Neuroscience Letters, 117 (1990) 358–362
Elsevier Scientific Publishers Ireland Ltd.

NSL 07157

Ammonia is endogenously generated in the brain in the presence of presumed and verified dementia of Alzheimer type

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(Received 12 April 1990; Revised version received 16 May 1990; Accepted 22 May 1990)

Key words: Dementia of Alzheimer type; Early onset DAT; Advanced DAT; Brain ammonia; Release

The healthy, mature, non-starved brain was found to take up a small amount of ammonia on average 7.22 ± 0.72 μg/100 g × min. In contrast, in patients thought to be suffering from incipient early-onset dementia of the Alzheimer type (DAT) the brain released a larger amount of ammonia on average 25.59 ± 16.17 μg/100 g × min. In advanced DAT states, an average of 2.73 ± 0.32 μg/100 g × min was released indicating the temporary nature of the severe loss of amino-N during the early stages of presumed DAT. Detrimental effects of endogenously formed ammonia on brain metabolism may affect the membrane potential, the excitability of neurons, and the energy metabolism. Ammonia may be assumed to be involved in the morphological changes in astrocytes and in the gliosis observed in early degeneration related to DAT. Endogenously generated brain ammonia thus may have a role in the cascade of cell damaging events in presumed incipient DAT.

Both molecular genetics and molecular biology have made considerable contributions to our understanding of dementia of the Alzheimer type (DAT) [17]. The genetic defect on the long arm of chromosome 21 [27] that is responsible for 'familial' DAT distinguishes this type of DAT from that in the majority of cases, but no genetic linkage to this chromosome has been found [26]. Furthermore, this genetic defect was found not to be identical with the A_4-amyloid precursor protein gene [28], being associated with the formation of amyloid in neuritic plaques. Thus, the etiopathogenesis of DAT (in the majority of cases) and the pathobiochemical mechanisms of its neuropathology remain obscure. Otherwise, several abnormalities with respect to brain glucose and its related metabolism have been described, even in the incipient state of the disease. In PET studies, cerebral glucose utilization was found to be predominantly reduced in the parietotemporal association cortex [10]. Overall cerebral glucose utilization was found to be diminished by around 50%, whereas oxygen utiliza-

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tion tended rather to be normal both in patients presumedly suffering from incipient early-onset DAT [16] and in states of beginning 'organic dementia' [20], indicating an imbalance in oxidative metabolism, which may not exist in obvious late-onset DAT when oxygen utilization of the brain was found to be reduced [11]. This abnormality in cerebral glucose metabolism was associated with a substantial loss of both amino acids and ammonia as reflected in cerebral arterio-venous differences [15, 24]. In this paper, we report on the functional significance of the cerebral metabolic rate (CMR) of ammonia and discuss its implication in abnormal cerebral metabolism and cellular damage in presumed DAT. Substantial differences between incipient and advanced states of the disease were demonstrated which are indicative of the temporary nature of severe proteolytic events.

The study includes four out of 20 patients (mean age 47 ± 4 years) clinically diagnosed as having incipient dementia, in all probability DAT of early-onset type (dementia history $\leqslant 6$ months); four of the 20 patients differed from those reported here insofar as a family history of DAT was found in any. Further diagnostic criteria applied to these patients have been detailed elsewhere [15]. In brief, inclusion criteria for this study on incipient dementia were: severe and persistent impairment of memory and cognition as verified by several subsequent neuropsychiatric examinations and by several dementia rating scales: personality changes not of abrupt but of rather rapid to insidious onset; more or less rapid progression of symptoms during the observation period; an ischemia score of 1–4 points; EEG showing general slowing with a loss of alpha frequencies, appearance of theta and delta waves, no focal disturbances; and incipient ventricular enlargement revealed by CT. The patients studied had received no drug treatment for several weeks prior to the investigation. The study also included seven patients suffering from advanced DAT which had been diagnosed earlier by clinical examination and was morphologically confirmed in 4 cases after death. These patients had all been suffering from dementia for more than 8 years and had been hospitalized in a general psychiatric hospital for at least 5 years because of severe and progressive dementia. They were unable to perform psychometric tests. In 6 out of the 7 cases, severe brain atrophy could be verified. Low doses of sedative drugs had to be given overnight in all cases. The results in these two patient groups were compared with the data recorded in a group of 15 healthy volunteers (medical students, mean age 25 ± 2 years) investigated earlier [13; see also below]. Arterial and mixed cerebral venous blood from the internal jugular bulb was sampled simultaneously. The ammonia concentration was measured by the method of Moore and Stein [22]. The CMR was calculated from the cerebral arterio-venous difference from ammonia and the global cerebral blood flow rate as measured by a modified Kety–Schmidt technique [4, 18]. Statistical analysis was performed by means of the t-test ($P \leqslant 0.05$). Homogeneity of variance was tested by the F-test. In the case of different variances, differences in the mean values were tested for statistical significance by means of a modified t-test [30].

The mean CMRs for ammonia released amounted to -25.59 μg/100 g $\times$ min in presumed incipient early-onset DAT, and to -2.73 μg/100 g $\times$ min in advanced states of DAT, whereas the amount taken up was $+7.22$ μg/100 g $\times$ min in healthy

TABLE I

ORIGINAL DATA, MEAN VALUES AND STANDARD DEVIATIONS OF CEREBRAL BLOOD FLOW (CBF), THE CEREBRAL ARTERIO-VENOUS DIFFERENCES (avD) FOR AMMONIA, AND THE CEREBRAL METABOLIC RATE (CMR) OF AMMONIA IN HEALTHY CONTROLS (mean age 25 years), PRESUMED INCIPIENT EARLY-ONSET DAT (mean age 44 years), AND ADVANCED DAT (mean age 72 years).

Control data from ref. 13. +, positive avD and uptake by the brain; −, negative avD and release from the brain.

Healthy controls ($n=15$)			Presumed early-onset DAT ($n=4$)			Advanced DAT ($n=7$)		
CBF (ml/100 g × min)	avD-NH_3 (μg/ml)	CMR-NH_3 (μg/100 g × min)	CBF (ml/100 g × min)	avD-NH_3 (μg/ml)	CMR-NH_3 μg/100 g × min)	CBF (ml/100 g × min)	avD-NH_3 (μg/ml)	CMR-NH_3 μg/100 g × min)
47.9	+0.117	+5.60	55.8	−0.850	−47.43	28.6	−0.097	−2.77
50.2	+0.129	+6.47	65.5	−0.425	−27.84	27.1	−0.105	−2.85
55.8	+0.152	+8.50	59.0	−0.272	−16.05	30.4	−0.098	−2.98
52.3	+0.152	+7.93	50.0	−0.221	−11.05	26.7	−0.084	−2.24
56.9	+0.132	+7.51				32.7	−0.091	−2.99
58.4	+0.117	+6.82				28.4	−0.082	−2.33
54.1	+0.129	+6.98				27.3	−0.109	−2.98
51.4	+0.146	+7.48						
48.8	+0.130	+6.35						
55.0	+0.128	+7.04						
49.8	+0.155	+7.72						
50.1	+0.159	+7.97						
55.6	+0.130	+7.24						
55.9	+0.133	+7.44						
50.6	+0.142	+7.21						
Mean								
52.9	+0.137	+7.22	57.8	−0.442*	−25.59*	28.7*	−0.095*	−2.73*
±S.E.M.								
3.3	0.013	0.72	6.5	0.285	16.17	2.1	0.010	0.32

*Statistically significantly different from healthy controls ($P \leq 0.05$).

young control subjects. The patients' data differed significantly from the control values (Table I). Thus, the total balance of free ammonia shifted from uptake in controls to release of different amounts from brains was probably affected by DAT. As detailed earlier, these changes can be assumed to be disease-related rather than age-related [15, 16]. In contrast to hyperammonemia in hepatic encephalopathy [7], the dementia patients in this study were normoammonemic, implying that the causes of the changes in CMR ammonia are intrinsic to the brain.

There is evidence that certain conditions induce ammonia formation in the brain. In vitro, ammonia was formed aerobically in a glucose-free medium, whereas formation was suppressed in the absence of oxygen [29]. In vivo, the cerebral ammonia con-

centration rose dramatically when CMR glucose decreased as a consequence of arterial hypoglycemia [23]. Besides the condition of reduced brain glucose availability, brain ischemia was associated with a severe increase of ammonia concentration in the cerebral cortex [9]. In DAT, the 50% reduction in the amount of available glucose in the brain [16] may give rise to increased ammonia production in the brain and thus in turn also to the enhanced release from the brain. It is important to emphasize, however, that a massive cerebral ammonia loss occurred in the incipient state of presumed DAT only. This loss was only moderately exacerbated in advanced states of the disease, indicating the temporary nature of this abnormality in the brain was probably affected by DAT.

The sources of the endogenously formed ammonia are not yet precisely known. Sources may lie in proteolysis [3, 15], catabolism of catecholamines [25], or the purine nucleotide cycle [21].

The manifold effects of ammonia include effects on both membrane potential and excitability of neurons [5]. Ammonia inhibits the binding of acetylcholine to synaptosomes [19] and reduces the phosphocreatine content of the brain [12]. Morphologically, Alzheimer type II astrocytes have been found in gray matter, showing an enlarged clear appearance with a large vesicular nucleus obviously resulting from cerebral toxicity during arterial hyperammonia. With respect to DAT, Alzheimer himself described glial cells with enlarged nuclei and an enhanced formation of fibrillary glial cells, particularly in deeper cortical layers [1]. More recently, laminar gliosis of layers II, III and V in association cortex has been reported in DAT [2], and frequently paired astrocytic nuclei and reactive fibrillary astrocytes have been found in the early degeneration preceding neuronal drop-out [6]. It is not yet known, however, whether or not early gliosis in the Alzheimer brain can also be induced by ammonia, as in hepatic encephalopathy. Otherwise, endogenously generated ammonia in increased concentrations must not be ignored as an additional detrimental factor [8] in cell-damaging events in the brain probably affected by DAT [14].

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