

mina-lime-celite and yielded, finally, four pigments which, on the basis of their adsorption affinities, could have belonged to the poly-*cis* lycopene series. However, none of them gave the very sensitive optical test described earlier, *viz.*, the conspicuous migration of the spectral bands, upon iodine catalysis, from 470 and 443.5 $m\mu$ to 502, 471.5 and 443 $m\mu$, for example, as observed in hexane solutions of polycopene in the Loewe-Schumm Grating Spectroscope.

Since this test became strongly positive upon the addition of 10 μg of polycopene to any of the four pigment solutions, no inhibitor of the steric rearrangement was present. Further controls showed that quantities of polycopene as small as 10 μg , when washed through a 50×24 cm. column with a few liters of benzene, could be easily recovered to the extent of 60% from the first liter of the chromatographic filtrate.

We conclude that, although about 40% of a lycopene sample is converted into a mixture of *cis* isomers by refluxing, the order of magnitude of poly-*cis* forms in such equilibria must be 1 part in 3 million (or more) parts of this total pigment.

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DI-DESOXYRIBONUCLEOTIDES¹

Sir:

Among the products of the action of pancreatic desoxyribonuclease upon highly polymerized calf thymus desoxyribonucleic acid (prepared according to the method of Mirsky and Pollister²) are a set of dinucleotides.³ It has been found possible to separate several of these into apparently homogeneous fractions by ion exchange chromatography.⁴ Figure 1 is a portion of an ion exchange elution diagram, indicating the resolution of the first three fractions, which came off the column after the mononucleotides.⁵

The ultraviolet absorption spectra of these fractions indicate that P1 contains only desoxycytidylic acid, and P3 contains desoxycytidylic and desoxyadenylic acids, in equimolar proportion. (P2 appears to be a mixture of nucleotides and is under further investigation.) These spectroscopic analyses of P1 and P3 have been completely confirmed by quantitative degradation to mono-

(1) Aided by a grant from the Rockefeller Foundation.

(2) A. E. Mirsky and A. W. Pollister, *J. Gen. Physiol.*, **30**, 117 (1946).

(3) Previous titrimetric work (J. A. Little and G. C. Butler, *J. Biol. Chem.*, **188**, 695 (1951) and reference therein) had demonstrated that the products of this degradation, are, on the average, tetranucleotides. Since the products described here amount to only some 7% of the total digest, our results are not considered to be in conflict with the earlier observations, but emphasize their average character.

(4) W. E. Cohn, *THIS JOURNAL*, **72**, 2811 (1950).

(5) R. L. Sinsheimer and J. F. Koerner, *Science*, **114**, 42 (1951).

(6) A column of Dowex-1 resin, 250-500 mesh, 18 cm. \times π sq. cm., initially in the acetate form, was used. The eluting medium was 0.05 *M* acetate buffer pH 4.3 plus an increasing concentration of chloride anion. Over the portion of the elution diagram shown, the Cl⁻ concentration varied from 0.02 to 0.05 *M*.

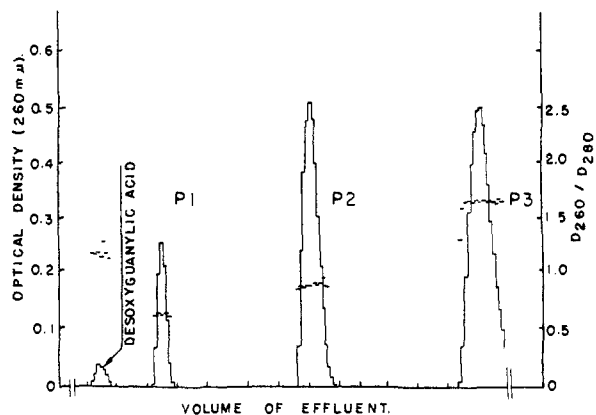


Fig. 1.—Elution diagram of first three dinucleotides.⁶

nucleotides by means of a purified phosphodiesterase preparation from rattlesnake venom,⁷ followed by quantitative determination of the mononucleotides by ion exchange separation and ultraviolet absorption measurement.

That these two fractions are dinucleotides is adduced from the fact that, in each case, exactly half of the total phosphorus of the fraction is monoesterified, being quantitatively removable by an alkaline phosphatase of bone, which previously had been freed of phosphodiesterase activity.⁸ Following the action of this enzyme, a product can be recovered by ion exchange fractionation (P3' of Fig. 2), which has an ultraviolet absorption similar to that of the dinucleotide, one-half the original phosphorus content, and which migrates considerably more rapidly in the ion exchange chromatogram, as illustrated in Fig. 2.

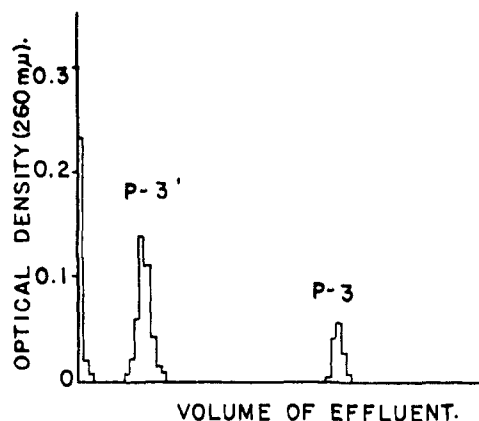


Fig. 2.—Elution diagram after incomplete degradation of P-3 by alkaline phosphatase.

In the case of P₃, the possibility of isomerism exists, and it has not been determined whether the fraction is truly homogeneous or is a mixture of the two isomers. For P1 (di-desoxycytidylic acid), this question does not arise.

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(7) R. L. Sinsheimer and J. F. Koerner, to be published.

(8) J. M. Gulland and E. M. Jackson, *Biochem. J.*, **32**, 590 (1938).