is a function of the RNA concentration, the reaction approaching completion when 0·5–1 molecule of Mg^{++} is present per molecule of RNA phosphorus. In Table I the results are therefore presented on the basis of a Mg^{++}/RNA-P ratio and show a parallelity between the formaldehyde reaction and the hypochromic effect. From a quantitative point of view it may be interesting to note that in the absence of salts the same number of formaldehyde molecules are bound as there are amino groups present and that this amount is reduced by about 65% in the presence of higher Mg^{++} concentrations. Similar values have been obtained with TMV-RNA in 10^{-2} to 10^{-1} *M* potassium chloride as well as with TMV-RNA prepared by means of metal containing phenol[2].

This strongly suggests that the amino groups of the purine and pyrimidine bases are involved in the formation of this more structured form of RNA. Mg^{++} has been shown to favour the formation of double and triple stranded helices through hydrogen bonds between base-pairing polyribonucleotides[3,4], probably through its effect on the negative changes of the phosphate groups. The amino groups lose their reactivity under these conditions[7]. From an analogy with these models it may be inferred that a base pairing of the purine and pyrimidine bases takes place in RNA also. If this were the case the results would indicate that about 80% of all the theoretically possible hydrogen bonds have been formed, since purines are present in about 18% excess over the pyrimidines in TMV-RNA[8]. Not all the bases, therefore, could become involved in hydrogen bonding. The unreactivity of the bases does not necessarily permit the conclusion, however, that hydrogen bonds have been formed since other ways are also conceivable by which the amino groups might be blocked. ZUBAY[9] has recently postulated that in denatured DNA Mg^{++} might chelate with the adenine and guanine bases. But it seems unlikely that this would be the way by which Mg^{++} changes the configuration of RNA, since similar changes in configuration have also been observed by higher concentrations of monovalent ions and since under these conditions the hypochromic effect is not reversed by versene[10].

Table II

Hypochromic effect in the presence of various formaldehyde and Mg^{++} concentrations (ε 260 mμ)

Formaldehyde concentration	H_2O	$3 \times 10^{-6} MgCl_2$	10^{-5} $MgCl_2$	10^{-4} $MgCl_2$
—	0·940	0·930	0·840	0·820
0·1%	0·940	0·930	0·840	0·820
0·2%	0·950	0·945	0·860	0·840
0·5%	0·980	0·970	0·900	0·875
1·0%	0·990	0·985	0·970	0·950

Yeast RNA[11] was incubated for 48 h at room temperature with the concentrations of formaldehyde and $MgCl_2$ indicated and the absorption at 260 mμ dermined. Glass distilled water was used throughout the experiment

Interestingly in our experiments we have found no changes in the ultraviolet spectrum of ribonucleic acids upon treatment with formaldehyde in contrast to the earlier report by FRAENKEL-CONRAT[12]. The difference can be accounted for by the different concentrations of formaldehyde which were used. The effect of various concentrations of formaldehyde upon the optical properties of nucleic acid is shown in Table II. Formaldehyde in concentrations of 0·1% which earlier experiments[7] have shown to suffice for complete reaction of all amino groups of the nucleic acid do not cause any increase in its ultraviolet absorption. An increase occurs only with higher concentrations and is especially evident in the presence of Mg^{++}.

It seems likely that at low concentrations formaldehyde reacts by forming a monomethylol derivative ($-NH-CH_2OH$) whereas at higher concentrations a dimethylol derivative ($-N=(CH_2OH)_2$) is formed which would be responsible for the spectroscopic changes. The larger increase in the presence of Mg^{++} could be an indication that formaldehyde at higher concentrations causes a reversal of the salt dependent change in configuration of RNA[13]. The absence of any spectroscopic changes at low concentrations indicates that under these conditions formaldehyde does not alter in any way the configuration of the RNA molecule. The stoichiometric relationship between the formaldehyde bound and the amino groups present as well as the evidence that under the conditions of the reaction (0·1%) only the monomethylol derivative is formed lend further support to the conclusion that in a salt free medium all amino groups are free and reactive and the reduction in formaldehyde binding by 65% upon the addition of Mg^{++} is a true expression of the number of amino groups which through hydrogen bonding or by some other means have become unreactive.

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M. STAEHELIN*

Virus Laboratory, University of California, Berkeley, September 3, 1959.

Zusammenfassung

Die Nukleinsäure des Tabakmosaikvirus kann in zwei Formen vorliegen. In salzfreiem Milieu sind sämtliche Aminogruppen frei und reagieren mit Formaldehyd, während in Gegenwart von Mg^{++} nur etwa 35% reaktionsfähig bleiben. Diese Veränderung, die parallel mit dem hypochromen Effekt einhergeht, deutet darauf hin, dass unter dem Einfluss von Mg^{++} die Basen gegenseitige Beziehungen eingehen und dadurch dem Molekül eine geordnete räumliche Konfiguration geben.

[8] K. K. REDDI, Biochem. biophys. Acta *23*, 208 (1957).

[9] G. ZUBAY, Biochem. biophys. Acta *32*, 233 (1959).

[10] H. FRAENKEL-CONRAT, Personal communication.

[11] A. M. CRESTFIELD, K. C. SMITH, and F. W. ALLEN, J. biol. Chem. *216*, 185 (1954).

[12] S. ZAMENHOF, G. GRIBOFF, and N. MARULLO, Biochem. biophys. Acta *13*, 459 (1954).

[13] This might explain why formaldehyde, although at 0·1% it does not react with DNA, leads to inactivation of the transforming principle of bacteria at 3%[11].

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Alkaloids of Apocynaceae IV[1]. Dregamine, a New Alkaloid from *Voacanga dregei* E. M.

In continuation of our studies of alkaloids from different species of the family *Apocynaceae*, we have examined a

[1] Alkaloids of Apocynaceae III, M. GORMAN, N. NEUSS, J. A. DEYRUP, and N. J. CONE, J. Amer. chem. Soc. (1959), in press.

new species of the genus *Voacanga*, *Voacanga dregei* E.M. We wish to report the isolation of a new alkaloid, dregamine, from the bark of this tree[2].

Dregamine is a representative of 2-acyl indole alkaloids[3]; it was isolated[4] by chromatography of alkaloids obtained by benzene extraction of the bark of the trunk put at our disposal through the cooperation of Mr. J. L. SIDEY (Pietermaritzburg, Natal, S. Africa), who also established the botanical authenticity of the plant material with herbarium specimen (fruit and leaves). Elution with benzene-chloroform mixture (3:1) gave the crude base which crystallized from methanol in long prisms; m.p. 106–109°C, resolidified, then m.p. 186–205°C (dec.); $[\alpha]_D^{26} = -93{\cdot}1°C$ ($CHCl_3$, C = 1).

Calculated for $C_{21}H_{28}O_3N_2$: C 70·76; H 7·92; N 7·86. Found: C 70·57; H 7·47; N 7·42. The hydrochloride was prepared in the conventional manner and recrystallized from methanol-ether, m. p. 249–250°C (dec.). Calculated for $C_{21}H_{28}O_3N_2 \cdot HCl$: C 64·19; H 7·74; N 7·13; Cl 9·02; OCH_3 (1) 7·90; (N)-CH_3 (1) 3·83. Found: C 64·49; H 7·62; N 7·00; Cl 8·98; OCH_3 8·12; (N)-CH_3 3·84.

The ultraviolet spectrum of dregamine is characterized by the following bands: λ_{max}^{EtOH} 239 mμ, $a_M = 15{,}200$; 316 mμ, $a_M = 18{,}600$ and very similar to that of 1-keto-1,2,3,4-tetrahydrocarbazole[3]. In addition to the 2-acyl indole moiety (major band at 6·05 μ)[1], the infrared spectrum indicated the presence of a carbomethoxyl moiety with absorption at 5·78 μ and 8·03 μ[5].

Other botanically related genera are currently under investigation in these laboratories.

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N. NEUSS and NANCY J. CONE

Lilly Research Laboratories, Indianapolis (Indiana), July 6, 1959.

Zusammenfassung

Dregamine, ein neues Alkaloid aus der Apocynaceae *Voacanga dregei* E. M. wurde isoliert und charakterisiert. Diese Verbindung stellt einen neuen Vertreter der Klasse der 2-Acyl-indole dar.

[2] After the completion of this work, SCHULER, VERBEEK, and WARREN, J. chem. Soc. *1958*, 4776, have reported the isolation of the known alkaloids vobtusine and voacangine from the bark of *Voacanga dregei* collected in the South Coast, Natal, S. Africa. We were unable to find these alkaloids in our plant material.

[3] M. GORMAN, N. NEUSS, and N. J. CONE, Amer. chem. Soc. nat. Meeting, San Francisco, Calif., April 1958. The spectral data reported for voacafrine and voacafricine [K. V. RAO, J. org. Chem. *23*, 1455 (1958)] are also indicative of the presence of a 2-acyl indole moiety in these two alkaloids from *Voacanga africana*.

[4] Small amounts of this alkaloid were also isolated from *Ervatamia coronaria*[3].

[5] Subsequent to the preparation of this manuscript, RENNER has described the isolation of vobasine and voacryptine from *V. africana*. The former alkaloid appears to be another representative of 2-acyl indoles[3] [U. RENNER, Exper. *15*, 185 (1959)].

A Simple Synthesis of Nicotinic Aldehyde

Practically all known routes leading to nicotinic aldehyde involve the reduction of various nicotinic acid derivatives. We have now found a simple and advantageous method for the synthesis of this valuable compound based on crotonaldehyde.

Recently we have shown[1,2] that the Vilsmeier-Haack reaction may be extended to the aliphatic series, in particular to carbonyl compounds. By this reaction, we have prepared a large number of β-dicarbonyl derivatives, such as the β-dialdehydes, β-chlorovinylaldehydes and some novel types of polyformyl derivatives. Most of these compounds had previously been difficult of access.

$CH_2=CH-CH=CH-N(CH_3)_2$ (I)

$CH_3-CH(N(CH_2)_2)-CH=CH-N(CH_3)_2$ (II)

$(CH_3)_2N-CH=C(CH=O)-CH=CH-CH=O$ (III)

$[(CH_3)_2N-CH=CH-CH=CH-CH-N(CH_3)_2]\ X$ (IV)

We have now applied the reagent prepared from dimethylformamide and phosgene to compounds I and II, which are readily available from crotonaldehyde in a single step[3,4]. This reaction led to the trialdehyde derivative III in about 60% yield. This derivative is even more simply obtained by formylation of the well known quaternary salt IV which is also believed to be an intermediate in the formylation of I and II. The trialdehyde derivative III very readily passes into nicotinic aldehyde in excellent yield, e.g. merely on heating with aqueous NH_4Cl.

Nicotinic aldehyde is thus available from crotonaldehyde in a simple three-step process in about 40% overall yield.

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Department of Organic Synthesis, Institute of Chemistry, Czechoslovak Academy of Science, Prague, July 17, 1959.

Zusammenfassung

Nikotinaldehyd wurde durch ein dreistufiges Verfahren aus Krotonaldehyd in einer Gesamtausbeute von 40% erhalten.

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[3] C. MANNICH, K. HANDKE, and K. ROTH, Chem. Ber. *69*, 2112 (1936).

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Demonstration of Poliomyelitis Virus in Homogenates and Ureadesoxycholate Lysates of Cells Exhibiting or Lacking Cytopathogenicity

A method for dissolution of tissue cultures (TC) with Ureadesoxycholate (UDC) was described previously[1]. Ap-

[1] E. KOVÁCS, Naturwissenschaften *45*, 339 (1958); Arch. Biochem. Biophys. *76*, 546 (1958).