# THE EFFECT OF HYDROGEN-ION CONCENTRATION ON THE ESTIMATION OF DIASTATIC POWER BY THE POLARIMETRIC METHOD

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The polarimetric Lintner method[1] was devised to shorten the classical method while retaining the Lintner scale. In its present form, however, no arrangement is made for controlling the hydrogen-ion concentration of the solution. Sherman, Thomas and Baldwin[2] have shown that the activity of highly purified malt diastase varies greatly with the hydrogen-ion concentration, and the work described below shows that the diastase in a malt infusion is equally sensitive. It is also shown that by use of a buffer, fluctuations in hydrogen-ion concentration are readily controlled. Accordingly, it is suggested that the polarimetric Lintner method be modified by so preparing the soluble starch solution that it contains a suitable buffer; a satisfactory mixture is shown to consist of 10 cc. of *N* acetic acid plus 10 cc. of *N* sodium acetate contained in each liter of soluble starch solution.

To determine the effect of hydrogen-ion concentration on the diastatic activity of an infusion of malt by the new polarimetric method, the following experiments were made. A solution of soluble starch was prepared containing 4 g. in 180 cc., and an infusion of malt diastase was made up by macerating 25 g. of ground distillers' malt with 500 cc. of water for one hour, frequently stirring the mixture and finally filtering it. The initial polarization was determined by diluting 180 cc. of starch solution to 200 cc. with water, adding 2 cc. of strong ammonium hydroxide and 2 cc. of diastase solution in the order named and polarizing. Portions of 180 cc. of each starch solution were placed in 200cc. flasks and the hydrogen-ion concentration of each adjusted by adding hydrochloric acid or sodium hydroxide. The solutions were then cooled to 21° in a water-bath, made up to 200 cc. and 2 cc. of the diastase solution was added to each. Each solution was digested at 21° for one hour when 101 cc. of each was transferred to a 100cc. flask graduated at 101 cc. One cc. of strong ammonium hydroxide was then added and the solution polarized after 25 minutes when the readings had become constant.

The Sörensen values ($P_H$) of the solutions remaining in the 200cc. flasks were then determined electrometrically and the Lintner values calculated from the formula, $L = \frac{100\ d}{t \times 2.18}$ where $d$ is the fall in polarization in a 4dm. tube and $t$ is the time in hours.

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[1] *J. Assoc. Official Agr. Chem.*, **7,** 364 (1924).

[2] Sherman, Thomas and Baldwin, This Journal, **41,** 231 (1919).

| *P*H | L | *P*H | L | *P*H | L |
|---|---|---|---|---|---|
| 3.14 | 39 | 4.26 | 117 | 5.56 | 119 |
| 3.31 | 60 | 4.31 | 122 | 5.85 | 122 |
| 3.45 | 80 | 4.51 | 124 | 6.02 | 119 |
| 3.67 | 101 | 5.06 | 122 | 6.64 | 119 |
| 4.02 | 112 | 5.07 | 126 | 7.15 | 117 |
| 4.07 | 115 | 5.44 | 122 | 8.28 | 60 |

When these results are plotted, the smooth curve obtained is similar in shape to that shown by Sherman, Thomas and Baldwin.[2] The optimum range is from 4.5 to 5.5 *P*H.

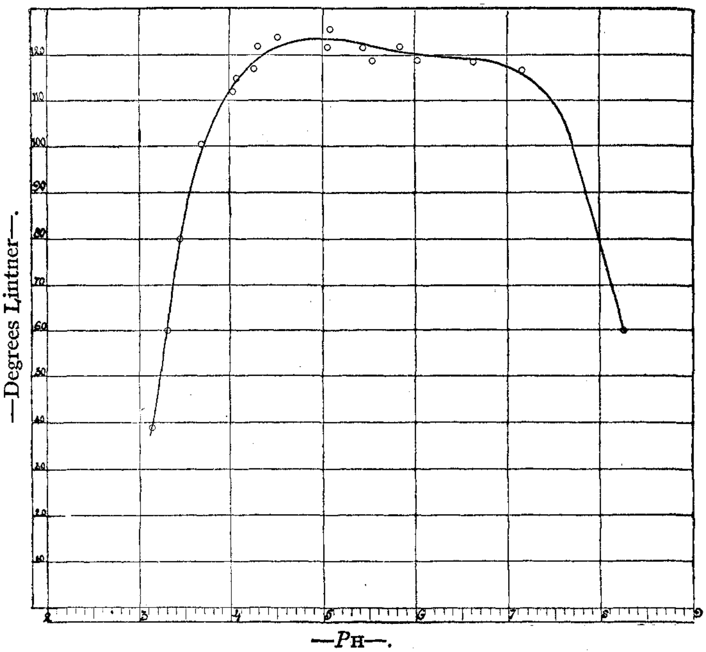

Fig. 1.—The effect of *P*H on the estimation of diastatic power of malt.

The following experiment was made to test an acceptable method of adjusting the *P*H of the starch solution. Four solutions were prepared.

Solutions A consisted of 180 cc. of starch solution containing 4 g. of soluble starch. To this were added 15 drops of 0.1 *N* hydrochloric acid and the volume was made up to 200 cc. with water at 21°. Two cc. of diastase infusion was then added, the solution kept at 21° for an hour when 101 cc. was removed and polarized after the addition of 1 cc. of ammonium hydroxide as described above. The Sörensen value of the solution remaining in the 200cc. flask was measured.

Solution B was made up and treated in the same manner, except that the volume was made up to 200 cc. with standard acetate solution. This was prepared by mixing 10 cc. of $N$ acetic acid and 10 cc. of $N$ sodium acetate and diluting to 100 cc.

Solution C was identical with Solution A except that 15 drops of 0.1 $N$ sodium hydroxide solution were used instead of the 0.1 $N$ hydrochloric acid; and Solution D was identical with Solution B except that 15 drops of 0.1 $N$ sodium hydroxide solution replaced the 0.1 $N$ hydrochloric acid. The results are given below.

| | $P_H$ | $L$ |
|---|---|---|
| Solution A—HCl added, not buffered | 3.57 | 103 |
| Solution B—HCl added, buffered | 4.68 | 135 |
| Solution C—NaOH added, not buffered | 7.15 | 117 |
| Solution D—NaOH added, buffered | 4.77 | 135 |

Thus the acetate solution changed the Sörensen value of the solutions to within the range required.

## Summary

The diastase in an infusion of malt was as sensitive to changes in hydrogen-ion concentration as the highly purified diastase used by Sherman, Thomas and Baldwin. The optimum $P_H$ range was 4.5 to 5.5. It is shown that for correct results by the polarimetric Lintner method, the mixture of soluble starch and diastase infusion must be held within the above range.

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

# TRIHYDROXY-METHYLANTHRAQUINONES. V. SYNTHESIS OF MORINDONE[1]

BY R. A. JACOBSON[2] WITH ROGER ADAMS

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Morindone, chiefly in the form of its glucoside, morindin, is the main constituent of *Morinda citrifolia* and M. *umbellata.*[3] It is an anthraquinone derivative which before the advent of synthetic dyes was rather extensively used in India for dying cloth. Both morindin and morindone were discovered as early as 1849,[4] and the similarity of the latter compound

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[1] Previous papers in this field are (a) Graves and Adams, THIS JOURNAL, **45,** 2439 (1923); (b) Gardner and Adams, *ibid.*, **45,** 2455 (1923); (c) Jacobson and Adams, *ibid.*, **46,** 1312 (1924); (d) *ibid.*, **46,** 2788 (1924).

[2] This communication is an abstract of a portion of a thesis submitted by R. A. Jacobson in partial fulfilment of the requirements for the degree of Doctor of Philosophy in Chemistry at the University of Illinois.

[3] (a) Oesterle and Tisza, *Arch. Pharm.*, **245,** 534 (1907). (b) Perkin, *Proc. Chem. Soc.*, **24,** 150 (1908). (c) Simonsen, *J. Chem. Soc.*, **113,** 766 (1918).

[4] Anderson, *Ann.*, **71,** 216 (1849).