afforded (R)-(−)-1-methylpropylamine, the absolute configuration of which has been established by several workers[9,10]. Thus, (+)-methyl-2-propynylamine can be ascribed the R configuration.

Zusammenfassung. Die optischen Antipoden der 3 sehr wirksamen Oxotremorinantagonisten, N-(1-Methyl-4-pyrrolidino-2-butinyl)pyrrolidon, N-(1-Methyl-4-pyrrolidino-2-butinyl)succinimid und N-(1-Methyl-4-perhydroazepino-2-butinyl)succinimid, sind hergestellt und auf ihre pharmakologische Aktivität geprüft worden: Die (+)-Antipoden, die R-Konfiguration haben, wurden als Träger der Aktivität erkannt, während die (−)-Antipoden praktisch unwirksam sind.

R. Dahlbom, Å. Lindquist, S. Lindgren, U. Svensson, B. Ringdahl and M. R. Blair jr.

Department of Organic Chemistry, Faculty of Pharmacy, University of Uppsala, Box 574, S-751 23 Uppsala (Sweden), and Research Laboratories, Astra Pharmaceutical Products Inc. Worcester (Massachusetts 01606, USA), 5 June 1974.

Increase in Monoamine Concentration in Rat Brain Following Melatonin Administration

Melatonin is a hormone which is secreted from the pineal gland of mammals in relation to illumination[1]. Previously, Anton-Tay, et al.[2] have shown that melatonin administered i.p. significantly increases brain serotonin concentration, but this change was not associated with changes in norepinephrine content. We decided to investigate the possibility that melatonin may alter brain norepinephrine and dopamine concentrations when administered by methods where sufficient brain levels of melatonin could be achieved prior to its metabolism to 6-hydroxymelatonin by the liver[3]. To accomplish this, melatonin was injected into the common carotid artery or the cisterna magna of rats.

Materials and methods. Male Sprague-Dawley rats (150–200 g) maintained on a 12-h light-dark cycle, 3 to a cage, with free access to Purina Rat Chow and water, were used in these experiments.

Rats to be injected intraarterially were anesthetized with sodium pentobarbital (60 mg/kg, i.p.). A ventral midline incision of approximately 1 cm was made, and the left common carotid artery was exposed. A cannula was inserted into the artery toward the brain, secured with sutures and the incision closed. Following a 24-h recovery period, rats were injected with either melatonin in saline or saline only. All injections were made between the hours of 10.00 and 12.00.

Rats to be injected intracisternally by the method of Schanberg et al.[4], were lightly anesthetized with ether and given specified amounts of melatonin (Sigma Chemical Company, St. Louis, Mo.) dissolved in 10 µl of distilled water. Control rats were injected with 10 µl of water only.

Rats were decapitated at the time specified after injection. Brains were removed and immediately frozen, halved through the mid-saggital plane, homogenized in 0.4 *N* perchloric acid containing disodium ethylenediaminetetraacetate and norepinephrine and dopamine content determined by the method of Shellenberger and Gordon[5].

Results and discussion. Cotzias et al.[6] found the i.p. injection of melatonin (400 mg/kg) in mice did not result in significant alterations in brain dopamine content. Anton-Tay et al.[2] reported that the i.p. administration of melatonin (2.8 mg/kg) did not alter rat brain norepinephrine concentrations. However, both of the above investigators did report a melatonin induced increase in brain serotonin levels[2,6].

In our experiments, melatonin was given by intraarterial injection and intracisternal injection; both routes of administration led to significant increases in rat brain dopamine and norepinephrine content. The intraarterial

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Effect of melatonin on rat brain catecholamines when administered by intraaterial or intracisternal injection[a]

		Dopamine (µg/g)	Norepinephrine (µg/g)
Intraarterial (controls)	T = 0[b]	0.71 ± 0.03	0.49 ± 0.01
	T = 1 h[c]	0.65 ± 0.03	0.52 ± 0.03
Intraarterial melatonin (250 µg/kg)	T = 0	0.75 ± 0.02	0.45 ± 0.03
	T = 1 h	1.40 ± 0.09[d]	0.68 ± 0.03[d]
Intracisternal (controls)	T = 0	0.64 ± 0.03	0.39 ± 0.01
	T = 1 h	0.76 ± 0.04	0.46 ± 0.03
Intracisternal melatonin (40 µg/kg)	T = 0	0.67 ± 0.08	0.45 ± 0.02
	T = 1 h	1.02 ± 0.06[d]	0.67 ± 0.03[d]

[a] $n = 4$ rats/group. [b] T = 0 are rats sacrificed immediately following injection. [c] T = 1 h are rats sacrificed 1 h following injection. [d] significantly different from appropriate controls as well as T = 0 melatonin treated rats ($p < 0.05$).

injection of melatonin (250 μg/kg) nearly doubled the dopamine content and significantly ($p < 0.05$) increased the norepinephrine concentration 1 h following its administration (Table). No significant changes in dopamine or norepinephrine content were observed when rats were injected with melatonin and sacrificed immediately.

In order to circumvent the blood-brain barrier, melatonin was given intracisternally[4]. Melatonin (40 μg/kg) given in this manner resulted in significant increases in both brain dopamine and norepinephrine (Table). The increase in dopamine and norepinephrine levels induced by melatonin given intracisternally was noted within 15 min after administration, achieved maximal levels between 30 min and 1 h, and returned to control levels approximately 2 h later. A slight, though insignificant increase in brain dopamine and norepinephrine content was noted 1 h after sham injection in control animals (Table). This is probably related to the ether anesthesia since the increase was not seen in control animals which were injected intraarterially in which ether was not used.

The intracisternal injection of 6-hydroxymelatonin (40 μg/kg), the main metabolite of melatonin did not result in significant changes in either dopamine or norepinephrine content one hour following its administration. The i.p. injection of melatonin is known to result in its rapid metabolism to 6-hydroxymelatonin[3]. Perhaps the inability of previous investigators[2,6] to show melatonin induced changes in brain catecholamine content may be due to its rapid conversion to 6-hydroxymelatonin which we find is inactive when given intracisternally.

In summary, our results demonstrate that melatonin given both intraarterially and intracisternally results in significant increases in both rat brain dopamine and norepinephrine concentration. Also, the intracisternal administration of comparable amounts of 6-hydroxymelatonin does not alter brain catecholamine content.

Zusammenfassung. Nachweis, dass Melatonin die Konzentration von Dopamin und Noradrenalin im Rattenhirn erhöht, wenn es intraarteriell oder intracisternal injiziert wird.

O. T. WENDEL, L. D. WATERBURY and L. A. PEARCE

Departments of Physiology and Neurology, Bowman Gray School of Medicine, Wake Forest University, Winston-Salem (North Carolina 27103, USA),
27 March 1974.

Tremorogenic Effect of Thyrotropin Releasing Hormone in Rats

In 1972 PRANGE et al.[1] reported on antidepressant activity of thyrotropin releasing hormone (TRH) in man. They assumed that the beneficial effect of TRH in depressive patients was not thyroid-mediated, but rather due to a direct central nervous stimulating activity[2]. These reports and interpretations have encouraged the study of the central nervous effects of TRH in experimental animals. PLOTNIKOFF's group[3] has described an L-DOPA-potentiating effect of TRH in pargyline pretreated mice. HINE et al.[4] have noted pharmacological effects of TRH in the 'conscious' dog which they suggested might be related to a general sympathetic activation by the hormone.

In the context of a study of the endocrinological activity of TRH, we have observed a number of effects which appeared immediately after the i.v. injection of the hormone. In additional experiments we attempted to define the mechanism and specificity of these effects.

Effects of TRH in the rat. Shortly after an i.v. injection of TRH (0.3 to 3.0 mg/kg) muscle tremor, excitation, tail lifting, and pilo-erection were observed in the non-anaesthetized rat. The tremorogenic effect only lasted a few minutes. In addition, immediately after the injection the amplitude of respiration increased and signs of augmented cutaneous blood flow appeared. In rats anaesthetized with pentobarbital (50 mg/kg i.p.). all these effects were more pronounced. The latency of onset, the amplitude and the duration of the tremor were dose-dependent (Table I and Figure 1a). After some min the tremor ceased, but it could still be elicited by touching the animal.

After the arousal phase, within 5 to 60 min after the injection, the animals returned to the inert sleeping position. The duration of anaesthesia was not shortened significantly. After waking, the animals appeared normal.

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Table I. Tremorogenic effect of TRH in rats. Latency and duration of tremor

TRH (mg/kg)	No. of rats reacting	Latent period until onset of tremor (sec)	Duration of spontaneous tremor (min)	Total time of spontaneous + elicitable tremor (min)
0.3	7/7	123 ± 12	3.4 ± 0.9	18.7 ± 4.9
1.0	5/5	54 ± 4	5.4 ± 0.5	16.4 ± 2.0
3.0	6/6	24 ± 1	21 ± 4	44 ± 8
		$\bar{X} \pm SE$		

Rats were anaesthetized with pentobarbital (50 mg/kg) by i.p. injection. TRH (pGlu-His-Pro-NH_2) was dissolved in 0.5 ml physiological saline and injected over exactly 2 min into the jugular vein. The onset of tremor was recorded in sec after beginning of the injection.