to the limit of the solvent migration. Consequently, an elution chromatogram of a portion of Solution I was made by diluting with acetone and running on a silica gel column (0.9 × 30 cm.) using acetone–water mixtures. The radioactivity of the fractions was measured by counting evaporated drops of the eluate on aluminum discs with a G.M. counter. One fraction (Solution IX) behaving in the column in a manner similar to authentic B_{12}, was then subjected to further study. This solution exhibited absorption maxima at 2780 and 3610 Å. No radioactivity could be detected in the organic phase when IX was extracted with dithizone in chloroform. A paper chromatogram of IX revealed a concentrated spot with little tailing, coincident for both the radio- and bioactivity. The R_f of this spot (0.79–0.80) was in agreement with a B_{12} standard run simultaneously.

The radioactivity of the original and purified samples was shown to be Co^{60} (5.3 y) by measurement of the gamma ray spectrum in a scintillation spectrometer. On a G.M. counter (geometry ~9%), Solution I had 689 ± 16 cpm./γ, solution IX 567 ± 30 cpm./γ, giving a retention value of ~80%. By employing serial dilutions of I and IX against standard B_{12}, it was determined that the *bio*-activity in Solution I was 80 ± 20% and in Solution IX was 100 ± 15% of the standard by weight.

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ENZYMATIC DISINTEGRATION OF WHEAT GERM DESOXYPENTOSE NUCLEIC ACID

Sir:

Previous work in this Laboratory[1] has shown that the degradation of calf thymus desoxypentose nucleic acid (DNA) by crystalline pancreatic desoxyribonuclease proceeded according to a complex pattern, resulting in the formation of dialyzable fragments and of a non-diffusible core which was characterized by greatly increased ratios of adenine to guanine, thymine to cytosine, and purines to pyrimidines.

A study of the generality of this phenomenon appeared important, since it offers an additional tool for the differentiation between DNA preparations of different origin and for the understanding of the relationship between nucleotide sequence and enzyme action. Wheat germ DNA, highly polymerized and entirely free of pentose nucleic acid, was employed. This DNA contains an appreciable quantity of a third pyrimidine, 5-methylcytosine,[2] and provides thereby one more marker, useful for the study of the enzymatic attack. The results summarized in Table I indicate the trend of degradation; "19% core" and "8% core" refer to the dialysis residues recovered after 81 and 92% of the DNA, respectively, had been converted to dialyzable products.

(1) S. Zamenhof and E. Chargaff, *J. Biol. Chem.*, **178**, 531 (1949); **187**, 1 (1950).
(2) G. R. Wyatt, *Biochem. J.*, **48**, 584 (1951).

TABLE I
Wheat Germ DNA; Intact Preparation and Enzymatically Produced Cores (as moles per 100 moles P)

Constituent	Intact DNA	19% Core	8% Core
Adenine	26.3	33.2	35.4
Guanine	21.8	20.0	19.8
Cytosine	16.2	11.8	10.3
5-Methylcytosine	5.8	4.3	3.6
Thymine	26.1	26.2	23.4
Total	96.2	95.5	92.5
Purine to pyrimidine ratio	1.00	1.26	1.48

TABLE II
Liberation of Adenine (as moles per 100 moles P)

Agent	Intact DNA	19% Core	8% Core
1 N H_2SO_4, 100°, 1 hr.	26.5	31.2	31.6
98% HCOOH, 175°, 2 hr.	26.3	33.3	35.4
7.5 N $HClO_4$, 100°, 1 hr.		33.2	

Several points appear of interest. The ratio of cytosine to 5-methylcytosine remained constant in all stages, *viz.*, 2.8. In the intact DNA, the sum of these two pyrimidines equalled the molar concentration of guanine, a relationship observed with respect to the ratio of guanine to cytosine in almost all DNA specimens studied.[3] As judged from the extent of its liberation by various hydrolyzing agents, adenine seems to occur in two types of linkage, one of which is enriched in the cores (Table II). The procedures employed for the isolation and purification of the DNA will soon be discussed in detail. The enzyme used was supplied by the Worthington Biochemical Laboratory, Freehold, N. J. The analytical methods have been described before.[4]

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(3) E. Chargaff, *Experientia*, **6**, 201 (1950); *J. Cell. Comp. Physiol.*, in press; *Federation Proc.*, **10**, in press.
(4) E. Vischer and E. Chargaff, *J. Biol. Chem.*, **176**, 703, 715 (1948); E. Chargaff, E. Vischer, R. Doniger, C. Green and F. Misani, *ibid.*, **177**, 405 (1949); E. Chargaff, R. Lipshitz, C. Green and M. E. Hodes, *ibid.*, **192**, in press.

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ON THE SYNTHESIS OF CORTISONE ACETATE

Sir:

We wish to report the synthesis of cortisone acetate from *allo*pregnane-3β-ol-11,20-dione acetate, I, made available recently from $\Delta^{5,6}$ steroids, devoid of functional groups in ring C.[1,2]

Hydroxylation of the *allo*pregnane I at the 17-position by conversion into its enol acetate and treatment with perbenzoic acid followed by caustic saponification[3] yielded *allo*pregnane-3β,17α-diol-

(1) Chamberlain, Ruyle, A. E. Erickson, Chemerda, Aliminosa, R. L. Erickson, Sita and Tishler, THIS JOURNAL, **73**, 2396 (1951).
(2) Stork, Romo, Rosenkranz and Djerassi, *ibid.*, **73**, 3546 (1951).
(3) Kritchevsky and Gallagher, *J. Biol. Chem.*, **179**, 507 (1949); Marshall, Kritchevsky, Lieberman and Gallagher, THIS JOURNAL, **70**, 1837 (1948).