of large amplitude) were of too long latencies to be taken into account. The recovery time of the response begins at 150 msec of ID, first in waking and rapid sleep. At 400 msec, the response attains 100% of the conditionant response amplitude in waking without theta (NA); the responses are still of lower amplitude in the states of slow sleep (SW, SP, IS) pointing to a longer recovery time for these states of sleep.

Discussion. From a general point of view, our results are in good agreement with those obtained in the cat by stimulation of the somesthetic radiations[2-6]. Nevertheless, there is no facilitation during R2 as observed in the cat by direct visual cortex stimulation[7]. This is probably linked to the absence of cortical phasic activities during rapid sleep in the rat[8]. The study of the 2nd evoked potential, also in agreement with the results obtained in the cat[2,9], shows a long-lasting cortical recovery process. The order of recovery (first in waking, then in rapid sleep and latter in slow sleep), unlike the 1st response, can be explained by the fact that the cortical inhibitory processes are as important as the *initial excitation is significant*[10]. Interesting processes probably linked to cortical[11,12] and sub-cortical[13] influences must occur to explain the curious evolution of the 2 evoked potentials and have to be studied first by antidromic stimulation of pyramidal tract and then by single unit approach.

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Endothelial injury by nicotine and its prevention[1]

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Summary. Nicotine administered i.v. or p.o. in doses above 0.0125 mg/kg to the rat caused a highly significant increase in circulating anuclear carcasses of endothelial cells estimated by an original method. This effect of nicotine was completely prevented by a prior oral administration of the flavonoids hydroxyethylrutosides (HR) or Mono-7-HR.

Smoking is generally accepted to be one of the main risk factors of coronary heart disease. Nevertheless, the mechanism of its damaging influence on blood vessels is far from clear. From this point of view, nicotine is the most conspicuous among the noxious tobacco products; but besides its vasoconstricting activity, no direct damaging effect has been shown conclusively.

A very sensitive experimental as well as clinical method has now been developed in our laboratory to demonstrate the endothelial injury[2]. The method is based on counting circulating anuclear carcasses of detached endothelial cells. In principle, 1 ml of platelet-rich plasma is mixed with 0.2 ml of 2.34 M adenosine-5'-diphosphate (Calbiochem) and mechanically shaken for 10 min. Plasma is centrifuged at 395×g for 20 min to remove platelet aggregates. The supernatant is centrifuged at 2100×g for 20 min and the sediment suspended in 0.1 ml physiological saline. From this suspension a Bürker's chamber is filled and the cells counted under phase contrast microscope in two ruled platforms corresponding to 0.9 µl each. The counting is repeated once more and the mean from 4 platform countings is taken as the result. The results are expressed in terms of the element count in the volume of 1 platform after the correction for dilution. The elements were identified mor-

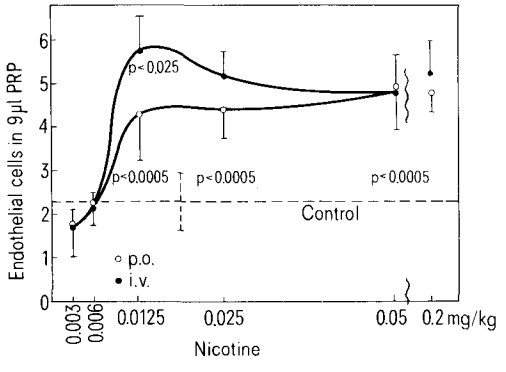

Fig. 1. The dose-dependence of the endothelial cell count increasing effect of nicotine in rats. Means (n = 10) ± SD are indicated. Control: saline i.v.

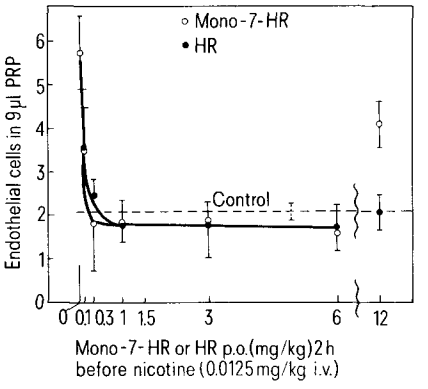

Fig. 2. The inhibition of the nicotine effect on endothelaemia by mono-7-hydroxyethylrutoside (Mono-7-HR) and by a mixture of hydroxyethylated rutosides (HR).

phologically on the basis of previous studies in which the cells were obtained directly from the surface of opened vessels under histological control and counted by the same method[2].

Albino female rats (Wistar), at the age of 3 months and weighing 180–220 g, were used in the present experiments. Nicotine was administered i.v. in the volume of 1 ml/200 g dissolved in physiological saline or orally 2 ml/200 g as aqueous solution by a gastric tube. Citrated blood was obtained from hearts of anaesthetized animals (sodium thiopental 20 mg/kg b.wt) for cell counting using siliconized collecting material. The time intervals between nicotine administration and blood collection were 5 min with the i.v. injection and 2 h with the oral administration.

Figure 1 shows a highly significant increase of cell counts after nicotine administered either i.v. or p.o. in a fraction of the dose corresponding to that absorbed during smoking of 1 cigarette (0.05 mg/kg b.wt). The statistical significances were calculated by the t-test, as in previous studies with larger groups of animals, the endothelaemic response to various stimuli has shown normal distribution. Similar repeated stimuli for detachement of cells produced serious changes in permeability of vessel walls resulting eventually in degenerative changes.

Several drugs have been shown to possess a protective effect on endothelium against various injurious agents. An outstanding position among them is occupied by flavonoids, particularly by their newer semisynthetic derivatives[3]. Consequently, 2 such derivatives were investigated for the ability to prevent increases of endothelial counts after nicotine. The lowest fully effective dose of i.v. administered nicotine (0.0125 mg/kg) was selected as a standard injury. The flavonoids, mono-7-hydroxyethylrutoside (mono-7-HR) and a mixture of hydroxyethylated rutosides (HR), were administered in various doses orally 2 h before the i.v. nicotine administration. Figure 2 shows that, even at a very low dose level representing one-twentieth of the clinical single dose, both drugs completely blocked the endothelaemia increase after nicotine.

1 Acknowledgment. Both flavonoid preparations were kindly supplied by Zyma SA, Nyon Switzerland.

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The effects of dopamine, noradrenaline and serotonin in the visual cortex of the cat

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Summary. The predominant effect of dopamine, norepinephrine and serotonin on the photically-evoked unitary activity was a prolonged inhibition of firing. These amines were also able to block acetylcholine-induced excitations and for longer periods of time than GABA.

Evidence supporting specific afferent pathways containing the biogenic amines dopamine (DA)[2-4], norepinephrine (NE)[5,6] and serotonin (5-HT)[7,8] has been reported in the mammalian CNS. These studies include histofluorescent microscopy, biochemical studies on the release of the amines[9] and radioautography at the light[10,11] and EM[12,14] levels, and would imply that the biogenic amines act as specific and classic neurotransmitters. However, the fact that such terminals are relatively few in number and lack specialized and specific synaptic contacts[12,13] suggests a 'modulatory' rather than a true neurotransmitter role[12-16]. In an attempt to determine the functional role of the biogenic amines in cerebral cortex we examined their effects upon cortical neurons in the visual (occipital) cortex and their interactions with the neurotransmitters acetylcholine (ACh) and γ-amino-n-butyric acid (GABA).

Methods. The experiments were performed on 15 adult cats which were initially anaesthetized with sodium methohexital (10 mg/kg i.v.). 'Encéphale isolé' preparations were prepared by making bilateral electrolytic lesions in A: +2.0 mm; L: 1.5 mm and H: −2.0 to −6.0 mm[17]. Standard microiontophoretic and extracellular recording techniques were employed[16,18]. 5-barrel micropipettes having an overall tip diameter of 4 to 8 µm were filled with the following agents: DA-Cl 0.5–0.8 M, pH 4.0; NE-Cl 0.5 M, pH 4.0; 5-HT oxalate 0.1 M, pH 4.0; ACh-Cl 1.0 M, pH 4.0 and GABA 0.5 M, pH 4.0. The central barrel containing 4 M NaCl (resistance 3–6 MΩ) was used for recording and one of the side barrels (2 M NaCl) as a balancing channel. In this study only visually-driven neurons were analyzed. These cells were found 300–1800 µm below the cortical surface and fired in response to a photic stimulus (Grass PS-20 photostimulator, frequency 0.5/sec or the beam of the oscilloscope).

Results and discussion. The biogenic amines DA, NE and 5-HT were found to inhibit the visually-evoked activity of the majority of cortical neurons (table). This inhibition, obtained with ejection currents of 50–100 nA during 20-

Effects of biogenic amines, GABA and acetylcholine on visually-driven cells*

Biogenic amine						GABA				Acetylcholine			
	Total	↑	↓	=	nt	↑	↓	=	nt	↑	↓	=	nt
Dopamine	35	0	22	13	0	0	34	0	1	19	13	2	0
Norepinephrine	84	1	55	22	6	0	78	0	6	47	17	10	10
Serotonin	42	6	16	14	6	0	42	0	0	11	5	20	6
Total	161	7	93	49	12	0	154	0	7	77	35	32	16

↑, Excited; ↓, Inhibited; =, No effect; nt, Not tested. *These cells were sampled between 300–1800 µm in the occipital (visual) cortex of the cat.