Results and discussion. Autopsy revealed hypertrophy of the adrenals in the treated animals. Microscopic examination indicated hypertrophic changes in the cells of both the cortical and the medullary regions of the adrenals of the treated animals; in addition, the cells of the zona fasciculata of the treated groups were distorted with hyalinated cytoplasm, the nuclei being in a condition of pyknosis; the nuclei of the cells of the medulla were vesicular, enlarged and less stainable (figures 1-3).

Cytometrical study indicated that in group C receiving 20 mg CS/kg, there was a highly significant rise in the cell nuclear diameter and cell diameter of both the cortex and the medulla, whereas in group B receiving 10 mg CS/kg, the same effect was observed only in the cells of the zona glomerulosa, zona fasciculata and the medulla. As regards zonal cellular width, while a highly significant fall was found in cells of the zona glomerulosa, zona fasciculata and zona reticularis in group B, the same effect was observed in group C only in cells of the zona fasciculata; a consistent feature was the sharp rise in the cellular width in the medulla of both groups B and C.

It is well known that the biochemical response of the adrenals to any stress is the increased secretion of catecholamines and glucocorticoids. Histological changes of the adrenals under both acute and chronic stress have also been recorded[8-10]. In the present investigation, the very sharp rise of the cell nuclear diameter, cell diameter and cellular width in the medulla recorded under the action of CS may be closely associated with hypersecretion of the catecholamines. The increase in the cell nuclear diameter and cell diameter of the cortex also may be connected with increased secretion of glucocorticoids; however, the consistent fall in the cellular width in the cortex is difficult to explain at this stage[11].

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A preliminary study on the behaviour and biochemical responses subsequent to the injection of 5,6-dihydroxytryptamine into the substantia nigra of the rat

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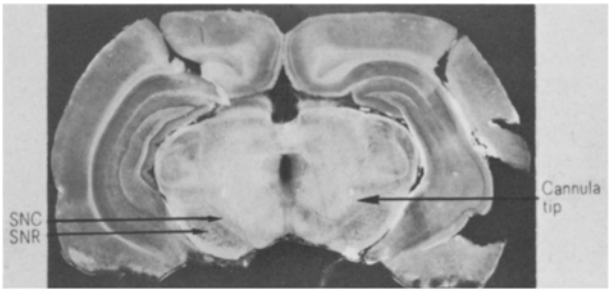
Summary. Injection of 5,7-dihydroxytryptamine into the substantia nigra of rats produces an increase of dopamine in the ipsilateral striatum, and when these animals are injected with amphetamine they do not exhibit any rotation. The mode of action of this neurotoxin is compared with that of 6-hydroxydopamine.

Recent studies suggest that ascending 5-hydroxytryptamine (5-HT) neurones may modulate the output of the nigrostriatal dopamine (DA) mechanism. The use of the rotating rat model[2] demonstrated that circling responses to both direct and indirect DA agonists are potentiated by drugs attenuating whole brain 5-HT transmission, while treatment enhancing 5-HT transmission decreases such rotations[3]. It is possible, therefore, that 5-HT neurones directly influence the DA neurones in the substantia nigra (SN), which would mean that neurones in the striatonigral and nigrostriatal loop are all directly or indirectly affected by the 5-HT neurone endings in the SN. To test this idea, the neurotoxins 5,7-dihydroxytryptamine (5,7-DHT) and 6-hydroxydopamine (6-OHDA) were injected unilaterally into the rat's SN and the animals observed for evidence of rotational locomotor activity characteristic of nigrostriatal activity[4]. The DA concentration and choline acetyltransferase (ChAc) activity in the corresponding corpora striata were determined as a biochemical measure of DA and acetylcholine (ACh) cell activity. It is known that 6-OHDA specifically degenerates catecholamine neurones[5], while 5,7-DHT specifically degenerates 5-HT and noradrenaline (NA) neurones[6].

Methods. Wistar albino rats, 200-260 g, were anaesthetized with nembutal. Either 5,7-DHT or 6-OHDA at a concentration of 0.016 M (in 1% ascorbic acid) was injected stereotaxically in a volume of 2 µl Hamilton syringe. The left SN received an equivalent volume of physiological saline containing 1% ascorbic acid. On recovery, the animals were placed immediately and 14 h later in rectangular grill boxes (20 × 20 × 20 cm), and observed for rotating and general behaviour; 2 weeks after the operations, the animals were injected i.p. with amphetamine (5 mg/kg) and placed once again in the boxes to observe their rotations and behaviour. The animals were then killed 3 weeks after chemical lesion with 6-OHDA or 5,7-DHT and the striata carefully dissected and assayed radiochemically for DA[8]



Frontal section of rat brain. SNC, substantia nigra pars compacta; SNR, substantia nigra pars reticularis.

Dopamine (DA) content and choline acetyltransferase (ChAc) activity in both striata of rats (means ± SEM) 3 weeks after the injection into the right substantia nigra of 2 µl/0.016 M of 6-hydroxydopamine (6-OHDA) or 5,7-dihydroxytryptamine (5,7-DHT) in 1% ascorbic acid. The contralateral substantia nigra received 2 µl of physiological saline containing ascorbic acid.

	n	DA content (µg/g wet wt)			ChAc activity (µmoles/g wet wt/30 min)		
		Ipsilateral	Contralateral	Δ% Ipsilateral to contralateral	Ipsilateral	Contralateral	Δ% Ipsilateral to contralateral
Control (intact)	4	5.44 ± 0.38 (100%)	5.92 ± 0.50 (100%)	+9%	9.80 ± 1.56 (100%)	9.44 ± 1.90 (100%)	−4%
6-OHDA	4	1.59 ± 0.22 (−71%)*	7.17 ± 0.25 (+21%)*	−78%	13.37 ± 1.91 (+37%)	10.50 ± 1.82 (+11%)	+27%**
5,7-DHT	4	7.90 ± 0.72 (+45%)*	5.82 ± 0.31 (−2%)	+36%**	11.14 ± 0.93 (+14%)	11.18 ± 1.20 (+18%)	−0.4%

* Difference between 6-OHDA or 5,7-DHT and control (100%) values (non-paired t-test); significant at $p \ll 0.05$. ** Difference between corresponding ipsi- and contralateral striata (paired t-test); significant at $p \ll 0.05$.

and ChAc[3]. The sites of injections were determined histologically using the remainder of the brains.

Results and discussion. After the operations, asymmetric body posture and occasional spontaneous slow circling movements towards the lesioned side were observed in both groups of rats. However, 14 h after the lesions the animals exhibited a clear-cut asymmetric behaviour and slow circling movements towards the opposite non-lesioned side. They were also hypoactive and the 'Straub tail' phenomenon was observed. There were no observable differences at this stage between the 6-OHDA lesioned or 5,7-DHT lesioned rats; 2 weeks after the operations and just before the injection of amphetamine no observable behavioural differences could be detected between operated and control animals. However, following amphetamine treatment the operated rats exhibited an exaggerated sniffing and also demonstrated episodic stereotyped licking and gnawing. Moreover 3 out of the 4 6-OHDA lesioned rats circled towards the lesioned side (peak frequency of 360° turns, 5–10 turns per min), whereas the 5,7-DHT lesioned or control rats did not. Between 20 and 40 min after amphetamine injection, the control and 6-OHDA treated animals defecated fairly strongly and this was not observed in the 5,7-DHT lesioned rats.

The ChAc and DA distribution in the left and right striata from rats stereotaxically injected with 6-OHDA or 5,7-DHT in their right SN is shown in the table. Histological analyses of the SN from all rats showed that the pars compacta was the injection site (cannula tip, see figure). Although the distribution of solution injected into the SN was not studied, it has been reported that substances injected into different brain regions remained strictly localized in a sphere with a diameter of less than 2 mm[10–12], and the area of unspecific necrotic lesion after 6-OHDA injection is even smaller[5]. Although there was a considerable variation in the striatal concentrations of DA (range 5–9 µg/g wet wt) and ChAc (range 8–16 µmoles/g wet wt/30 min), bilateral differences in striatal contents or activity of these substances in control animals were less than 10%. The table clearly shows that the 6-OHDA induced ipsilateral turning after amphetamine treatment was accompanied by a pronounced striatal DA decrease in the ipsilateral lesioned side, and a slight though significant increase in the contralateral DA content and an increase in the ipsilateral ChAc activity. In contrast, injection of 5,7-DHT into the right SN caused an increase in DA content in the ipsilateral striata and no change in the ChAc activity.

The present results reveal that there is a variation between 6-OHDA and 5,7-DHT in their mode of action at the level of SN. 6-OHDA destroys the dopaminergic nigro-striatal pathway (NSP) and causes a drop in ipsilateral striatal DA content together with a rise in ChAc activity on the same side. These changes were accompanied by a slight increase in the DA content in the opposite striatum. This may be interpreted as a compensatory effect, as it is known that a multitude of links exist between bilateral extrapyramidal nuclei (Wolfahrt[13]). The rise in the ChAc activity in the striatum ipsilateral to the degenerated NSP supports the idea that NSP has an inhibitory influence on the cholinergic interneurones within the striatum[14,15]. This is in agreement with the 'nigro-basal ganglia-nigral loop' theory[16]. The injection of 5,7-DHT into SN could theoretically provoke the destruction of both 5-HT and NA neurones, because desimipramine was not used to protect the latter[17]. However, NA neurones do not seem to play a role in either the SN or striatum, whereas 5-HT neurones are known to project from the raphe nuclei to both the striatum and SN and impinge on the striatal neurones and NSP cells[18,19]. The marked rise in the ipsilateral DA content within the neostriatum after the destruction of nigral 5-HT terminals with 5,7-DHT injection thus strongly supports the idea of an inhibitory role for these terminals on the dopaminergic neurones. A satisfactory explanation cannot be given for the observed enhancement of amphetamine stereotype in both 6-OHDA and 5,7-DHT lesioned animals, although the possibility that the slight augmentation of striatal ChAc activity seen in both striata after the lesions may be responsible for this effect cannot be excluded.

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