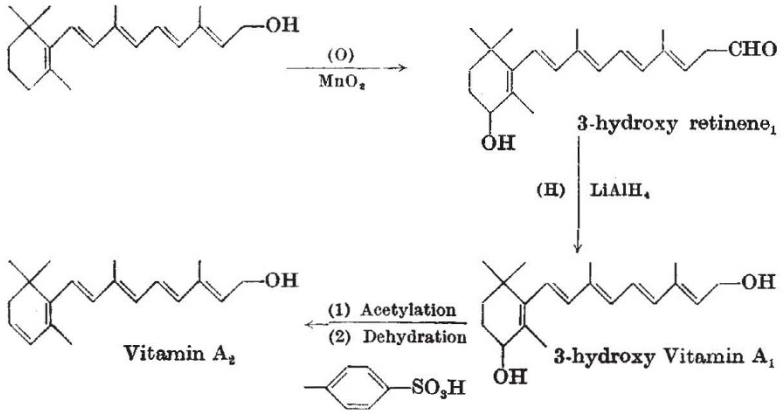
A New Synthesis of Vitamin A_2: Conversion of Vitamin A_1 to A_2

THE earlier confusion regarding the structure of vitamin A_2 was removed by its synthesis[1], which conclusively proved it to have the dehydrovitamin A_1 formula suggested by Morton, Salah and Stubbs[2]. In this synthesis an extra double bond was introduced into the β-ionone ring of the methylester of vitamin A_1 acid, by bromination with *N*-bromosuccinimide, followed by dehydrobromination with 4-phenyl morpholine. Reduction with lithium aluminium hydride gave a product which closely agreed with the spectroscopic properties of vitamin A_2. Recently, Henbest, Jones and Owen[3] carried out an elegant conversion of vitamin A_1 to A_2 by treating the aldehyde of vitamin A_1 (retinene$_1$) with *N*-bromosuccinimide, to obtain retinene$_2$, which by reduction with lithium aluminium hydride was converted to vitamin A_2.

The present communication records a new synthesis of vitamin A_2 and also demonstrates the conversion of vitamin A_1 to vitamin A_2 through 3-hydroxy retinene$_1$.

Retinene$_1$ was shown to be best prepared by the oxidation of vitamin A_1 by the elegant and convenient manganese dioxide method[4]. It has been shown by Wald[5] that a rapid method of effecting the oxidation is to pass a solution of the alcohol in light petroleum ether through a tube containing the oxidizing agent. We have now observed that by using a specific variety of manganese dioxide in the column and by controlling the conditions, it is quite possible to effect a preponderance of 3-hydroxy retinene$_1$, in the oxidation of vitamin A_1 alcohol on the column of manganese dioxide. Full details of this work will be given elsewhere. The mixed oxidation products were chromatographed on weakened alumina, and the main band after extrusion and elution with ethanol gave an absorption maximum at 375 mμ, characteristic of 3-hydroxy retinene$_1$ (ref. 3). Further synthetic work was carried out, using 3-hydroxy retinene$_1$ as the starting material. It was carefully reduced, using the calculated quantity of lithium aluminium hydride, to 3-hydroxy vitamin A_1. This new substance gave an absorption maximum at 324 mμ. The hydroxyvitamin A_1 was first acetylated (selective acetylation) and then dehydrated with *p*-toluene sulphonic acid[6]. After chromatographic purification[7], the yellow band which was extruded and eluted with ethanol, gave an ultra-violet absorption spectrum characteristic of vitamin A_2 (refs. 8 and 9) (having absorption maxima at 351 and 287 mμ). The reactions involved are as follows:



Further work is in progress to characterize more fully the products obtained in the different steps in this synthesis.

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Genetics and Biochemistry of Riboflavin Auxotrophs of *Aspergillus nidulans*

BIOCHEMICAL and genetical investigations are being made on seven riboflavin-less mutants of *Aspergillus nidulans*, isolated in Prof. G. Pontecorvo's laboratory, in the University of Glasgow. A brief account of the important results obtained is presented in this communication.

Genetic analyses were made only on ribo$_3$, ribo$_4$, ribo$_5$, ribo$_6$ and ribo$_7$ since ribo$_1$ and ribo$_2$ have been located earlier in chromosomes 1 and 8, respectively[1]. It is found that ribo$_4$ is allelic to ribo$_2$ and ribo$_7$ to ribo$_1$, whereas ribo$_3$, ribo$_5$ and ribo$_6$ are not allelic to one another nor to ribo$_1$ and ribo$_2$. Genetic analysis made by means of mitotic recombination and meiotic crossing-over showed that the non-allelic ribo$_3$ and ribo$_5$ are in the fifth chromosome and ribo$_6$ in the second chromosome.

Since the five non-allelic riboflavin-less mutants ribo$_1$, ribo$_2$, ribo$_3$, ribo$_5$ and ribo$_6$ are due to single gene-mutations, each would therefore be expected to differ from the normal in only one biochemical reaction in the biosynthesis of riboflavin. It is found that all these auxotrophs are independent of temperature and the three-colour markers green, yellow and white show very similar responses to riboflavin for growth. Whereas, there is maximal growth in the case of ribo$_3$ at a concentration of 0·1 μgm. of riboflavin per ml. of the medium, the responses of the mutants ribo$_1$, ribo$_2$, ribo$_5$ and ribo$_6$ to riboflavin are considerably poorer at low concentration. However, they reach a normal weight with greater amount of riboflavin. The response to flavin mononucleotide (FMN) by these auxotrophs is very slight, and to flavin dinucleotide (FAD), negligible.

Attempts made to find growth response to riboflavin-like substances for the vitamin activity showed that lumichrome, a structural analogue of riboflavin, inhibits growth of these auxotrophs in a competitive manner. Among the different compounds tried as possible precursors of the vitamin, it was found that a pyrimidine compound such as alloxan and a purine (xanthine) have riboflavin activity