process, could be greatly improved through purification of this compound by vacuum distillation rather than by recrystallization from water. The crude 3,5-dimethyl-4-cyanophenol was recrystallized from benzene; it formed glistening, white scales; m. p., on non-standardized thermometer, 177.5–177.7°.

Because of reports[19,21] that 3,5-dimethyl-4-cyanophenol crystallizes from benzene in small needles and melts at about 175°, the identity of this compound was confirmed by analysis.[22]

Anal. Calcd. for C_9H_9ON: C, 73.38; H, 6.16; N, 9.52. Found: C, 73.55; H, 6.38; N, 9.46.

2,6-Dimethyl-4-cyanophenol.—2,6-Dimethyl-4-cyanophenol was prepared from mesitol by the method of Thiele and Eichwede[13] and recrystallized from medium-boiling petroleum ether; m. p. 124.0–124.4°.

(21) Houben and Fischer, *Ber.*, **66**, 339 (1933).

(22) The carbon-hydrogen analysis was made by Mr. James G. Burt.

Summary

1. The pK_a's of the following compounds have been determined at 25°: 3,5-dimethylphenol, 2,6-dimethylphenol, 3,5-dimethyl-4-nitrophenol, 2,6-dimethyl-4-nitrophenol, 3,5-dimethyl-4-cyanophenol, 2,6-dimethyl-4-cyanophenol.

2. The pK_a's at 25° have been redetermined for the following compounds: phenol, *p*-nitrophenol, *p*-cyanophenol.

3. The results provide evidence that the total effect produced by a para nitro or para cyano group on the acid strength of phenol is due about equally to an electrostatic interaction and to resonance.

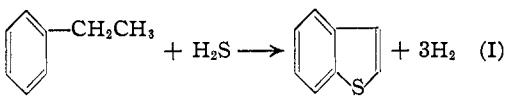
CHICAGO, ILLINOIS RECEIVED APRIL 5, 1948

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, POMONA COLLEGE]

Catalytic Synthesis of Thianaphthene from Ethylbenzene

BY CORWIN HANSCH AND FRED HAWTHORNE

In a recent paper, Moore and Greensfelder[1] published the procedure for a new synthesis of thianaphthene from styrene and hydrogen sulfide. While this procedure is an excellent one for the preparation of thianaphthene itself, it would not be as convenient for the preparation of substituted thianaphthenes because of the lack of availability of the proper styrenes as starting materials. Thus, it seemed that the dehydrogenation of an alkylbenzene in the presence of hydrogen sulfide might be accomplished over a single catalyst with formation of the thianaphthene in one step according to Equation I.



(I)

The present paper reports the results of such an attempt.

Experimental

All experiments were carried out in a Pyrex catalyst tube in a continuous flow system. The apparatus used was similar to that described by Hoog, Verheus and Zuiderweg.[2]

The hydrogen sulfide used in this work was commercial grade used directly from the cylinder. Eastman Kodak Co. white label ethylbenzene was distilled before using.

Catalyst Preparations. I. Chromium on Aluminum Oxide.—To a boiling solution of 36.4 g. of chromic anhydride in 400 ml. of distilled water was added 200 g. of ALORCO alumina,[3] Grade H40, Type R2200, 8–14 mesh. The mixture was dried at 100°.

II. Chromium and Nickel on Aluminum Oxide.—Chromic anhydride (7.6 g.) and 12.5 g. of $Ni(NO_3)_2 \cdot 6H_2O$ were dissolved in 50 cc. of water and the solution brought to boiling. To this solution was added 50 g. of activated alumina with vigorous stirring. The mixture was then dried in an oven at 100°.

For the preparation of thianaphthene, the straight chromium catalyst[4] was reduced *in situ* with a slow stream of hydrogen, for one hour at the temperature at which dehydrogenation was to be made, then a stream of hydrogen sulfide passed over the catalyst for fifteen to twenty minutes at the same temperature. Ethylbenzene was then introduced at a uniform rate. A space velocity ratio of about 9:1 of hydrogen sulfide and ethylbenzene was found to give the highest conversion. Most of the liquid products were separated in a Liebig condenser. A small amount of liquid entrained in the large volume of hydrogen sulfide and hydrogen was separated by passing the gases through a U-tube filled with glass wool and cooled in an ice-bath. Using this technique, it was possible to obtain excellent material balances in all runs. The thianaphthene was isolated by distillation and identified by a comparison of it and its picrate with that of a sample prepared by a known procedure.[5]

Discussion

Table I shows the effect of temperature and space velocity on the conversion of ethylbenzene to thianaphthene.

TABLE I

The space velocity of hydrogen sulfide in all runs was 1400 cc./cc./hr.

Catalyst	Temp., °C.	Space velocity,[a] ethylbenzene cc./cc./hr.	% Conversion to thianaphthene
I	550	160	9.3
I	575	160	18.5
I	575	260	13.2
II	575	160	17.0
II	610	245	13.2
II	625	160	18.5

[a] Calculated as cc. of vapor at S.T.P.

The above runs were made for periods of four hours. It was observed that, although the rate of dehydrogenation (as estimated by hydrogen evolution) was more rapid

(1) THIS JOURNAL, **69**, 2008 (1947).

(2) Hoog, Verheus and Zuiderweg, *Trans. Faraday Soc.*, **35**, 995 (1939).

(3) This type of alumina was used exclusively in the research and was supplied through the courtesy of the Aluminum Ore Company.

(4) The nickel-chromium catalysts were reduced for two hours.

(5) Hansch and Lindwall, *J. Org. Chem.*, **10**, 381 (1945).

at temperatures of 600° and above, the activity of the catalyst decreased much more rapidly than at the optimum temperature 575°. Very little difference in activity or life of the two catalysts reported in this paper was noted. Several other catalysts of different ratios of chromium and nickel were prepared; they also showed very little difference in activity. With space velocity ratio of 4:1 of hydrogen sulfide to ethylbenzene, only low yields (~5%) of thianaphthene were obtained.

Preliminary results with other alkylbenzenes indicate that the reaction may be general, however, the yields do not appear to be as good as in the case of ethylbenzene.

Acknowledgment.—The authors are very much indebted to the Research Corporation for a Frederick Gardner Cottrell grant-in-aid which supported this research.

Summary

Thianaphthene has been prepared in 18.5% conversion from hydrogen sulfide and ethylbenzene using a chromia on alumina catalyst at 575°.

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[CONTRIBUTION FROM THE GATES AND CRELLIN LABORATORIES OF CHEMISTRY, CALIFORNIA INSTITUTE OF TECHNOLOGY, PASADENA, NO. 1178]

The Purification and Properties of Antibody against *p*-Azophenylarsonic Acid and Molecular Weight Studies from Light Scattering Data

BY DAN H. CAMBPELL, ROBERT H. BLAKER AND ARTHUR B. PARDEE[1a]

It is becoming increasingly evident that many fundamental problems dealing with the structure and behavior of antibody molecules must be studied with purified antibody preparations in solution of known composition rather than in complex solutions such as serum. Methods which are devised for the isolation and purification of antibodies on a practical scale are hence of considerable interest and importance. The following report describes a method for the isolation and purification of antibody against *p*-azophenylarsonic acid in which the antibody is removed from the antiserum by specific precipitation with a polyhaptenic dye and recovered from a solution of the dissociated antigen–antibody complex.

The recovery of antibodies from specific antigen–antibody complexes has been accomplished by a variety of methods.[1b] Perhaps the best known is the one described by Heidelberger and Kendall[2] and Heidelberger and Kabat,[3] in which 15% sodium chloride solutions were used to produce a shift in the antigen–antibody ratio of specific precipitates of SSS or of intact Pneumococcus and antipneumococcus serums favoring the liberation of antibody. Liu and Wu[4] were able to obtain as good if not better yields of antibody preparations by acid dissociation of similar antigen–antibody complexes at about *p*H 4.0 with subsequent isolation of antibody by salt precipitation or removal of antigen by centrifugation if bacterial cells were used. Recently, a report has been made by Haurowitz, *et al.*,[5] which describes the isolation and purification of antibody against *p*-aminobenzylamine, anthranilic, arsanilic, and sulfanilic acids by the use of methods somewhat similar to those used by us in the present investigation. The principal difference was their use of an acid-insoluble conjugated protein for a precipitating antigen. Our own investigations of a number of antigen–antibody systems have indicated that, in general, acid dissociation is the method of choice, at least for the systems involving ovalbumin, polysaccharide, and arsanilic acid antigens. The last of these is a particularly good system since simple polyhaptenic dye antigens can be used for specific precipitating agents. The physical properties of such antigens are so different from those of the antibody proteins that the dissociated complexes can usually be separated into the antigen and antibody components without difficulty. Certain dye antigens have the added advantage that they have a low solubility under acid conditions and hence upon dissociation of the antigen–antibody precipitate the antibody dissolves and the antigen remains behind as an insoluble acid.

Purification of Antibody.—Several methods were studied for the dissociation of antibody from antigen–antibody complexes and its subsequent recovery from the dissociated mixture. For example, treatment of precipitates by alkali at *p*H 9.0–10.0 resulted in considerable dissociation, as evidenced by solution of the precipitates, but the yields of antigen-free protein were low because of the high solubility of the antigen and its tendency to remain attached to the protein. Furthermore, some denaturation of antibody protein always occurred and the purity of antibody as based on the ratio of specifically precipitable protein to total protein usually gave values of only 10 to 20%. Another method which was used with some success was dissociation of dye–antigen complexes with a simple hapten such as arsanilic acid and subsequent dialysis against the hapten until the solution was free of the dye antigen. This

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(1b) Dan H. Campbell and Frank Lanni, "The Amino Acids and Proteins," edited by D. M. Greenberg, Chapt. XII, "Immunology of Proteins," Thomas Publishing Co., in press.

(2) M. Heidelberger and F. E. Kendall, *J. Exptl. Med.*, **64**, 161 (1936).

(3) M. Heidelberger and E. A. Kabat, *ibid.*, **67**, 181 (1938).

(4) S. C. Liu and H. Wu, *Proc. Soc. Exptl. Biol. Med.*, **41**, 144 (1939).

(5) F. Haurowitz, Sh. Tekman, Miervet Bilen and Paula Schwerin, *Biochem. J.*, **41**, 305 (1947).