Production of anomalous compounds in paper electrophoresis by high concentrations of base*

We wish to report the appearance of an anomalous compound during the electrophoretic analysis of several reaction mixtures encountered in the synthesis of amino alcohols. This compound is attributed to the reaction of moderate concentrations of base with ninhydrin in the process of developing the electrophoretograms.

Method

A sample of the reaction mixture obtained in the reduction of glycylglycine with lithium aluminum hydride was placed on a 1.5 × 50 cm strip of S & S 597 filter paper previously saturated with potassium acid phthalate–sodium hydroxide buffer of pH 6.0 and of ionic strength 0.045. The electrophoretic separation was conducted at 600 V for 1 h on a Reco electrophoresis apparatus (model E-800-2). At the end of this time the paper strip was dried and the color developed with 0.25% ninhydrin in butanol saturated with water.

Results and discussion

The reaction mixture was shown to consist of at least four compounds giving a color with ninhydrin: unreacted glycylglycine, glycine, ethanolamine and an unknown compound which gave a yellow color with ninhydrin. The "yellow compound", as it will henceforth be called, had a negative charge at pH 6.0, migrating at approximately the same rate as ethanolamine, but in the opposite direction.

Identification of the components of the "yellow compound" was undertaken on a small quantity of this material which had been purified by means of a starch block separation. It was noted that the purified "yellow compound" did not have the same migration properties as it had in the mixture; that is, the purified "yellow compound" moved a short distance toward the cathode whereas the impure "yellow compound" moved rapidly toward the anode under identical electrophoretic conditions. An electrophoretic analysis of the acidic and basic hydrolysis mixtures of this compound indicated the presence of only one spot which occupied a position just slightly on the cathode side of the origin. No color area could be detected in the electrophoretic analysis of an unneutralized acid hydrolysis mixture.

The "yellow compound" was observed on the electrophoretograms of two other types of reaction mixtures dealing with the synthesis of N-glycylaminoethanol. In each case the pH of the mixture was on the alkaline side of neutrality.

A chance check of the effect of base on the electrophoretic migration of ethanol-

* Portion of a thesis presented by ELIZABETH C. SMITH as partial fulfillment of the requirements for the degree of Doctor of Philosophy. This report has been authorized for publication on October 9, 1958, as paper No. 2299 in the journal series of the Pennsylvania Agricultural Experiment Station.

amine showed some rather interesting results. Not only was a spot visible for ethanolamine in its normal position, but there was also a color area in the position of that assigned to the "yellow compound". Further experimentation showed that sodium hydroxide was indeed responsible for the yellow color appearing on this electrophoretogram. In addition to sodium hydroxide, barium hydroxide, lithium hydroxide and potassium hydroxide also produced a spot corresponding in color and in position to that originally assigned to the "yellow compound". Lithium hydroxide was probably responsible for the production of this spot in the case of the reduction of glycylglycine with lithium aluminum hydride. The anomalous spot produced by these basic compounds was very similar to the "slow moving spot" described by WALDRON-EDWARD[1]. In this case the anomaly was caused by traces of sulfate.

Additional study indicated that the color formation was independent of the type of paper used. The exact concentration of base required for the production of the "yellow compound" is unknown. However, this spot appeared on electrophoretograms when 5–10 μl of 10% sodium hydroxide were placed on the paper strip.

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[1] DEIRDRE M. WALDRON-EDWARD, *Chem. & Ind. (London)*, (1954) 104.

Received February 13th, 1959

BOOK REVIEWS

Analytical Chemistry, Some New Techniques, by A. G. JONES. Butterworths Scientific Publications, London, and Academic Press Inc., New York, 1959, 268 pages, price 40 s.

The author states that he selected eight topics of special interest to him and presented them to non-specialists as introductions to these techniques without giving a complete survey. The eight topics are: Flame Photometry (47 pages), Differential Spectrophotometry (28 pages), Gas Chromatography (43 pages), The Use of Ion Exchangers in Analytical Chemistry (36 pages), Acid–Base Titrations in Non-aqueous Media (22 pages), Coulometric Titrations (20 pages), Differential Refractometry (18 pages) and The Determination of Oxygen and Hydrogen in Metals (28 pages).

The reviewer will confine himself to the chapters on gas chromatography and ion exchangers which are both excellent. The author demonstrates that these two topics can be fully explained and illustrated with numerous applications in as little as about 40 pages each, provided, of course, that the whole literature is not discussed. The