REGRESSION OF LYMPHOSARCOMA TRANSPLANTS IN MICE

Sir:

Chemotherapy with cortisone acetate (CORTONE MERCK) as an adjunct to a riboflavin deficient diet brings about regression of well-established (13 day) transplants of lymphosarcoma 6C3HED in mice of the C_3H strain. Compound A acetate also shows significant activity.

Heilman and Kendall[1] observed an arrest in the growth of lymphosarcoma transplants following therapy with Compound E; however, the neoplasms usually recurred in a few days or weeks following withdrawal of the drug and the animals succumbed despite the resumption of treatment.

Regression of lymphosarcoma transplants occurred in mice subjected to riboflavin deficiency and such animals were refractory to subsequent inoculations with the tumor.[2]

Combination therapy consisting of administering cortisone acetate to mice with a restricted intake of riboflavin might be expected to enhance regression. Accordingly, mice[3] with well-established (13 day post-transplant) lymphosarcomas (10,000 cell subcutaneous inoculation) were transferred from a stock to a riboflavin deficient ration and treatment with 500γ or 1 mg. of cortisone acetate daily was instituted (50 mice were employed on each level). A reduction in the size of the tumors was noted by the second day and tumor tissue was non-palpable after four days of treatment. An early arrest (but not regression) of tumor growth was observed in 10 mice given 250γ of cortisone acetate daily. Measurable sites were still present in controls (a total of 108 mice) maintained on the riboflavin deficient diet but without additional therapy. Regression was also noted in four days when 1 mg. of cortisone acetate was administered daily to mice (10 in each group) maintained on a purified diet supplemented with 4, 6 or 8γ of riboflavin daily. Many animals succumbed from incidental causes such as Tyzzer's disease or Salmonella infection. Death may have been ascribable in some cases to a toxemia produced by the rapid resorption of necrotic tissue. The few surviving animals were refractory to a second transplant of the tumor. Although the growth of transplants in 40 mice receiving a natural food ration and 1 mg. of cortisone acetate daily was definitely suppressed, the effect was only transitory and the animals succumbed from their tumors.

Compound A acetate[4] was somewhat less effective than cortisone acetate as a carcinolytic agent. Two levels were employed, namely, 1 mg. (20 mice) and 4 mg. (10 mice). Dihydrocortisone acetate, pregnenolone, 21-acetoxy-pregnenolene, DCA, progesterone, 11-keto-progesterone, Compound S acetate, adrenosterone, and 3,11,20-triketo-4,21-diacetoxy-17-hydroxy-pregnane were inactive when administered to 10 mice in each group at a level of 1 mg. daily for six days.

MERCK INSTITUTE FOR THERAPEUTIC RESEARCH
RAHWAY, N. J.

GLADYS A. EMERSON
ELIZABETH WURTZ
MARY E. ZANETTI

(1) Heilman and Kendall, *Endocrinology*, **34**, 416 (1944).

(2) Stoerk and Emerson, *Proc. Soc. Exp. Biol. and Med.*, **70**, 703 (1949).

(3) The C_3H mice employed in these tests were 3–5 months of age. The sexes were equally divided in each group.

(4) Dr. George W. Wolley, Head of the Division of Steroid Biology of the Sloan-Kettering Institute, has found that Compound A acetate showed activity when administered at a level of 4 mg. daily in lymphatic leukemia P1534, lymphosarcoma 6C3HED and in normal mice (*Proc. Soc. Exp. Biol. and Med.*, **74**, 286 (1950)).

THE SYNTHESIS OF RACEMIC β-Δ^6-DIHYDRODESOXYCODEINE METHYL ETHER

Sir:

Hydrogenation over copper–chromium oxide of 3,4-dimethoxy-9,10-dioxo-13-cyanomethyl-5,8,-9,10,13,14-hexahydrophenanthrene (I)[1] yields the ketolactam II[2] (m. p. 263–264.5°, found, C, 68.62; H, 5.77; ultraviolet λ_{max} 281 mμ, log ϵ 4.16, *IR* λ_{max} 3.00, 5.88, 5.97 μ) which on Wolff–Kishner reduction and remethylation yields the lactam III (m. p. 210–212.5°, found, C, 72.44; H, 7.09; OCH_3, 20.31; ultraviolet λ_{max} 282, log ϵ 3.17). Reduction of III with lithium aluminum hydride followed by methylation with formaldehyde–formic acid yields the racemic base IV, (oil, found, C, 76.66; H, 8.44) purified through its picrate (m. p. 198.5–200°, found, C, 56.55; H, 5.76).

We have prepared both epimers at C_{14} of Δ^6-dihydrodesoxycodeine (IVa, IVb) for comparison with this material. β-Dihydrothebainone[3] on hydrogenation yields the corresponding alcohol (m. p. 165.5–166°, $\alpha^{30}D$ −23° (*c* 0.920, alc.) found, C, 71.34; H, 8.36; methiodide m. p. 264–265°, found, C, 51.15; H, 6.51) which on methylation yields the methyl ether (m. p. 152.5–153.5°, $\alpha^{30}D$ −9° (*c* 0.643, alc.) found, C, 71.93; H, 8.77; OCH_3, 19.44; methiodide m. p. 243–245°, found, C, 52.30; H, 6.57; picrate, m. p. 190–191°, found, C, 55.27; H, 5.85). On conversion to the tosylate and detosylation with boiling collidine[4] this ether affords β-Δ^6-dihydrodesoxycodeine methyl ether[5] (IVa) (oil, found, C, 76.23; H, 8.27) purified through its picrate, m. p. 210–212° (found, C, 56.51; H, 5.72).

Dihydrothebainol[6] by a similar series of transformations (methyl ether, oil, $\alpha^{27}D$ −28° (*c* 1.519, alc.) found, C, 71.42; H, 8.73; OCH_3, 18.88; methiodide m. p. 279–281°, found, C, 52.35; H, 6.48; hydrobromide (through which the base was purified) m. p. 254.5–255°, $\alpha^{28}D$

(1) M. Gates, THIS JOURNAL, **72**, 228 (1950).

(2) Compare *ibid.*, **72**, 1141 (1950).

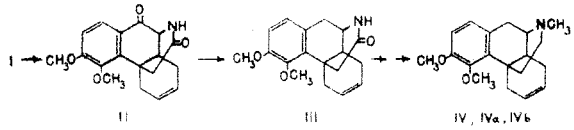
(3) L. F. Small and G. L. Browning, Jr., *J. Org. Chem.*, **3**, 618 (1939).

(4) J. von Euw and T. Reichstein, *Helv. Chim. Acta*, **29**, 654 (1946).

(5) The prefix β refers to a configuration at C_{14} epimeric with that of morphine; *cf.* Small and Browning, ref. 3.

(6) A. Skita, F. F. Nord, J. Reichert and P. Stukart, *Ber.*, **54**, 1562 (1921).

+ 34°, (*c* 0.447, alc.) found, C, 57.21; H, 7.29; tosylate, oil, methiodide, m. p. 165–166°, found, C, 52.45; H, 6.04) yields Δ^6-dihydrodesoxycodeine methyl ether (IVb) (oil, found, C, 75.83; H, 8.44) purified through its characteristic fumarate (2 moles of base to 3 of acid) m. p. 233–235°, found, C, 63.41; H, 6.58).



II III IV, IVa, IVb

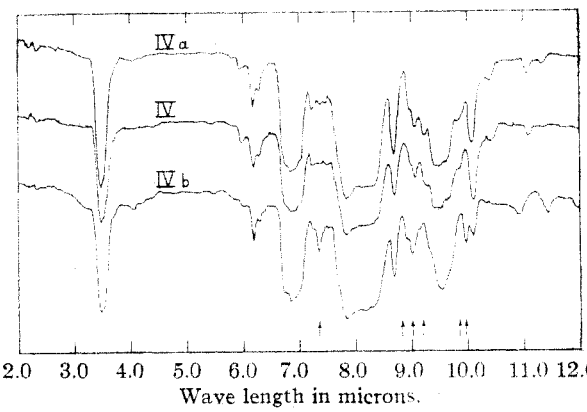


Fig. 1.—Infrared absorption spectra of: IVa, β-Δ^6-dihydrodesoxycodeine methyl ether; IV, synthetic *d,l*-β-Δ^6-dihydrodesoxycodeine methyl ether; IVb, Δ^6-dihydrodesoxycodeine methyl ether; approximately 1.7% in chloroform.

The infrared absorption spectra of IV and IVa (Fig. 1) are virtually superimposable and demonstrate conclusively that IV is the racemic modification of IVa. The spectrum of IVb is similar but unmistakeable differences are shown at the points indicated.

This synthesis provides an unequivocal demonstration that the point of attachment of the ethanamine side chain in the morphine alkaloids is at C_{13}.

DEPARTMENT OF CHEMISTRY
UNIVERSITY OF ROCHESTER MARSHALL GATES
ROCHESTER 3, N. Y. GILG TSCHUDI

RECEIVED AUGUST 23, 1950

DIFFUSION COEFFICIENT OF SUCROSE IN SUPERSATURATED SOLUTION

Sir:

In an important paper, English and Dole[1] have recently reported measurements by the Gouy method of the diffusion coefficient of sucrose in highly concentrated solutions. This communication is in no sense a criticism of English and Dole's beautiful measurements; it is intended merely to remove an apparent misconception as to the basis and applicability of the so-called Gordon equation, their Eq. 1

$$D = D^0(1 + d \ln f/d \ln c)(\eta^0/\eta)$$

This relation, as far as the relative viscosity factor is concerned, is entirely empirical, and was originally intended[2] merely as a possibly convenient device for interpolation and extrapolation; it would therefore seem inadvisable to use it as a criterion for normal diffusion. That it cannot be valid over wide ranges of concentration can be readily demonstrated.

Consider a system of two non-electrolytes, miscible in all proportions. The equation may be written for component No. 1 (see Eq. 4a of ref. 2)

$$D_1 = (D_1^0/RT)(n_1\partial\mu_1/\partial n_1)(V/n_2\bar{V}_2)(\eta_2^0/\eta) \quad (A)$$

Here D_1^0 is the limiting diffusion coefficient of 1 in pure 2, μ_1 is the thermodynamic potential of 1 in the solution, η_2^0 is the viscosity of pure 2, and $\bar{V}_2$ is the partial volume of 2 in the solution. A similar expression may be written down for the diffusion coefficient D_2 of component 2 in the solution merely by interchanging 2 and 1 in Eq. A. Now in the Gouy method, the flow is measured relative to a fixed frame of reference, so that D_1 and D_2 must be identical. A comparison of the two expressions shows that this can only be true if $D_1^0\eta_2^0/\bar{V}_2 = D_2^0\eta_1^0/\bar{V}_1$—a condition that obviously cannot generally hold.

(2) Gordon, *J. Chem. Phys.*, **5**, 523 (1937).

CHEMISTRY DEPARTMENT
UNIVERSITY OF TORONTO
TORONTO, CANADA A. R. GORDON

RECEIVED AUGUST 17, 1950

COLCHICINE. STRUCTURE OF WINDAUS' ANHYDRIDE[1,2]

Sir:

It has been shown by Windaus[3] that the oxidation of N-benzoyltrimethylcolchicinic acid with alkaline permanganate affords a compound, $C_{23}H_{21}O_7N$, designated as N-benzoylcolchinic anhydride (I). The Windaus structure for (I) was that of a dihydronaphthalene derivative; however, recent work[4] indicates that deaminocolchinic anhydride (II), obtained by the deamination of I is not a substituted naphthalene.

We have carried out the Cook degradation of N-benzoylcolchinic anhydride with phosphorus pentoxide in boiling xylene to obtain deaminocolchinic anhydride (m. p. 172.5–173°; reported[4] m. p. 170–171°). (*Anal.* Calcd. for $C_{16}H_{14}O_6$: C, 63.57; H, 4.67. Found: C, 63.86; H, 4.51.) This, on hydrogenation over a palladium catalyst, gave dihydrodeaminocolchinic anhydride (m. p. 119.5–120°) which was identical with synthesized 2,3,4-trimethoxybenzsuber-5-ene-5,6-dicarboxylic

(1) The work carried out at the University of Pennsylvania was aided by a Grant-in-Aid from the American Cancer Society recommended by the Committee on Growth of the National Research Council.

(2) This investigation was supported (in part) by a research grant from the National Cancer Institute, of the National Institutes of Health, Public Health Service.

(3) Windaus, *Ann.*, **439**, 59 (1924).

(4) Cook, Johnston and Loudon, *J. Chem. Soc.*, 537 (1950).