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Catecholamine Content and Response to Norepinephrine of Middle Cerebral Artery[1] (38179)

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It is generally agreed that there is abundant autonomic innervation to the cerebral vascular bed (1). Adrenergic nerve terminals in these vessels have been morphologically demonstrated by fluorescence (2) and electron (3) microscopy. Most investigators, however, attach minimal functional significance to this innervation (3), and pharmacologic effects of adrenergic transmitters are negligible when applied to cerebral vessels *in vivo* (4) and *in vitro* (5) or perfused *in vivo* (6). Only at nonphysiologic concentrations of norepinephrine (7) or abnormally high rates of sympathetic nerve stimulation (8) can significant adrenergic responses be elicited in these vessels.

Thus, the paradoxical situation arises in which adrenergic nerves have been anatomically demonstrated yet have no apparent physiological importance. Since fluorescence microscopy allows only a semiquantitative estimate of the amount of adrenergic transmitter present in the vascular wall, this study was carried out to analyze quantitatively the norepinephrine (NE) and epinephrine (E) content in cerebral vessels. In addition, the reactivity of cerebral arteries to NE was compared to that of an extracerebral vessel.

Methods. Whole bovine brains and tongues were obtained at a local slaughterhouse within 20 min after death and transported to the laboratory packed in ice. Middle cerebral arteries (MCA) were freed from their point of attachment at the circle of Willis for a distance of approximately 2 cm along the basal surface of the brain. Lingual arteries (LA) with outside diameters comparable to the MCA were dissected free in an area approximately 9 cm from the anterior portion of the tongue. Both vessels were flushed of clotted blood with cold saline, and adhering fat and connective tissue were removed. Samples were blotted dry and weighed, then homogenized in 20 ml of 10% trichloroacetic acid containing 0.6 ml of 10% EDTA (ethylenediaminetetraacetic acid, disodium salt). The procedure for catecholamine analysis has been described in a previous report (9).

Only one LA was required for each analysis (av wt = 942 mg); however, it was necessary to pool MCA from 5 different brains for each analysis (av pooled wt = 805 mg). Recovery after addition of 0.5 μg NE averaged 93% ($n = 4$); values in Table I are uncorrected for recovery. Data were analyzed by Student's t-test for group comparison; a P value of less than 0.05 was used as the criterion for significance.

Cumulative dose-response curves to NE were obtained on samples of both arteries. For these experiments brains and tongues were transported in a Krebs-bicarbonate solution and then treated according to

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TABLE I. Catecholamine Content (μg/g wet wt) of Bovine Middle Cerebral and Lingual Arteries.

Sample no.	Middle cerebral NE	Middle cerebral E	Lingual NE	Lingual E
1	1.35	0.06	0.25	0.07
2	1.15	0.01	0.21	0.09
3	1.95	0.02	0.26	1.09
4	0.69	0	0.23	0
5	0.26	0.02	0.30	0
6	1.24	0	0.43	0
7	0.85	0.29	0.10	0.10
8	0.34	0.01	—	—
9	0.76	0.04	0.24	0
10	0.71	0.17	0.25	0.02
11	0.54	0.59	—	—
12	1.51	0	0.19	0.01
13	1.40	0.74	—	—
Mean	0.98[a]	0.15[b]	0.25[a]	0.14[b]
±SE	±0.14	±0.07	±0.03	±0.11

[a] $P < 0.001$.
[b] $P > 0.9$.

standard procedures for spiral cut artery strips *in vitro* (10). Equilibrating tensions were 1 g for MCA, 2 g for LA. NE was was used as appropriate dilutions of a commercial preparation (Levophed).

Results. Table I repesents the data for NE and E analysis. The MCA contains about 4 times as much NE as the LA. Only 2 LA samples had a NE content that exceeded the lowest sample content in the MCA. E contents were almost identical in both vessels, accounting for only 13% of the total catecholamines in the MCA but 36% in the LA. This latter figure probably represents a falsely elevated estimate due to the unusually high content of one LA sample (#3). Only one LA and 2 MCA samples had E concentrations greater than or equal to that of NE.

The NE dose-response curves (Fig. 1) confirm the remarkably small response of the MCA relative to the LA. The difference in maximum developed tensions is highly significant (LA: 2.33 ± 0.28 g (SE), MCA: 0.25 ± 0.05 g; $P < .001$).

Discussion. This study provides the first definitive analysis of adrenergic transmitter contents in cerebral arteries. von Euler (11) found "quite a high activity" of a "specific sympathomimetic ergone" (NE) in extracts from bovine meninges and cerebral vessels, but did not quantitate his results.

The only other reliable study of catecholamines in bovine vessels is that of von Euler and Lishajko (12), who reported values for NE of 0.36 μg/g in splenic arteries and 0.35 μg/g in "large" pulmonary vessels. Our determination of NE in the LA is in agreement with these levels, but the content in the MCA is much greater. Adrenergic-mediated vasoconstriction has

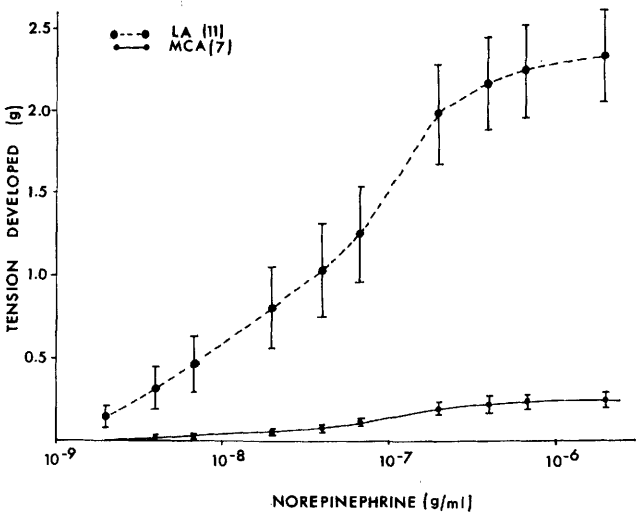

FIG. 1. Comparison of NE response in LA and MCA. Differences between responses at all doses are significant at $P < .05$ or less. Values are mean ± SE. ( ) = number of strips.

been adequately demonstrated in the abdominal viscera (13), skeletal muscle (14), and the lung (15). These findings suggest a possible inverse relationship between the NE content of some vascular beds and the degree of functional sympathetic vascular innervation.

NE is the major neurochemical transmitter released from sympathetic nerve endings (16). Several studies have shown that E accounts for only a very small percentage of the total catecholamine content in vascular tissues. Faredin *et al.* (17) report that E accounts for only 5–8% of total catecholamines in several arteries of the dog, while de la Lande (18) gives an average of 8.3% E in the central artery of the rabbit ear. The percent E in the MCA is only slightly above these values, while that in the LA, though high in this series of experiments due to a single markedly elevated sample value, would most likely normally fall in this range also.

The LA is much more responsive to NE stimulation than is the MCA, a fact which poses an inverse relationship between NE content and vascular reactivity to NE. A similar relationship has been noted by Maling *et al.* (19) in the aortae of rats, rabbits, and cats. Recently, Pegram *et al.* (20) have reported a direct relationship between the sensitivity to NE of several rabbit veins and their density of adrenergic innervation. These vessels were pretreated with propranolol and desmethylimipramine, however, which would permit a greater contractile response than might normally be expected.

One can only speculate as to the physiological significance of the high NE content in cerebral vessels which are only minimally responsive to this agent. Although the etiology of cerebral vasospasm is unknown, present evidence suggests that serotonin or a heat-stable component of blood is the primary stimulus (21). However, since α-blockade is partially effective in relieving cerebral vasospasm (22), NE may play a small part in this response. James *et al.* (23) have suggested that NE released from cerebral sympathetic nerves may function as a fine control over the response of cerebral vessels during conditions of hypoxia or hypercapnia.

Summary. The norepinephrine (NE) and epinephrine (E) contents of bovine middle cerebral (MCA) and lingual (LA) arteries were determined fluorometrically. Although the amount of E was almost identical in the 2 vessels, the MCA contained almost four times as much NE as the LA. Cumulative NE dose-response curves revealed a much greater reactivity of the LA than the MCA. An inverse relationship is demonstrated between NE content and vascular reactivity to this agent, and a similar relationship is suggested between NE content and the degree of functional innervation in some vascular beds.

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