The oxidation of a charcoal was shown to increase with the temperature of oxidation at an accelerating rate.

NEW YORK, N. Y.

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THE ABSORPTION SPECTRUM OF GELATIN AS A FUNCTION OF THE HYDROGEN-ION CONCENTRATION

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Loeb[1] has shown in numerous experiments that the iso-electric point of the gel form of gelatin lies at *P*H 4.7.

Thomas and Kelly,[2] as well as Porter,[3] have shown that the iso-electric point of collagen, or hide protein, has approximately the same value as that for gelatin, at room temperature. But the work of Davis and Oakes indicates an iso-electric point for the sol form of gelatin at about *P*H 8. In experiments upon the bating of calf skin at 40°, with varying hydrogen-ion concentrations, Wilson and Daub[4] observed that a point of minimum swelling occurred in the region of *P*H 8, but not at 4.7. The work of Smith[5] and of Lloyd[6] suggests that the change from the gel to the sol form takes place both with rise of temperature and with rise of Sörensen (*P*H) value.

The recent work of Kelly[7] on the rate of tanning as a function of hydrogen-ion concentration has an important bearing upon this problem. It was found that the rate of combination of hide protein and tannin decreases to a minimum at *P*H 5, increases to a maximum at about 8, and then decreases rapidly towards zero, with increasing Sörensen values. Since tannin is negatively charged, this work would seem to indicate that collagen carries a positive charge between the iso-electric points of its two forms. Between the iso-electric points of the two forms of any protein, any of the gel form present would have a negative charge and any of the sol form a positive charge, the net charge of the system being determined by the preponderance of one of the forms.

The similarity of behavior of collagen and gelatin indicates the possibility of a wider use of data on gelatin in tannery problems. The experiments also suggest that only the sol form of collagen is convertible into gelatin and that the sol forms of the two proteins may actually be identical. The fact that the transition from gel to sol form occurs near the neutral point makes the work of great importance in biological problems.

The object of the investigation to be described here was to observe

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[1] Loeb, "Proteins and the Theory of Colloidal Behavior," McGraw-Hill Book Co., 1922.

[2] Thomas and Kelly, THIS JOURNAL, **44**, 195 (1922).

[3] Porter, *J. Soc. Leather Trades Chem.*, **5**, 259 (1921).

[4] Wilson and Daub, *J. Ind. Eng. Chem.*, **13**, 1157 (1921).

[5] Smith, THIS JOURNAL, **41**, 135 (1919); *J. Eng. Ind. Chem.*, **12**, 878 (1920).

[6] Lloyd, *J. Biochem.*, **14**, 147 (1920).

[7] Margaret W. Kelly, "The Hydrogen Ion and Ion Factors in the Fixation of Tanning by Hide Substances." Read before the Leather Division at the meeting of the American Chemical Society, Pittsburgh, September, **1922**.

what relationship exists, if any, between the wave length of maximum absorption in the ultraviolet and the hydrogen-ion concentration of 1% gelatin solutions.

Experimental Part

Materials.—Normal solutions of sodium hydroxide and hydrochloric acid, 0.0078 *N* acetic acid, and Gold Label granulated gelatin were used.

Preparation of Iso-electric Gelatin.—Sixty g. of Gold Label granulated gelatin was treated in a 4-liter flask with 4 liters of 0.0078 *N* acetic acid at 5°. The mixture was stirred frequently during the course of one hour. The supernatant liquid was decanted, the gelatin re-treated with the acid. A third treatment was given the gelatin as before, at one-hour intervals. The acid was then removed on a large Büchner funnel, the swollen gelatin transferred to 3 liters of distilled water at 5°, and the mixture agitated from time to time. Thirty minutes later the supernatant water was decanted, and fresh water added. This treatment was given at intervals of 30 minutes, with four successive treatments. The gelatin was drained dry under pressure in a large Büchner funnel, transferred to a liter flask, 700 cc. of distilled water was added, and the gelatin brought into solution by immersing the flask in a constant-temperature bath at 50°. An ash determination, made by taking an aliquot part of this solution, drying it in a constant temperature oven at 105° and incinerating it, showed 0.02% of ash.

Method.—The hydrogen-ion concentrations of 1% gelatin solutions, made by taking the requisite amount of the purified stock solution, and treating with various amounts of *N* hydrochloric acid and *N* sodium hydroxide solution, were determined by means of the hydrogen electrode using a Type K Leeds and Northrup potentiometer.

Absorption Measurements.—The 1% solutions, described above, at 20° were placed in a small quartz cell measuring 1.5 cm. in thickness, and the absorption spectra taken by means of an ultraviolet spectrophotometer. As a source of ultraviolet light, an iron arc was used operating on 120 volts, 4 amperes, d. c., between electrodes 1 cm. in diameter.

Using the law for light absorption, $I = I_0 e^{-kd}$, we get $\log I/I_0 = -kd$. "Density" is defined as $\log (I/I_0)$ where I_0 is the intensity of incident light, and I the intensity of light after passing through the gelatin solution.

Light from an iron arc is divided into two beams which reach the slit of the ultraviolet spectrograph by two paths of equal optical density. The two paths are crossed by sector wheels, one of variable aperture, the other of fixed aperture. The two beams are rendered parallel and after passing the quartz prism they give two spectra on the photographic plate, one directly above the other. With the absorbing medium in place, a series of exposures is made with the aperture of the variable sector wheel decreasing in logarithmic decrements. The two spectra are compared side by side on the photographic plate.

Where two lines have the same density, the absorptions have been the same, and may be calculated from the slit of the sector wheel.

$$D = \log (I/I_0) = \log \left\{ \frac{1}{\text{area of sector slit expressed in fractions of the light intensity striking the gelatin}} \right\};$$

when $D = 0$, $\log (I/I_0) = \log (1/1) = \log 1$; ($\log 1 = 0$).

Here the sector wheel is wide open and both beams after passing gelatin and sector wheel have the same intensity, that is, the gelatin is transparent.

When $D = 1$, $\log (I/I_0) = \log (1/0.1) = \log 10$; ($\log 10 = 1$).

The gelatin absorbs $^9/_{10}$ of the light and the sector wheel absorbs $^9/_{10}$ of the light and both have equal action on the photographic plate.

When $D = 2$, $\log (I/I_0) = \log (1/0.01) = \log 100$; ($\log 100 = 2$).

Similarly, wave lengths for densities of 0.6, 0.4 and 0.2 represent the position of absorption bands where 0.75, 0.6 and 0.4 of the light is absorbed.

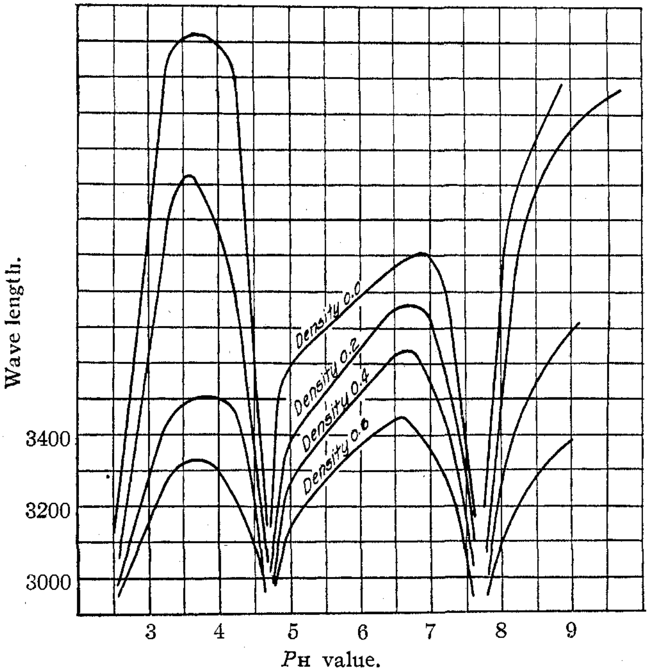

Fig. 1.

Exposures were made upon Wratten and Wainwright panchromatic emulsions, developed in the recommended pyrogallol developer for three minutes at 18°. Ordinary procedure was followed in fixing, washing and drying the plates.

Results of Experimentation.—The points of equal intensity for the two beams of light were read from the plates,[8] and by reference to Kayser's

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[8] This is best done by the photometer, but approximate results may be obtained by direct visual examination.

"Handbuch" the wave lengths for these lines were ascertained. These wave lengths plotted against the hydrogen-ion concentration of the solutions are shown in the accompanying graph.

The curves show two points of maxima at approximately a *P*H of 5.5 and 6.6. These coincide surprisingly well with the two points of maxima found by Loeb and by Wilson and Kern, respectively. Two points of minima are also observed, one at *P*H 4.69, a value coinciding with the so-called iso-electric point found in the literature, and a second at *P*H 7.65. This figure differs but slightly from the value found by Wilson and Kern in their work on the swelling of gelatin (*P*H 7.7).

Summary

The authors have made measurements of the absorption of light by gelatin sols as a function of the hydrogen-ion concentration (expressed in Sörensen values), using a spectrophotometric method, and have expressed these results in the form of curves in which Sörensen values are plotted against wave lengths for definite fractions of light absorbed, the fractions being 0.75, 0.6, 0.4 and 0 (transparent).

The curves for the different fractions absorbed are of the same general shape, as would be expected. The curious thing noted is the sharp shift of the absorption band toward the ultraviolet as the Sörensen values of 4.69 and 7.65 are approached. Just why this occurs we are unable to say. It might perhaps be argued that the second minimum point may represent the so-called "iso-electric point" of the "sol" form of gelatin which has caused so much confusion in the interpretation of results obtained with this substance. Just how a given colloidal system may have two iso-electric points is difficult to see, if the orthodox definition of iso-electric point is accepted.

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[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE UNIVERSITY OF COLORADO]

THE DETERMINATION OF SULFUR BY MEANS OF A THERMOMETRIC TITRATION[1]

BY PAUL M. DEAN AND OTTO O. WATTS

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Dutoit and Grobet[2] have described a method of indicating the formation of compounds in solution by means of a thermometric titration. These investigators were of the opinion that the method could not be applied to volumetric analysis. In order to determine the applicability of thermometric titrations to precipitation reactions the following experiments were carried out.

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[1] Extract from a thesis submitted by Otto O. Watts in partial fulfilment of the requirements for the degree of Master of Arts in Chemistry at the University of Colorado.

[2] Dutoit and Grobet, *J. chim. phys.*, **19**, 324 (1922).