the complex and acid extraction and treatment with base of the acidic aqueous fraction gave the product as a free base, which was isolated by ether extraction. The ether solution was dried and stripped of solvent. Fractionation of this residue gave 11 g. (58% yield) of product, boiling at 87–93° (0.15 mm.).

The hydrochloride salt of the amine prepared by the addition of isopropanolic hydrogen chloride to an ethereal solution of the base melted at 199–200°.

Anal. Calcd. for $C_{13}H_{17}NO \cdot HCl$: C, 64.85; H, 7.53. Found: C, 64.95; H, 7.78.

The hexahydro - 9a - dibenzofuranamines prepared by this method have been listed in Table V.

4a,5,6,7,8,8a-Hexahydro-N-(3-methoxypropyl)-4H-indeno-[1,2 - *b*]thiophen - 8a - amine Hydrochloride (XXX).—This compound was prepared by the reaction of 12 g. (0.0435 mole) of 2-(2-bromo-3-thenyl)cyclohexanone (*vide infra*) with 8 g. of 3-methoxypropylamine in benzene (benzenesulfonic acid as catalyst), followed by treatment of the bromo-imine with butyllithium at −70°, as described for the preparation of XXVIII. There was obtained 7 g. (54%) of product, m.p. 145–146° (methanol/ether).

Anal. Calcd. for $C_{15}H_{23}NOS \cdot HCl$: C, 59.70; H, 8.01. Found: C, 59.73; H, 8.25.

Acknowledgment.—The authors are indebted to Dr. Vandenbelt and Messrs. E. Schoeb and R. B. Scott for spectral data and interpretations, and to Mr. C. E. Childs and associates for analytical results. Special thanks are due to Dr. R. Parcell for the many helpful discussions and suggestions during the course of this work.

Synthetic Approaches to Quinoxaline Antibiotics. Synthesis of Bisquinoxaloyl Derivatives[1a]

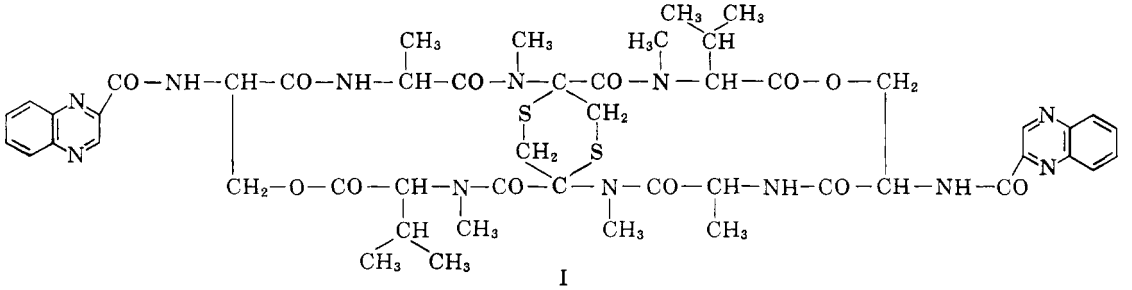
HENRY C. KOPPEL, IRWIN L. HONIGBERG, ROBERT H. SPRINGER, AND C. C. CHENG[1b]

Midwest Research Institute, Kansas City 10, Missouri

Received November 13, 1962

The synthetic approach to the preparation of some antibiotics composed of two quinoxaline rings connected through polypeptide linkages has been studied. Model compounds of the types R—CO—NH$(CH_2)_x$—NH—CO—R and R—CO—NH—CH_2—CO—NH$(CH_2)_x$—NH—CO—CH_2—NH—CO—R, where R = quinoxalinyl group, have been synthesized.

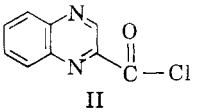
A number of polypeptide antibiotics such as levomycin,[2] actinoleukin,[3] echinomycin,[4] quinomycin,[5] etc., containing the quinoxaline moiety have recently been reported. The structure of echinomycin (I), which was found to be identical with quinomycin A,[6] is composed of two quinoxaline rings linked by peptide chains containing the amino acids D-serine, L-alanine, L-N-methylcysteine, and L-N-methylvaline.



I

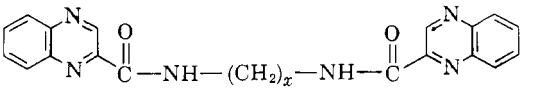
These antibiotics have not yet been synthesized. In order to understand the importance of the peptide link and the requirement for the amino acid sequence in this type of compound,[7] the synthesis of hitherto unreported bisquinoxaloyl (bisquinoxalinecarbonyl) derivatives was investigated.

2-Quinoxalinecarboxylic acid, prepared by the oxidation of 2-methylquinoxaline,[8] was chlorinated in thionyl chloride to yield 2-quinoxaloyl chloride (II). Interaction of two equivalents of II with one equivalent



II

of the appropriate diamine in an inert solvent, in the presence of triethylamine, readily yielded the N,N′-polymethylenebis-2-quinoxalinecarboxamides (III).



III

(1) (a) This investigation was supported by the Cancer Chemotherapy National Service Center (contract SA-43-ph-3025), National Cancer Institute, National Institutes of Health, U. S. Public Health Service. (b) To whom all inquiries should be directed.

(2) H. E. Carter, C. P. Schaffner, and D. Gottlieb, *Arch. Biochem. Biophys.*, **53**, 282 (1954).

(3) M. Ueda, Y. Tanigawa, Y. Okami, and H. Umezawa, *J. Antibiotics*, **7**, 125 (1954).

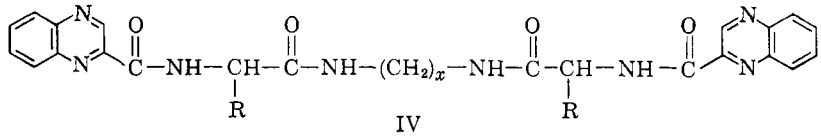
(4) (a) R. Corbaz, L. Ettilinger, E. Gäumann, W. Keller-Schierlein, F. Kradolfer, L. Neipp, V. Prelog, P. Reusser, and H. Zähner, *Helv. Chim. Acta*, **40**, 199 (1957); (b) W. Keller-Schierlein, M. L. Mihailović, and V. Prelog, *ibid.*, **42**, 305 (1959).

(5) (a) T. Yoshida, K. Katagiri, and S. Yokozawa, *J. Antibiotics*, **14A**, 330 (1961); (b) J. I. Shōji and K. Katagiri, *ibid.*, **14A**, 335 (1961).

(6) K. Katagiri, and K. Sugiura, "Antimicrobial Agents and Chemotherapy—1961," M. Finland and G. M. Savage, ed., Braun-Brumfield, Inc., Ann Arbor, Mich., 1962, p. 162.

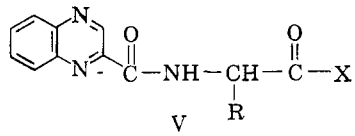
(7) (a) Most of these quinoxaline antibiotics were found to have interesting biological activity, see ref. 2–6; (b) D. A. Hall, *Biochem. J.*, **40**, xlii (1946), has stated that the growth of *Streptococcus lactis* R. is inhibited by quinoxaline. R. M. Acheson, *J. Chem. Soc.*, 4731 (1956), also reported the growth-inhibitory effect of some quinoxaline derivatives on *Lactobacillus casei*.

(8) B. R. Brown, *J. Chem. Soc.*, 2577 (1949).



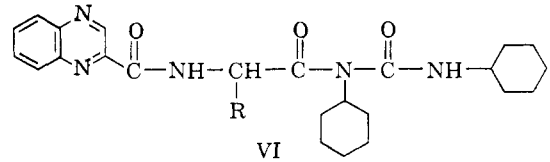
IV

Compounds of type IV should be prepared by a normal peptide-forming reaction. Many classical methods of peptide synthesis have been reported,[9] yet almost all seemed to have drawbacks that precluded their use for the preparation of this type of bisquinoxaloyl derivative. Attempts to bring about the reaction of α,ω-diaminoalkanes with the ethyl ester of N-2-quinoxalinecarbonylglycine (V. R = H, X = OC_2H_5), preparation of the corresponding acid chloride from N-2-quinoxalinecarbonylglycine (V. R = H, X = OH), and reaction of the acyl azide (V. R = H, X = N_3) with α,ω-diaminoalkanes, etc., were all unsuccessful owing to the unreactive character or insolubility of this type of quinoxalinyl intermediate.



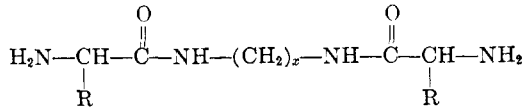
V

Utilization of carbodiimide is also a standard method for peptide synthesis.[9,10] When a mixture of two equivalents of N-2-quinoxalinecarbonylglycine (V. R = H, X = OH) and one equivalent of 1,3-diaminopropane was stirred in the presence of two equivalents of N,N'-dicyclohexylcarbodiimide, the desired product IV (R = H, x = 3) was not obtained, but the reaction took a different route to form a substituted N-acylurea (VI. R = H). N-2-Quinoxalinecarbonyl-*dl*-serine (V. R = CH_2OH, X = OH) and α,ω-diaminoalkanes with this carbodiimide, similarly, gave the corresponding N-acylurea (VI. R = CH_2OH). The formation of this type of N-acylurea from carbodiimide has been discussed by Khorana[11a] and Sheehan.[11b]



VI

Since the usual acylating methods had failed, synthesis of the inner "core" of IV, illustrated by structure VII, followed by reaction with two moles of quinoxaloyl chloride, should yield the desired compounds (IV). This proved to be the case: two equivalents of carbobenzoxyglycine (VIII. R = H), triethylamine, and ethyl chlorocarbonate were stirred at low temperature in toluene, the resulting product was then treated *in situ* with one equivalent of diamine to form N,N'-[polymethylenebis(iminocarbonylmethylene)]bisbenzoxycarboxamide (IX. R = H). Catalytic hydrogenation of IX (R = H) readily yielded VII (R = H), the "core." The latter compound was then treated with two equivalents of 2-quinoxaloyl chloride (II) to yield the desired compound IV (R = H).[12] Similarly, by using the carbobenzoxy derivative of other amino acids (VIII. R = $CH(CH_3)_2$, CH_2—C_6H_5, etc.), the corresponding product, IV, could be obtained.



VII

This synthesis thus proved to be a useful method for the preparation of bisquinoxaloyl derivatives with peptide linkages,[13] which may be capable of extension to the synthesis of more complex molecules such as the quinoxaline antibiotics.

Experimental[14]

2-Quinoxaloyl Chloride (II).—A mixture of 20 g. of 2-quinoxalinecarboxylic acid[8] in 100 ml. of thionyl chloride was refluxed for 40 min. Excess thionyl chloride was distilled under reduced pressure from the resulting dark brown solution. The solid residue was dissolved in 250 ml. of hot toluene, treated with charcoal, and filtered. The volume of the filtrate was reduced to 50 ml. and chilled. The crystalline product was recrystallized from a large volume of petroleum ether (b.p. 60–70°) to give 16 g. (79%) of long, white needles, m.p. 112–113°.

Anal. Calcd. for $C_9H_5ClN_2O$: C, 56.1; H, 2.60; N, 14.5. Found: C, 56.1; H, 2.89; N, 14.2.

General Synthesis of N,N'-Polymethylenebis-2-quinoxalinecarboxamides (III).—To a stirred and refluxing solution of one equivalent of II in tetrahydrofuran was added dropwise a solution of one equivalent of triethylamine and one-half equivalent of the appropriate diamine. A precipitate appeared almost immediately. After the addition, the mixture was refluxed for 15 min., cooled, and filtered. The crude product was washed successively with water, dilute hydrochloric acid, water, dilute sodium carbonate solution, water, and finally acetone. It then was recrystallized from the appropriate solvent (see Table I).

Ethyl Ester of N-2-Quinoxalinecarbonylglycine (V. R = H, X = OC_2H_5).—To a stirred mixture of 13.9 g. (0.1 mole) of the ethyl ester of glycine hydrochloride in 75 ml. water and 200 ml. of methylene chloride cooled to 10° was added dropwise a solution of 19.3 g. (0.1 mole) of II in 75 ml. of methylene chloride. During this addition 5.2 g. (0.13 mole) of magnesium oxide was added to the reaction mixture in three equal portions. After the addition was complete, the reaction mixture was allowed to stir for 30 min., then 5 ml. of pyridine was added and the stirring was continued for another 5 min. The reaction mixture was then acidified to pH 4 with dilute hydrochloric acid and the methylene chloride layer was separated, washed successively with dilute hydrochloric acid, water, dilute sodium carbonate, and water, and then dried over anhydrous sodium sulfate. Evaporation of the solvent under reduced pressure yielded a light pink residue, which was recrystallized from a mixture of benzene and heptane to give 22.4 g. (87% yield) of light pink needles, m.p. 89–91°.

Anal. Calcd. for $C_{18}H_{13}N_3O_3$: C, 60.2; H, 5.02; N, 16.2. Found: C, 60.1; H, 5.19; N, 16.0.

(9) For leading references, see an excellent review by J. P. Greenstein and M. Winitz, "Chemistry of the Amino Acids," Vol. II, John Wiley and Sons, Inc., New York, N. Y., 1961.

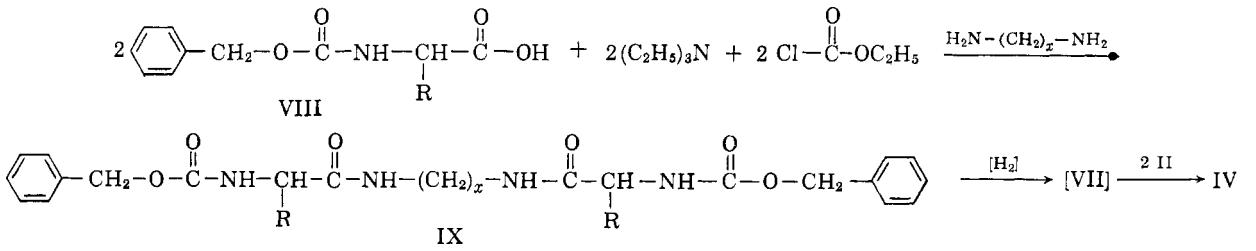
(10) J. C. Sheehan and G. P. Hess, *J. Am. Chem. Soc.*, **77**, 1067 (1955).

(11) (a) H. G. Khorana, *Chem. Ind.* (London), 1087 (1955); (b) J. C. Sheehan, M. Goodman, and G. P. Hess, *J. Am. Chem. Soc.*, **78**, 1367 (1956).

(12) This method is a utilization of the mixed carboxylic–carbonic acid anhydride procedure described by J. R. Vaughan, Jr., and R. L. Osato, *ibid.*, **74**, 676 (1952).

(13) In contrast to the type of compounds prepared in this laboratory, a recent publication described the preparation of succinamino peptides in which the arrangement of the amino acids in the peptides is as follows: —CO—CHR—NH—CO—CH_2—CH_2—CO—NH—CHR—CO—. This arrangement is, interestingly enough, just in the reversed order as compared with ours. See S. Berse, L. Piché, L. Lachance, and G. Laflamme, *J. Org. Chem.*, **27**, 3489 (1962).

(14) All melting points were taken on a Thomas–Hoover melting point apparatus.

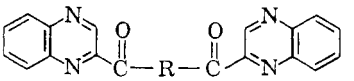


VIII

IX

TABLE I

BISQUINOXALOYL DERIVATIVES



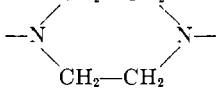
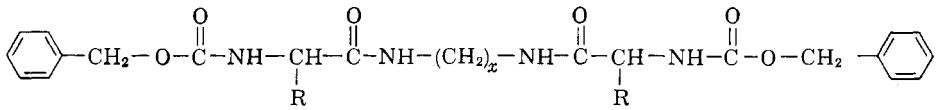
R	Recrystallization solvents	M.p., °C.	Yield, %	Formula	Analyses—Calcd. C	H	N	Found C	H	N
—NH—$(CH_2)_3$—NH—	Dimethylformamide–water	225–227	40	$C_{21}H_{18}N_6O_2$	65.3	4.66	21.7	65.2	4.70	21.4
—NH—$(CH_2)_5$—NH—	Ethyl acetate	186–187	49	$C_{23}H_{22}N_6O_2$	66.7	5.31	20.3	66.6	5.25	20.0
—NH—$(CH_2)_6$—NH—	Ethanol–water	153–154	59	$C_{24}H_{24}N_6O_2$	67.3	5.61	19.6	67.4	5.61	19.4
—NH—$(CH_2)_{10}$—NH—	Methanol	109 dec.	50	$C_{28}H_{32}N_6O_2$	69.4	6.61	17.4	69.3	6.50	17.4
	Dimethylformamide–water	>250 dec.	60	$C_{22}H_{18}N_6O_2$	66.3	4.52	21.1	66.1	4.68	21.2

TABLE II

N,N′-[POLYMETHYLENEBIS(IMINOCARBONYLMETHYLENE)]BISBENZOXYCARBOXAMIDES



R	x	Recrystallization solvents	M.p., °C.	Yield, %	Formula	Analyses—Calcd. C	H	N	Found C	H	N
H	3	95% Ethanol	177–178	62	$C_{23}H_{28}N_4O_6$	60.5	6.14	12.3	60.6	6.34	12.1
H	5	95% Ethanol–methanol	174–175	51	$C_{25}H_{32}N_4O_6$	62.0	6.61	11.6	61.9	6.79	11.3
H	6	Ethanol–water	170–171	50	$C_{26}H_{64}N_4O_6$	62.7	6.83	11.3	62.6	7.20	11.3
H	10	Dimethylformamide–95% ethanol	171–172	44	$C_{30}H_{42}N_4O_6$	65.0	7.58	10.1	64.8	7.72	10.1
$CH(CH_3)_2$	3	95% Ethanol	>175 dec.	49	$C_{29}H_{40}N_4O_6$	64.4	7.41	10.4	64.5	7.72	10.3
$CH(CH_3)_2$	6	Dimethylformamide–methanol	194–196	60	$C_{32}H_{46}N_4O_6$	66.0	7.90	9.6	66.2	8.22	9.8
CH_2—C_6H_5	5	Dimethylformamide–water	170 dec.	54	$C_{39}H_{44}N_4O_6$	70.4	6.63	8.4	70.0	6.60	8.5

N-2-Quinoxalinecarbonylglycine (V. R = H, X = OH).—To a stirred solution of 5.3 g. (0.07 mole) of glycine, 2.8 g. (0.07 mole) of sodium hydroxide in 50 ml. of water and 100 ml. of acetone cooled to 10° was added simultaneously and dropwise a solution of 13.0 g. (0.07 mole) of II in 100 ml. of acetone and a solution of 2.8 g. (0.07 mole) of sodium hydroxide in 50 ml. water. Toward the end of the addition a precipitate appeared and the pH of the reaction mixture fell to 6. This was readjusted to pH 9 by the addition of dilute sodium hydroxide. The reaction mixture was stirred for 1 hr. at room temperature and its pH was adjusted to 6 with dilute hydrochloric acid, at which time a clear solution resulted. Acetone was distilled under reduced pressure and the residual solution was acidified to pH 1 with concentrated hydrochloric acid. On cooling, a white solid product precipitated. This was recrystallized from water to give 9.0 g. (56% yield) of white needles which sublimed at 215° and decomposed at 226°.

Anal. Calcd. for $C_{11}H_9N_3O_3$: C, 57.1; H, 3.90; N, 18.2. Found: C, 57.3; H, 4.16; N, 18.0.

N-2-Quinoxalinecarbonyl-*dl*-serine (V. R = CH_2OH, X = OH) was similarly prepared from 25.2 g. (0.24 mole) of *dl*-serine and 47.0 g. (0.24 mole) of II to give, after recrystallization from a mixture of dimethylformamide and water, 37.0 g. (59% yield) of white needles, m.p. 224° dec.

Anal. Calcd. for $C_{12}H_{11}N_3O_4$: C, 55.2; H, 4.21; N, 16.1. Found: C, 54.8; H, 4.22; N, 16.4.

N-2-Quinoxalinecarbonylglycine Hydrazide (V. R = H, X = $NHNH_2$).—To a solution of 7.5 g. (0.03 mole) of ethyl ester of N-2-quinoxalinecarbonylglycine (V. R = H, X = OC_2H_5) in 125 ml. of ethanol at 70° was added 1.92 g. (0.06 mole) of anhydrous hydrazine. The mixture was stirred for 1 hr. and cooled and the resulting solid was filtered and washed with cold ethanol and ether. Recrystallization from ethanol afforded 6.5 g. (90% yield) of white crystals which decomposed at 215°.

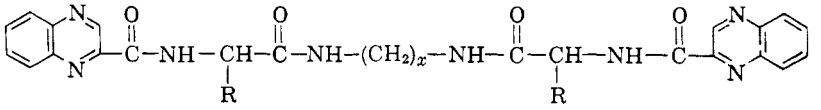
Anal. Calcd. for $C_{11}H_{11}N_5O_2$: C, 53.9; H, 4.49; N, 28.6. Found: C, 53.8; H, 4.53; N, 28.6.

N,N′-Dicyclohexyl-N-(N″-2-quinoxalinecarbonyl-*dl*-seryl)-urea (VI. R = CH_2OH).—To a solution of 13.0 g. (0.05 mole) of N-2-quinoxalinylcarbonyl-*dl*-serine (V. R = CH_2OH, X = OH) in 150 ml. of dimethylformamide was added 1.8 g. (0.025 mole) of 1,3-diaminopropane followed by 10.3 g. (0.05 mole) of N,N′-dicyclohexylcarbodiimide.[15] The solution was stirred for 4 hr. and a small amount of solid was removed by filtration. The filtrate was added to 400 ml. of water. An oily substance which formed gradually solidified upon stirring. This was collected and recrystallized from methanol to give 7.0 g. (30% yield) of the product as white crystals, m.p. 169–171°.

(15) E. Schmidt, F. Hitzler, and E. Lahde, *Ber.*, **71**, 1933 (1938). Purchased from Mann Research Laboratories, Inc., New York, N. Y.

TABLE III

N,N′-{POLYMETHYLENEBIS[IMINOCARBONYL(SUBSTITUTED-METHYLENE)]}BIS-2-N″-QUINOXALINECARBOXAMIDES



R	x	Recrystallization solvents	M.p., °C.	Yield, %	Formula	Analyses Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
H	3	*p*-Dioxane	>200 dec.	37	$C_{25}H_{24}N_8O_4$	60.0	4.80	22.4	59.6	5.18	22.6
H	5	Dimethylformamide–water	>240 dec.	10	$C_{27}H_{28}N_8O_4$	61.4	5.30	21.2	61.4	5.12	21.0
H	6	Dimethylformamide–water	257 dec.	11	$C_{28}H_{30}N_8O_4$	62.0	5.54	20.6	62.1	5.80	20.2
H	10	Dimethylformamide–water	228–229	8	$C_{32}H_{38}N_8O_4$	64.2	6.35	18.7	64.0	6.52	18.8
CH_2—C_6H_5	5	Methanol	>170 dec.	12	$C_{41}H_{40}N_8O_4$	69.5	5.65	15.8	69.8	5.84	15.4

Anal. Calcd. for $C_{25}H_{33}N_5O_4$: C, 64.2; H, 7.07; N, 14.9. Found: C, 64.1; H, 7.36; N, 14.5.

General Synthesis of N,N′-[Polymethylenebis(iminocarbonylmethylene)]bisbenzoxycarboxamides (IX).—To a solution of one equivalent of the appropriate N-carbobenzoxyamino acid[16] and one equivalent of triethylamine in 250 ml. of toluene at −5° was added, with stirring, one equivalent of ethyl chlorocarbonate in 25 ml. of toluene. The resulting jell was held at −5° for 30 min., then one-half equivalent of the appropriate diamine in 25 ml. of toluene was added. Some evolution of carbon dioxide was noted. The mixture, which gradually thickened, was allowed to stand at room temperature overnight. The solvent was then distilled *in vacuo* and the residue was washed successively with water, dilute hydrochloric acid, water, dilute sodium carbonate, and water, followed by recrystallization from the appropriate solvent (see Table II).

General Synthesis of N,N′-{Polymethylenebis[iminocarbonyl(substituted-methylene)]}bis-2-N″-quinoxalinecarboxamides (IV).—A mixture of 0.05 mole of IX, 1 g. of 10% palladium on charcoal and 300 ml. of 95% ethanol containing 20 drops of glacial acetic acid was hydrogenated at 70° and 70 p.s.i. for 4 hr. The catalyst was removed by filtration from the clear reaction mixture and washed with methanol. The solvent from the combined washing and filtrate was distilled under reduced pressure. The thick oily residue was taken up in 50 ml. of 95% ethanol, and to this solution was added 0.2 mole of triethylamine. The resulting solution was added dropwise to a refluxing and stirred solution of 0.1 mole of II in 400 ml. of tetrahydrofuran. Almost immediately a precipitate appeared. The reflux period was extended for 15 min. and the reaction mixture was cooled and filtered. The crude solid product was washed successively with water, dilute hydrochloric acid, water, dilute sodium carbonate solution, water, and acetone, and purified by recrystallization (see Table III). These compounds are usually waxy in nature and hence extremely difficult to purify, which accounts for the relatively low over-all yields from IX.

Acknowledgment.—The authors wish to express their appreciation to Professor Roland K. Robins of Arizona State University for his suggestions and encouragement and to Mr. Hal P. Van Fossen and Mrs. Phyllis G. Lewis for their valuable assistance in performing the analytical measurements.

(16) (a) M. Bergmann and L. Zervas, *Ber.*, **65**, 1192 (1932); (b) S. W. Fox, M. Fling, H. Wax, and C. W. Pettinga, *J. Am. Chem. Soc.*, **72**, 1862 (1950).

Reactions of the Perfluoroalkylnitriles. IV. Preparation and Characterization of Some N′-(Perfluoroacylimidoyl)perfluoroalkylamidines and Their Metal Chelates[1]

HENRY C. BROWN AND PAUL D. SCHUMAN

Department of Chemistry and Department of Chemical Engineering, University of Florida, Gainesville, Florida

Received October 15, 1962

Thermal condensation of perfluoroalkylamidines with the elimination of ammonia to yield 2,4,6-tris(perfluoroalkyl)-1,3,5-triazines was shown to proceed through an initial formation of the stable N′-(perfluoroacylimidoyl)perfluoroalkylimidines, $R_FC(:NH)N{=}C(NH_2)R_F$. Although originally isolated as their metal chelates, these compounds were easily synthesized from the reaction of perfluoroalkylamidines, $R_FC(:NH)NH_2$, with perfluoroalkylnitriles, and were prepared by this method for a detailed study of their properties. The N′-(perfluoroacylimidoyl)perfluoroalkylamidines appeared to exist predominantly in the "enolic" form; infrared and electronic spectra of these compounds and their metal chelates are discussed.

Thermal condensation of the perfluoroalkylamidines has been shown[2] to produce 2,4,6-tris(perfluoroalkyl)-1,3,5-triazines by the elimination of ammonia. This type of reaction was used subsequently to prepare a series of polymers containing *sym*-triazine rings connected by perfluoroalkyl chains that were stable at very high temperatures.[3]

Since the condensation reaction obviously involves three molecules of the perfluoroalkylamidine, the mechanism of the reaction is not readily apparent. This paper describes the study of the mechanism of the thermal condensation of the perfluoroalkylamidines and defines the nature of the first intermediate formed.

(1) This research was supported by the U. S. Air Force, Air Research and Development Command under contract AF 33(616)7971 monitored by Non-Metallic Materials Laboratory, Aeronautical Systems Division, Wright-Patterson Air Force Base, Ohio. Presented in part at the 140th National Meeting of the American Chemical Society, Chicago, Illinois, September, 1961. Preceding paper in this series by H. C. Brown and D. Pilipovich, *J. Am. Chem. Soc.*, **82**, 4700 (1960). This paper represents a portion of the dissertation presented by Paul D. Schuman to the Graduate School of the University of Florida in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

(2) W. L. Reilly and H. C. Brown, *J. Org. Chem.*, **22**, 698 (1957).

(3) H. C. Brown, *J. Polymer Sci.*, **44**, 9 (1960).