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Regional concentrations of noradrenaline and dopamine in the frontal cortex of the rat: dopaminergic innervation of the prefrontal subareas and lateralization of prefrontal dopamine

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Catecholamine levels in the two subareas of the prefrontal cortex and in one non-prefrontal region of the rat frontal lobe were measured radioenzymatically. In contrast with noradrenaline (NA), the distribution of dopamine (DA) in the frontal lobe is markedly heterogeneous. DA levels of the orbitofrontal and medial prefrontal subarea are, respectively, 3 and 4 times higher than those of a non-prefrontal region of the frontal lobe, confirming the expectation of neuroanatomical findings. Furthermore, it appears that at the population level, DA levels of the medial prefrontal subarea are lateralized, the left hemisphere being significantly higher than the right hemisphere.

During the past 5 years neuroanatomical studies in various mammalian species have demonstrated that the dopaminergic mesocortical system projects in those areas of the frontal lobe which also receive projections from the nucleus dorsomedialis thalami[1]. On the basis of these thalamic projections these areas are termed prefrontal cortex, PFC[10], which in the rat comprises a medial and an orbitofrontal subarea[7]. Quantitative, biochemical studies have recently shown that the dopamine (DA) levels of the prefrontal areas are relatively high. Palkovits et al.[8] measured DA and noradrenaline (NA) levels in the medial subarea of the PFC and in several non-prefrontal areas in the frontal lobe. While NA appeared to be homogeneously distributed over these areas (as in other cortical areas), the concentration in the medial subarea was considerably higher than in the other frontal areas. Tassin et al.[11] measured DA levels in both PFC subareas and in one non-prefrontal area, situated at the dorsolateral aspect of the frontal lobe. DA levels in this latter area were markedly lower than in the two PFC-subareas.

However, the data of Palkovits et al.[8] were based on relatively large frontal cortical samples, not confined to neuroanatomical boundaries, while Tassin et al. did not measure NA. In this study we have measured both NA and DA in approximately the same areas in the frontal cortex as Tassin et al.[11] In view of data indicating a lateralization of striatal DA[9], we made bilateral measurements in order to determine whether a lateralization of DA levels also occurs in the DA-rich subareas of the PFC.

Male rats of the WEzob-strain (Wistar derived) were used, weighing between 310 and 380 g. They had been housed with an ovariectomized female under a partly reversed dark/light schedule (lights on at 13.30 h, off at 1.30 h) and were fed ad libitum with standard rodent food pellets (Hope Farms, Woerden). They were sacrificed during the latter 2 h of the dark period and the first 3 h of the light period. Following decapitation the brains were rapidly extirpated and frozen on dry-ice. Serial, coronal 300 μm sections were made with a cryostat at —10 °C. Using a cooled stainless needle with an i.d. of 1 mm, bilateral punches were made in 2 successive sections ranging approximately between 10,500 and 11,100 μm anterior of the stereotaxic 0-point (König and Klippel[6]). In each section the needle was positioned in the medial and orbitofrontal PFC subareas and a dorsolateral non-prefrontal area. Punch sites

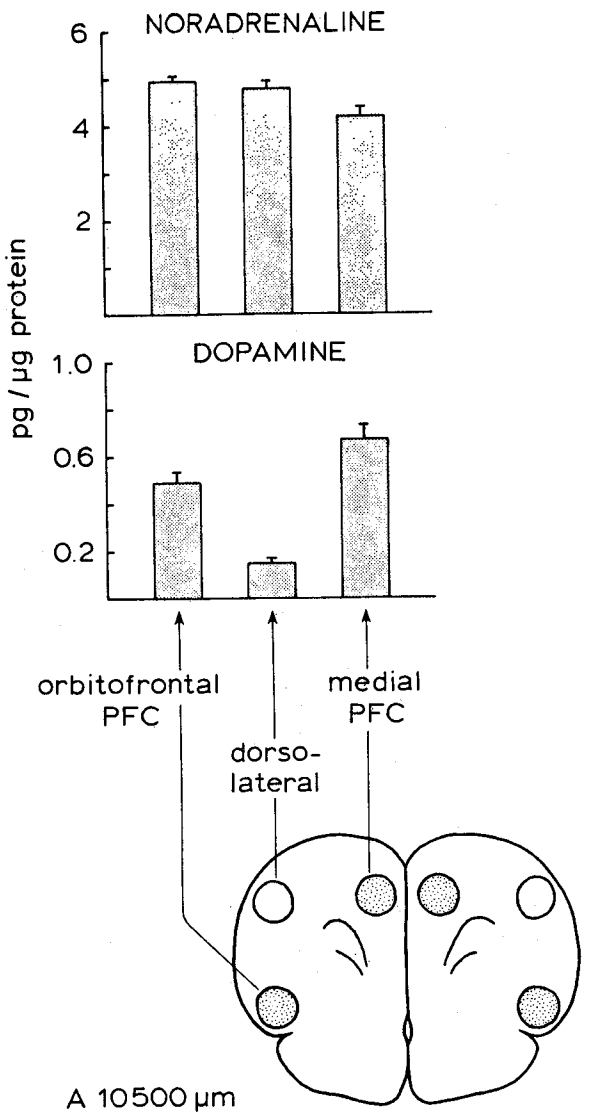

Fig. 1. Regional catecholamine concentration (mean ± S.E.) in the frontal lobe of male WEzob rats (n = 17). Noradrenaline and dopamine levels were measured in the same samples.

are illustrated in Fig. 1. Thus each sample consisted of 2 punches in the same area of two successive sections in one hemisphere. Tissue was homogenized in 60 µl of 0.2 N perchloric acid at 0 °C. Catecholamine concentrations were measured radioenzymatically[12] and expressed as pg/µg protein. S-adenosyl-L-methionine-(^{3}H) of highest available specific activity (70 Ci/mmole, NEN Chemicals) was used which resulted in an assay sensitivity of 1–2 pg. Mean interference by NA in the assay for DA amounted to 0.8% and was corrected for.

The results of the quantitative measurements of both NA and DA in each of the 3 investigated areas, both hemispheres taken together, are presented in Fig. 1. One notices a markedly heterogeneous distribution of DA, in contrast with NA, while DA levels are considerably lower than NA levels, the DA/NA ratio varying between 0.03 and 0.16. DA levels of both prefrontal subareas exceed those of the non-prefrontal area by a factor of 3–4. A comparison of the DA levels of both prefrontal subareas shows that the levels of the medial subarea are significantly higher than those of the orbitofrontal subarea ($P<0.01$, Newman–Keuls test). A slight but significant difference in opposite direction is found in the NA levels, being higher in the orbitofrontal than in the medial subarea ($P<0.01$, Newman–Keuls test).

The regional difference in DA levels found in this study is in agreement with the results of Tassin et al.[11]. However, the absolute DA levels in both PFC subareas are approximately 40% lower than in Tassin et al.'s study*. A comparison with the results of Palkovits et al.[8] is thwarted by the fact, that in that study no punches were made, but longitudinal strips were cut out covering a range of up to 2 mm or more. The DA levels of the medial subarea of the PFC found by Palkovits et al. were approximately twice as high as in this study. However, their sample was taken at a slightly more caudal and ventral level, which may explain the difference, since DA levels in the ventral part are approximately 3 times higher than in the dorsal part[11]. One of the non-prefrontal areas of the frontal lobe in which Palkovits et al.[8] took samples was situated between the orbitofrontal subarea of the PFC and the dorsolateral area of this study (area 3 in ref. 8), another was situated lateral to the dorsolateral aspect of the medial subarea of the PFC[7] (area 2 in ref. 8). Both areas were situated caudal to our punch sites, the latter (area 2) covering a range of 2.4 mm. DA levels of area 3 were comparable with our dorsolateral area, whereas the DA levels of area 2 even equalled those of our medial subarea of the PFC. This last finding is not in

*Tassin et al. made two punches in the medial subarea, a dorsal and a ventral one. Since our punch in this subarea was approximately intermediate between these sites, we have used the mean value of their two punches for comparison.

line with our expectation, but it is worth mentioning in this context, that contrary to Palkovits et al.[8], Tassin et al.[11] found a negligible DA concentration in region 2 at a level just caudal (approximately 0.7–1.2 mm) to our punch sites. In addition to the above considerations on differences in the absolute DA levels, we must realize that perhaps strain differences exist in the levels of catecholamines. Significant strain differences in brain DA levels of up to 20% have already been reported in mice by Kempf et al.[5] and recently Wolf et al.[13] have demonstrated strain differences in the density of dopamine receptors in rats. Moreover, in Tassin et al.'s study DA levels were not corrected for NA interference.

Thus we can conclude that the previous studies[8,11] and this study have demonstrated, that DA levels in prefrontal subareas are higher than in other frontal areas, confirming the expectation of neuroanatomical findings[1]. DA levels of the medial subarea exceed those of the orbitofrontal subarea. Such regional differences in the frontal lobe are not evident in the NA levels.

In addition to the results described above, we have examined possible interhemispheric differences in NA and/or DA levels. The mean concentrations of both NA and DA in the 3 areas are given in Table I separately for left and right hemisphere. No interhemispheric differences can be detected in the NA concentrations of the 3 areas investigated. However, with regard to DA a marked asymmetry is witnessed in the medial subarea of the PFC: DA levels being significantly higher in the left hemisphere ($P<0.02$, Student's t-test, two-tailed). This DA asymmetry cannot be due to a bias in the dissection procedure. Protein concentrations of the punches in the medial subarea of the PFC clearly demonstrate, that there had been no left–right difference in the amount of tissue in the samples (mean ±S.E. values of left and right samples, 39.1±1.2 and 40.1± 0.9 μg protein, respectively). Visual inspection of the sections revealed that the sites of the medial punches in both hemispheres were very similar with respect to the distance from dorsal and medial boundaries of the cortex. In the few cases that the plane of sectioning had been somewhat oblique, corrections were made by making the punches in the other hemisphere either in a more rostral or caudal section.

A left–right asymmetry of the frontal cortex at the population level has so far only been demonstrated in frontal cortical energy metabolism of female Sprague–Dawley rats by Glick et al.[3]. This bias is in the same direction as the medial prefrontal DA asymmetry reported in our study. From Glick's study it also appeared, that rats' side preferences in circling behaviour were higher if frontal cortex activity was higher on the contralateral side. A side preference in circling behavior has also been correlated with a higher striatal DA level on the contralateral side, but only after amphetamine treatment in female (not in male) Sprague–Dawley rats[2,9]. On the basis of these data, Glick and Ross[4] suggested an interaction of the left–right asymmetry in frontal cortex with the DA nigrostriatal asymmetry. Robinson et al.[9] have measured DA levels of the medial prefrontal subarea (not in other areas of the frontal lobe) in Sprague–Dawley rats of both sexes either

TABLE I

Regional catecholamine concentrations in left and right frontal lobe of male WEzob rats

Values represent mean (± S.E.) levels in pg/μg protein (n = 17).

	Noradrenaline		*Dopamine*	
	Left	*Right*	*Left*	*Right*
Dorsolateral part of the frontal lobe	4.873 ± 0.267	4.755 ± 0.314	0.149 ± 0.017	0.155 ± 0.023
Medial subarea of the PFC	4.418 ± 0.212	3.991 ± 0.213	0.800 ± 0.083*	0.547 ± 0.076
Orbitofrontal subarea of the PFC	4.939 ± 0.230	4.904 ± 0.216	0.511 ± 0.077	0.473 ± 0.049

* DA levels, left > right ($P < 0.02$, Student's t-test, two-tailed).

untreated or treated with amphetamine. However, in contrast to our data reported above, they did not demonstrate a left–right asymmetry in DA levels of the medial prefrontal subarea at the population level.

The functional implications of the interhemispheric difference in the DA levels of the medial PFC found in this study remain a subject for further experiments.

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