of the base. Weak bases gave salts of the general formula $X.H_2MoO_4.MoO_3$, where X represents one molecule of a mono-acid base. In other words, a substituted mono-ammonium salt of a dimolybdic acid was formed. With strong bases, especially aliphatic bases, however, another molecule of water was added to the salt. The general formula, $X.H_2MoO_4.MoO_3.H_2O$, was indicated.

With tungstic acid a similar relationship developed. The salts tended to add more water of crystallization as the degree of strength of the base increased. Also, the ratio of the base to the tungstic oxide decreased with the increase of strength of the base.

Summary

The methods of preparation and some properties of some of the alkyl and aryl substituted ammonium molybdates and tungstates have been investigated. The aliphatic amines gave well-defined, crystalline, water-soluble molybdates and tungstates with little difficulty. On the contrary, the aromatic amines formed precipitates that were only slightly soluble in water. Large yields of pure crystals were not possible on account of the ease of hydrolysis of these aromatic compounds. The yield of the salts varied greatly with the temperature, pressure, solvents and the concentration of the reactants.

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THE INFLUENCE OF HYDROGEN-ION CONCENTRATION ON THE OPTICAL ROTATION OF GELATIN

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The optical rotation of gelatin has been studied by C. R. Smith, and reported by him in two important papers.[1] The purpose of Smith's work was (1) to observe the optical rotation of gelatin at various temperatures and (2) to determine the minimum gelatin concentration at which gelation takes place on cooling, this gelatin concentration being determined by an application of his findings on the relation of rotation to concentration.

Smith reported that the specific rotation, $[\alpha]_D$, of a 2% solution of the highest grade of commercial gelatin at 35°, and above was —141°, and that the specific rotation at 15° and below was —313°. Between these temperatures the rotation varied in a regular manner. On the basis of these and other findings, Smith concluded that the change in rotation is due to a reversible change between the gel and the sol, the gel having a specific rotation of —313° and being the only form stable at temperatures below 10°, while the sol has a specific rotation of —141° and is the only form

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[1] This Journal, **41,** 135 (1919); *J. Ind. Eng. Chem.,* **12,** 878 (1920).

stable above 35°. The intermediate rotation values obtained between these temperatures are attributed to an equilibrium between the two forms.

The present investigation was undertaken to determine the influence of variation in the hydrogen-ion concentration on the specific rotation of gelatin. It was intended to study the problem at several temperatures from about 10° to 40°, so as to cover the entire range from pure gel to pure sol; to perform the work on gelatins containing known and various amounts of hydrolytic products; to use various concentrations of gelatin; and to observe the influence of various impurities. Unfortunately, this program cannot now be carried out by the authors, and they invite the attention of research men to this interesting field. The present report is admittedly incomplete, covering only the one condition, namely, the influence of hydrogen-ion concentration on the optical rotation of a 2% solution of originally iso-electric and ash-free gelatin at 30°. It is felt, however, that publication is justified because of certain points discovered, and because attention is directed thereby to a fruitful field for study.

The gelatin used was practically ash-free (0.015% of ash) and iso-electric (*P*H of aqueous solution = 4.7) and was obtained through the courtesy of the Eastman Kodak Company. The method of preparation insures that it consists almost entirely of unhydrolyzed gelatin. One stage of the purification process, following the electrolyzing of the jelly, consists in the precipitation of the 8 or 10% gelatin solution with an equal volume of pure acetone. This precipitation, according to Sheppard, Sweet and Benedict[2] "not only further reduces the ash content, but removes the greater part of any hydrolysates (gelatose, gelatin peptones, etc.)." Two per cent. solutions were prepared by soaking the gelatin in cold water or electrolyte until fully swollen, raising the temperature to 60° in a water-bath to effect solution, and cooling quickly to 32°. Careful correction of concentration was made, the solution poured into 1-dcm. polariscope tubes at the same temperature, and immersed in a water-bath held at 30° ± 0.005°.

Readings were taken on several check solutions in a Schmidt and Haensch polariscope. This instrument was calibrated carefully by sugar solutions of definite concentration and temperature, and calibration and correction curves and factors were deduced.

Immediately on the completion of the readings for rotation, the solution was subjected to a determination of hydrogen-ion concentration, by the usual potentiometric procedure, at the same temperature.

The hydrogen-ion concentration was caused to vary by small increments from *P*H 0.3 to *P*H 13.4 by the addition of the proper amounts of hydrochloric acid or sodium hydroxide. A few readings obtained in strongly alkaline solutions were found to be very erratic, but it is believed that this was due to the hydrolysis which took place under these conditions. On standardizing the technique, this difficulty was overcome to a point where it is felt that the solid portion of the curve reported is correct to the limits of legitimate experimental error. Further work must be done at *P*H values greater than 12 before a precise curve may be drawn. By reference to a previous study reported by one of us,[3] and also to a report of J. H. Northrop[4] it seems improbable that appreciable hydrolysis could occur between *P*H 2 and 10 in the brief period (less than five minutes) of heating at 60°. Beyond these values it may be significant. Northrop

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[2] THIS JOURNAL, **44,** 1859 (1922).

[3] Bogue, *Ind. Eng. Chem.*, **15,** 1154 (1923).

[4] Northrop, *J. Gen. Physiol.*, **3,** 715 (1921).

found, for example, that at 65° the hydrolyzed material of gelatin, as determined by the formol titration method, was doubled in about one and one-half days in the presence of hydrochloric acid, *P*H 1.65 to 2.70, or by citric acid, *P*H 2.2, and in three days in the presence of citrate carbonate and hydrochloric acid of *P*H 8.5. The effect on rotation of time of standing at 30° between *P*H 2 and 10 was found to be negligible up to periods of about six hours. It is believed, therefore, that the sol-gel equilibrium is attained rapidly under the conditions of the experiment, and that at 30° the effect of time on hydrolysis at any *P*H value between 2 and 10 is insignificant so far as the element of time enters into the value of the determinations.

The results obtained in this study are given in Table I, and the curve showing specific rotation plotted on the ordinate and *P*H on the abscissa is given in Fig. 1. A few interesting relations are shown by an examination of these data. It is obvious that the rotation varies in a definite manner with the hydrogen-ion concentration. A sharp break is found at the iso-electric condition *P*H 4.7 at which point the rotation is least, namely, —104°. The rotation rises rapidly on either side of this point, reaching a value of —134° at *P*H 2.9, and —139° at *P*H 7.1. Beyond these values the rotation again decreases. This makes evident that it is not permissible to refer to the specific rotation of gelatin unless the hydrogen-ion concentration is stated, as well as the temperature and gelatin concentration. The specific rotation of ash-free and iso-electric gelatin of *P*H 4.7, in 2% concentration at 30°, is found to be —104 angular degrees, with a probable

TABLE I

SPECIFIC ROTATION AT VARIOUS SÖRENSEN VALUES

*P*H	0.3	.7	.8	1.0	1.4	1.8	2.9	3.4	3.5	4.2	4.3	4.4	4.7
$[\alpha]_D^{30}$	−90	−102	−110	−117	−128	−131	−134	−125	−125	−115	−120	−107	−104
*P*H	4.8	4.8	4.9	7.1	10.0	11.7	11.8	12.2	12.7	12.8	13.0	13.4	
$[\alpha]_D^{30}$	−129	−117	−122	−139	−135	−122	−124	−71	−86	−101	−116	−69	

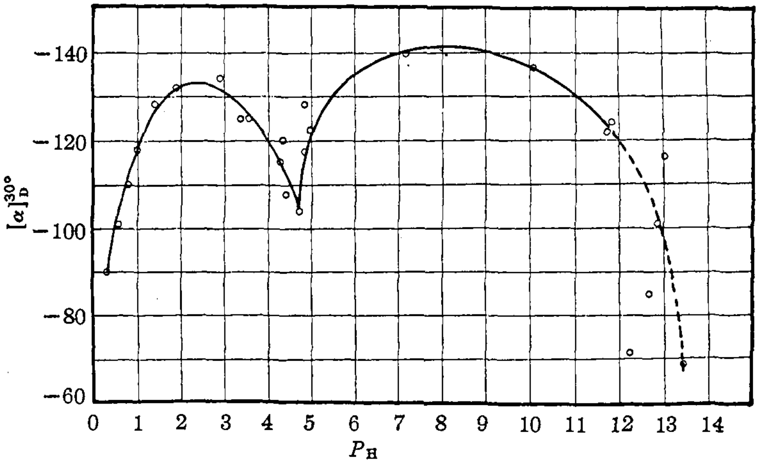

Fig. 1.—Specific rotation of 2 per cent. solutions of ash-free originally iso-electric gelatin at 30°, at varying *P*H.

error of ±4°. Smith's value for commercial gelatin interpolated to the same temperature, and for the same concentration, is about —124°, but the Sörensen value is not stated. Our curve shows this value at a *P*H of about 5.2, which is slightly less than that found for most commercial gelatins. The use of optical rotation for calculating the gelatin concentration of a solution must take into consideration this variability.

The data lend evidence also to the controversy on the matter of a second iso-electric point of gelatin[5] but do not support a belief in the existence of this second point. J. A. Wilson has suggested that the two points of maximum swelling of calf skin and of gelatin which he observed at 7° at *P*H 4.7 and 7.7 may be due to the existence of two iso-electric points, the one at *P*H 4.7 being that of the gel and the one at *P*H 7.7 being that of the sol. This necessitates the hypothesis that hydrogen-ion concentration as well as temperature may function to change one form into the other. C. R. Smith finds that the specific rotation of the gel form (at 15° and below) of gelatin is —313°, which is a value of an altogether different order from that found in this study at any hydrogen-ion concentration at 30°. From this evidence it seems improbable that any large amount of the gel form is produced at 30° at any hydrogen-ion concentration studied herein. There was found no break in the curve at *P*H 7.7, nor at any *P*H value other than 4.7 which is the generally accepted iso-electric condition for gelatin. Since we find that at 30° the sol is present in very large excess over the gel, then if, as Wilson suggests, the iso-electric point of the sol is at *P*H 7.7, a break would be expected in the curve at *P*H 7.7 and none at 4.7 unless reversion occurred. The data reported would seem to indicate that no reversion to gel occurs at any *P*H at 30°, but the break is found at *P*H 4.7 and not at *P*H 7.7. This supports the contention that the iso-electric point of the sol as well as of the gel is at *P*H 4.7.

Summary

Determinations of the specific rotation of 2% solutions of originally iso-electric and ash-free gelatin at 30° at varying hydrogen-ion concentrations from *P*H 0.3 to *P*H 13.4 have been made. The results show that the specific rotation of the solution varies with the hydrogen-ion concentration, being low at high concentrations of acid or alkali, rising as the acidity or alkalinity is decreased, and exhibiting a minimum value at the iso-electric condition, *P*H 4.7. No indication of a second iso-electric point at *P*H 7.7 is observed. The specific rotation of pure ash-free and iso-electric gelatin of *P*H 4.7 in 2% concentration at 30° is found to be —104 angular degrees, with a probable error of ±4 degrees.

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[5] Wilson and Kern, THIS JOURNAL, **44**, 2633 (1922); **45**, 3139 (1923). Higley and Mathews, *ibid.*, **46**, 852 (1924).