution was added and the column was finally filled with water. The heaters were turned on and the run started. Samples were taken at twenty-four-hour intervals from the water boiler and analyzed according to the procedure described in Experiment 1. After the sixth sample had been collected, the run was stopped and the chloroform phase collected and analyzed. The specific rotations of the mandelic acid solutions obtained from the samples were  $-6.2$ ,  $-4.0$ ,  $-2.0$ ,  $-1.3$ ,  $+0.5$ ,  $+3.7^\circ$ . The chloroform phase yielded a solution of mandelic acid having a specific rotation of  $+16.0^\circ$ . The specific rotation of pure *d*-mandelic acid is  $157^\circ$ , so that this latter solution contained 55% of the *d*-form and 45% of the *l*-form.

**Experiment 3—Partial Resolution of *dl*-Acetylmandelic Acid by the Distribution of Brucine *dl*-Acetylmandelate between Water and Chloroform.**—Acetylmandelic acid was prepared by the action of excess acetyl chloride on *dl*-mandelic acid.<sup>6</sup> Equivalent weights of the acid (6.0 g.) and of brucine (12.2 g.) were dissolved in hot water (400 ml.) and the aqueous solution was cooled and shaken for one hour with 16 ml. of a solution of brucine *dl*-acetylmandelate (6.18 g.) in chloroform. A four-fold extraction was carried out.

Acetylmandelic acid was obtained from the aqueous phase of the first funnel by the procedure described in Experiment 1, except that the residue after the removal of ether was taken up with alcohol. The chloroform phase of the fourth funnel was extracted with excess dilute ammonia (1 *N*) and the ammoniacal extract treated to yield an alcoholic solution of acetylmandelic acid. For the acetylmandelic acid from the water phase  $\alpha = -0.40$ ,  $c = 2.43$ ,  $l = 1$ ,  $[\alpha]^{20D} = -16.4^\circ$  and for the acid from the chloroform phase  $\alpha = +0.47$ ,  $c = 5.8$ ,  $l = 1$ ,  $[\alpha]^{20D} = +8.1^\circ$ .

**Experiment 4—Partial Resolution of *o*-Nitromandelic Acid.**—*o*-Nitromandelic acid was prepared by hydrolysis of the cyanohydrin made by treating a suspension of *o*-nitrobenzaldehyde in acetic acid with potassium cyanide.<sup>7</sup> A solution of brucine *o*-nitromandelate (6.0 g.) in water (200 ml.) was prepared by dissolving equivalent weights of the acid and base in hot water. The aqueous solution was shaken for one hour with 23 ml. of chloroform. Alcoholic solutions of *o*-nitromandelic acid were obtained by the procedures described in the preceding experiment. The specific rotation of the acid obtained from the aqueous phase was  $+1.4^\circ$  and for the acid from the chloroform phase  $-2.1^\circ$ .

**Experiment 5—Partial Resolution of *o*-Nitro-acetylmandelic Acid.**—This acid was prepared by acetylating *o*-nitromandelic acid with excess acetyl chloride. The melting point of the crystals was  $100^\circ$ . An aqueous solution (300 ml.) of brucine *o*-nitro-acetylmandelate (2.66 g.) was shaken for one hour with 20 ml. of chloroform. Alcoholic solutions of the free acid were obtained by the procedures of Experiment 3, and had specific rotations of  $-2.3$  and  $+1.6^\circ$ . The solution from the aqueous phase was levorotatory.

**Experiment 6—Partial Resolution of  $\alpha$ -Formamido- $\alpha$ -phenylacetic Acid.**— $\alpha$ -Amino- $\alpha$ -phenylacetic acid (15.1 g.) was formylated by treatment with a mixture of 90% formic acid (180 ml.) and acetic anhydride (60 ml.). The mixture was allowed to stand for two hours and was then diluted with water (400 ml.). The dilute solution was evaporated under vacuum (18 mm.) to dryness. The residue was dissolved in hot water (150 ml.) and crystals of

$\alpha$ -formamido- $\alpha$ -phenylacetic acid precipitated when the solution was kept at  $0^\circ$  for six hours. The crystals were purified by re-crystallization from hot water and when dry melted at  $185^\circ$ . An aqueous solution (200 ml.) of brucine  $\alpha$ -formamido- $\alpha$ -phenylacetate (6.4 g.) was shaken for one hour with 100 ml. of chloroform. The procedures described in Experiment 3 were used to give alcoholic solutions of the acid which had specific rotations of  $-2.3$  and  $+1.6^\circ$ . The solution from the aqueous phase was levorotatory.

### Discussion

The results of the preceding experiments show that it is possible to obtain partial resolution of enantiomorphs by liquid-liquid extraction. Although only diastereoisomeric salts were studied, there appears to be no reason why the method should not be applicable to any diastereoisomers. However, the distribution of a salt between water and a non-aqueous solvent in which little ionization occurs may very well prove to be the most favorable case, for the relative ease of forming ion pairs in the non-aqueous solution will presumably be different for the two diastereoisomers, but in the aqueous solution the interaction of the free ions will be very nearly the same.

The practical utilization of this method is dependent upon the development of an efficient countercurrent liquid-liquid extraction apparatus. It is estimated that a column having an efficiency of from 60 to 300 theoretical stages would be needed to effect 90% resolution of the salts studied.

**Acknowledgment.**—It is a pleasure to express our gratitude to Mr. J. K. Lilly, who generously defrayed the cost of this investigation.

### Summary

Partial resolution of five racemic acids was obtained by fractional distribution of the brucine salts of these acids between water and chloroform.

Ten per cent. resolutions of mandelic acid and acetylmandelic acid were obtained by multiple extractions in separatory funnels.

A countercurrent liquid-liquid extraction column was constructed and utilized in the resolution of mandelic acid.

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(6) "Organic Syntheses," Coll. Vol. 1, 12 (1932).

(7) McKenzie and Stewart, *J. Chem. Soc.*, 104 (1935).