

The melting point was 104° C. This salt appeared to be insoluble in vegetable oils and very little soluble in ethylene glycol and di-ethylene-glycol.

Attempts to prepare stable neutral lead and mercury salts of menthylsuccinic acid did not prove to be successful, if the above-mentioned method of preparation was employed. The lead salts obtained were basic salts and no definite salt could be isolated. The mercury compound obtained was found to be a menthylsuccinic acid mercuric chloride. When prepared from $\frac{1}{6}$ mol of sodium menthyl-succinate solution and $\frac{1}{10}$ mol HgCl in aqueous solution, a yield of 41.1 Gm. of a white salt was obtained. After two recrystallizations from methanol a product with the following assay was obtained.

Found:

40.37% Hg

7.14% Cl

Calculated:

40.83%

7.22% for

$\text{CH}_2.\text{CO}_2\text{C}_{10}\text{H}_{19}$

|
 $\text{CH}_2.\text{CO}_2.\text{HgCl}$

This salt was easily soluble in methanol, acetone and iso-propanol.

SUMMARY.

1. The bismuth, manganese, silver and mercury salts of menthylsuccinic acid were prepared and investigated for their faculty to dissolve in vegetable oils.
2. The bismuth and manganese salts were soluble, while the silver and mercury salts obtained were not.
3. The toxicity of the bismuth compound was approximately 200 mg. of metallic bismuth per Kg. body weight, when injected intramuscularly.

LITERATURE REFERENCES.

- (1) Zelikow, *J. Russ. Phys.-Chem. Gesell.*, 34, 724; *Ber.*, 37, 1379 (1904).
- (2) Arth, *Ann. chim. phys.* (6), 7, 433 (1886).
- (3) Lauter and Braun, *JOUR. A. PH. A.*, 25, 394 (1936).

THE SOLUBILITY OF LYOPHILE GELATINS.*

BY L. F. TICE.¹

The ordinary commercial gelatins commonly employed are not, generally speaking, soluble to any appreciable extent in water at 25° C. When placed in water such gelatins absorb water to a considerable degree and become greatly swollen, but the individual particles originally present do not lose their identity nor have their number reduced. What has been interpreted as partial solubility by some workers is probably best explained as the exosmosis of lower molecular weight hydrolysis products from the swollen gelatin particle and their subsequent presence in the external aqueous medium. This seems borne out by the fact that gelatins of very low Bloom test, which are considerably hydrolyzed during manufacture, are actually found to slowly but completely dissolve in water at 25° C.

* Presented before the Scientific Section, A. PH. A., New York meeting, 1937.

¹ Department of Pharmacy, Philadelphia College of Pharmacy and Science.

During the past year the writer has had the opportunity to study several samples of gelatin prepared in dry solid form by the so-called "Lyophile" process. It is the purpose of this paper to report the unusual features of this material and to discuss the effect of a number of variables on its solubility in water.

The "Lyophile" process has been developed by the efforts of several workers. Reichel, Masucci and Boyer (1) perfected a method for its application to serums and biologicals in bulk. Flosdorf and Mudd (2) modified the apparatus used in the production of bulk material in such a manner as to make it applicable to individual ampul packages. This apparatus has since been improved and perfected by Dr. John Reichel and his co-workers at the research laboratories of Sharp & Dohme. The process in brief consists of the rapid freezing of an aqueous solution followed by dehydration of the frozen material under high vacuum. The solution is frozen at -78°C . in a shell in very large Pyrex or metal bulbs which are connected to a large bulb condenser at a distance from the containers. This condenser is surrounded by a refrigerant bath of solid CO_2 in acetone or methyl cellosolve. The material is kept in the frozen state solely by the rapid sublimation *in vacuo* of water vapor from its surface. The outer surfaces of the containers are surrounded merely by the air of the room at ordinary temperature during all of the dehydration process.

In preparing a series of lyophile gelatins, three samples of commercial gelatin were selected as follows:

	Precursor.	Jelly Strength.	pH.
#1	Bone	250 Bloom	6.0
#2	Porkskin	250 Bloom	4.4
#3	Porkskin	50 Bloom	4.4

Gelatins #1 and #2 represent high test gelatins from the two different precursors whereas #3 was a low test gelatin from the same precursor as #2. A corresponding low test gelatin from bone was not employed due to the unsatisfactory color and clarity observed in such material. Using these representative gelatins, warm solutions of varying concentrations were prepared and these were then frozen and dried by the lyophile process. In each case solutions whose concentrations were beyond a certain point could not be processed due to gelation of the material before it could be frozen. As would be expected, the low test gelatin could be prepared in higher concentration than the high test samples without gelation resulting during freezing.

The physical properties of lyophile gelatin are quite unusual. It is a pure white, extremely porous solid, easily compressed, very hygroscopic but not deliquescent, and exhibiting solubility features entirely different from ordinary gelatin. When examined microscopically, it appears to possess a mesh-like structure, the size of the openings decreasing with increased concentration of the solution processed. It is not believed that this observed structure is peculiar to gelatin since other materials yield porous bodies in lyophile form which are not subject to gelation. Lyophile gelatin is very bulky and porous making its apparent density extremely small. This is particularly true in cases where very dilute solutions were dried, the more concentrated solutions giving a denser product.

By far the most interesting feature of several of these gelatins was their complete and rapid solubility in water at $25\text{--}28^{\circ}\text{C}$. When one considers that in each case, regardless of the solution processed, the resultant product was pure dry gelatin this solubility is indeed unusual. If all of the lyophile gelatins had been uniformly soluble or if only those prepared from a low test gelatin were soluble the explanation would have been relatively simple, since it would be easily explained on the basis of increased surface area only. This not being the case an attempt was made to correlate the many variables in so far as their effect on the solubility was concerned.

In all of the series of gelatins prepared some of the gelatin was soluble in water; the difference noted was that in some cases it was completely soluble in a rela-

tively short time whereas in others hydrated masses were formed which persisted for a long period without going into solution.

The following experimental samples were prepared in bulk for examination:

A	0.2% gelatin #1	H	1.0% gelatin #2
B	0.5% gelatin #1	I	2.0% gelatin #2
C	1.0% gelatin #1	J	0.1% gelatin #3
D	2.0% gelatin #1	K	0.2% gelatin #3
E	0.1% gelatin #2	L	0.5% gelatin #3
F	0.2% gelatin #2	M	1.0% gelatin #3
G	0.5% gelatin #2	N	2.0% gelatin #3
		O	5.0% gelatin #3

In addition, several identical samples were processed in individual "vacuoles" since, in this type of evacuated ampul, solution of the material may be accomplished without preliminary exposure to air or moisture. This was done to determine the effect, if any, of these variables on the product.

The results are summarized as follows:

In Samples J to O, inclusive, wherein the low Bloom porkskin was used, all were easily soluble in water at 25° C. This would be expected due to the inherent solubility of the initial gelatin in water at the same temperature. The effect here is plainly one of increased surface, resulting in easy and rapid solution.

With the samples prepared with #1 and #2 gelatin, however, considerable differences were noted compared with both #3 and each other. Samples E and F corresponding to 0.1% and 0.2% of #2 were easily soluble in water at 25° C. while the higher concentrations required standing over night at 25° C. in order to completely dissolve. At 28° C. Samples G and H, representing 0.5% and 1.0% of #2, were easily soluble; and at 30° C. Sample I dissolved without difficulty. With samples prepared with gelatin #1 very poor solubility was found. For example, A prepared from 0.2% of #1 required a temperature of 28° and considerable time for solution to be complete. B, C and D were not completely soluble except when the temperature approached 35° C.

The solubility of samples made with gelatin #2 which was not observed in the case of the initial commercial material even on long standing, is probably explainable on the basis of increased surface area coupled with the theory that the rapid freezing prior to dehydration did not permit extensive aggregation of the gelatin molecules to the extent that an insoluble aggregate was reached. Since gelation is largely a process of molecular aggregation to form a network structure and one requiring an appreciable time for completion, then such aggregation would be materially prevented if cooling was rapid and the concentration of gelatin in solution was relatively low.

The greater difficulty experienced in dissolving those samples prepared from more concentrated solutions and, consequently, containing many more gelatin molecules is probably best explained by assuming that a greater degree of aggregation had taken place prior to complete freezing.

The difference in behavior of samples from gelatin #1, as compared with those from gelatin #2, may be explained by the fact that the p_H of #1 gelatin (6.0) lies so near to its isoelectric point, approximately 4.7, that it possessed less affinity for water and was, consequently, less soluble than corresponding samples made from #2 whose p_H (4.4) is considerably removed from its isoelectric zone of about 7.5.

In addition to the foregoing results the following observations were also made concerning the behavior of lyophile gelatin:

1. The ease of solution is increased when the ratio of solvent to gelatin is small. Since lyophile gelatin is practically anhydrous and possesses a marked affinity for moisture, when only a small amount of water is employed a measurable elevation in temperature results, thus facilitating solution.

2. The use of rapid agitation during the addition of water greatly aids solution. This is a factor favoring the individual evacuated ampuls as when water is injected into them extremely vigorous agitation results.

3. Compression of the porous material does not alter its solubility but retards the rate somewhat due to a decrease in surface area exposed to the solvent.

4. Exposure to air was found to reduce the ease with which a given sample could be dissolved.

5. The lyophile process does not apparently alter the jelly strength of the gelatin so treated. When 5 per cent gels of the initial gelatins were prepared and compared with 5 per cent gels made with the lyophile product no difference in jelly strength could be detected.

6. Although samples prepared in "vacuoles" were, in general, easier to dissolve than identical bulk samples, in some few cases small films of insoluble material were observed which would not dissolve even upon heating. The significance of these is being investigated.

With ordinary gelatins, after a solution is made some considerable reduction in temperature is required in order for gelation to take place. However, by the use of a soluble lyophile gelatin a phenomenon of considerable interest to the colloid chemist may be accomplished, namely, what is in effect an isothermic sol→gel transformation. Five grams of a lyophile gelatin (sample F) are slowly dissolved in water at 25° C. in such a manner as to avoid any appreciable rise in temperature. The resulting solution when allowed to stand at the same temperature will slowly set to form a firm jelly. One might go even a step further and prepare a sol at a given temperature, then raise the temperature a few degrees and have a gel form. Obviously, the temperature at which the solution is allowed to stand must not approach too closely 35° C., the temperature at which gelatin can exist only in the sol state.

CONCLUSIONS.

1. The properties of lyophile gelatin have been investigated and in particular its solubility behavior.

2. Some explanations are advanced for the effect of several variables on the solubility of lyophile gelatin.

3. The change of gelatin from the dry solid state→sol→gel with the temperature being maintained constant is described.

ACKNOWLEDGMENT.

This work has been supported by a research grant from Sharp & Dohme and conducted with the collaboration and coöperation of their research staff.

REFERENCES.

- (1) Research Laboratories, Sharp & Dohme, paper unpublished.
- (2) *J. Immunol.*, 29, 389-425 (1935).

DEGRADATION STUDIES ON DIBENZOYL DISULFIDE.*

BY E. S. COOK¹ AND KARL BAMBACH.¹

The use of dibenzoyl disulfide as an antipuritic (1) has led to the recent study of satisfactory methods of synthesis (2), (3). We were further interested in studying the degradation of dibenzoyl disulfide by various methods in order to obtain quantitative conversion to a simple derivative which might supplement the routine Parr bomb sulfur analysis. Several procedures were tried before a successful quantitative degradation to benzoic acid was accomplished. This degradation was mentioned briefly in the paper of Shelton and Rider (2) but it seems worth while to pre-

* Presented before the Scientific Section, A. P. H. A., New York meeting, 1937.

¹ From the Research Laboratories of The Wm. S. Merrell Company, Cincinnati, Ohio.