## A Paradoxical Solubility Phenomenon with Gelatin

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During the course of an experimental study of the electrochemical properties of proteins in glacial organic acids, it was noticed that, using the same stock gelatin, solutions in glacial acetic acid were sometimes readily obtained and at other times the gelatin showed very little tendency to dissolve even on heating or with vigorous and prolonged agitation. This curious behavior was finally traced to the fact that the gelatin was soluble in the acetic acid when the concentration was greater than a certain limiting level but was insoluble below this limit. In other words, gelatin in acetic acid offers the paradoxical example of a substance that is soluble in a concentrated solution but is insoluble in dilute solution. Above the limiting solubility value as far as could be judged, it was possible to prepare gelatin solutions of any concentration.

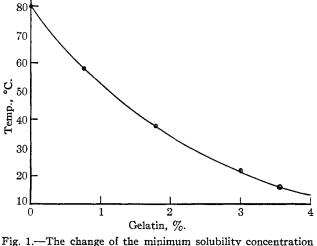


Fig. 1.—The change of the minimum solubility concentration of gelatin in acetic acid with temperature.

As might be expected, the limiting solubility value was found to vary somewhat with different samples of gelatin and also to change with the temperature, so that as the temperature was raised, a point was reached at which the gelatin became completely soluble in all proportions in the acetic acid.

The phenomenon can be most readily demonstrated by first preparing a concentrated solution of the gelatin in acetic acid and then diluting with more acetic acid. The gelatin flocculates out when it is diluted to below the limiting solubility value. This is the most convenient way of studying the subject and was used by us to determine quantitatively the limiting solubility values. The material used mostly was Eastman Kodak Com-

## Notes

pany's electrolytically de-ashed gelatin with an ash of 0.035% and a moisture content of 12%. The acetic acid was Baker c. p. containing 99.5% of the acid. The curve in Fig. 1, giving the change of the lower level of solubility with temperature, was determined on this gelatin. The experiments were carried out by warming a stock solution of the gelatin and the glacial acetic acid to the desired temperature in a water-bath and then adding portions of the acid to the gelatin solution until a standard turbidity developed. This turbidity was determined using the same glass vessel in every case and adding acetic acid until the lettering of a selected printed page could no longer be distinguished through the liquid. As Fig. 1 shows, for this sample of gelatin the solubility limit was 3.1% at  $20^\circ$ , and at  $25^\circ$ 2.7%. The temperature of complete miscibility was determined as  $80^\circ$ .

It was suspected that the moisture content of the gelatin might be the source of this curious behavior and a certain small amount of water is required to disperse gelatin in acetic acid. However, this did not prove to be the case, since on adding up to 5% of water to the acetic acid, which is far more than is introduced with the gelatin, the phenomenon still persisted.

As a test of the reversibility of this paradoxical solubility behavior, the following experiment was carried out. A sample of gelatin thrown out from solution by diluting with acetic acid was collected, dialyzed in a collodion membrane to remove the acid, and was then flocculated from the aqueous solution with alcohol. After being thoroughly dried in a vacuum desiccator, the solubility of this recovered gelatin was tested. It was found to have the same features as the original gelatin, namely, it dissolved to form a concentrated solution and was flocculated out on being diluted with more acetic acid.

Other proteins tested by us proved to be completely insoluble in acetic acid.

DIVISION OF BIOCHEMISTRY UNIVERSITY OF CALIFORNIA MEDICAL SCHOOL BERKELEY, CALIFORNIA RECEIVED APRIL 18, 1933 PUBLISHED JULY 6, 1933