Zusammenfassung. Es ist gelungen, *Nemalion multifidum*, eine marine Floridee, in axenische Kultur zu bekommen. Die Alge wurde in einer synthetischen Nährlösung gezüchtet. Wenn sie ohne Vitamine 2 Monate kultiviert wurde, konnte ein Zusatz einer Vitaminmischung den Zuwachs verdoppeln. Die wirksamen Vitamine waren am Anfang B_{12} oder B_6; wenn die Alge aber längere Zeit in Hungerkultur gezüchtet worden war, konnte nur die B_{12}-Analoge, Factor Ib, den Zuwachs erhöhen.

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6-Hydroxylation: An Important Metabolic Route for α-Methyltryptamine

D,L-α-methyl tryptamine (αMT) has been found to produce LSD_{25} like effect in normal volunteers[1]. We have found that the effect of 20 mg αMT is comparable in intensity to the psychotomimetic effect of about 60 mg N,N-diethyltryptamine (DET) in man with the difference that the perceptual distortions are more and the sympathomimetic vegetative changes are less pronounced than those produced by DET[2]. Since DET has been found to produce the behavioral changes through the 6-hydroxylated metabolite[3], the question arises whether or not αMT also forms 6-hydroxylated metabolites.

Although the biochemical effects of αMT on serotonin metabolism has been extensively studied[4–6], very little is known about the metabolism of the compound itself. Since the side chain of αMT is identical with the side chain of amphetamine ($-CH_2 \cdot \underset{\displaystyle CH_3}{\underset{|}{CH}} \cdot NH_2$), the compound was expected to undergo the same microsomal deamination as amphetamine[7].

Incubating αMT with rat liver microsomes, soluble supernatant fraction, TPN and Mg^{++}[7] at 37°C resulted in the formation of 3-indolylacetone as evidenced by the paper chromatographic identification of the metabolite in the neutral ether extract with synthetically prepared 3-indolylacetone[8]. Both the metabolite and the synthetic compound gave the same Rf value in three different solvent systems and produced the same purple color reaction with Ehrlich's *p*-dimethylaminobenzaldehyde. The chromatograms of the neutral ether extract also showed another new compound which gave a fast developing blue reaction with Ehrlich's reagent and instant red color with diazotized sulfanilic acid in HCl specific for 6-hydroxy indole derivatives[9]. The Rf values of this new compound (0.83 in *n*-butanol-acetic acid and 0.90 in isopropanol-ammonia) and the color reactions were identical with those of the single metabolite obtained from synthetic 3-indolylacetone in the same microsomal enzyme system. Therefore, this compound is most probably 6-hydroxy-3- indolylacetone.

The pH of the enzymatic reaction mixture (previously extracted with ether) was then adjusted to 9.5 and extracted with *n*-butanol. When the butanol extract was shaken with dilute acetic acid, the aqueous layer contained, besides unchanged αMT[10], a new apparently basic indole derivative (Rf 0.48 in butanol-acetic acid, 0.66 in butanol-ammonia) which gave the color reactions characteristic for 6-hydroxy indoles. It is most likely that this compound is 6-hydroxy-α-methyltryptamine (6-HO-αMT).

Additional evidence for the identity of 6-HO-αMT was obtained by an experiment in which the isolated compound was again incubated with the microsomal enzyme system and yielded 6-hydroxy-3-indolylacetone by deamination.

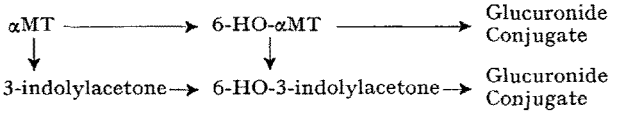
Thus the three major enzymatic products were tentatively identified as 3-indolylacetone, 6-HO-αMT and 6-hydroxy-3-indolylacetone.

The same metabolites have been found to be formed in intact animals and excreted in the urine mainly as glucuronide conjugates.

Male albino rats were injected with αMT (5 mg/kg) intraperitoneally and the urines were collected for 48 h in metabolic cages. Aliquots of the urine samples were incubated with bacterial β-glucuronidase (Sigma) in order to hydrolyze the glucuronide conjugates.

The urine samples were chromatographed without previous extraction. The chromatograms of the samples not treated with β-glucuronidase showed the presence of some unchanged αMT, a spot corresponding to 3-indolylacetone, a small spot corresponding to 6-HO-αMT and a large presumably double spot with low Rf values (0.04 to 0.20 in butanol-acetic acid and 0.20–0.45 in isopropanol-NH_3) and with typical 6-hydroxy-indole color reactions. On the chromatograms of the glucuronidase treated urine samples, this large double spot with low Rf values was absent but the spot corresponding to 6-HO-αMT was tremendously increased and another compound appeared which proved to be identical with the enzymatically prepared 6-hydroxy-3-indolylacetone.

From the evidence described above the following tentative scheme for the metabolism of αMT may be drawn:



Since 6-hydroxylated indole derivatives are more potent pharmacologically than the non-hydroxylated parent compounds[11], it is possible that one or both of the 6-hydroxy metabolites play a part in the behavioral effect of αMT.

Zusammenfassung. Der Stoffwechsel der psychotropischen Verbindung α-Methyltryptamin wurde untersucht. Die drei hauptsächlichsten Metabolite wurden *in vitro* und *in vivo* als 6-Hydroxy-α-methyltryptamin, 3-Indolylaceton und 6-Hydroxy-3-indolylaceton identifiziert.

S. SZARA

Clinical Neuropharmacology Research Center, Washington (D.C.), October 4, 1960.

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