

Dialysis of supernatant and Ro 4-4602 against phosphate buffer did not significantly reduce the inhibition of 3,4-dihydroxyphenylalanine decarboxylation (inhibition without dialysis: $89 \pm 4\%$; inhibition after dialysis: $78 \pm 2\%$; $p > 0.01$).

In vivo: Intraperitoneal injection of 30–200 $\mu\text{M/kg}$ Ro 4-4602 induced maximal inhibition of 3,4-dihydroxyphenylalanine decarboxylation in brain and kidney within 20–60 min. The decarboxylase activity was restored after about 3 days with and without addition of pyridoxal-5'-phosphate. Within 1 h a 50% inhibition of decarboxylase was obtained by 42 $\mu\text{M/kg}$ Ro 4-4602 (corresponding to about 12 mg/kg) in brain (Figure) and by 0.5 $\mu\text{M/kg}$ Ro 4-4602 in kidney. Addition of pyridoxal-5'-phosphate to the incubation medium had no influence on the effect of the inhibitor.

Monoamine oxidase activity in brain was not inhibited significantly ($p < 0.01$) within 16 h after i.p. application of doses as high as 1.0 mM/kg (293 mg/kg) Ro 4-4602. According to the above-mentioned results, Ro 4-4602 probably inhibits decarboxylase *in vivo*. It can, however, not be excluded that an *in vitro* effect might be involved. Ro 4-4602 present in the blood and the extracellular fluid possibly causes or enhances decarboxylase inhibition

only after homogenization of the tissues, whereas *in vivo* the penetration of the drug to the site of the enzyme might be hindered.

Preliminary measurements of the 5-hydroxytryptamine and catecholamine content of various organs showed, however, that Ro 4-4602 has at least some activity *in vivo*. Thus, after single and repeated doses of the compound the level of the amines in different tissues decreased (Table). Furthermore, the increase of 5-hydroxytryptamine in heart and brain induced by 5-hydroxytryptophan could be completely inhibited by Ro 4-4602²³. These effects are probably produced by inhibition of the decarboxylation of the monoamine precursors, such as 5-hydroxytryptophan and 3,4-dihydroxyphenylalanine. It remains to be elucidated why repeated high doses of Ro 4-4602 decreased the content of endogenous monoamines only moderately. The following explanations have to be considered: (a) Only a small fraction of the decarboxylase activity available in normal tissues might be sufficient for the physiological rate of decarboxylation. (b) The amine stores cannot be completely depleted despite marked reduction of monoamine formation.

Zusammenfassung. N-(DL-Seryl)-N'-(2,3,4-trihydroxybenzyl)hydrazin (Ro 4-4602) bewirkt *in vitro* starke Hemmung der Decarboxylase, die durch Pyridoxal-5'-phosphat nicht und durch Dialyse nur wenig vermindert werden kann. Nach i.p. Applikation der Substanz kommt es zu starker Verminderung der Decarboxylase-Aktivität und zu mässiger Herabsetzung von 5-Hydroxytryptamin und Noradrenalin in verschiedenen Organen. Monoaminoxidase des Gehirns wird *in vivo* durch Ro 4-4602 nicht beeinflusst.

W. P. BURKARD, K. F. GEY, and A. PLETSCHER

Medizinische Forschungsabteilung der F. Hoffmann-La Roche & Co. A.G., Basel (Switzerland), June 1, 1962.

Effect of repeated i.p. administration of Ro 4-4602 (6 × 500 mg/kg within 4 days) on catecholamine and 5-hydroxytryptamine content of various organs.

		Endogenous monoamines in % of controls	
		Catecholamines	5-Hydroxytryptamine
Mice	Brain	88 ± 5^a	58 ± 6
	Heart	48 ± 5	
Guinea pigs	Brain	64 ± 5	64 ± 6
	Heart	41 ± 10	
	Duodenum		65 ± 8
	Adrenals	26 ± 5	

^a $0.01 < p < 0.05$; p of all other values < 0.01 .

²³ A. PLETSCHER et al., in preparation (1962).

The Golgi Apparatus in the Male Germ-Cells of *Vaginula* (Pulmonata)

NATH¹, in his review of work on spermatogenesis, refers to my paper on the spermatogenesis of *Vaginula*² (a pulmonate) and states that the Golgi body described by me as such is not Golgi body 'as all the other previous workers on the spermatogenesis of the pulmonate gastropods have described the Golgi body as a dictyosome'. He refers to his earlier view³ that the inclusion I have described as Golgi body is a chromatoid body. It is difficult to make out what has led NATH to hold this view. Is it because I find this body to be spherical rather than plate-like? If the body I have described is not a Golgi body, then this organelle must be represented by some other cytoplasmic inclusion. The illustrations in my fuller paper⁴, to which NATH does not refer, make it abundantly clear that there is no other cytoplasmic body which can be taken to represent the Golgi apparatus. Surely NATH would not go to the length of thinking that there is no Golgi body in these cells at all. Also, my sketch of the Golgi apparatus of *Vaginula*⁴ shows it

to be structurally similar to the Golgi elements of other pulmonates as described by GATENBY, BAKER et al. It is correct that I find it spherical rather than a flat platelet, but so does BAKER⁵, in *Helix*. I have again studied the spermatocytes of *Vaginula* with phase-contrast microscopy and find I can only confirm my previous description.

NATH further states that to the best of his knowledge I am the only worker 'who has denied any connection between the acrosome and the Golgi complex'. This is incorrect. One of the works listed by NATH in his references is BAKER's paper on *Helix*⁵, in which he clearly states that there is no connection between the Golgi apparatus and acrosome-formation. BAKER⁵ (p. 303) states 'the

¹ V. NATH, Int. Rev. Cyt. 5, 395 (1956).

² M. D. L. SRIVASTAVA, Nature 172, 689 (1953).

³ V. NATH and H. C. CHOPRA, Res. Bull. East Punjab Univ. 74, 91 (1955).

⁴ M. D. L. SRIVASTAVA, Z. Zellforsch. 40, 313 (1954).

⁵ J. R. BAKER, Quart. J. microscop. Sci. 90, 293 (1949).

very small acrosome meanwhile appears at the front end of the spermatid, *without any apparent connection with the Golgi bodies* (Italics mine). Further, NATH refers in some detail to ROGUE's⁶ paper on the cytoplasmic inclusions of the male germ-cells of *Helix*. This author finds no connection between acrosome-formation and any of the cytoplasmic inclusions (the so-called paranuclear bodies of this author, which must include the Golgi bodies also). NATH actually quotes ROGUE's own words to this effect.

As stated above, I have reinvestigated the male germ-cells of *Vaginula* by phase-contrast microscopy and I still find no evidence of the participation of any cytoplasmic inclusions in the acrosome-formation. This participation is not in evidence in any of GATENBY's figures^{7,8}, nor is it proved by NATH's¹ photographs despite his claim to have demonstrated it. At present, NATH, following Dr. BAKER⁹, does not accept that there is anything like a Golgi apparatus in animal cells, but it makes no difference to this discussion, whatever be the other name (lipochondria or any other term) that

NATH applies now to the structure which he used to name Golgi apparatus formerly.

Zusammenfassung. Der Verfasser macht am Golgi-körper der männlichen Geschlechtszellen von *Vaginula* eine Nachuntersuchung mit dem Phasenkontrastmikroskop und bestätigt, dass zwischen Golgikörperbildner und Akrosomgenese kein verwandtschaftlicher Zusammenhang besteht. Damit werden entgegenstehende Auffassungen von NATH widerlegt.

M. D. L. SRIVASTAVA

Department of Zoology, University of Allahabad (India), May 11, 1962.

⁶ A. L. ROGUE, J. Roy. microscop. Soc. 74, 188 (1954).

⁷ J. B. GATENBY, Quart. J. microscop. Sci. 62, 555 (1917).

⁸ J. B. GATENBY, Quart. J. microscop. Sci. 63, 445 (1919).

⁹ J. R. BAKER, Proc. Linn. Soc. London 162, 67 (1950).

Rotatory Dispersion Curves of some Decalones of Microbiological Origin¹

The Octant Rule² provides a theoretical basis for the study of optical rotatory dispersion curves of ketones. The application of this rule to extensive collections of data for cyclohexanone types from Professor DJERASSI's laboratory

Decalones and related compounds
Amplitudes of rotatory dispersion curves (in methanol)

No.	Substituents	Amplitude <i>a</i>	Amplitude contributions Δa for substituents named	Refer- ences
(9 <i>R</i>)- <i>trans</i> -1-Decalones				
I	None	−40		⁸
II	(4 <i>R</i>)-4-OH	−43	Δa -OH (II-I) = −3	^{5,12,a}
III	(4 <i>S</i>)-4-OH	−37	Δa -OH (III-I) = +3	^{5,12,a}
IV	(5 <i>R</i>)-5-OH	−61	Δa -OH (IV-I) = −21	^{6,12,a}
V	(5 <i>S</i>)-5-OH	−50	Δa -OH (V-I) = −10	^{6,12,a}
(9 <i>R</i>)-9-Methyl- <i>trans</i> -Decalones				
VI	None	+27		⁹
VII	(5 <i>R</i>)-5-OH	+8	Δa -OH (VII-VI) = −19 Δa -Me (VII-IV) = +69	¹³
(9 <i>R</i>)-10-Methyl- <i>trans</i> -1-Decalones				
VIII	None	−32 ^b		¹⁰
IX	(5 <i>R</i>)-5-OH	−81	Δa -Me (IX-IV) = −20	¹³
X	(5 <i>S</i>)-5-OH	−72	Δa -Me (X-V) = −22	¹³
(9 <i>R</i>)- <i>trans</i> - Δ^6 -1-Octalones				
XI	(4 <i>R</i>)-4-OH	−6		¹⁴
XII	(4 <i>S</i>)-4-OH	−6		¹⁴
(9 <i>R</i>)- <i>cis</i> -1-Decalones				
XIII	None	−14		⁸
XIV	(4 <i>R</i>)-4-OH	−14	Δa -OH (XIV-XIII) = 0	^{5,12,a}
XV	(5 <i>R</i>)-5-OH	−2	Δa -OH (XV-XIII) = +12	^{6,12,a}

^a Curves for these compounds had previously been measured by Professor C. DJERASSI (then at Wayne State University, Detroit) but complete amplitudes were not obtained.

^b Amplitude incomplete; estimated from mixture of *cis* and *trans* isomers.

and from our own laboratory, has been described elsewhere³. It is becoming increasingly apparent that the amplitude⁴ of a Cotton effect curve is a useful measure of the asymmetry of the surroundings of the carbonyl group.

The decalone derivatives prepared by microbiological oxidations and reductions at the ETH, Zürich, in the laboratory of Professor V. PRELOG⁵⁻⁷, provide an excellent series, from which reference values have been obtained illustrating the semi-quantitative application of the Octant Rule.

The amplitudes obtained in this work, together with some values previously obtained by DJERASSI et al.⁸⁻¹⁰, are collected in the Table. All results are expressed for the 9*R*-series for the sake of ready comparison (although some were actually measured with the 9*S*-compounds).

The conclusions may be briefly presented as follows: (i) The differences Δa (II-I) and Δa (III-I) represent the contribution to the Cotton effect of hydroxyl groups in a symmetry plane (at C-4). They are, as expected, negligible. (ii) The differences Δa (IV-I) and Δa (V-I) relate to hydroxyl groups in a negative octant. They are, as expected, negative. (iii) The difference Δa (VII-VI) is Δa for an axial methyl group in the 2-position relative to a cyclohexanone carbonyl. The value (+69) agrees well with

¹ Paper V of the series 'Optical Rotatory Dispersion'. For Paper IV see C. DJERASSI and W. KLYNE, Proc. Nat. Acad. Sci. Washington, in press (1962).

² W. MOFFITT, R. B. WOODWARD, A. MOSCOWITZ, W. KLYNE, and C. DJERASSI, J. Amer. chem. Soc. 83, 4013 (1961).

³ C. DJERASSI and W. KLYNE, J. chem. Soc. 1962, in press.

⁴ Amplitude (*a*) = 10^{−2} (Molecular rotation at *extremum* (peak or trough) of longer wavelength *minus* molecular rotation at *extremum* of shorter wavelength). The measurements were made, in methanol solution, on a Rudolph spectropolarimeter generously provided by the Wellcome Trust.

⁵ P. BAUMANN and V. PRELOG, Helv. chim. Acta 41, 2362 (1958).

⁶ P. BAUMANN and V. PRELOG, Helv. chim. Acta 41, 2379 (1958).

⁷ V. PRELOG et al., Helv. chim. Acta 39, 748 (1956); 41, 1416, 1424, 1428, 2396 (1958); 42, 736, 1239, 1862, 2624 (1959).

⁸ C. DJERASSI and J. STAUNTON, J. Amer. chem. Soc. 83, 736 (1961).

⁹ C. DJERASSI, R. RINIKER, and B. RINIKER, J. Amer. chem. Soc. 78, 6362 (1956).

¹⁰ C. DJERASSI and D. MARSHALL, J. Amer. chem. Soc. 80, 3986 (1958).