For the detection of the 4-hydroxyphenazone, the extracts were converted to trimethylsilyl ethers before injection on the column, since we were unable to obtain satisfactory results with the free compound. The retention times of the compounds relative to phenazone ($t_R = 1{\cdot}0$) using the same g.l.c. conditions as above, except N_2 at 20 ml min^{-1} was the carrier gas, were: norphenazone 0·32, trimethylsilyl 4-hydroxyphenazone 0·9. The retention time of phenazone under these conditions was 9·6 min. Approximately 6% of the total dose of phenazone was excreted as norphenazone in the 24 h following ingestion of the drug. [In subjects taking phenazone after a course of phenobarbitone, the amount of norphenazone produced was not affected even though there was a considerable increase in the amount of the 4-hydroxyphenazone excreted.]

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Pharmacogenetics Division.
Department of Medicine,
University of Liverpool,
Liverpool, L69 3BX, U.K.
September 5, 1972.

J. D. BATY
D. A. PRICE EVANS

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Effects of intracerebral injections of 6-hydroxydopamine on sleep and waking in the rat

Monoamines in the central nervous system are considered to play an important role in the onset and maintenance of the different forms of sleep (Jouvet, 1969). Noradrenergic neurons are implicated not only in the induction of paradoxical sleep (PS) (Roussel, 1967) but have also received attention of being possible mediators of waking (Jones, Bobillier & Jouvet, 1969; Jones, 1969, 1972). Suppression of PS occurred (Jouvet, 1967) after destruction of the noradrenaline-containing neurons in the locus coeruleus (Dahlström & Fuxe, 1964). The ascending axons of these neurons probably branch and give off collaterals to the cerebellar cortex, reticular formation, the colliculi and the thalamus on their way to the cortical areas of the forebrain (Loizou, 1969; Ungerstedt, 1971a; Olson & Fuxe, 1971).

Recent studies indicate, that sedative drugs like barbiturates and benzodiazepine derivatives decrease the turnover of cortical noradrenaline (Taylor & Laverty, 1969; Corrodi, Fuxe & others, 1971; Lidbrink, Corrodi & others, 1972a), suggesting a way by which these drugs may partly exert hypnotic as well as PS suppressant effects. We have now followed sleep and waking in the rat after intracerebral injections of 6-hydroxydopamine (6-OH-DA) (Ungerstedt, 1968; 1971b) in the mesencephalon causing small and selective lesions of the so called dorsal noradrenaline pathway which contains the axons from the locus coeruleus (Ungerstedt, 1971a; see also Fuxe, Hökfelt & Ungerstedt, 1970).

Male Sprague-Dawley rats (150 g) were operated in a stereotaxic instrument under halothane-N_2O–O_2 anaesthesia. 6-OH-DA dihydrobromide (Regis) dissolved in saline containing ascorbic acid (0·2 mg ml^{-1}) or the vehicle alone was injected bilaterally into the caudal mesencephalon in the vicinity of the dorsal noradrenaline pathway (the level according to König & Klippel (1963): anterior: 0·6 mm; lateral = 1·3 mm; ventral: 1·0, 1·3 or 1·5 mm. The amount of 6-OH-DA was 8 μg in 4 μl (for details, see Ungerstedt, 1971a). Small holes were drilled in the skull above the frontal and parietal cortex, where 4 female connecting pins and 3 screws were fitted. Two electrodes were placed in the neck muscle and connected to two female pins held in a position behind the cortical electrodes. The set-up was fixed to the skull with dental cement. The operation lasted 1 h. The recording of the eeg and emg started at 4 p.m. on the same day and was continued during the following 8 days. The time course after such 6-OH-DA induced lesions on noradrenaline levels and degeneration processes of the cortical noradrenaline nerve terminals has been followed by Lidbrink, Jonsson & Fuxe (1972b). The eeg records were distinguished according to Michel, Klein & others, (1961) and Jones (1969) into 4 different stages of activity: waking (W), cortical low voltage fast activity and a high muscle tone; slow wave sleep (SWS) 1, low voltage fast activity interrupted by high amplitude slow waves and a moderate to high emg activity; SWS 2, continued high voltage slow waves and a marked decrease of muscle tone; PS, a waking eeg with a complete disappearance of emg activity except for some twitches. Each minute of the record was scored as belonging to one of these four stages. The amount of time spent in each stage was then calculated as % of total (8 days) recording time.

The animals injected with 6-OH-DA spent less time in W compared to the saline-treated controls during the entire period (Table 1). Instead there was a significant increase in SWS 1 and a small increase in SWS 2. PS was not significantly altered by 6-OH-DA. When analysing the effects day by day it was noted that in the 6-OH-DA-treated animal, W increased somewhat for each day compared to the controls—indicating that the effects were transient.

Injections of 6-OH-DA close to the dorsal noradrenaline pathway in the dorsomedial tegmentum in the caudal mesencephalon causes the noradrenaline nerve terminals of the cerebral cortex to disappear (Ungerstedt, 1971a). The immediate effect is probably an arrest of the flow of nervous impulses, since the depletion of cortical noradrenaline seen after α-methyltyrosine is greatly reduced (Lidbrink, Jonsson & Fuxe, unpublished). The lesion also leads to a transient increase in noradrenaline in the nerve terminals, probably due to continued synthesis and a diminished release (Lidbrink, Jonsson & Fuxe, 1972b). The unspecific damage to the brain tissue produced by 6-OH-DA consists of a small round area about 0·3 mm in

Table 1. *The effect of 6-OH-DA injected into the dorsal ascending noradrenaline pathway on sleep and waking in the rat.* 6-OH-DA (8 μg 4 μl^{-1}) or the vehicle was injected bilaterally into the dorsomedial tegmentum in the caudal mesencephalon. The eeg recording started at 4 p.m. on the same day and continued through 8 days. The values for sleep and waking are expressed as percent of total time.

Treatment	n	W*	SWS 1*	SWS 2*	PS*
Saline	5	33·2 ± 0·3	13·5 ± 0·9	43·2 ± 0·9	10·1 ± 0·2
6-OH-DA	7	25·8 ± 0·9†	19·0 ± 0·4†	45·9 ± 0·9	9·3 ± 0·6

* For explanation of symbols see text.
† $P < 0·001$ (Student's *t*-test)

diameter with degenerating cells and autofluorescent pigments close to or within the dorsal pathway (Ungerstedt, 1971b; Lidbrink & others unpublished). This is observed only during a limited period, and a month after the lesion no pathological changes can be seen in the fluorescence microscope.

The changes in waking and sleep found here after this type of lesion, suggest that ascending noradrenaline neurons participate in the regulation of the sleep-wakefulness cycle. The decreased W observed is in accordance with the hypothesis of Jones, Bobillier & Jouvet, (1969) (see also Jones, 1969; 1972) that noradrenaline is important for maintenance of cortical arousal. Furthermore, drugs, which elevate the noradrenaline levels, e.g. dopa (Delorme, 1966) or facilitate noradrenaline neurotransmission, e.g. amphetamine (Kornetsky, Mirsky & others, 1959) produce increased arousal, whereas drugs which lower the noradrenaline levels, e.g. α-methyl-*p*-tyrosine (Peyrethon-Dusan, 1968; Feist, 1970) or decrease noradrenaline neurotransmission, e.g. phenothiazines, (Longo, 1962) suppress W.

When lesions in the pons involving the locus coeruleus are performed, both the tonic and phasic activities of PS diminish (Rossi, Favale & others, 1961; Jouvet, 1962; 1967; Roussel, 1967; Buguet, 1969). In the present paper transection of the ascending axons from this nucleus with 6-OH-DA was found not to decrease the amount of time spent in PS, suggesting that the noradrenaline nerve terminals cranial to the lesion are not involved in the genesis of cortical activation during PS. A lesion of this type has been shown not to produce a retrograde degeneration of the cellbodies, probably because the axons diverge to the cerebellum and reticular formation caudally to the lesion (Olson & Fuxe, 1971; Ungerstedt, 1971a).

The technique described here, of using compounds like 6-OH-DA intracerebrally, which rather selectively destroy one of the monoamine systems may prove beneficial in resolving some of the problems in the monoamine theory of sleep. The present results indicate that maintenance of W is dependant upon the "coerulo-cortical" noradrenaline pathway, since its destruction results in decreases of W and increases in SWS.

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Department of Histology
Karolinska Institutet,
104 01 Stockholm, Sweden.

PETER LIDBRINK
KJELL FUXE

September 29, 1972

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Lorazepam on visuo-motor co-ordination and visual function in man

In any study of centrally acting drugs in man involving a response to a visual stimulus, the possibility of a drug effect on the peripheral visual apparatus must be considered. This is especially true for benzodiazepine drugs. Miller (1962) reported that chlordiazepoxide in doses of 20 mg daily produced significant exophoria and reduced visual acuity. Hedges, Turner & Harry (1971) showed that there was a significant dose-related reduction in critical flicker frequency, disc-dotting and reaction times by the benzodiazepine drug, lorazepam, in doses of 0·5, 1·0 and 2·0 mg, the maximum effect being seen at 4 or 6 h.

We have examined the effect of lorazepam in normal volunteers to determine if a dose sufficient to produce significant impairment of hand-eye co-ordination (a recognized test in the evaluation of centrally acting drugs—Molson, Mackey & others, 1966; Large, Wayte & Turner, 1971), was associated with change in tests of visual function.

Six healthy volunteers (aged 19–21 years) with normal colour vision and visual acuities of 6/4·5 or better in both eyes, and in good health, who were receiving no other medication, were given lorazepam 1·0 and 2·0 mg and a placebo in tablet form in random order based on two latin-square designs, under double-blind conditions, each treatment being separated by at least one week. The investigations were made at the same time in the afternoon after a standard light lunch. Subjects avoided coffee, tea, alcohol and nicotine on the test days. Tests were made before and at $1\frac{1}{2}$ and 3 h after the treatment. The subjects had been familiarized with the procedures before the investigation. In the hand-eye co-ordination test they had reached a plateau of performance to minimize further learning effects.

The tests were of (a) refraction (b) visual acuity (c) amplitude of accommodation (d) oculomotor balance (e) visual fields (f) hand-eye co-ordination, and were made