Changes in the Configuration of Synaptic Membranes of the Human Brain in Aging and Vascular Pathology

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It is common knowledge that synapses exhibit a pronounced structural diversity, manifested, in particular, in changes of the shape and curvature of the synaptic membranes. One example are the synapses on invaginated processes, described as "convex" and "concave" depending on which synaptic zone (pre- or postsynaptic) they invaginate. Synapses in which a postsynaptic process plunges into the presynapse are specified as "convex," while synapses with the presynaptic zone invaginated into a postsynaptic element are "concave." In recent years interest in such contacts has greatly increased [7,12,14,15]. Some authorities consider the "convex" synapses as "positive" and the "concave" as "negative" [11], while others identify them correspondingly as "frown" and "smile" synapses [16]. This matter has been discussed comprehensively in two papers [4,14], and yet it is evident from these reports that the causes of the change in shape of synapses and their functional role still remain unclear. Furthermore, the majority of studies of the changes in curvature of synaptic membranes have been performed using experimental materials.

The aim of the present investigation was an analysis of the ultrastructure of synapses with different types of curvature of the synaptic membranes in the human brain in normal aging and vascular pathology.

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MATERIALS AND METHODS

The nucleus caudatus and some areas of the cerebral cortex of individuals who had died of cardiac ischemia at the age of 36, 39, 70,72, 73, and 80 and of 73- and 83-year-old persons whose death was not due to vascular pathology were examined. The material was collected 2.5-4.5 h after death and was subjected to routine electron microscopy and embedded in Epon-812. The slides were studied under Hitachi H-600 and HIIE microscopes.

RESULTS

The investigation revealed a significant number of atypical bowl-shaped synapses situated on invaginated processes. A specific feature of these contacts is a convex or concave surface of the synaptic membranes (Fig. 1). The degree of curvature of the latter is variable and directly proportional to the degree of invagination of the process. The area of the active zone of these synapses also varies; often it occupies the whole surface of the membranes in contact, more seldom it looks like a dotted line, and sometimes perforated contacts are noted. There are mixed-type contacts in both convex and concave synapses (Fig. 2, *d, e*) and their

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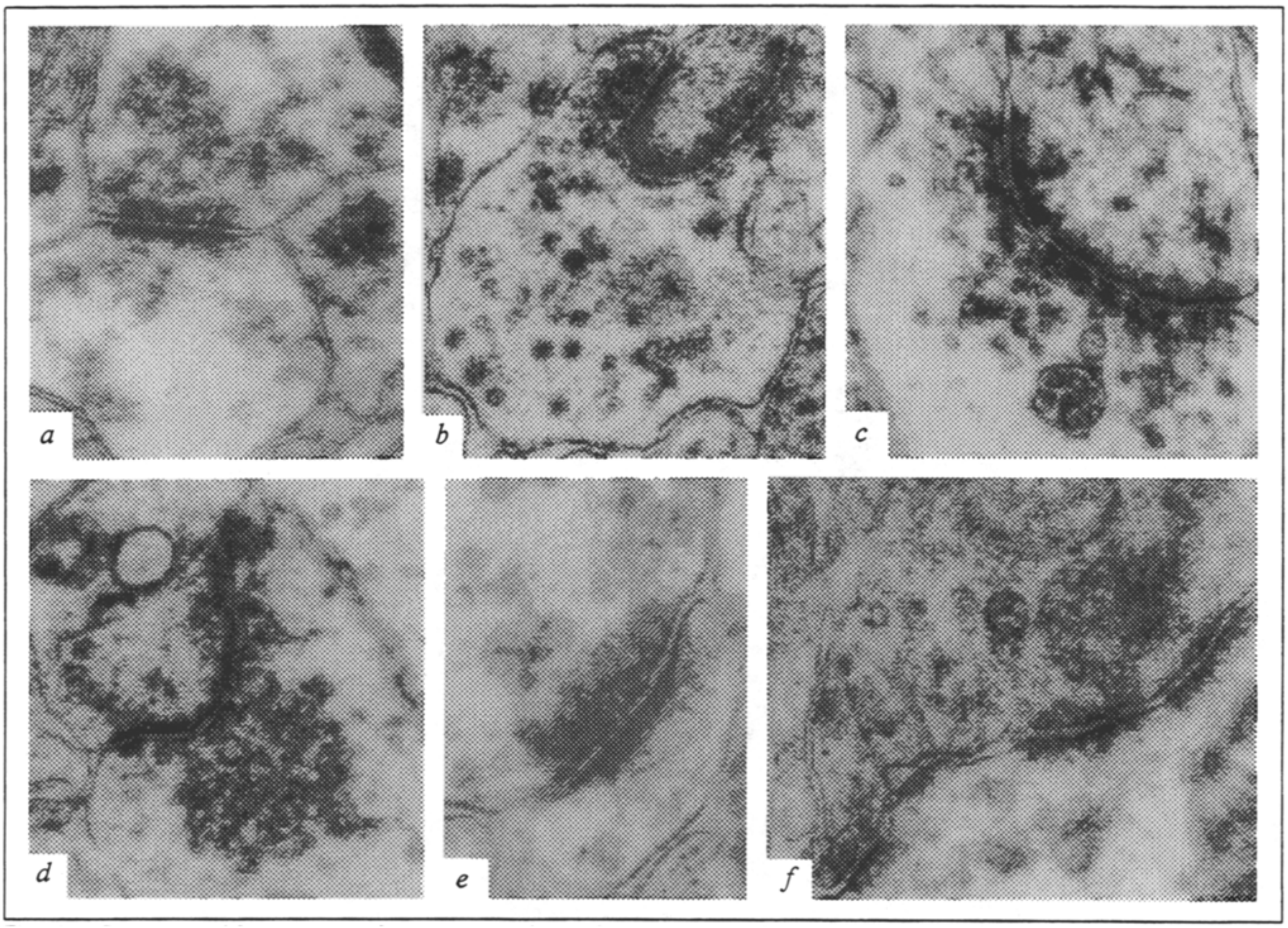

Fig. 1. Synapses with convex and concave surface of synaptic membranes. *a*) ultrastructure of synapse with plane surface of synaptic membranes (frontal region of cortex of a person aged 83 years); *b, d*) convex contacts: *b*) nucleus caudatus in a person aged 70 years; *c, d*) temporal region of cortex of a person aged 36; *e, f*) concave contacts in temporal lobe: *e*) in a person aged 83; *f*)in a person aged 36.

active zone consists of asymmetric or symmetric and desmosome-like thickenings of the synaptic membranes. A desmosome may be situated at some distance from an asymmetric contact (Fig. 2, *e*). Quantitatively, the number of convex synapses dominates over the synapses with a concave surface of the membranes.

Together with the simple shapes of convex and concave synapses more complex contacts were noted (Figs. 2 and 3). These are synapses with two or more active zones. At least two variants of such contacts may be recognized. In the first variant both active zones are situated on one of the surfaces of the synaptic membranes: convex or concave, in which case the synaptic membrane appears "two humped" (Fig. 2, *a, b*). Among the latter are synapses on dendritic spines which are classified as convex contacts. Some of them look "comblike" as a result of a change of spine shape (Fig. 2, *a*). As is shown in the microphotograph, the spine contains an electron-dense matrix, the spine apparatus is deformed, and there are no synaptic vesicles in the region of the active contact zone. In the second variant of complex contacts one of the active zones is situated on the convex and the other on the concave surface of the synaptic membrane, and in this case the latter has the form of a sinusoid (Fig. 3). Sometimes a combination of a convex or concave synapse with a straight one was noted in the material (Fig. 3, *e*). Such contacts often looked like dotted lines (Fig. 3, *a, c-f*).

Submicroscopically there were destructive alterations in both types of synapses noted to one degree or another in both portions forming the synapse. The presynaptic terminal is as a rule hypertrophic, and the number of synaptic vesicles and their localization in the presynapse are very diverse. An agglutination of synaptic vesicles was often noted in the region of the presynaptic membrane or far away from it. Some presynaptic processes were electron-dense and completely filled with altered synaptic vesicles (Fig 2, *c*).

Thus, the convex and concave contacts are very different from routine plane synapses due to

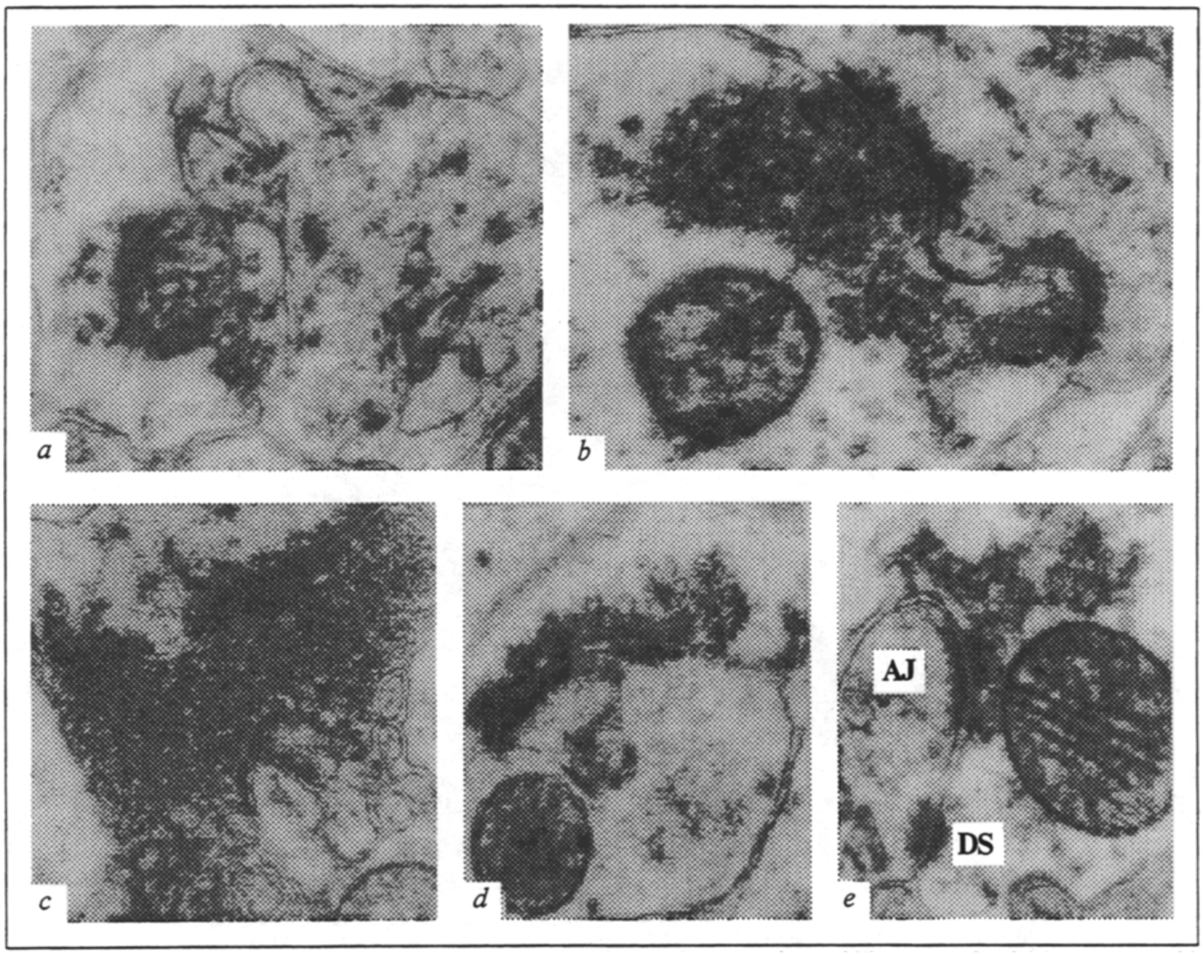

Fig. 2. Complex changes of the curvature of synaptic membranes. *a*) comblike axo–dendritic synapse; b) two–humped axodendritic contact; *c*) accumulation of synaptic vesicles in a presynapse; *d*) mixed contact consisting of a desmosome (DS) and asymmetric junction (AJ); a combination of a plane synapse and a convex synapse; *e*) the desmosome is distant from the asymmetric junction; *a–c*) nucleus caudatus in a person aged 70; *d*) temporal region of cortex of a person aged 36; *e*) frontal region of cortex of a person aged 73.

the curvature of the synaptic membranes, which, as already noted, may have a convex or a concave surface of the active zone, may be sinusoidal, or exhibit another type of contour. But for the existence of synapses with complicated changes of the synaptic membrane, it might be assumed that the formation of such contacts is the result of invagination of the processes. The change in curvature is most probably due to an imbalance in the processes in the pre- and postsynaptic membranes - possibly a disturbance of the ionic-molecular equilibrium, of the processes of regulation and autoregulation, etc. A likely cause is the drop of the axoplasmic flow observed in neurons in aging [2].

The direction of the curvature of synaptic membranes seems to be of functional importance [4,14]. Dyson and Jones [9] consider "positive" synapses as functionally active and "negative" ones to be nonfunctioning. The majority of concave or "negative" synapses observed by us belong to desmosomic contacts without synaptic vesicles in the active zone, and they may probably be classified as inactive synapses. However, these also include synapses with an impaired synaptic conduction (in our case, the convex and concave contacts containing agglutinated synaptic vesicles situated far from the presynaptic membrane). Thus, synaptic curvature is not in itself a marker of the functional activity of synapses. For example, there are synapses with sinusoidal changes of the membranes, where there is a combination of convex and concave surfaces of the active zones in one synapse, or, according to one hypothesis [11], there is both a "positive" (functional) and a "negative" (nonfunctional) synapse. In our view, the major role in the functional activity of a synapse is played by the state of the synaptic vesicles and the change of the curvature of the synaptic membranes, reflecting the plastic properties of the synapse. The same idea is shared by other authors [4,11]. It is also confirmed by our discovery of "two-humped"

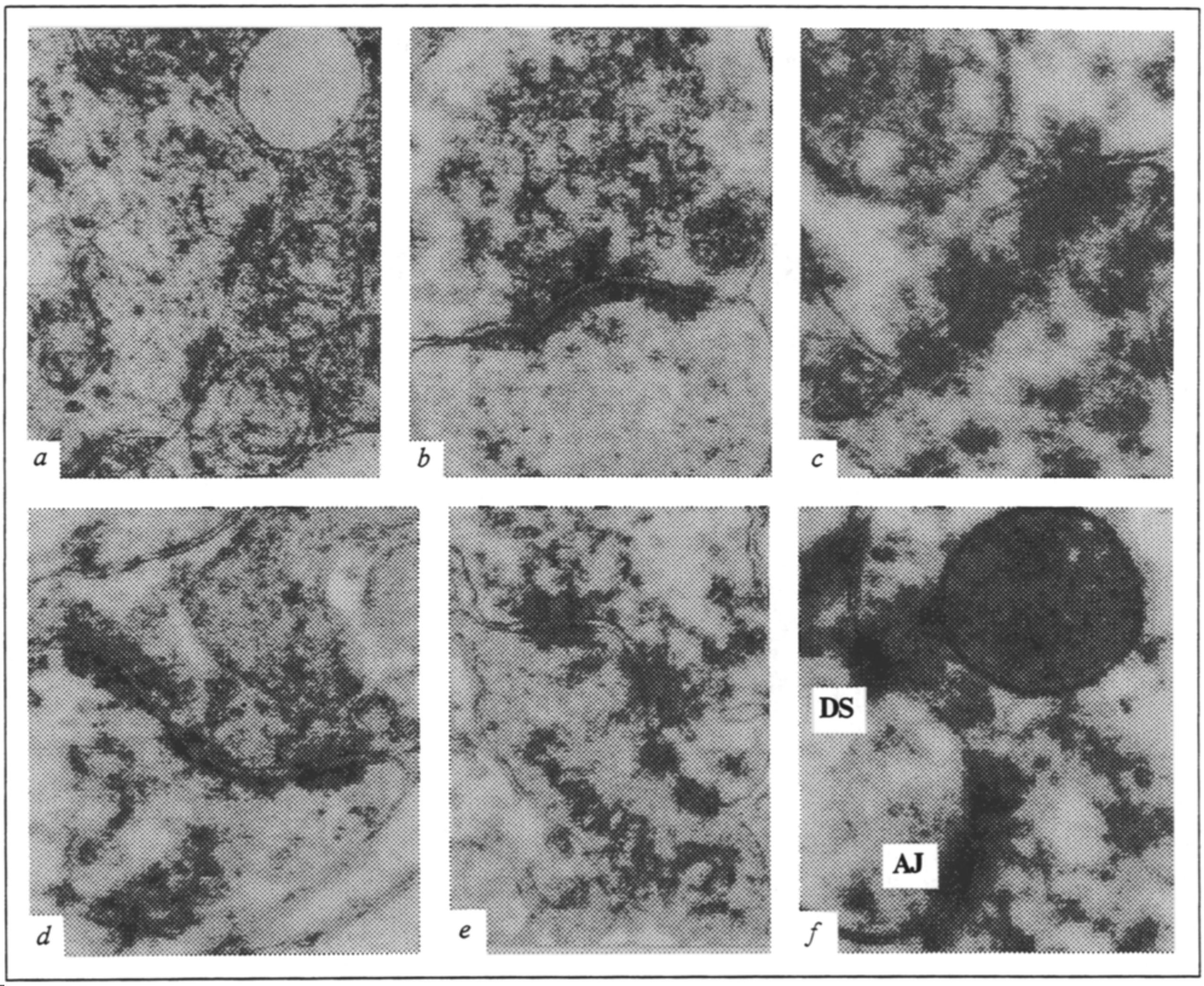

Fig. 3 Dotlike synapses with changed curvature of synaptic membranes: a combination of convex and concave surfaces of synaptic membranes. *a*) nucleus caudatus of a person aged 70; *b, f*) temporal region of cortex of a person aged 36; *c*) frontal region of cortex of a person aged 83; *d*) frontal region of cortex of a person aged 39; *e*) frontal region of cortex of a person aged 72. DJ: desmosome–like junction; AC: axodendritic contact.

convex axodendritic synapses like spinules, described elsewhere [17]. Such contacts are derived from the invagination of a spine in a presynaptic terminal and are feedback areas which are drawn into the process of synaptic plasticity or reorganization [9].

We found that the number of convex synapses exceeded that of concave contacts, and this, along with the overall decrease of the number of synapses in aging [5] and vascular pathology [10], may be considered as an index of reactivity to synaptic depletion. It is further corroborated by data [3] on the stimulation of plastic changes in preserved contacts by the loss of synapses in aging. Some authors point to the possible transformation of certain types of synapses into others [7,12], while others evaluate the increase of the number of curved contacts and decrease of straight ones as a sign of partial depletion of the synaptic pool [1].

REFERENCES

1. V. V. Semchenko, S. S. Stepanov, and A. Yu. Savchenko, *Zh. Nevropat. Psikhiatr.*, **84**, № 7, 1038-1042 (1984).
2. V. V. Frol'kis, *Aging and Increasing the Life Span* [in Russian], Leningrad (1988).
3. I. Adams, *Brain Res.*, **424**, № 2, 343-351 (1987).
4. R. K. S. Calverley and D. G. Jones, *Ibid.*, **15**, № 3, pp. 215-249 (1990).
5. B. G. Cragg, *Brain*, **98**, 81-90 (1975).
6. A. J. Cronin, T. P. Sutula, and N. L. Desmond, *Soc. Neurosci. Abstr.*, **13**, 947 (1987).
7. R. M. Devon and D. G. Jones, *Dev. Neurosci.*, **4**, 351-362 (1981).
8. M. B. A. Djamgoz, J. E. G. Downing, M. Kirsch, *et al.*, *Neurocytol.*, **17**, 701-710 (1988).
9. S. E. Dyson and D. G. Jones, *Dev. Brain Res.* **13**, 125-137 (1984).
10. H. Haug, *Clin. Neuropath.*, **8**, № 5, 233 (1989).
11. D. G. Jones and R. M. Devon, *Brain Res.*, **147**, 17-63 (1978).
12. D. G. Jones and S. E. Dyson, *Ibid.*, **208**, 97-111 (1981).
13. E. J. Markus, T. L. Petit, and J. C. LeBoutilier, *Dev. Brain Res.*, **35**, № 2, 239-248 (1987).

14. E. J. Markus and T. L. Petit, *Synapse*, **3**, 1-11 (1989).
15. T. L. Petit and E. J. Markus, *Neuroplasticity: Learning and Memory*, Eds. N.W. Milgram *et al.*, New York (1987), pp.87-124.
16. T. L. Petit, in: *Neurology and Neurobiology, Neural Plasticity: A Lifetime Approach*, Eds. T. L. Petit and G. O. Ivy, Vol. 36, New York (1988), pp. 201-234.
17. A. Routtenberg and S. Tarrant, *Anat. Rec.*, **181**, 467 (1975).

Changes in Kupffer Cells after Reversible Ischemia in Rat Liver

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Occlusion of the portal vein results in the deposit of intestinal endotoxin (lipopolysaccharide) in the portal vein system. An increase of the lipopolysaccharide content in the blood may trigger the release of hepatic macrophage products from the cells, which may damage the liver parenchyma.

The aim of the present investigation was to study Kupffer cells (KC) after a 30-min reversible normothermal total ischemia in rat liver.

MATERIALS AND METHODS

For the study, 35 male Wistar rats weighing 200-220 g were used. The rats were kept fasting 12-14 h before the experiment, but water was given ad libitum. The operation was performed under nembutal anesthesia (40 mg/kg). Each experimental series consisted of 5 animals. In the operation the common bile duct was separated from the hepatoduodenal ligament and the latter was compressed for 30 min to produce ischemia of the liver. Rats were decapitated in the 30th min of normothermal ischemia before removal of the clamp and 2, 12, and 24 h and 3 and 7 days after recirculation. Sham-operated animals were the control. The tissue samples were processed for electron microscopy as described previously [2]. For ultrastructural study of KC, photographs were taken under an electron microscope with a power of 3,500. The morphometric indexes of organelles were measured at ×17,000 magnification using an open test grid. The data were processed statistically using an Elektronika DZ-28 computer. The differences between values were significant at $p<0.05$ (Student t test [1]).

RESULTS

After 30 min of ischemia (acute stage) the volume of secondary lysosomes in KC increased by 64% in relation to the control (Table 1). Active adhesion of monocytes to the endothelium was noted, and these monocytes accounted for 43% of the KC population (5% normally) (Fig. 1).

Recirculation during 2 h after 30-min ischemia resulted in a further increase of adhesion of KC bone marrow precursors to the endothelium. In this period the 3-fold increase of the volume of secondary lysosomes was predominantly due to erythrophagosomes (Figs. 2 and 3) and the latter were found in 64% of KC (normally in 2.5%) (Fig. 2). Massive erythrophagia caused atrophic

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