# Correlation of the Oil-Water Distribution Ratios of Some Substituted Acids with Their Bacteriostatic Properties[1]

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As Degering and Goshorn (1) have shown that the bacterial effect of phenylalkanoic acids increases with increasing oil solubility, this work was undertaken to see if such a correlation holds for other acids and for solutions other than aqueous.

## Experimental

Twenty-five milliliters of the 0.005 M acid solution was placed in an oil sample bottle and 25 ml. of U.S.P. cottonseed oil added. This mixture was shaken at room temperature for 18 hours. The oil and water emulsion was separated by centrifuging at approximately 2000 r.p.m. Ten milliliters of the aqueous layer was titrated with 0.0096

Table I.—The Distribution Ratios of Phenylalkanoic Acids in 5% Ethylene Glycol,* in 10% Ethanol,* and in Water, and Their Bactericidal Properties in Water

| Acid | Ratio | | | Bactericidal Tests† (1) | |
|---|---|---|---|---|---|
| | 5% Glycol | 10% Ethanol | Water (1) | Colon. | Aureus |
| Alpha Series— | | | | | |
| Benzoic | 5.1 | 5.0 | .......... | 1/900 | 1/100 |
| Phenylacetic | 2.0 | 1.8 | 1.8 | 1/800 | 1/700 |
| Phenylpropionic | 6.2 | 5.8 | 5.6 | 1/950 | 1/650 |
| Phenylbutyric | 20.9 | 12.6 | 14.4 | 1/1600 | 1/1600 |
| Phenylvaleric | 74.0 | 32.1 | 26.4 | 1/2000 | 1/2300 |
| Omega Series— | | | | | |
| Phenylpropionic | 9.1 | 6.8 | 6.6 | 1/1000 | 1/1200 |
| Phenylbutyric | 25.2 | 19.3 | 14.8 | 1/1300 | 1/1400 |
| Phenylvaleric | 42.7 | 52.0 | 28.8 | 1/2500 | 1/2500 |

*Highest bacteriostatically inactive concentrations (4).
†Maximum effective dilution is expressed in gm./ml.

[1]This is the sixth of a series of articles on the effect of pH and substituent groups on the bacteriostatic and bactericidal properties of certain antiseptics. The other articles are: 1. Antiseptic and bactericidal action of benzoic acid and inorganic salts. Ind. and Eng. Chem. **30**:646; 2. Preparation of alpha-phenylalkanoic acids and a study of their bactericidal and physical properties. Journ. Amer. Phar. Assoc. **27**:865-870; 3. Bactericidal properties of commercial antiseptics. Ind. and Eng. Chem. **31**:742; 4. The effect of para-substituents on the bacteriostatic properties of phenylacetic acid. Journ. Amer. Phar. Assoc. **28**:514-519; 5. A further study of the effect of pH on the bactericidal properties of some commercial antiseptics. Ind. and Eng. Chem. Vol. 43 (1940).

sodium hydroxide, using phenolphthalein as the indicator. Nitrogen was bubbled into the solution to prevent carbon dioxide absorption. Two samples of each acid were shaken, and two titrations were made on each sample. The results checked to 0.10 ml. of base. From the known normality of the original solution and the calculated normality of the extracted solution the ratio can easily be calculated. A blank was run and the normality of the acid solution produced by the solvent in contact with the oil subtracted from the titrated normality.

TABLE II.—Substituted Mandelic Acids—Aqueous Solutions*

| Acid | Ratio | Bact. Tests† (2) |
|---|---|---|
| Mandelic | 0.51 | 1 |
| *p*-methylmandelic | 0.28 | 1-2 |
| *p*-ethylmandelic | 0.67 | 1-4 |
| *p*-n-propylmandelic | 2.12 | 1 or less |
| *p*-iso-propylmandelic | 1.78 | 1 |
| *p*-n-butylmandelic | 5.66 | 1 or less |
| *p*-sec-butylmandelic | 3.85 | 1-2 |
| *p*-ter-butylmandelic | 3.19 | less than 1 |
| *p*-ter-amylmandelic | 7.76 | 1 or less |
| 2,4,6-trimethylmandelic | 0.78 | 1.4 |
| *p*-chloromandelic | 0.56 | 2.4 |

*Prepared by J. L. Riebsomer of DePauw University.
†Based on mandelic acid as 1.

TABLE III.—Para-Substituted Phenylacetic Acids—Aqueous Solutions*

| Acids | Ratio | Bacteriostatic Tests† | |
|---|---|---|---|
| | | Staph. Aureus | Esch. Coli. |
| Hydroxyphenylacetic | 0.09 | Less than 1/1000 | |
| Aminophenylacetic | 0.13 | 1/1500 | 1/2000 |
| N-Chloroacetylaminophenylacetic | 0.25 | 1/3000 | 1/3000 |
| N-Acetylaminophenylacetic | 0.28 | Less than 1/1000 | |
| Phenylacetic | 1.85 | 1/1000 | 1/1500 |
| Nitrophenylacetic | 2.71 | 1/2000 | 1/610 |
| Methoxyphenylacetic | 2.79 | 1/1000 | 1/1500 |
| Ethoxyphenylacetic | 8.28 | 1/1000 | Less than 1/1000 |
| Ethylphenylacetic | 36.1 | 1/2500 | 1/1000 |
| Chlorophenylacetic | 23.7 | 1/3000 | 1/1000 |
| Iodophenylacetic | 42.9 | 1/4000 | Less than 1/1000 |
| Bromophenylacetic | 73.1 | 1/5000 | Less than 1/1000 |

*Acids prepared by W. A. Bittenbender.
†Tests indicate the highest dilution negative growth and were made by Professor P. A. Tetrault.

## Conclusions

From Table I it is readily seen that, with one exception which is well within the accuracy of the tests, the relative oil-solvent distribution ratio parallels the bactericidal action. Benzoic acid seems to be out of place but follows the general rule that the first member of an homologous series shows unusual properties.

In Table II there is practically no correlation between the distribution ratios and the bacteriological tests except in the first three members. Even here mandelic acid shows a higher distribution ratio but lower bacterial action. This table and Table III bring out the point that the ratios of alkyl-, alkoxy-, and similar substituted compounds cannot be compared with compounds substituted with halogens, or with nitro-, hydroxy-, and other polar groups.

Table III presents fairly consistent data. The ratios show that the *p*-hydroxy-derivative will be the weakest, and this is borne out by the tests. It also shows *p*-nitrophenylacetic acid to be better than phenylacetic acid. Most striking of all the predictions is the curious behavior of the halogen-substituted compounds. The bromo-phenylacetic acid is shown to be better than either the chloro- or the iodophenylacetic acid. The results with *Esch. coli* are quite varied.

In general it can be seen that the oil-water distribution ratio is helpful in predicting the relative bacteriostatic action of certain types of closely related compounds, thus serving as a confirmatory check on standard bacteriostatic or bactericidal tests, but the principle does not appear to be applicable to all types of bacteriostatic agents. The correlation seems to apply, in general, to members of the same homologous series or to members of the same solubility group.

## Bibliography

1. Goshorn, R. H., and Ed. F. Degering, 1938. Preparation of alpha-phenylalkanoic acids and a study of their bactericidal and physical properties. Journ. Amer. Pharm. Assoc. **27**:865.

2. Riebsomer, J. L., et al, 1938. The preparation of substituted mandelic acids and their bacteriological effects. Journ. Amer. Chem. Soc. **60**:1015, 2974.

3. Tetrault, P. A. Private communication.