

reversing its direction of rotation) and by adding a cooling cell to the water bath to oppose the action of the heating element. Several typical plots of time *versus* the temperature of the reaction system obtained under these operating conditions are illustrated in Fig. 3. The constancy of the rates of heating and cooling is evident.

The programmed thermoregulator appears to have usefulness in a variety of applications where accurate control of rates of heating or cooling are required. The performance of the apparatus compares favorably with that specified for the more costly program devices which are available commercially.

Hydrogenation of 1-Naphthol with Rhodium Catalyst

By MORRIS FREIFELDER and GEORGE R. STONE

The low-pressure hydrogenation of 1-naphthol with rhodium is found to cause perhydrogenation. Use of this is made to produce *cis,cis* 1-decalol.

IN THE COURSE of studying the effect of rhodium catalysts in the hydrogenation of organic compounds, we investigated the reduction of 1-naphthol (I) under low-pressure conditions. Hydrogenation was interrupted after 2 *M* equivalents were absorbed and the resultant reaction mixture subjected to vapor-phase chromatography. A major peak consisting of about 70% of the total was identified as 5,6,7,8-tetrahydro-1-naphthol (II). In addition to a small amount of decalin and a few per cent of what appeared to be 1-tetralone, about 20% of another product was seen in the chromatograph. From its position it was suspected to be a decalol.

In view of this tendency toward overhydrogenation, it seemed worthwhile to study the reaction further. The mild conditions employed should lead to a predominance of *cis* products. It was hoped that *cis,cis* 1-decalol (III) would be obtained. If so, a less time consuming and more direct method than those described in the literature would be available.

Gutsche and Peter have prepared III in a three-step synthesis (1). Their material was of excellent quality judging from its melting point, but the conversion of II to III with freshly prepared platinum oxide required 4 days. Zimmerman and Mais, starting with I, also found that a long reduction period was necessary (2). The same slow uptake of hydrogen was encountered in another reduction in which decalin and mixed decalols were obtained as well as III (3). With Raney nickel at elevated temperature and pressure, mixtures were obtained (4).

Hydrogenation of I with rhodium catalyst at 60° and 3 Atm. did lead to III. Vapor-phase chromatography showed about 5% of decalin and almost 10% of a product which we assumed to be the intermediate 1-decalone, and a major peak (84.5%). On recrystallization of the residue from this reduction III was obtained. After several reductions at low pressure, it appeared more desirable to run the reaction at high pressure and at room temperature to convert the intermediate 1-decalone completely (if indeed it was present) and to cut down on hydrogenolysis. When the hydrogenation was carried out at 130 Atm., presumably the only product obtained

was III. On chromatography, a small amount of impurity (*ca.* 3%) was found. Infrared examination showed the absence of OH and aromatic ring and the presence of a carbonyl peak at 5.78 μ . In the absence of any conjugation, the impurity was assumed to be 1-decalone.

EXPERIMENTAL

5,6,7,8-Tetrahydro-1-naphthol.—A solution of 14.4 Gm. (0.1 mole) of 1-naphthol in 50 ml. of analytical reagent methyl alcohol was hydrogenated under 3 Atm. pressure in the presence of 4.3 Gm. of 5% rhodium on alumina catalyst.¹ Uptake of 2 *M* equivalents was complete in 5 hours. The reaction was interrupted and the solution filtered from the catalyst. After concentration, the residue solidified on cooling. Vapor-phase chromatography² showed 0.72% decalin, 20.4% of decalol or decalols, 69% of a tetrahydronaphthol, plus about 7% of two unknowns. The mixture on infrared examination³ showed presence of OH, aromatic ring, and a carbonyl peak at 5.9+ μ (1-tetralone shows a peak at 5.98 μ).

The mixture was then dissolved in benzene and extracted with 100 ml. of 50% sodium hydroxide solution. The alkaline solution was acidified and extracted with benzene. The benzene extract was dried over anhydrous magnesium sulfate, filtered, and concentrated. The residue was treated with hexane and allowed to stand to crystallize. The product obtained melted at 68° (described 68.5 to 69°) (1) and was shown to be 5,6,7,8-tetrahydro-1-naphthol by mixed melting point and infrared absorption compared to a known standard.

***cis,cis* 1-Decalol.**—The hydrogenation of 0.1 mole of 1-naphthol was carried out at 60° and 3 Atm. in the presence of 5.8 Gm. of 5% rhodium on carbon.¹ Uptake for 5 H₂ was complete in 12 hours. After removal of catalyst and solvent, the residue obtained solidified and melted at 77–86° with softening at 70°. Recrystallization from heptane raised the melting point to 90 to 91.5° (described 93°) (1). The crude material was examined by means of vapor-phase chromatography. Results showed 5.4% decalin, 9.6% of an unidentified fraction (possibly 1-decalone), and 84.3% of III.

A second set of runs was carried out as above. The residue was distilled. Decalin was again obtained, the remainder of the material distilled over a wide range, 210–237° at atmospheric pressure.

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¹ Available from Engelhard Industries, Newark, N. J.

² Carried out by P. F. Helgren of this laboratory.

³ Carried out by W. Washburn and A. Kammer of this laboratory.

Reduction at Higher Pressure.—A solution of 72.0 Gm. (0.5 mole) of I in 250 ml. of 95% ethyl alcohol was hydrogenated at room temperature and 130 Atm. in the presence of 30 Gm. of 5% rhodium on carbon. Reaction was slightly exothermic, the temperature rising to 46°. Uptake was complete in about 1 hour. The catalyst was removed, the solution concentrated, and the residue distilled. It boiled at 237–240° [lit. 246° at 745 mm. (5)] at atmospheric pressure and solidified quickly. The product weighed 74.5 Gm. and melted at 89–91°.

To determine the purity of the product, 2.5 Gm. was dissolved in a mixture of 20 ml. of 99% benzene and 1% ether, put over a silica gel column, and eluted with a 95% benzene and 5% ether mixture. Fractions were collected in 30-ml. portions. These were followed by means of thin-layer chromatography. In one fraction a minor component was

⁴ Performed by D. E. Dickson, Regis Chemical Co., Chicago, Ill.

seen. It was isolated and found by infrared examination to have a peak at 5.78 μ , indicative of an unconjugated carbonyl group. Since there was no evidence of aromaticity, it was assumed to be 1-decalone. One more fraction produced a mixture in which the major component predominated (85%). Thereafter only *cis,cis* 1-decalol was obtained, 1.5 Gm., m.p. 91–92°.

In a larger run (1.0 mole) at 200 Atm. pressure, the temperature rose to 54–55°. In this instance the yield of distilled III was lower (76.5%).

REFERENCES

- (1) Gutsche, C. D., and Peter, H. H., *J. Am. Chem. Soc.*, **77**, 5971 (1955).
- (2) Zimmerman, H. F., and Mais, A., *ibid.*, **81**, 3644 (1959).
- (3) Dauben, W. G., Tweit, R. C., and Mannerskantz, C., *ibid.*, **76**, 4420 (1954).
- (4) Hüchel, W., and Maucher, D., *Ann.*, **645**, 141 (1961); Hüchel, W., and Feltkamp, H., *ibid.*, **649**, 21 (1961).
- (5) Hüchel, W., Danneel, R., Gross, A., and Naab, H., *ibid.*, **502**, 99 (1933).

Lipid Content of *Chlorella* "Aerated" with a CO₂-Nitrogen versus a CO₂-Air Mixture

By ROBERTSON PRATT and EVELYN JOHNSON

Contrary to some earlier reports, substitution of nitrogen for air in the CO₂-air mixture conventionally used to "aerate" cultures of *Chlorella* had little effect on dry weight or on protein and lipid content of *C. pyrenoidosa* harvested after 8 weeks, whether the cells were provided with a good nutrient solution or with one deficient in nitrogen. In a manganese-deficient medium, the yield of dry weight and the protein and lipid content of the cells produced was approximately the same whether a CO₂-air or a CO₂-nitrogen gas stream was used, but the dry weight and lipid content were markedly lower and the protein content was higher than in cultures developed in the standard complete medium.

IT HAS BEEN REPORTED (1, 2) and confirmed (3) that cells of *Chlorella pyrenoidosa* have a higher lipid content when cultured in a nitrogen-deficient mineral nutrient solution than when cultured in one with an adequate concentration of nitrogen. However, the highest level of lipid found by the later authors was markedly lower than the 65% in 4 weeks and approximately 75% in 10 weeks, reported by the earlier workers (1, 2). Seeking sources of the discrepancy is difficult because of the lack of specific details with respect to environmental conditions in the earlier papers. Since composition of the "aeration" stream might have been an important variable, this has now been investigated and is the principal subject of this report.

The "aeration" mixture customarily used in *Chlorella* experiments in this laboratory and elsewhere for many years consists of 5% carbon dioxide and 95% air. However, a personal communication from one of the earlier authors states that they "found it essential to use 5% CO₂ in nitrogen in order to maintain cultures in good shape for the long time needed to obtain the highest lipid content" and that "we too observed clumping of cells in old cultures in SM (nitrogen-deficient) medium grown with CO₂ in air... But even in the 70–80

day old, very high lipid cultures grown with CO₂ in N₂, the cells remained freely suspendible and showed negligible sticking together" (4).

RESULTS

In three experiments (each run in duplicate) averaged in Table IA, substitution of CO₂-nitrogen for the CO₂-air stream had only minimal effect on the dry weight of cells produced and on their protein and lipid content, whether the alga was cultured in the standard medium or in the nitrogen-deficient one. Regardless of which gas mixture was provided, the lipid content (as per cent of dry weight) was about 19% higher, and the protein level was about 10% lower in the medium deficient in mineral nitrogen than in the standard. Under the experimental conditions employed, we found no evidence that composition of the culture solution affected the response of the cells to substitution of nitrogen for air in the gas stream or to support the cautiously worded suggestion that *Chlorella* may "under the special conditions producing high R-value (namely, mineral nitrogen starvation and supplied with 5% CO₂ in nitrogen), fix atmospheric nitrogen" (1).

It has been stressed that high lipid values were never attained when the "residual fixed nitrogen concentration was greater than 0.001 molar" in the medium (1). Although the conditions provided in the present experiments are believed to have

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