

ported that antibiotic treatment of mastitis by intramammary infusion resulted in milk containing sufficient antibiotic to result in the production of inferior cheese as well as other dairy products.

The addition of a 4% level of a vitamin B₁₂ supplement (3.3 mg vitamin B₁₂ daily) to the ration of lactating cows also did not result in any increase in the vitamin B₁₂ level of the milk when assayed microbiologically (10).

Further details of this work will be published elsewhere.

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Desmethylecolchicine, a Constituent of USP Colchicine¹

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In purifying a batch of USP colchicine for biological studies we have found that the material contains an appreciable amount (ca 4%) of a second alkaloid, as well as minor amounts of other constituents. In view of this we believe that biological investigators using USP colchicine would be well advised to repurify the material.

Colchicine is generally purified by adsorbing the impure alkaloid on alumina and eluting the pure colchicine with chloroform (1). The column of alumina thus eluted often retains an intense yellow color. If methanol chloroform (1:99) is then used as the eluting agent a compound may be recovered (up to 4% based on the weight of impure colchicine) having the properties of desmethylecolchicine (one of the methoxyl groups in ring A demethylated). The evidence for this consists in the facts that the compound C₂₁H₂₃O₆N, (a) contains three methoxyl groups, (b) yields colchicine on treatment with diazomethane, and (c) gives a color with ferric chloride only after it has been heated in dilute HCl (presence of an enol ether).

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TABLE 1

	Desmethylecolchicine	Substance C (2)
Melting point	176°–190°; 275.5°–277°	176°–182°
[α] _D ²⁰ (CHCl ₃)	–143.6°* (C=2) –152.0°* (C=1)	–130.7°† (C=2.0118)
λ (log ε)		
In alcohol	243.5 μ (4.47) 353 μ (4.21)	245 μ (4.54) 352.5 μ (4.27)
In 0.05 N alkali	249 μ 376.5 μ	250 μ 379 μ
Acetyl derivative mp	225°–226.5°	231°–233° (2) 225° (3)
[α] _D ²⁰ (CHCl ₃)	–90.7° (C=0.7)	–115° (C=0.671)

* Dried at 155°.

† Dried at 50°.

Desmethylecolchicine is obtained as yellow prisms from ethyl acetate-ether containing a trace of chloroform. It turns to a glass at ca 176°–190°, recrystallizes at ca 200°–210°, and melts finally at 275.5°–277°. No chemical changes are involved in these transitions. The compound retains solvents and had to be dried to constant weight at 155° in *vacuo* before the following analytical data were obtained:

Anal calcd for C₂₁H₂₃O₆N: C, 65.44; H, 6.01; 3CH₃O, 24.15.

Found: C, 65.42; H, 5.81; CH₃O, 24.14.

Santavy and Reichstein (2) recently reported the isolation of a number of new constituents from the seeds of *Colchicum autumnale*, including a desmethylecolchicine designated by them as "Substance C." Substance C gave the same reactions as the desmethylecolchicine described here. A comparison of the physical properties of the compounds and of certain derivatives is shown in Table 1.

Although the data for desmethylecolchicine and Substance C agree only fairly well in some particulars, the agreement of the absorption spectra in neutral and alkaline solution is good. We believe there is sufficient over-all correspondence to assume the compounds identical. It should be noted that a double melting point may have been overlooked in the case of Substance C.

The biological effects of desmethylecolchicine are now being studied.

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