129. *Some Derivatives from Colchicine*¹⁾²⁾

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On the structure of colchicine, Dewar³⁾ proposed that its C-ring is corresponding to the methyl ether of tropolone, and as is well known, colchicine possesses some specific biological actions, such as the interference of cell mitosis, that deep interest is being shown on this compound.

Many workers have reported on various kinds of N-substituted colchicine derivatives in connection with chemotherapy of cancer and other diseases. The present writers utilized various reactions carried out with tropolones on colchicine (I), which seemed of interest from biological standpoint, and these attempts are briefly described in the present paper.

Some time ago, various kinds of mercaptotropolones and mercaptotropolones⁴⁾ were obtained in this laboratory by the reaction of halotropolones and their methyl ethers with thiols. This reaction was first taken up, and an attempt was made to obtain thiocolchicine (II) by the reaction of colchicine (I)⁵⁾ and sodium mercaptide. However, the objective (II) was not obtained and only the rearrangement products, allocolchicine and allocolchicine⁶⁾⁷⁾ were obtained in 10% and 80% yield. When (I) and equimolar amount of sodium hydrosulfide were reacted under the same conditions, besides the recovery of 20% of (I), levorotatory acidic substance was obtained as an amorphous power of m.p. 175–190° (decomp.).⁸⁾ This substance was very hard to crystallize and was therefore purified by repeated reprecipitation. Since its ultraviolet absorption spectrum, shown in Fig. 1, possesses an absorption in a longer wave length range, i.e. 390 m μ , the substance was assumed to still retain the seven-membered C-ring corresponding to tropolone ring. With the analytical values obtained, the substance was assumed to be thiocolchicine (III). Yield, 60% (Anal. Calcd. for C₂₁H₂₃O₅NS·H₂O: C, 60.13; H, 6.01; N, 3.34. Found: C, 60.75; H, 5.91; N, 4.48).⁹⁾

Treatment of (III) with diazomethane gave a substance assumed to be (II) but this substance also failed to crystallize. Purification was effected by repeated chromatography through alumina to a neutral substance as yellow powder of m.p. ca. 130°. Yield, 60% (Anal. Calcd. for C₂₂H₂₅O₅NS·H₂O: C, 60.96; H, 6.28; N, 3.23. Found: C, 61.25; H, 6.51; N, 4.61). Treatment of (III) with diethyl

sulfate and chromatography through alumina gave an ethyl ether (IV) as yellow powder, m.p. *ca.* 120°. Yield, 50% (Anal. Calcd. for $C_{23}H_{27}O_5NS$: C, 64.32; H, 6.34; N, 3.26. Found: C, 63.80; H, 6.90; N, 4.12).

Fig. 1. Ultraviolet absorption spectra in methanol

- (a) Colchine (I) (.....)
- (b) Thiocolchicine (III) (———)
- (c) Thiocolchicine (II) (-----)
- (d) Hydrazone (VI) (- · - · -)

Application of hydrazine hydrate to the methyl ether of tropolones is known to give easily crystalline hydrazino compounds.^{10,11,12)} This reaction was applied on colchicine (I). A mixture of (I) and 80% hydrazine hydrate in dil. methanol was allowed to stand overnight from which a resinous substance was obtained. This resinous substance was dissolved in methanol, acetone added, and the mixture was refluxed for 5 minutes from which yellow needles, m.p. 223–224°, $[\alpha]_D^{15} - 21.7^\circ$,¹³⁾ were obtained. The analytical values of these crystals corresponded to a kind of hydrazone, an acetone condensate shown by (VI). Yield, 60% (Anal. Calcd. for $C_{24}H_{29}O_5N_3 \cdot H_2O$: C, 63.00; H, 6.83; N, 9.13. Found: C, 63.12; H, 7.02; N, 9.35).

From these results, the foregoing resinous substance was assumed to be an expected hydrazino compound shown by (V). (V) also undergoes condensation with methyl ethyl ketone, under the same conditions as in the case of (VI), to give a hydrazone (VII), m.p. 197.5–199°, $[\alpha]_D^{13.5} - 19.3^\circ$ (Anal. Calcd. for $C_{25}H_{31}O_5N_3 \cdot H_2O$: C, 63.67; H, 7.05; N, 8.91. Found: C, 63.73; H, 6.95; N, 9.48).

It has recently been found in this laboratory that the condensation of guanidine or thiourea with tropolone methyl ether respectively yielded 2-amino-¹⁴⁾ and 2-mercapto-1,3-diazazulene.¹⁵⁾ This condensation was found to proceed comparatively smoothly with

colchicine (I) when the guanidine solution, obtained by the neutralization of guanidine hydrochloride with equimolar amount of ethanolic sodium hydroxide, and (I) in absolute ethanol were refluxed for 3.5 hours. The vitreous product was purified by its derivation to the picrate, liberation, and by chromatography through alumina from which yellow microprisms, m.p. 213–215° (decomp.), $[\alpha]_D^{18} -33.9^\circ$, were obtained. Fig. 2 shows the ultraviolet absorption spectra of this substance and of 2-amino-1,3-diazazulene for comparison and it can be seen that there is some similarity between the two.¹⁴⁾ From the similarity of the spectra and analytical values, the substance obtained by this condensation was assumed to be 1,3-diazazulene derivative (VIII), yield, 75% (Anal. Calcd. for C₂₂H₂₄O₄N₄·2H₂O: C, 59.45; H, 6.35; N, 12.61. Found: C, 59.13; H, 6.03; N, 12.80). Chloroplatinate: Orange microprisms, m.p. over 300°. Picrate: Yellow needles, m.p. 251–252° (decomp.) (Anal. Calcd. for C₂₈H₂₇O₁₁N₇: C, 52.75; H, 4.27; N, 15.38. Found: C, 52.43; H, 4.41; N, 15.82). During the formation of (VIII), a small amount of reddish orange powder of m.p. ca. 180° are obtained as a by-product and detailed examinations are being made on this substance.

The use of sodium methoxide in place of sodium hydroxide in the foregoing condensation resulted in the rearrangement of the majority and the yield of (VIII) decreased markedly.

Acylation of (VIII), such as acetylation and benzylation, and methylation by diazomethane was found to be very difficult, (VIII) being recovered. Such facts suggest that the amino group in the D-ring, five-membered heteroring newly formed by condensation, does not possess a primary amine characteristics and has a tendency to react in an imino-form as shown in (VIII-B). Such an example has also been witnessed in the case of 1,3-diazazulene.¹⁴⁾ When a mixture of (VIII) and 6N HCl was warmed on a water bath for 1 hour, desacetylated compound (IX) was obtained as pale yellow crystals of m.p. over 300°, in almost quantitative yield (Anal. Calcd.

for $C_{20}H_{22}O_3N_4$: C, 65.55; H, 6.05; N, 15.29. Found: C, 65.26; H, 6.00; N, 15.31). Picrate: Yellowish orange microprisms, m.p. 171–173° (decomp.) (Anal. Calcd. for $C_{26}H_{25}O_{10}N_7$: N, 16.45. Found: N, 16.20). This desacetylated compound (IX) can also be obtained by the reaction of guanidine and a mixture of the two isomeric methyl ethers of trimethylcolchicine acid.

When a mixture of (IX) and acetic anhydride was evaporated to dryness on a water bath, pale yellow crystals of m.p. 210–215° (decomp.) were obtained whose nitrogen determination and ultra-violet absorption spectrum were identical with those of (VIII).

Fig. 2. Ultraviolet absorption spectra in methanol

- (a) 2-Amino-1,3-diazazulene (.....)
- (b) Condensation product of colchicine with guanidine (VIII) (—)
- (c) 2-Mercapto-1,3-diazazulene (-----)
- (d) Condensation product of colchicine with thiourea (X) (— · — · —)

A mixture of an absolute ethanolic solution of colchicine (I) and absolute ethanolic solution of thiourea containing 0.3 moles equivalent of sodium ethoxide was refluxed on a water bath for 3 hours from which 30% of allocolchicine ethyl ester^{6,7)} and reddish orange microprisms, m.p. 215–217° (decomp.), were obtained. The latter was assumed to be a substance (X) corresponding to the derivative of 2-mercapto-1,3-diazazulene¹⁵⁾ from its analytical values and ultraviolet absorption spectrum. Yield, 60% (Anal. Calcd. for $C_{22}H_{23}O_4N_3S \cdot 2H_2O$: C, 57.25; H, 5.90; N, 9.09. Found: C, 56.97; H, 5.88; N, 8.73). Chloroplatinate: Reddish orange, microprisms, m.p. over 300°. (X) does not give any picrate.

In this condensation, use of sodium ethoxide as a catalyst is a disadvantage since it is likely to induce rearrangement reaction, as was the case in the reaction of colchicine (I) and guanidine. In spite of such a fact, sodium ethoxide was used because the

reaction failed to proceed when sodium hydroxide was used or when a catalyst was not used in the formation of 2-mercapto-1,3,-diazazulene and the condensation occurred only when sodium alkoxide was used.¹⁵⁾

The foregoing reactions on the derivatives of colchicine have indicated the interesting fact that the C-ring of colchicine behaves in a quite similar manner to its parent structure, tropolone methyl ether.

Biological tests of the various derivatives described herein are being carried out by our collaborators,¹⁶⁾ and the results will be published in later reports.

References

- 1) Gist of the present studies was read as a paper at the 7th Annual Meeting of the Chemical Society of Japan in Tokyo, April 1, 1954. Detailed report will be published in the forthcoming Science Reports of the Tōhoku University.
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