Oxygenation Experiments. General Procedure.—To carry out these experiments, a 5-mm. Pyrex sample tube was prepared which was long enough to fit inside a 10-mm. Pyrex glass tube. This latter one resembled an absorption tube used ordinarily in carbon and hydrogen determinations, but was smaller in size. The sample tube was constricted at one end and a small glass wool plug was inserted inside this part. The outer tube, which was equipped with glass valves at each end, was just long enough to accommodate the sample tube. The volume of the outside tube was, therefore, decreased to a minimum.

Both the sample tube and the absorption tube were properly tared by means of an identical set. Taring was accomplished after both tubes had been swept with pure nitrogen and the glass valves at each end of the tubes had been closed.

A small portion (approx. *10 mg.*) of the imidazole hemochrome was then introduced into the sample tube with a micro-spatula. The sample tube was fitted inside the absorption tube and was then placed inside a small, electrically heated furnace. The tube was evacuated to a mercury pressure of 5 mm., heated and kept at 60–65° for two hours. At the end of this period, pure nitrogen was allowed to pass through the tube for two hours, while the temperature was kept at 60–65°. This operation was necessary to remove any oxygen which may have been taken up by the sample. The tube was then weighed. Next, dry air was passed through the sample for eight hours, at room temperature, at a rate of 20 cc. per minute, after which the tube was swept with pure nitrogen for one minute and again weighed. Desorption of the oxygen was carried out by heating the sample in the tube to 60–65° and passing pure nitrogen through it for six hours while the 60–65° temperature was maintained. The sample then was weighed. A second and a third cycling were carried out as described above.

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COMMUNICATIONS TO THE EDITOR

THE CRYSTAL STRUCTURE OF TOSYL-L-PROLYL-L-HYDROXYPROLINE MONOHYDRATE

Sir:

Hydrolysis studies[1,2] have shown that the sequence -gly-pro-hypro- is a common one in collagen and gelatin and this evidence has been used as a restrictive factor in building models of collagen.[3,4] Hence the structure analysis of a peptide involving the sequence -pro-hypro- would be of interest. The compound tosyl-L-prolyl-L-hydroxyproline was available to us[5] and additional interest lay in testing the usefulness of a "marker group" (in this case *p*-toluenesulfonyl) in the X-ray determination of relatively complex structures.

Tosyl-L-prolyl-L-hydroxyproline monohydrate,[6] M = 400.44, crystallizes from water as plates, frequently twinned. The crystal system is monoclinic, a = 6.291, b = 7.689, c = 19.640 Å., β = 99°27.5′. The space group is P2, with Z = 2, the density calculated being 1.419, measured 1.415.

With information derived from partial three-dimensional data, image-seeking processes[8] were applied to the sharpened-up zero-layer Patterson function $P_0(u, w)$. Guided by the use of models of the component molecular units, tosyl,[9] prolyl[10] and hydroxyproline,[11] a reasonable disposition in the *ac* projection area was achieved. The reliability index, ($R = \Sigma\,|F_0 - F_c|/\Sigma|F_0|$), initially 0.52, has been reduced to 0.18 for the observed *h0l* structure amplitudes.[12] The corresponding electron-density distribution is shown in Fig. 1 with

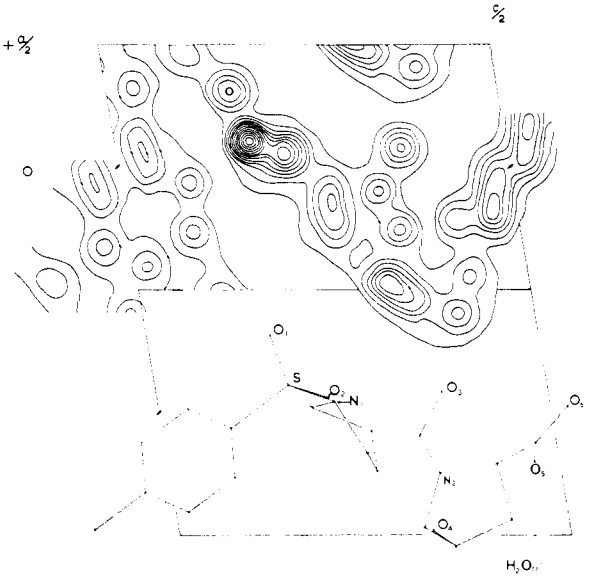

Fig. 1.—Crystal structure of tosyl-L-polyl-L-hydroxyprolin monohydrate.

the molecular interpretation adjacent.[13] The y parameters are being determined from the three-dimensional data.

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(5) A. F. Beecham, THIS JOURNAL, **79**, 3262 (1957).

(6) Difference maps[7] revealed the presence of an atom additional to those in the molecular formula.[8] Assumed to be a water molecule, independent physical measurements confirmed this. *Anal.* Calcd. for $C_{17}H_{22}N_2O_6S\cdot H_2O$: C, 50.99; H, 6.04; N, 7.00; O, 27.97; S, 8.01. Found: C, 50.93; H, 6.12; N, 6.48; O, 27.80; S, 7.94.

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(8) M. J. Buerger, *ibid.*, **4**, 531 (1951).

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(10) A. McL. Mathieson and H. K. Welsh, *Acta Cryst.*, **5**, 599 (1952).

(11) (a) J. Zussman, *ibid.*, **4**, 72, 493 (1951); (b) J. Donohue and K. Trueblood, *ibid.*, **5**, 419 (1952).

(12) Y. C. Leung and R. E. Marsh, *ibid.*, **10**, 815 (1957); **11**, 17 (1958).

(13) A table of provisional x, z parameters of the 27 atoms has been deposited as Document Number 5599 with the ADI Auxiliary Publications Project, Photoduplications Service, Library of Congress, Washington 25, D. C. A copy may be secured by citing the Document Number and by remitting $1.25 for photoprints, or $1.25 for 35 mm. microfilm in advance by check or money order payable to: Chief, Photoduplication Service, Library of Congress.

The molecule may be considered as built of four main planar or approximately planar groups, *p*-methyl-thiophenyl, prolyl, peptide + pyrrolidine and carboxyl, all of which tend to lie at approximately right angles to one another. In the case of the prolyl ring it would appear that the carbon atom opposite the N–C_α bond does not show any tendency to occupy alternate sites as noted by Leung and Marsh in the analysis of leucylprolylglycine monohydrate.[12] Neither does it maintain the position observed in proline[10] or hydroxyproline,[11] *i.e., trans* with respect to the carboxyl group. In this compound it appears to have swung to the same side as the peptide C=O, providing further evidence of the flexibility of the pyrrolidine ring system in the prolyl group.

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RECEIVED MAY 5, 1958

PHOTOSYNTHESIS OF GALACTOLIPIDS

Sir:

An appreciable portion of the products of brief photosynthesis in $C^{14}O_2$ is lipid in nature.[1] Chromatograms of these products exhibit several separable lipids[2] which had been assumed to be fatty acid-labeled phosphatides. However, we wish to report that the phosphatides constitute but a fraction of these labeled products. We have examined the lipids of *Chlorella* and find that the glycolipid concentration exceeds that of the phospholipids by a factor of four. These glycolipids include the β-D-galactosyl and the α-D-galactosyl-(1 → 6)-β-D-galactosyl monoglycerides which had been identified in wheat flour lipids by Carter, *et al.*,[3] and 3′-*O*-oleyl-glyceryl-1′β-D-galactopyranoside-6-sulfate.[4] Glycerolphosphatides[5] and lesser amounts of a galactotriosyl monoglyceride are observed.

Radiograms of deacylated[5,6] products of five minutes and of thirty seconds of photosynthesis in $C^{14}O_2$ by *Chlorella* revealed that the galactolipids are labeled very rapidly with C^{14}. In five minutes photosynthesis over half of the C^{14} in the lipids was found in the galactose moieties. These lipids had the following C^{14} distribution: fatty acids, 40%; galactosylglycerol, 39%; galactosylgalactosylglycerol, 10%; galactosyl-6-sulfate glycerol, 2%; diglycerophosphate, 3%; glycerol, 4%. The galactolipids were identified by examination of radiograms of the deacylated lipids and then acid hydrolysis of the uniformly-labeled galactosyl glycerols to yield simple ratios of C^{14} in galactose and glycerol. Glyceryl β-D-galactoside and its digalactose homolog cochromatographed precisely with authentic samples generously provided by Professor H. E. Carter. The glyceryl galactotrioside was found in a chromatographic position (R_f = 0.50 in phenol–water and R_f = 0.14 in butanol–propionic acid–water[2]) characteristic of a third member of the homologous series of galactosyl glycerides. Hydrolysis by thirty minutes in 3 *N* hydrochloric acid at 100° of the uniformly labeled unknown formed during eight days of photosynthesis gave galactose and glycerol in an activity ratio of 6.1 to 1, thus indicating that there are three galactose units bound to a monoglyceride in the original lipid.

The only precursor available in appreciable concentration in *Chlorella* for biosynthesis of these glycolipids is uridine diphosphate galactose.[7,8] Biosynthesis of galactosyl glycerides is probably analogous to that of the uridine diphosphate galactose-mediated saccharide syntheses.[9]

We are indebted to Mr. M. S. Brown for valuable assistance. This work was supported by the Atomic Energy Commission and the Pennsylvania Agricultural Experiment Station.

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RECEIVED JULY 24, 1958

STEREOCHEMISTRY OF DIELS-ALDER ADDUCTS. I. THE REARRANGEMENT OF 2-*exo*-BROMONORBORNANE-2-*endo*-CARBOXAMIDE

Sir:

A recent communication[1] describing the rearrangement of 2-*exo*-bromonorbornane-2-*endo*-carboxylic acid (I, R = OH) and its methyl ester (I, R = OCH_3) upon catalytic or chemical hydrogenolysis prompts us to report our observations upon the rearrangement of the corresponding carboxamide. 2 - *exo* - Bromonorbornane - 2 - *endo*-carboxamide (I, R = NH_2) does not rearrange upon hydrogenolysis and yields only norbornane-2-*endo*carboxamide (III, R = NH_2), identified by analysis, mixed melting point and comparison of the infrared spectrum with that of an authentic sample. When I (R = NH_2) was heated above its melting point, resolidification took place and a second melting point was observed. The isomeric bromocarboxamide (II, R = NH_2) obtained from the melt gave norbornane-1-carboxamide (IV, R = NH_2) upon hydrogenolysis. The rearrangement of I (R = NH_2) to II (R = NH_2) was also catalyzed by alcoholic alkali. On the basis of Wagner–Meerwein rearrangements undergone by 2,2- disubstituted bicyclo[2,2,1]heptane derivatives[2–4] we have tentatively designated the rearrangement product II (R = NH_2) as 2-*exo*-bromo-

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