In conclusion, our results indicate that amphetamine gains access to the brain by a combination of 2 independent mechanisms: free diffusion and a carrier-mediated process. The amphetamine carrier is saturable, pH dependent, and participates in the uptake of methylphenidate and β-phenethylamine. The rapid onset of action of amphetamine after intravenous administration may reflect the simultaneous operation of these uptake mechanisms.

Zusammenfassung. Untersuchungen über die die Penetration von Amphetaminen ins Gehirn bestimmenden Faktoren. Entwicklung neuer Aspekte im Hinblick auf den Transport von Amphetaminen über die Blut-Hirn-Schranke mittels eines Trägers.

W. M. PARDRIDGE and J. D. CONNOR

Department of Pharmacology, College of Medicine, The Pennsylvania State University Milton S. Hershey Medical Center Hershey (Pennsylvania 17033, USA), 14 August 1972.

A Study of the Mechanism of Lowering the Sensitivity of Smooth Muscle to Noradrenalin with Sympathetic Hyperinnervation

Nerve growth factor (NGF) is known to induce the hypertrophy and hyperplasy of sympathetic and embryonal sensory cells[1–3]. The injection of NGF to new-born mice results in an increase in the amount and dimensions of the sympathetic ganglia neurons[2,3], a thickening of sympathetic trunks due to an increase of the fiber number, and also an augmentation of the density of sympathetic terminals in the effector organ[2,4,5]. As compared with the normal, the noradrenalin content of the peripheral organs is augmented in such animals[6]. Thus, by means of NGF, animals can be obtained in which organs are hyperinnervated by sympathetic terminals. An investigation of the intestine smooth muscle with sympathetic hyperinnervation revealed that the sensitivity of this muscle to noradrenalin is lowered as compared with the control[7]. It has been assumed that denervation may not only increase the sensitivity of the effector, according to the denervation law, but decrease at hyperinnervation[7].

Adrenergetic terminals are known for their capacity to uptake catecholamines from the environment. Therefore, some amount of the injected agent is uptaken by the terminals after noradrenalin injection and the noradrenalin concentration in the area of the smooth muscle membrane decreases. An augmented response of the smooth muscle to humoral stimulus may occur as a result of the destruction of sympathetic fibers, e.g. due to their degeneration, even if the sensitivity of the postsynaptic membrane has remained unchanged[8]. It is clear that the effect of lowering the sensitivity on account of the terminals will be more pronounced if the numbers of terminals on the periphery has increased. Against a cocaine background the terminals lose their capacity to take up-catecholamines[8–10]. Therefore, a comparative study of sensitivity in a normal and hyperinnervated smooth muscle in response to noradrenalin against a cocaine background permits us to determine whether the sensitivity of the effector organ cells to this agent differs or not.

Material and method. In the present work we used the NGF preparation from the Wellcome, Kent, England. The preparation was injected to young mice at a rate of 500 biological units per 1 g of body weight for 15 days, after which the animals were tested.

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Mean noradrenalin doses ($M \pm m$; in μg) causing standard responses of the smooth muscle in normal and hypersympathetized animals in intact preparations and after cocaine treatment

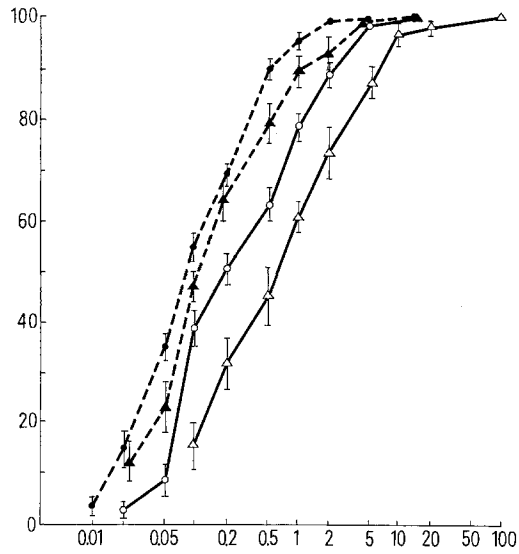
Response	Norm	Hypersympathetization	Cocaine	
			Norm	Hypersympathetization
Threshold	0.865 + 0.097 − 0.088 $n = 27$	1.693 + 0.349 − 0.288 $n = 14$ $p < 0.01$	0.322 + 0.038 − 0.034 $n = 25$	0.473 + 0.075 − 0.065 $n = 16$ $p < 0.05$
50% of the maximal	1.914 + 0.375 − 0.311 $n = 22$	5.009 + 1.122 − 0.915 $n = 12$ $p < 0.01$	0.726 + 0.095 − 0.084 $n = 18$	1.446 + 0.269 − 0.227 $n = 10$ $p < 0.01$
Maximal	43.29 + 5.37 − 4.78 $n = 28$	149.0 + 39.0 − 30.9 $n = 13$ $p < 0.001$	11.11 + 1.91 − 1.62 $n = 25$	40.90 + 11.67 − 9.08 $n = 15$ $p < 0.001$

A macroscopic study revealed a hypertrophy of sympathetic ganglia. The use of the impregnation autoradiography, and also the Nissle, Mazia and Brachet methods, revealed that the hypertrophy of the stellate ganglion of the sympathetic chain is associated with the hypertrophy of neurons with a manifested increase of their dimensions. These nerve cells show an increase of protein and RNA synthesis. Most of the neurons contain 2–4 nucleoli.

These data indicate that in our experiments NGF exhibited a well-known stimulating effect on the development of sympathetic system.

Two series of experiments were performed both on animals with a hypertrophied sympathetic system and normal animals with a view of studying the sensitivity of the smooth muscle to noradrenalin. The experimental technique was similar to that described above[7].

Results. The first series of experiments confirmed the earlier results obtained[7] concerning the lower sensitivity of the intestine smooth muscle to noradrenalin in animals with sympathetic hyperinnervation as compared with the normal. In this experimental series, we used 14 animals injected with NGF and 28 normal ones. The results are shown in Figure and in the Table. The Figure shows that the dose-effect curve for hypersympathetized animals is shifted to the right as compared with the control. In the Table medium values of the doses are given which cause a threshold response, the response comprising 50% of the maximal and the maximal response for both cases. In every case the differences are statistically significant. The second series of experiments performed in 16 experimental and 25 control animals was similar to the preceding one. The only differency was that to the Krebs solution, into which the preparation was put, cocaine was added at a concentration of 1×10^{-5}. A special series of experiments revealed that the treatment with the doses resulted in a maximal effect. A subsequent concentration increase failed to produce an augmentation of sensitivity to noradrenalin. The results of the experiments are presented in the Figure and in the Table.



The dose-effect curves for the intestine smooth muscle in normal (circles) animals and in animals with sympathetic hyperinnervation (triangles) before (white circles and triangles, continuous lines) and after (black circles and triangles, broken lines) cocaine treatment. Ordinate, the response value in % of the maximum; Abscissae, noradrenalin concentration (μg/ml, log. scale).

Discussion. As is clear from the obtained data, on cacaine treatment the curve for dose-effect in normal and hyperinnervated animals is shifted to the left, i.e. the sensitivity in both cases is augmented. However, after cacaine treatment the curve for hyperinnervated animals is situated more to the right than in the case of normal animals. The differences in the sensitivity of smooth muscles in normal and experimental animals after cocaine treatment have also been revealed by comparing the doses causing the threshold response, the response amounting to 50% of the maximal and the maximal response (Table). The data obtained give grounds to believe that in a state of hyperinnervation the sensitivity of the smooth intestine muscle to noradrenalin is lower than in the norm.

Since differences are also revealed upon cocaine treatment, when the sympathetic terminals lose the ability to take up catecholamines from the environment, there is therefore reason to believe that a decrease insensitivity is fore reason to believe that a decrease in sensitivity is associated not only with an excess of sympathetic terminals but is dependent on the properties of the postsynaptic membrane at hyperinnervation. According to the published data there are some indications that an increase in sensitivity following cocaine treatment is due not only to the inactivation of the effector terminals, but to its capability of increasing the sensitivity of the postsynaptic membrane [11,12]. However, even if cocaine reveals such property, this cannot affect the conclusions based upon our experimental material. Indeed, had the sensitivity of the smooth muscles proper been similar in a state of hyperinnervation and in norm we should not have been able to trace any differences arising after the action of cocaine. However, as has been shown, such differences do exist.

ВЫВОДЫ. При исследовании гладкой мышцы кишечника, гипериннервированной симпатическими волокнами, показано, что чувствительность ее к норадреналину ниже, чем у нормальной. Более низкая чувствительность наблюдается и на фоне действия кокаина. Предполагается, что при избытке симпатических окончаний в ткани чувствительность гладкомышечных клеток к медиатору понижена.

N. A. Kuzmicheva[13], I. M. Rodionov[13], O. V. Volkova[14] and M. Z. Chunaeva[14]

Department of Animal Physiology, Faculty of Biology and Pedology, Moskwa Lomonosov State University, Moskwa W–234 (USSR), and Department of Histology and Embryology, Faculty of Pediatrics, IInd Medical Institute, Moskwa (USSR), 14 June 1972.

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