Effects of L-DOPA Treatment on Indole Metabolism in Parkinson's Disease

Large doses of L-dopa are used in the treatment of Parkinson's disease for the relief of akinesa, rigor and tremor[1]. This report describes effects of this treatment on urinary indoles.

Methods. 24 h urinary excretion of 5-hydroxyindole-acetic acid (5-HIAA), total indole-3-acetic acid (total IAA) and tryptamine were determined by quantitative methods[2-4] in parkinsonian patients on 2 to 3 successive days before (1st period) and at least 4 weeks after onset of L-dopa therapy (2nd period). In all patients the initial dose of L-dopa was 0.5 g/day and was increased to 3 g/day within 4 weeks. At the time of the 2nd investigation period in this group of patients the daily L-dopa dosis varied from 3 to 4.5 g/day. In both investigation periods the patients received 1-bicyclo-heptenyl-1-phenyl-3-piperidino-propanol-(1) (Akineton®, 3 × 2 mg/day). In addition to the chemical analyses described above 5-hydroxyindole-acetic acid was determined by thin layer chromatography in 2 patients using chloroform, methanol, acetic acid (75/20/5) as solvent and Ehrlich's reagent as spray.

Results. The results are summarized in the Table. They show a significant reduction in the urinary excretion of 5-hydroxyindole-acetic acid during L-dopa administration, while at the same time urinary total indole-3-acetic acid and tryptamine were not affected by the treatment ($p > 0.2$). Thin layer chromatography confirmed the decrease of 5-hydroxy-acetic acid during L-dopa administration.

Discussion. EVERETT and BORCHERDING[5] investigated the effects of L-dopa on brain amines in mice and reported a remarkable decrease of serotonin 30 min after i.p. injection of the drug. Because of a simultaneous increase of brain 5-hydroxyindole-acetic acid, these authors interpreted the results in terms of an increase in the release and metabolism of serotonin due to displacement by dopamine. Our findings of decreased urinary excretions of 5-hydroxyindole-acetic acid during longterm L-dopa treatment indicate that release and displacement of serotonin from its storage sites apparently do not represent the only effects of L-dopa on serotonin metabolism, but additional mechanismus must be considered. The unchanged urinary excretions of total indole-3-acetic acid and tryptamine suggest that absorption of tryptophan is not impaired. In addition, the observed normal excretions of urinary tryptamine give no evidence of decreased decarboxylase or monoamine oxidase activities. Although the biochemical mechanisms responsible for the decreased excretions of 5-hydroxyindole-acetic acid during L-dopa treatment are not clear as yet, it might be speculated that they are due either to decreased activity of tryptophan-5-hydroxylase or to increased activity of tryptophan pyrrolase diverting tryptophan metabolism away from the 5-hydroxytryptophan pathway.

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Investigation-periods	5 HIAA (mg/day)	Total IAA (mg/day)	Tryptamine (μg/day)
1. Control-period	6.4 ± 2.13 (20)	6.5 ± 1.83 (23)	64 ± 39.2 (15)
2. L-dopa (3–4.5 g/day)	1.7 ± 1.19[a] (18)	7.1 ± 2.92 (18)	81 ± 30.2 (10)

24 h urinary excretions of 5 HIAA and Total IAA were determined in 8 patients, tryptamine in 5 patients. Total number of 24 h urinary samples analyzed are shown in parentheses. [a] Different from control $p < 0.005$.

Zusammenfassung. Orale L-Dopa-Behandlung bewirkt bei Patienten mit Parkinsonismus eine signifikante Minderung der Harnausscheidungen von 5-Hydroxyindolessigsäure.

G. G. BRUNE and K.-W. PFLUGHAUPT

Neurologische Universitätsklinik und -Poliklinik, Josef-Schneider-Strasse 2, D-87 Würzburg (Germany), 4 December 1970.

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Length Measurement of Gut Segments for Mucosal Transport Studies

In studies on absorption or transport functions of gut segments in vivo and in vitro it is necessary to standardize transport to some reference quantity of the gut segment in order to compare results in several pieces of gut. Ideally the reference quantity should be some direct measure of the mucosal layer itself and length measurement of gut segments is the simplest direct estimate which can be made although it is apparently often considered too imprecise to be useful. Various workers have used protein content or wet or dry tissue weight as reference quantities in calcium transport studies but some difficulties with these have been noted[1] and it has been reported[2] that standardization according to simple length measurements can, under some conditions at least, give more meaningful results. In this laboratory calcium transport in rat gut segments has been referred to length measurements done

under conditions in which the gut appears relatively quiescent viz: In vivo, in animals under deep ether anaesthesia, when the gut appears to be relaxed; and in vitro, in buffer at 25 °C when the gut appears partially contracted. The following experiment was done to determine the variability and relation of lengths measured in vivo and in vitro by comparing these to the lengths of the same segments when relaxed completely by inhibitors.

Methods. 20 male Sprague-Dawley rats (175–200 g) were anaesthetized with an O_2 : ether vapour mixture to deep surgical anaesthesia and the abdomens opened. By means of a piece of flexible, fine, rubber tubing, 8.0 cm lengths of the duodenum, proximal jejunum, middle of the small gut, distal ileum and colon were measured off and excised. The gut contents were flushed out and the mesentery was dissected away. The segments were put into Krebs-Henseleit buffer[3] containing 0.3% glucose and gassed with O_2:CO_2 (95:5), at 25 °C. After 5 min the lengths of the gut segments were measured in the buffer while stretching them just sufficiently to straighten out any curves. The buffer was then warmed to 37 °C and after 10 min the lengths were taken again. The segments were then relaxed by adding to the buffer firstly a mixture containing the following inhibitors: Methysergide 20 µg, Hyoscine 10 µg, Promethazine 10 µg, Hexamethonium 2 mg and Lignocaine 200 µg; and secondly, after 10 min, EDTA pH 7.4, 5 m*M* (all final concentrations/l). After a further 10 min the gut segments were measured again. The ratios of the lengths of each segment under the first 3 conditions to the relaxed length were individually worked out and the geometric means and S.D. for the various gut regions and for each experimental condition were determined from these.

Ratios of gut lengths measured in vivo and in vitro to relaxed lengths

	In vivo length (= 8.0 cm) Relaxed length	25 °C length Relaxed length	37 °C length Relaxed length
Duodenum	0.97 ± 0.02	0.78 ± 0.02	0.72 ± 0.06
Jejunum	0.93 ± 0.04	0.81 ± 0.02	0.74 ± 0.05
Mid gut	0.92 ± 0.03	0.79 ± 0.04	0.80 ± 0.09
Ileum	0.91 ± 0.04	0.79 ± 0.03	0.84 ± 0.07
Colon	0.98 ± 0.02	0.82 ± 0.04	0.68 ± 0.07
Overall ratios	0.94 ± 0.03	0.80 ± 0.03	0.75 ± 0.06

Values for each gut region are geometric means ± S.D. for segments from 20 rats.

The results shown in the Table indicate that as far as the relaxed lengths may be considered as an absolute measure of gut lengths then the lengths measured at 25 °C were fairly constantly about 0.80 or 80% of this length, and the in vivo lengths (i.e. 8.0 cm), though varying slightly more between gut regions, were about 0.94 or 94% of the relaxed lengths. Length measurement under one or other of these conditions would appear to be constant enough for standardization in most transport studies. The length variation of gut segments in vitro at 37 °C however was much greater and suggests that such measurements would be less useful[4].

Résume. La longueur des segments d'intestin de rat, mesurée in vivo sous une anesthésie profonde à l'ether et in vitro, dans une solution tampon à 25 °C, atteint environ le 94% et le 80% de la longueur des segments totalement relâchés. L'une ou l'autre de ces mesures semble assez constante pour être employée dans la standardisation des méthodes en usage dans la plupart des études sur le transport mucosal.

D. D'A. Webling

Biochemistry Department, University of Western Ontario, London (Ontario, Canada), 19 November 1970.

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A Histochemical Study of the Pectoralis Muscle of the South Indian Flying Lizard, *Draco dussumieri*

The vertebrate skeletal muscle has been the material for careful investigation by several workers. These muscles in different vertebrates, apart from having morphological differences, vary also in the physiological and biochemical characteristics. They are often referred to as the 'red' and 'white' muscles. The existence of a mixed type of muscle composed of narrow, red and broad, white fibres is well-known among vertebrates. These 2 morphologically different fibres vary in their metabolite utilization as well. George et al.[1] attributed the difference in muscle fibres to their functional adaptation at the molecular level. Grinyer and George[2] suggested that the red fibres are 'slow twitch' fibres and the white 'fast twitch' fibres. The histochemical studies on the pectoralis muscle of the South Indian Flying Lizard, *Draco dussumieri*, revealed a number of interesting peculiarities and the results of that study are reported in this short communication.

The pectoralis muscle of *Draco* is a mixed muscle consisting of narrow, intermediate and broad fibres. Diameter ranges from 35–45 µm in the narrow, 50–65 µm in the intermediate and 75–100 µm in the broad fibres. There is a small band of narrow fibres towards the centre of the muscle, surrounded by broad and intermediate ones. Some narrow fibres are found scattered at random towards the periphery. The number of narrow fibres is considerably less per unit area.

Since myoglobin content was very poor, distinction of fibres into red and white was difficult. The central narrow fibres possessed lesser amount of fat than the

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