somewhat more stable to alkaline hydrolysis as determined by *Lactobacillus arabinosus* assay than by *Neurospora crassa* assay.

Biotin in Relation to the Metabolic Activity of *Aspergillus niger*.—A 500-ml. amount of *Aspergillus niger* medium (no added pimelic acid) was brought to *p*H 2.5 which is the *p*H that results from the growth of *Aspergillus niger* under the conditions that have been described. Biotin in an amount of 25 μg. was added (0.050 μg./ml.), the mixture was autoclaved and then shaken at 30° for 5 days. At the end of this time differential assay (*Neurospora crassa* and *Lactobacillus arabinosus*) as well as bioautography (paper chromatography with butanol–acetic acid–water and localization of activity with *Neurospora crassa*) indicated unchanged biotin (*Neurospora crassa* assay 0.045 μg./ml. (90% recovery), *Lactobacillus arabinosus* assay 0.045 μg./ml. (90% recovery), R_f 0.87). In a second experiment 25 μg. of biotin (0.050 μg./ml.) was added to 500 ml. of *Aspergillus niger* culture medium. Growth was carried out in the usual way. At the end of this time differential assay as well as bioautography indicated biotin *l*-sulfoxide equivalent to the biotin added (*Neurospora crassa* assay, 0.046 μg. as biotin/ml. (92% recovery), *Lactobacillus arabinosus* assay, 0.0053 μg. as biotin/ml. (11% recovery, biotin *l*-sulfoxide is only about 5% as active as biotin for *Lactobacillus arabinosus*), R_f 0.47). In a third experiment *Aspergillus niger* was grown with aeration at 30° for 5 days. After growth, 25 μg. of biotin (0.050 μg./ml.) was added to the fermentation and the mixture was immediately autoclaved to stop growth and enzymatic activity. The mixture was then shaken at 30° for 5 additional days. At the end of this time differential assay as well as bioautography indicated unchanged biotin (*Neurospora crassa* assay, 0.051 μg./ml. (102% recovery), *Lactobacillus arabinosus* assay 0.051 μg./ml. (102% recovery), R_f 0.86).

WEST POINT, PENNA.
RAHWAY, NEW JERSEY

[CONTRIBUTION FROM ROHM AND HAAS COMPANY]

5-Alkoxymethylenerhodanines and their Reactions with Rhodanines[1]

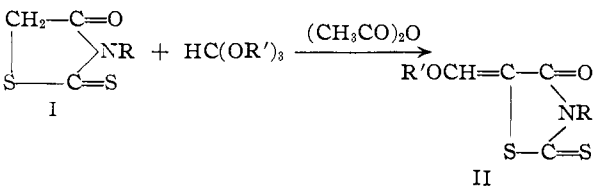
BY CHIEN-PEN LO AND W. J. CROXALL

RECEIVED MARCH 8, 1954

Reaction of 5-unsubstituted rhodanines (I) with alkyl orthoformates in the presence of acetic anhydride yields 5-alkoxymethylenerhodanines (II). Condensation of I and II in the presence of a tertiary amine gives the amine salt of 5,5-methylidynebisrhodanines which upon treatment with hydrochloric acid yield the 5,5′-methylidynebisrhodanines. The acidic property of the latter compounds is believed to be due to the stabilization of the enolate ion by resonance. That the enolate ions are actually hybrids of two extreme resonant forms is supported by experiments. This two-step synthesis furnishes a satisfactory method of preparation of 3,3′-unsymmetrically substituted 5,5′-methylidynebisrhodanines which have not been previously reported in the literature.

Alkyl orthoformates react with active methylene compounds such as malonic, acetoacetic, cyanoacetic esters and others to give either the alkoxymethylene derivatives or the methylidynebis compounds according to the experimental conditions used.[2] The reactivity of the methylene group in rhodanine and 3-substituted rhodanines has long been established. For example, they react with aldehydes and ketones to give 5-alkylidenerhodanines,[3] with isatin to give rhodanine-($\Delta^{5,3'}$-oxindole),[4] with *p*-nitrosodimethylaniline to give 5-(*p*-dimethylaminophenylimino)-rhodanines[5] and with diphenylformamidine to give 5-anilinomethylenerhodanine.[6]

We have found that rhodanine condensed with ethyl orthoformate in the presence of acetic anhydride to yield 5-ethoxymethylenerhodanine (II, R = H; R′ = C_2H_5).



The assignment of the structure of this product was based on analysis and molecular weight determination. The use of methyl orthoformate in the above reaction yielded 5-methoxymethylenerhodanine (II, R = H; R′ = CH_3). This definitely eliminated the possible methylidynebisrhodanine structure. The generality of this reaction was demonstrated by its successful application to a number of 3-substituted rhodanines including 3,3-ethylenebisrhodanine. The 5-alkoxymethylenerhodanines thus prepared are characterized in Table I.

It was reported by Kendall and Fry[7] that the reaction of alkyl orthoformates and rhodanines in the presence of a tertiary amine yielded dyestuffs to which they assigned the structure of 5,5-methylidynebisrhodanines. Since the 5-alkoxymethylene rhodanines obtained above are the logical intermediates for the formation of the compounds of Kendall and Fry, they are expected to react with another molecule of rhodanine with the formation of 5,5′-methylidynebisrhodanines. While the method of Kendall and Fry would yield only symmetrical compounds, ours, if successful, should also be applicable to the synthesis of 3,3′-unsymmetrically substituted 5,5′-methylidynebisrhodanines.

(1) Presented at the Miniature Meeting of the Philadelphia Section of the American Chemical Society, January 29, 1953.

(2) (a) For a review and bibliography on earlier work, see H. W. Post, "The Chemistry of the Aliphatic Orthoesters," Reinhold Publ. Corp., New York, N. Y., 1943, p. 81; (b) R. C. Jones, THIS JOURNAL, **73**, 3684 (1951).

(3) Some of the pertinent references are (a) M. Nencki, *Ber.*, **17**, 2277 (1884); (b) C. Gränacher, M. Gero, A. Ofner, A. Klopfenstein and E. Schlatter, *Helv. Chim. Acta*, **6**, 458 (1923); (c) P. Julian and B. Sturgis, THIS JOURNAL, **57**, 1126 (1935); (d) F. C. Brown, C. K. Bradsher, S. G. McCallum and M. Potter, *J. Org. Chem.*, **15**, 174 (1950); (e) F. C. Brown, C. K. Bradsher, S. M. Bond and M. Potter, THIS JOURNAL, **73**, 2357 (1951).

(4) (a) R. Andreasch, *Monatsh.*, **38**, 138 (1917); (b) C. Gränacher and A. Mahal, *Helv. Chim. Acta*, **6**, 467 (1923); (c) R. V. Jones and H. R. Henze, THIS JOURNAL, **64**, 1669 (1942).

(5) F. Kučera, *Monatsh.*, **35**, 137 (1914).

(6) F. B. Dains and A. E. Stephenson, THIS JOURNAL, **38**, 1843 (1916).

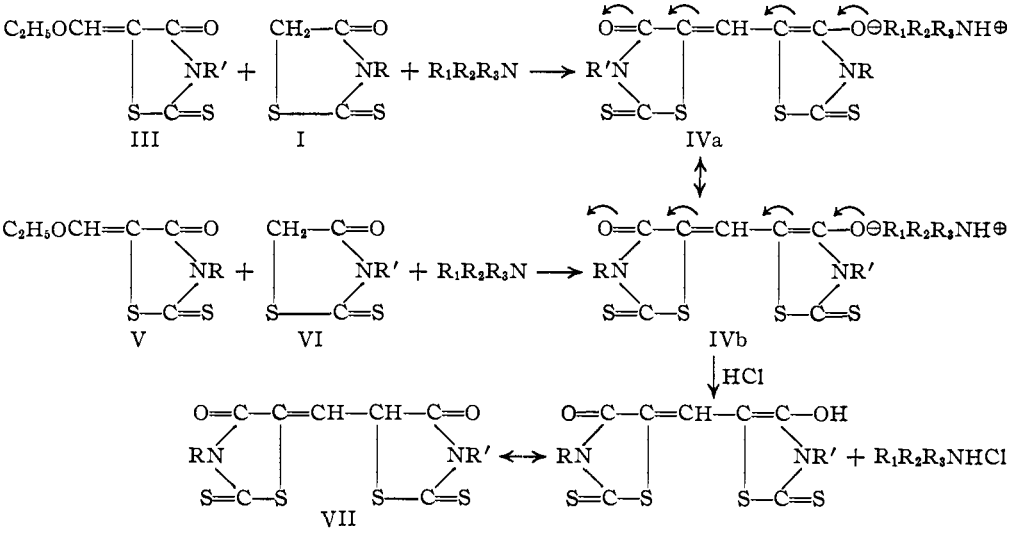
(7) J. D. Kendall and D. J. Fry, British Patent 540,577 (1941).

In a preliminary experiment, the reaction of 5-ethoxymethylene-3-methylrhodanine with 3-methylrhodanine was chosen in order to obtain a product which has been described by Kendall and Fry. When these two components were heated in acetone in the presence of one mole equivalent of triethylamine, a deep purple crystalline product was obtained. It decomposed at the same temperature (230°) as that reported by Kendall and Fry. However, analysis showed that it contained 10.3% nitrogen and 31.8% sulfur, whereas 5,5′-methylidynebis-(3-methylrhodanine) required 9.2% nitrogen and 42.1% sulfur. It was therefore concluded that this could not be the 5,5′-methylidynebis-(3-methylrhodanine). On the other hand, this analysis corresponded to the composition of this compound plus one molecule of triethylamine. To further substantiate this interesting finding, the reaction was repeated with the use of different tertiary amines. In every case, the analysis indicated the presence of one molecule of the amine used. The results are shown in Table II.

These products are believed to be the amine salts of 5,5′-methylidynebis-(3-methylrhodanine).[8] The acidic property of the 5,5′-methylidynebis-(3-alkylrhodanines) appears to be due to the stabilization of the enolate ions by resonance. That the product is an amine salt is further supported by the following experiments. When 5-ethoxymethylene-3-methylrhodanine (III, R′ = CH_3) was treated with 3-phenylrhodanine (I, R = C_6H_5) in the presence of the triethylamine, there was obtained a product which was identical with that obtained from 5-ethoxymethylene-3-phenylrhodanine (V, R = C_6H_5) and 3-methylrhodanine (VI, R′ = CH_3) under the same conditions. These results appear to indicate that the enolate ion of the product is a hybrid of the extreme resonant forms IVa and IVb.

When one mole of 3,3′-ethylenebis-(5-ethoxymethylenerhodanine) was treated with two moles of 3-substituted rhodanines in the presence of two moles of triethylamine, the products were found to contain two molecules of the amine as expected.

Treatment of the amine salts of 3,3′-disubstituted 5,5′-methylidynebisrhodanines with hot acetic acid yielded in most cases mixtures of the 5,5′-methylidynebisrhodanine and the unchanged amine salt. In one case, the triethylamine salt of 3-methyl-3′-phenyl-5,5′-methylidynebisrhodanine, the amine salt could be recrystallized from acetic acid. The free 5,5′-methylidynebisrhodanines VII were obtained in pure form by treatment of the amine salts with hydrochloric acid. For example, 5,5′-methylidynebis-(3-methylrhodanine) (VII, R = R′ = CH_3) was obtained from the reaction of hydrochloric acid on its triethylamine or benzyldimethylamine salt. Thus this furnishes a general method for the synthesis of 3,3′-disubstituted 5,5′-methylidynebisrhodanines. Furthermore, if a 3-substituted 5-alkoxymethylenerhodanine was condensed with a rhodanine having a different 3-substituent, the final product would be 3,3′-unsymmetrically substituted 5,5′-methylidynebisrhodanines which have not been previously reported in the literature.

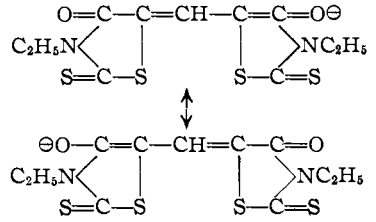


The 5-alkoxymethylenerhodanines reported here possess a reactive alkoxy group and are capable of reacting with a variety of compounds. Their reactions with amines and other compounds will be reported later.

Experimental[9]

The 3-substituted rhodanines were prepared by the general method described in reference 10. The 3-(3′,5′,5′-

(8) After this work was completed, there appeared an article by L. G. S. Brooker, G. H. Keyes, R. H. Sprague, R. H. van Dyke, E. van Lare, G. van Zandt, F. L. White, H. W. J. Cressman and S. G. Dent (THIS JOURNAL, **73**, 5332 (1951)) in which they reported the triethylamine salt of 5,5′-methylidynebis-(3-ethylrhodanine) and mentioned the resonance

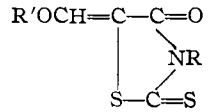


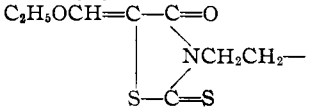
(9) All melting points are uncorrected.

(10) C. E. Redemann, R. N. Icke and G. A. Alles, *Org. Syntheses*, **27**, 73 (1947).

TABLE I

5-ALKOXYMETHYLENERHODANINES



R	R'	M.p., °C.	Yield, %	Formula	Nitrogen, % Calcd.	Nitrogen, % Found	Sulfur, % Calcd.	Sulfur, % Found
H	CH_3	197–198	35	$C_5H_5NO_2S_2$	8.0	8.1	36.6	37.0
H	C_2H_5	157–158	60	$C_6H_7NO_2S_2$[a]	7.4	7.3	33.8	34.3
CH_3	C_2H_5	132–133	57	$C_7H_9NO_2S_2$	6.9	6.8	31.5	31.2
C_9H_{19}[b]	C_2H_5	39–41[c]	47	$C_{15}H_{25}NO_2S_2$	4.5	4.2	20.3	20.6
C_6H_5	C_2H_5	153–155	68	$C_{12}H_{11}NO_2S_2$	5.3	5.3	24.2	24.7
	C_2H_5	207–209	59	$C_{14}H_{16}N_2O_4S_4$	7.0	6.9	31.8	31.8

[a] Calcd. for $C_6H_7NO_2S_2$: C, 38.1; H, 3.7; mol. wt., 189. Found: C, 38.4; H, 4.0; mol. wt., 190. [b] 3,5,5-Trimethylhexyl. [c] A mixture of this and the starting material, 5-(3′,5′,5′-trimethylhexyl)-rhodanine, melted at room temperature (27°).

trimethylhexyl)-rhodanine, a new compound, was obtained in 59% yield from 3,5,5-trimethylhexylamine. It had a melting point of 41–43°.

Anal. Calcd. for $C_{12}H_{21}NOS_2$: N, 5.4. Found: N, 5.3.

The preparation of 5-alkoxymethylenerhodanines is represented by the following example.

5-Ethoxymethylenerhodanine.—A mixture of rhodanine (140 g.), ethyl orthoformate (200 ml.) and acetic anhydride (300 ml.) was heated under reflux for 17.5 hours. The wine-red crystals which separated upon cooling were collected and washed with acetic acid. The dried product weighed 118 g. and melted at 152–155°. It was recrystallized from acetic acid; m.p. 157–158°.

Table I summarizes the 5-ethoxymethylenerhodanines prepared.

Tertiary Amine Salt of 5,5′-Methylidynebis-(3-methylrhodanine).—The general method of preparation of these amine salts is illustrated by the following example.

Triethylamine Salt of 5,5′-Methylidynebis-(3-methylrhodanine).—A mixture of 5-ethoxymethylene-3-methylrhodanine (4 g.), 3-methylrhodanine (3 g.), triethylamine (3 ml.) and acetone (20 ml.) was heated under reflux for one hour. After cooling, the solid was collected and washed with benzene. The dried product melted at 228–229° (with decomposition) and weighed 5.5 g.

The other amine salts similarly prepared are summarized in Table II. All the amine salts of 5,5′-methylidynebis-(3-methylrhodanine) are purple except the benzyldimethylamine salt which is green. They are crystalline solids which melt with decomposition.

TABLE II

AMINE SALTS OF 5,5′-METHYLIDYNEBIS-(3-METHYLRHODANINE)

Amine	M.p., °C.[a]	Yield, %	Nitrogen, % Calcd.	Nitrogen, % Found
Triethylamine	228–229	69	10.4	10.3[b]
2-Ethylhexyldimethylamine	146–147	44	9.1	9.0
Benzyldimethylamine	216–217	34	9.6	9.5
Triethanolamine	188–190	45	9.2	8.8[c]
N-Methylmorpholine	243–244	10	10.4	10.0

[a] With decomposition. [b] Calcd. for $C_{15}H_{23}N_3O_2S_4$: S, 31.6. Found: S, 31.8. [c] Calcd. for $C_{15}H_{23}N_3O_5S_4$: S, 28.2. Found: S, 28.4.

5,5′-Methylidynebis-(3-methylrhodanine). (a) **From Triethylamine Salt.**—To a suspension of triethylamine salt of 5,5′-methylidynebis-(3-methylrhodanine) (3.3 g.) in acetic acid (100 ml.) was added concentrated hydrochloric acid (1 ml.). The mixture was heated on a steam-bath to complete the reaction. After cooling, the solid was collected and washed with acetic acid. The product was a red solid which melted at 173–174° and weighed 2.5 g. (quantitative).

Anal. Calcd. for $C_9H_8N_2O_2S_4$: N, 9.2; S, 42.1. Found: N, 9.1; S, 41.8.

(b) **From Benzyldimethylamine Salt.**—The benzyldimethylamine salt of 5,5′-methylidynebis-(3-methylrhodanine) (1 g.) was treated with hydrochloric acid as above. There was obtained 0.7 g. of a red solid which melted at 173–174°. A mixture with the product obtained in (a) melted at the same temperature.

3-Methyl-3′-phenyl-5,5′-methylidynebisrhodanine. (a) **From Ethoxymethylene-3-methylrhodanine and 3-Phenylrhodanine.**—A mixture of 5-ethoxymethylene-3-methylrhodanine (4 g.), 3-phenylrhodanine (4 g.), triethylamine (3 ml.) and acetone (20 ml.) was heated under reflux for 1.5 hours. The triethylamine salt of 3-methyl-3′-phenyl-5,5′-methylidynebisrhodanine (6.4 g., 88%) was obtained as purple crystals which had a m.p. of 230–231° dec.

Anal. Calcd. for $C_{20}H_{25}N_3O_2S_4$: N, 9.0; S, 27.4. Found: N, 8.7; S, 27.7.

This salt was treated with hydrochloric acid as above. The 3-methyl-3′-phenyl-5,5′-methylidynebisrhodanine (2.3 g., 98%) was obtained as a red solid; m.p. 177.5–178.5°.

Anal. Calcd. for $C_{14}H_{10}N_2O_2S_4$: N, 7.7; S, 35.0. Found: N, 7.5; S, 35.5.

(b) **From 5-Ethoxymethylene-3-phenylrhodanine and 3-Methylrhodanine.**—The reaction of these two compounds in the presence of triethylamine as in (a) yielded the triethylamine salt of 3-methyl-3′-phenyl-5,5′-methylidynebisrhodanine (94%); m.p. and mixed m.p. with that obtained in (a), 231° dec.

This product upon treatment with hydrochloric acid yielded the 3-methyl-3′-phenyl-5,5′-methylidynebisrhodanine, m.p. and mixed m.p. with that obtained in (a), 178–179°.

Bis-triethylamine Salt of 3,3′-Ethylenebis-5-(2″-thiono-4″-keto-3″-methyl-5″-thiazolidylmethylenerhodanine).—A mixture of 3,3′-ethylenebis-(5-ethoxymethylenerhodanine) (3 g.), 3-methylrhodanine (3 g.) and triethylamine (3 ml.) and acetone (30 ml.) was treated as above. The bistriethylamine salt was obtained as a purple solid (4 g., 50%) melting at 235–236° dec.

Anal. Calcd. for $C_{30}H_{44}N_6O_4S_8$: N, 10.4; S, 31.7. Found: N, 9.9; S, 31.7.

This bistriethylamine salt upon treatment with hydrochloric acid yielded the **3,3′-ethylenebis-5-(2″-thiono-4″-keto-3″-methyl-5″-thiazolidylmethylenerhodanine)**, m.p. 122°.

Anal. Calcd. for $C_{18}H_{14}N_4O_4S_8$: N, 9.3; S, 42.3. Found: N, 8.9; S, 41.8.

Bis-triethylamine Salt of 3,3′-Ethylenebis-5-(2″-thiono-4″-keto-3″-phenyl-5″-thiazolidylmethylenerhodanine).—A mixture of 3,3′-ethylenebis-(5-ethoxymethylenerhodanine) (3 g.), 3-phenylrhodanine (4 g.), triethylamine (3 g.) and acetone (30 ml.) reacted as above. The bistriethylamine salt was obtained as a brown solid (3.4 g., 37%) melting at 253–254° dec.

Anal. Calcd. for $C_{40}H_{48}N_6O_4S_8$: N, 9.0; S, 27.5. Found: N, 8.8; S, 27.6.

This bistriethylamine salt upon treatment with hydrochloric acid yielded the **3,3′-ethylenebis-5-(2″-thiono-4″-**

keto-3″-phenyl-5″-thiazolidylmethylenerhodanine) as a red solid, m.p. 197° dec.

Anal. Calcd. for $C_{28}H_{18}N_4O_4S_3$: N, 7.7; S, 35.2. Found: N, 7.7; S, 34.7.

Acknowledgment.—The authors are indebted to Mr. T. P. Callan and his staff for chemical analyses.

PHILADELPHIA 37, PENNSYLVANIA

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, FLORIDA STATE UNIVERSITY]

The Role of the Solvent in Radical Decomposition Reactions: *p*-Nitrophenylazotris-(*p*-anisyl)-methane

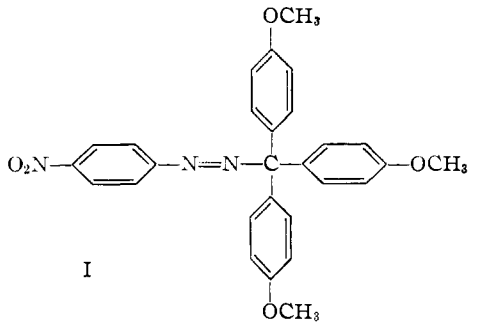
BY MENDEL D. COHEN,[1] JOHN E. LEFFLER[2] AND LIBERO M. BARBATO[3]

RECEIVED APRIL 5, 1954

The decomposition of *p*-nitrophenylazotris-(*p*-anisyl)-methane has been studied in a series of solvents at several temperatures. The changes observed in the activation parameters can be explained in terms of differential solvation of the initial and transition states. In spite of the highly asymmetrical structure of this compound, there appears to be no shift to a polar mechanism in the solvents studied.

Introduction

The decomposition of *p*-nitrophenylazotris-(*p*-anisyl)-methane (I) is of twofold theoretical interest. The substituents present in this molecule should make its interaction with the solvent different from that of phenylazotriphenylmethane[4] and thus afford a test of the complexing theory of special solvent effects. The polar nature of the substituents might, but apparently did not, shift the mechanism from a radical to a polar one.[5] The decomposition therefore helps to locate the structural boundaries separating radical from polar mechanisms.



I

Interpretation of the Results

The decomposition of the azo compound has been followed by the change in pressure of the nitrogen evolved. The rate constants are presented in Table I and the activation parameters in Table II. The trend in activation enthalpies does not parallel that expected for the enthalpies of complex formation of the solvent with the azo compound. This is in contrast to the behavior of phenylazotriphenylmethane, whose enthalpy of activation appeared to include the enthalpy necessary to remove solvent from the solvated ground state molecules.[4] Nor is there any parallelism between the activation parameters and the solubility of the substituted azo compound. The solubilities will also be found in Table II.

(1) Office of Naval Research Post-Doctoral Research Associate, 1952–1953. Present address: Weizman Institute, Rehovoth, Israel.

(2) To whom requests for reprints should be sent.

(3) Office of Naval Research Post-Doctoral Research Associate, 1953–1954.

(4) M. G. Alder and J. E. Leffler, THIS JOURNAL, **76**, 1425 (1954).

(5) J. E. Leffler, *ibid.*, **72**, 67 (1950).

TABLE I

FIRST-ORDER RATE CONSTANTS FOR THE DECOMPOSITION OF *p*-NITROPHENYLAZOTRIS-(*p*-ANISYL)-METHANE IN VARIOUS SOLVENTS

Temp., °K.	Benzonitrile C_0[a]	Benzonitrile k[b]	Benzonitrile-veratrole[c] C_0[a]	Benzonitrile-veratrole[c] k[b]	Decalin C_0[a]	Decalin k[b]	Veratrole C_0[a]	Veratrole k[b]
360.31	5.32	304	2.71	267	4.37	241	3.11	284
	2.61	296	5.38	267	1.86	232	4.41	291
	3.81	288			3.75	223		
	4.16	290			1.84	232		
350.76	2.65	100.8	3.83	87.5	2.77	73.8	3.33	89.6
	2.01	104.1	4.55	90.1	2.94	73.3	3.33	93.7
	5.36	104.8	3.66	94.6	1.93	74.0	5.11	87.9
	4.39	101.4	4.33	90.0	3.23	75.1	4.02	88.1
335.08	5.53	14.0	4.35	13.0	1.96	9.96	3.35	12.25
	4.16	15.7	3.15	13.2	3.85	8.77	3.13	11.33
	1.73	14.6	5.96	12.3	4.78	10.63	4.08	9.91
	3.69	14.4	5.61	12.4	4.43	9.08	3.75	12.64
					2.33	10.82	1.30	11.26
							4.18	10.67

[a] Initial concentration in moles/liter times 10^3. [b] Rate constant in sec.$^{-1}$ times 10^5. [c] 49.4% benzonitrile by weight.

TABLE II

SOLUBILITIES AND ACTIVATION PARAMETERS FOR THE DECOMPOSITION OF *p*-NITROPHENYLAZOTRIS-(*p*-ANISYL)-METHANE

Solvent	Enthalpy of activation, kcal./mole	Entropy of activation, cal./mole degree	Solubility, g./100 cc.
Benzonitrile	27.86 ± 0.16	6.88 ± 0.46	2.9
Mixed benzonitrile (49.4% by weight) and veratrole	28.35 ± .16	8.06 ± .48	1.15
Decalin	29.37 ± .25	10.58 ± .71	0.19
Veratrole	30.04 ± .31	12.86 ± .90	9.24

Solvent Effects on the Activation Parameters.—In order to interpret the present solvent effects by means of the complexing theory, it is necessary to assume that some of the solvents solvate the transition state more and that others solvate the ground state more. For steric reasons, it seems likely that solvation of both types will be confined largely to the *p*-nitrophenylazo part of the molecule. There will be resonance contributions to the ground state corresponding to a negative charge on the oxygen of the nitro group and a positive charge on the benzene ring or adjacent azo group. This partial positive charge, or analogous structures in the solvent complex, will cause the ground state to be solvated by electron-rich molecules such as