form inosinic acid in these extracts,[4] inhibits Compound I formation by about 50%. These results suggest that Compound I is nicotinic acid mononucleotide and is formed by condensation of PRPP with NA.

To characterize these compounds, a large-scale incubation of erythrocyte acetone powder extracts with nicotinic acid-7-C^{14} was performed. A neutralized perchloric acid filtrate was then passed through a Dowex 1 formate column; 15-ml. aliquots were collected and assayed for radioactivity and absorption at 260 mμ. The various fractions were pooled and components identified by paper chromatography. Radioactive fractions were adsorbed on a Norit A column and, after washing with water, were eluted with 10% aqueous isoamyl alcohol. Ribose and phosphate analyses and absorption spectra suggested that Compounds I and II were the nicotinic acid analogs of NMN (desamido-NMN) and DPN (desamido-DPN), respectively. Compound II was not active with alcohol dehydrogenase and did not form a fluorescent acetone addition product.[5] Further work on the identification of these compounds is in progress.

TABLE I

SYNTHESIS OF DPN FROM DESAMIDO-DPN

Complete incubation system consisted of 1.7 μmoles of desamido DPN, 20 μmoles of glutamine, 2 μmoles of ATP, 5 μmoles of ATP, 40 μmoles of P_i buffer, *p*H 7.4, and 4 cc. of dialyzed yeast autolysate. Total volume 1.0 cc. after incubation of 100 minutes 1 cc. of H_2O was added and the protein denatured at 100° for 1.5 minutes; 1-cc. aliquots were taken for alcohol dehydrogenase assay of DPN formed.

Omission and/or Additions	μmoles DPN synthesized
None	0.116
−Desamido DPN	.028
−ATP	.011
$-Mg^{++}$	.014
−Glutamine	.026
−Glutamine + NH_4Cl 20 μm.	.015
−Glutamine + glutamate 20 μm.	.019
"O time"	.007

Dialyzed yeast autolysates have been found slowly to synthesize DPN upon addition of glutamine, ATP, PRPP, Mg^{++}, F^- and NA but not NAm. With nicotinic acid-7-C^{14} in the medium, accumulation of compounds I and II was again observed. Compound II prepared with erythrocytes or yeast exhibited an R_f in 70% ethanol, 30% 1 *M* ammonium acetate, *p*H 5.0 identical to that of an authentic sample of the nicotinic acid analog of DPN kindly supplied to us by Dr. N. O. Kaplan. This solvent system adequately separates Compound II from DPN. Synthesis of Compound II from NA by yeast autolysate is dependent on ATP, PRPP and Mg^{++}. R5P is 10% as effective as PRPP. Glutamine, ATP and Mg^{++} are required for synthesis of DPN from desamido-DPN (Table I). NH_4^+ and glutamate are inactive as amide donors. Further investigations of the mechanisms of these enzymatic syntheses, purification of the enzymes, and a study of their distribution in bacterial and mammalian systems are in progress.

(5) N. Levitas, J. Robinson, F. Rosen and W. A. Perlzweig, *J. Biol. Chem.*, **167**, 511 (1947).

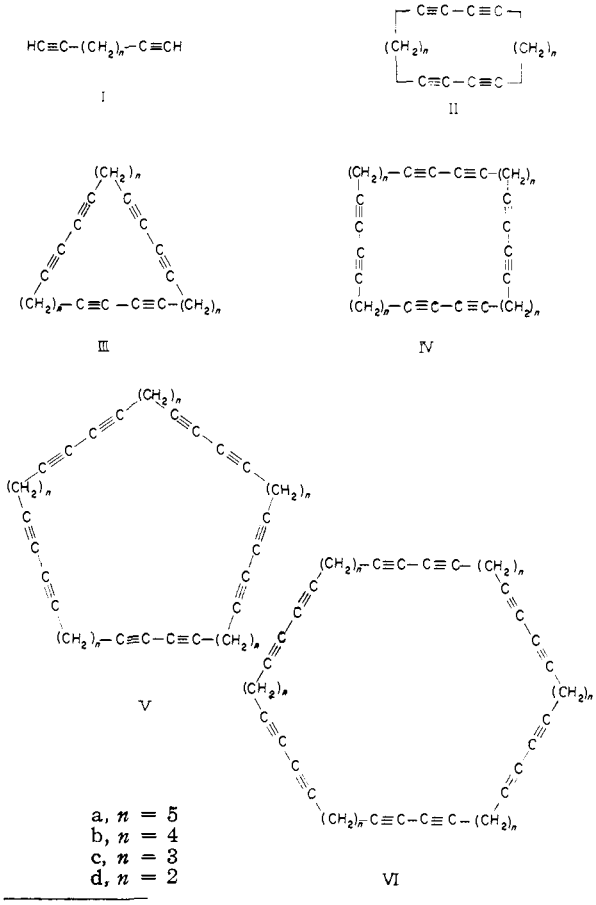
DEPARTMENT OF BIOCHEMISTRY JACK PREISS
DUKE UNIVERSITY SCHOOL OF MEDICINE
DURHAM, NORTH CAROLINA PHILIP HANDLER

RECEIVED JUNE 14, 1957

UNSATURATED MACROCYCLIC COMPOUNDS. V.[1] LARGE RING POLY-ACETYLENES

Sir:

We have shown recently that the oxidation of terminal diacetylenes of type I with oxygen in the presence of cuprous chloride and ammonium chloride in aqueous ethanol leads to the cyclic dimers II besides linear compounds.[1,2] We have studied this type of coupling of terminal diacetylenes under a variety of conditions, especially in homogeneous media. The surprising discovery has been made that when diacetylenes of type I in pyridine solution are simply heated with neutral cupric acetate,[3] not only the cyclic dimers II are formed, but also the cyclic trimers III, tetramers IV, pentamers V, hexamers VI and higher cyclic



(1) Part IV, F. Sondheimer, Y. Amiel and R. Wolovsky, THIS JOURNAL, in press.

(2) (a) F. Sondheimer and Y. Amiel, *ibid.*, **78**, 4178 (1956); *ibid.*, in press; (b) Y. Amiel, F. Sondheimer and R. Wolovsky, *Proc. Chem. Soc.*, 22 (1957).

(3) This sort of oxidation has been shown to result in the smooth coupling of simple acetylenes and to give the cyclic monomer, and dimer in the case of I, $n = 10$ (and of a diacetylenic diester) when carried out under conditions of high dilution (G. Eglinton and A. R. Galbraith, *Chem. and Ind.*, 737 (1956)).

polyacetylenes. Except in the case of Id, only small amounts of the linear coupling products result.

For example 1 part of nona-1,8-diyne (Ia) in 100 parts of pyridine was heated and stirred with 15 parts of neutral cupric acetate at 55° for 3 hours. The product on chromatography on 200 parts of alumina into *ca.* 300 fractions gave successively: (a) the C_{18}-tetrayne IIa (10%), m.p. 210–212° (identical with that made previously[1,2b]); (b) the C_{27}-hexayne IIIa (13%), m.p. 125–126° (hydrogenated to $C_{27}H_{54}$,[4] m.p. 47–48°, mol. wt., 374[5]; calcd. 378); (c) the C_{36}-octayne IVa (11%), m.p. 135–136° (hydrogenated to $C_{36}H_{72}$,[4] m.p. 70–71°, mol. wt., 517; calcd., 504); (d) the C_{45}-decayne Va (4%), m.p. 144–145° (hydrogenated to $C_{45}H_{90}$,[4] m.p. 78.5–79°, mol. wt., 618; calcd., 630); (e) probably the C_{54}-dodecayne VIa (4%), m.p. 144–145° (hydrogenated to $C_{54}H_{108}$,[4] m.p. 90–91°, mol. wt. not determined due to insolubility).

A similar coupling of octa-1,7-diyne (Ib) gave: (a) the C_{16}-tetrayne IIb (9%), m.p. 162–163° (identical with that made previously[2a]); (b) the C_{24}-hexayne IIIb (14%), m.p. 173–174° (hydrogenated to $C_{24}H_{48}$, m.p. 46.5–47°, mol. wt., 330; reported: m.p. 46–47°, mol. wt., 336); (c) the C_{32}-octayne IVb (8%), m.p. 154–155° (hydrogenated to $C_{32}H_{64}$, m.p. 58–59°, mol. wt., 429; reported: m.p. 59–60°, mol. wt., 448); (d) the C_{40}-decayne Vb (9%), m.p. 155–157° (hydrogenated to $C_{40}H_{80}$,[4] m.p., 74–75°, mol. wt., 568; calcd. 560). Coupling of hepta-1,6-diyne (Ic) did not give the C_{14}-tetrayne IIc (if formed, it probably decomposed),[1,2b] but the C_{21}-hexayne IIIc (3%), m.p. 174–175° (hydrogenated to $C_{21}H_{42}$,[4] m.p. 63–64°, mol. wt., 289; calcd. 294) and the C_{28}-octayne IVc (4%), m.p. 213–214° (hydrogenated to $C_{28}H_{56}$, m.p. 47–48°, mol. wt., 389; reported: m.p. 47–48°, mol. wt., 398).

Coupling of hexa-1,5-diyne (Id) and chromatography into 450 fractions gave: (a) the linear dimer (9%), m.p. 98–99° (identical with that made previously[1,2b]); (b) the C_{18}-hexayne IIId (6%), decomposes on heating (hydrogenated to $C_{18}H_{36}$, m.p. 72–73°, identical to that obtained from IIa); (c) the C_{24}-octayne IVd (6%), decomposes on heating (hydrogenated to $C_{24}H_{48}$, m.p. 46–47°, identical to that obtained from IIIb); (d) the C_{30}-decayne Vd (6%), decomposes on heating (hydrogenated to $C_{30}H_{60}$, m.p. 57–58°; mol. wt., 416; reported m.p. 57–58°, mol. wt., 420); (e) probably the cyclic C_{42}-tetradecayne, decomposes on heating (hydrogenated to $C_{42}H_{84}$,[4] m.p. 75–76°, mol. wt., 598; calcd., 588).

All the above cyclic polyacetylenes were highly crystalline. They were shown to differ from each other since they gave mutual depressions in m.p. Their cyclic nature was shown by the absence of acetylenic hydrogen (no band at *ca.* 3300 cm.$^{-1}$ in the infrared, no precipitate with silver nitrate) and by the absence of terminal methyl groups (no band at *ca.* 1380 cm.$^{-1}$) in the corresponding saturated compounds. The latter gave satisfactory analytical results, but most of the polyacetylenes exploded on attempted combustion and gave low carbon values. In all the four series studied, cyclic polyacetylenes of higher mol. wt. than those described were also obtained; their structures are now under investigation.

The present method makes available in one step highly unsaturated large ring alicyclic hydrocarbons (and by hydrogenation the corresponding saturated ones) over a wide range of size, including considerably larger ones than the thirty-four membered cycle which is the biggest alicyclic ring prepared previously.

DANIEL SIEFF RESEARCH INSTITUTE
WEIZMANN INSTITUTE OF SCIENCE
REHOVOTH, ISRAEL
FRANZ SONDHEIMER
YAACOV AMIEL
REUVEN WOLOVSKY

RECEIVED JUNE 17, 1957

(4) This is a previously unknown cycloalkane.

(5) All molecular weights were determined by the Rast method in camphene.

THE CHANGE OF OPTICAL ACTIVITY OF POLY-γ-BENZYL-L-GLUTAMATE IN AN ELECTRIC FIELD

Sir:

The optical activity of a molecule is different along different directions. Therefore we expect the optical activity of a solution of these molecules to change if the molecules are oriented. However, this effect has not been described. We wish to report the change of optical activity of a solution of poly-γ-benzyl-L-glutamate (PBLG) caused by orientation of the molecules in an electric field.

An equation relating the change in optical activity to molecular parameters at infinite dilution has been derived.[1] For a helical molecule the

$$\frac{[\alpha]_E - [\alpha]_0}{E^2} = \frac{2}{45}([\alpha_{33}) - [\alpha_{11}])[p_3^2 + (q_{33} - q_{11})]$$

change in the specific optical activity in an electric field[2] E is proportional to the difference in optical activity for light incident parallel to the helical axis $[\alpha_{33}]$ and perpendicular to the helical axis $[\alpha_{11}]$. The proportionality factor is the electrical orientation term which involves the permanent and induced dipole moments of the molecule. The specific optical activity in the absence of a field $[\alpha]_0$ is just $(2[\alpha_{11}] + [\alpha_{33}])/3$.

In order to measure the change of optical activity in an electric field, the light must be incident parallel to the field. Only along this direction is the refractive index and optical absorption the same for all directions of the plane of polarization.[3]

A one-cm. cell with transparent conducting glass[5] electrodes was used. The optical rotation with the electric field off, then on, was measured from λ = 330 to 550 mμ in a Rudolph automatic spectropolarimeter[6] for two concentrations of M_w = 64,000 PBLG[7] in ethylene dichloride. The

(1) I. Tinoco, Jr., and W. G. Hammerle, *J. Phys. Chem.*, **60**, 1619 (1956).

(2) The units of E are e.s.u./cm. (1 e.s.u. = 300 volts).

(3) The change of refractive index in an electric field is electrical birefringence or Kerr effect.[4] The change of optical absorption is electrical dichroism; it has not been reported.

(4) C. G. LeFèvre and R. J. W. LeFèvre, *Revs. Pure Appl. Chem.*, **5**, 261 (1955).

(5) We wish to thank Dr. E. M. Greist of Corning Glass Works for kindly furnishing this glass (E-C #7740).

(6) We wish to thank Professors J. B. Nielands and H. K. Schachman, and Mr. B. Burnham for the use of this instrument.

(7) We wish to thank Dr. E. R. Blout for kindly supplying a sample of PBLG (#ES-508) for these measurements.