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Neuritic plaque-like structures in the rat cerebellum following prolonged alcohol consumption[1]

M.M. Paula-Barbosa and M.A. Tavares

Department of Anatomy, Oporto School of Medicine, Oporto (Portugal), 18 February 1983

Summary. Primitive neuritic plaques were observed in the inner third of the molecular layer of the cerebellar cortex of rats following chronic alcohol consumption. Neurites were identified as dystrophic parallel fiber boutons. Amyloid material dispersed among neurites was not clearly recognized, dystrophic some fibrils were frequently seen among them. Astrocytic processes were noted in the periphery of the plaque. Microglial reaction, however, was non-existent. The rarity of these lesions in the rat cerebellum and their probable relation to long periods of alcohol consumption is discussed.

Neuritic plaques are a common finding in brains from patients with Alzheimer disease and aged persons[2,3]. Similar lesions have been observed in old dogs[4] and monkeys[5] and, after experimental induction, in a few other animal species[6-8]. It appears that neuritic plaques are rarely present in animals[9,10]; the most careful searches have been unable to detect their presence in many animal species[11]. It would seem that, for unknown reasons, these lesions are conspicuously age-dependent in man[9,10]. They are more abundant in the cerebral cortex, mainly in the frontal and temporal lobes, in the hippocampus and the amygdaloid nucleus[12], and infrequent in the cerebellum. The majority of plaques observed in the latter are morphologically different from those found in the cerebral cortex[13] and are probably related to forms of generalized amyloid angiopathy[14]. Classic neuritic plaques, however, have been described in the cerebellum of patients with familiar forms of Alzheimer disease[13], in aged patients with Down syndrome and in some cases of cerebellar atrophy in chronic alcoholics[15].

The purpose of this report is to describe the presence of primitive neuritic plaques in the cerebellum of rats following prolonged alcohol treatment; plaques which are probably related to the widespread alcohol-induced cerebellar deterioration.

Material and methods. 8-week-old male Sprague Dawley rats, weighing 200-220 g, were separated into 10 different groups of 6 animals each. Half of the groups were alcohol-fed for periods of 1, 3, 6, 12 and 18 months, and the others used as controls for the same periods.

Ethanol-treated animals were given unrestricted access to a 20% aqueous ethanol solution as the only available source of liquid. Food and fluid intake were measured every other day, and the amounts consumed were calculated for the alcohol-fed animals. Controls were given the same amounts of food and fluid, with sucrose isocalorically replacing ethanol. Details of this procedure have been described elsewhere[16].

The methods described by Palay and Chan-Palay[17] were used for fixation of the nervous system. Tissue blocks from the cerebellar vermal lobules 4-6 were embedded in epon[18]. Semi-thick sections were stained with toluidine blue and ultra-thin sections double-stained with uranyl acetate and lead citrate.

Results and discussion. In semi-thick sections stained with toluidine blue, aggregates of rods and dots, probably corresponding to abnormal neurites were often seen, after 12 months of alcohol treatment, in the inner third of the molecular layer (fig. 1).

At the ultrastructural level scattered single neurites were observed in all cerebellar cortical layers after 3 months of alcohol treatment (5-month-old animals). These neurites were seen in all sections studied; their number however, was greater in the molecular layer (fig. 2). Following 6 months of alcohol consumption (8-month-old animals), neuritic plaques of the primitive type[19] could be seen in the inner third of the molecular layer (fig. 3). This location is similar to that of the plaques described in the cerebellum of chronic alcoholics[15]. The number and complexity of the plaques are dependent on the duration of the experiment, Plaques with diameters between 20 and 30 µm were frequently seen after 12 months of alcohol treatment (14-month-old rats).

Plaques were formed by a variable number of neurites which were markedly distended by an accumulation of dense and lamellar bodies, lipofuscin granules and tubulovesicular profiles. Paired helical filaments were never noted, which is in agreement with previous descriptions in which it is affirmed that in the central nervous system these do not exist in animals[3,9,20]. However, it must be stressed that paired helical filaments have been demonstrated in neurons of the spinal ganglia in rats after chronic alcohol administration[21]. The careful study of the neurites showed that almost all could be identified as being parallel fiber boutons. In spite of the existing dystrophy, synaptic specializations were easily identifiable, as well as the Purkinje cell spines with which they maintained synaptic contacts (fig. 3). The presynaptic origin of neurites from plaques is in keeping with previous descriptions[22]. Dendrites and their spines in the proximity of the plaque seemed to be well-preserved, as opposed to those described in the cerebral cortex of patients with Alzheimer disease[23,24].

Although a central core of amyloid was never observed, filamentous material randomly oriented and dispersed among neurites, and whose size resembled amyloid filaments, was frequently seen (fig. 3). A similar finding has been reported by Terry and Wisniewsky[19] who showed that

small amounts of this substance are present when plaques have more than 5 neurites.

Reactive glial cell processes could be clearly identified at the periphery of the plaques; microglial cells, however, were never seen. No relationship could be established between plaques and blood vessels, as other authors have found[25].

The present results provide evidence that chronic alcohol ingestion induces the formation of cerebellar neuritic plaques of the primitive type after 6 months of alcohol consumption; this fits the assumption that chronic and excessive alcohol intake produces marked degenerative changes in the central nervous system, most impressive in the cerebellum[16,26]. As these lesions could not be found in the pair-fed controls, one could rule out aging and coexisting conditions which normally accompany excessive alcohol ingestion[27] as causal factors for plaque formation. On the other hand, the hypothesis that alcohol must be related to the formation of these lesions in the cerebellum is supported by the finding of neuritic plaques in alcoholics[15]

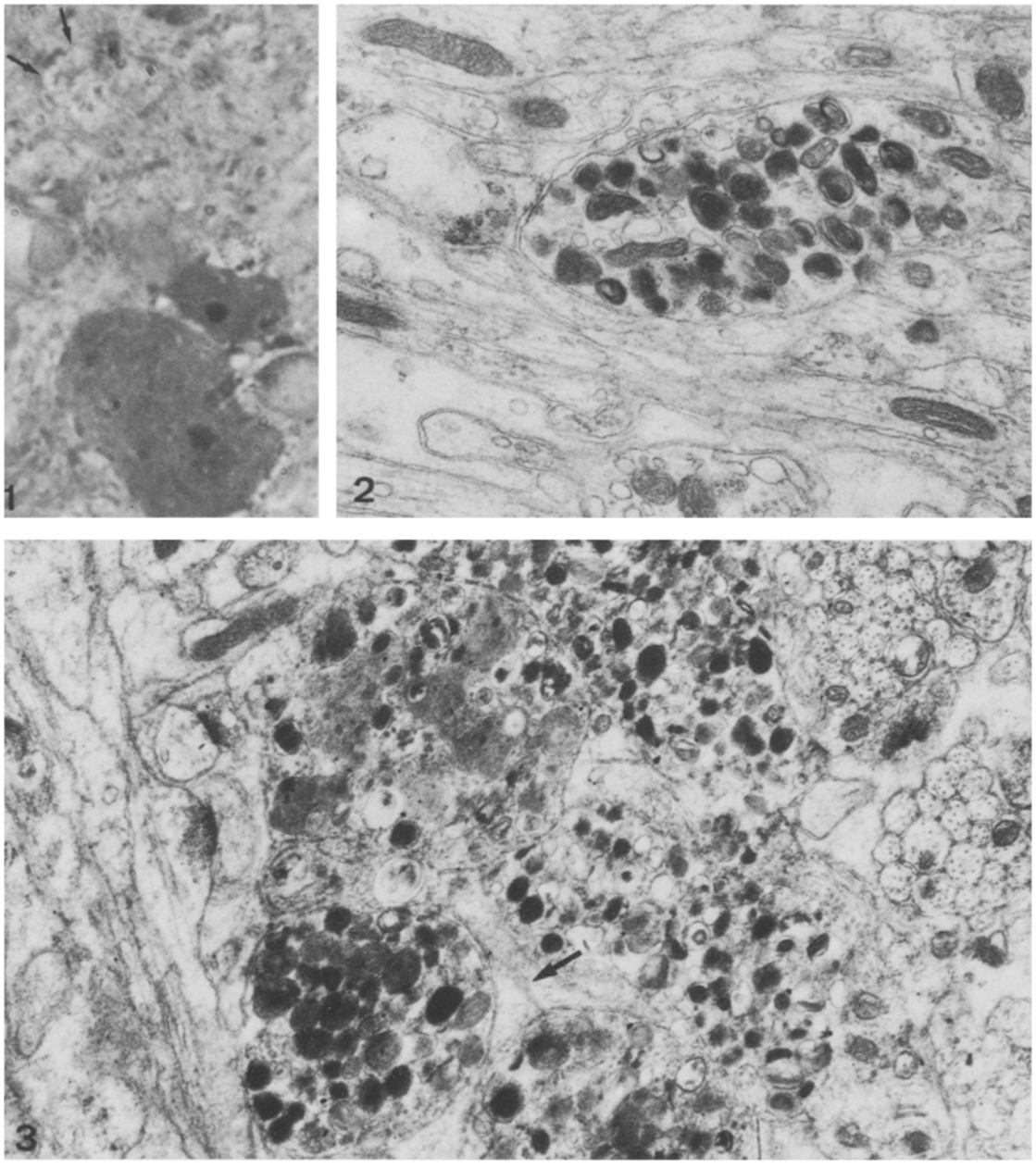

Figure 1. Neuritic plaque-like lesion seen in a semi-thick section from the cerebellar cortex molecular layer of a twelve-month alcohol-fed rat (arrows). The dots probably represent altered neurites. Toluidine blue. × 1200.

Figure 2. Isolated dystrophic neurite with dense bodies amid parallel fibres from the cerebellar cortex of a 3-month alcohol-fed rat. × 18,000.

Figure 3. A primitive neuritic plaque in the inner third of the molecular layer from the cerebellar cortex of a 6-month alcohol-fed rat. Neurites are distended by aggregates of dense and lamellar bodies, mitochondria and lipofuscin granules. Fibrillar material can be seen among neurites (arrow). The surrounding neuropile is well-preserved. × 24,000.

in the cerebellum, a rare location for such dystrophic changes even in those cases where cerebral cortex alterations are numerous[13,15].

Although wisps of material with the ultrastructural characteristics of amyloid fibrils could be frequently seen among neurites, the so-called primitive type of plaque[19] was the one observed. These dystrophic neurite aggregates are different from spheroids seen in gracilis and cuneatus nuclei following vitamin E deficiency or in aged animals[6,28,29]. They are like the experimental neuritic plaques seen in animals in which aluminium injection within the hemispheres was combined with cortical undercutting[7]. The fact that there was no well defined amyloid core, and also no microglial reaction in these lesions, supports those authors[8] who suggest that microglia play an important role in the processing and production of amyloid in neuritic plaques. To our knowledge, no neuritic plaques of any type have yet been described in the rat cerebellar cortex following prolonged alcohol consumption. Although a discussion of the pathogenesis and associated functional implications of these findings are beyond the scope of this report, it must be stressed that these lesions are probably the end-result of a dying-back process of parallel fiber boutons due to alcohol-induced granule cell dysfunction[16] and of other alterations of the cerebellar cortex milieu which create the environmental characteristics which predispose to neurite formation, as has been suggested under different circumstances[30].

Moreover, keeping in mind that neuritic plaques are related to aging and that a precocious and progressive accumulation of lipofuscin granules in the cytoplasm of Purkinje cells has been recently described under the same experimental conditions[31,32], it is tempting to suggest that prolonged alcohol intake might interact with, and probably accelerate, those mechanisms involved in the normal biological aging of the cerebellar cortex.

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Acute changes in dopamine metabolism in the medio-basal hypothalamus following adrenalectomy

D.H.G. Versteeg[1,2], I. Van Zoest and E.R. De Kloet

Rudolf Magnus Institute for Pharmacology, Medical Faculty, University of Utrecht, Vondellaan 6, NL-3521 GD Utrecht (The Netherlands), 7 October 1982

Summary. During the first hour following adrenalectomy the α-MPT-induced disappearance of dopamine was increased in the arcuate nucleus compared to that in sham-operated rats. In a number of other brain regions of both adrenalectomized and sham-adrenalectomized rats only stress-induced changes were observed in catecholamine utilization. These data suggest that corticosterone selectively modulates dopamine utilization in the medio-basal hypothalamus.

One h after bilateral adrenalectomy the turnover of serotonin in the dorsal hippocampus is significantly reduced when compared to that of sham-operated rats[3]. A low dose of corticosterone, given immediately after adrenalectomy, restores both serotonin turnover and steroid receptor occupancy in the dorsal hippocampus, whereas dexamethasone fails to do so[3,4]. The specificity of the serotonin response in the dorsal hippocampus corresponds to the properties of the glucocorticoid receptor system in rat hippocampal neurons[3,4]. The present experiments were carried out to