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Characteristic Residual Neuropathological Features of Japanese B Encephalitis

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Summary. Characteristic residual (12–67 years) neuropathological features of 4 verified or suspected cases of Japanese B encephalitis (JBE) are reported. These features are summarized as: 1. unique distribution pattern of the main lesions, i.e. combination of lesions in the thalamus, substantia nigra and Ammon's horn. Lesions in the thalamus consistently involved, in a linear fashion, lamina medullaris medialis with nucleus intralaminalis and adjacent portions of the nucleus lateralis thalami. Lesions in the substantia nigra usually occupied the middle parts of zona compacta. These lesions were usually symmetrical, though unequal in extent. 2. Unique nature of the lesions, especially those in the thalamus and substantia nigra. Characteristic "light circumscribed foci (LCF)", which consisted of small rarefied areas, with few cellular and fibrous elements, surrounded by dense gliomesenchymal scarring, were observed there and occasionally in cerebral cortices. Lesions were thought to be vestiges of "circumscribed necrotic foci" reported in the CNS of acute stage of JBE. Additional characteristic features in the thalamic lesions were calcified and binucleated nerve cells. Alzheimer's neurofibrillary tangles were not found. Authors consider that the distribution and nature of the lesions are of diagnostic value.

Key words: Japanese B encephalitis – Chronic residues of encephalitis – Binucleated nerve cell – Calcified nerve cell.

Introduction

The residual neuropathological features of Japanese B encephalitis (JBE) have been poorly documented. Reports of the condition are few (Hamada and Ishii, 1966; Shiraki, 1963; Shinohara et al., 1973) although reports of the acute stage were fairly numerous (Hayashi, 1931; Zimmerman, 1946; Haymaker, 1947; Suwa, 1950; Kanno, 1959; Takeya, 1962; Shiraki, 1963).

We examined 4 cases of serologically verified and suspected JBE, survival from which ranged from 12–67 years, and found that the residual lesions of JBE were very characteristic in both the localization and the nature of the lesions.

Materials and Methods

Brains of 4 cases of chronic residual JBE were studied. Clinical data of these cases is summarized in Table 1. Autopsy was performed 4–20 h after death. Brains were fixed in 10% formalin or alcohol. Celloidin sections from alcohol fixed blocks and paraffin sections from formalin fixed material were cut. H.-E., Nissl, PAS, Carmine, myelin (Sugamo and Woelke), Luxol fast blue, Sudan III, Holzer, Cajal, Bielschowsky, and Bodian stain were used.

Clinicopathology of 4 Cases

Case 1. S.K. (Matsuzawa No. 1310). 17 years old boy. The patient contracted JBE in summer of 1950, at the age of 4 years and 9 months. The onset of the illness was characterized by convulsions, high fever, and comatose state which lasted for 6 days. Cerebrospinal fluid examination on 12 days revealed positive complement fixation reaction for JBE. Residual neurological symptoms were left spastic hemiparesis, muscle rigidity in right upper extremity, dysarthria, mental retardation, tonic seizures with loss of consciousness, and twighlight state. At the age of 15, he was admitted to the Matsuzawa Hospital because of violence and other derailed conducts. The patient died of emaciation due to occasional fits of high fever in Nov. 1962. The total course of the illness was 12 years.

Autopsy Findings. The brain weighed 1070 g. Atrophy of the right frontotemporal lobes, the right Ammon's horn and basal ganglia were marked. The substantia nigra (SN) was normally coloured. Small scars were scattered on the inferior surface of the left cerebellar hemisphere. *Microscopically*, the lesions in the thalamus and SN were the most characteristic. Scar tissue in the right thalamus involved linearly lamina medullaris medialis (LMM) with nucleus intralaminalis and adjacent portions of the nucleus lateralis

Table 1. Clinical data on cases of chronic residues of JBE

Case	Age	Sex	Age at affliction	Serologic test	Survival	Symptoms
1. S. K.	17		4Y9M	+	12Y	High fever, coma, convulsions
2. S. S.	28		6Y	+	22Y	High fever, coma
3. K. F.	78		11Y	Not verified	67Y	"Yoshihara Flu" (detail unknown)
4. A. Y.	19		3Y3M	Not verified	16Y	High fever, coma, convulsions

thalami (Fig. 1a). Inside the scar, several small circumscribed round or oval patches of light, rarefied, poorly cellulated foci, "light circumscribed foci (LCF)", were prominent. Similar but smaller lesions were found also in the left ventromedial parts of the LMM thalami. In these foci, most nerve cells were lost but the remaining ones were atrophic, calcified, or often binucleated. The same "LCF" were also found in the dense glio-mesenchymal scar in the rostral part of the right SN (Fig. 7). Serial sections disclosed the "LCF" in SN continued columnwise from rostral to caudalwards. Similar but less extensive lesions were also found in the left SN. Glial nodules were not found except a dubious one in the red nucleus, and perivascular cuffing was minimal. Alzheimer's neurofibrillary tangles were not observed. A cystic scar extended from the surface to the subcortical white matter in the right frontal gyrus. Diffuse, laminar or pseudolaminar loss of neurons accompanied by progressive astrocytes was scattered in the frontal, temporal, and precentral cortices. The right Ammon's horn showed a glio-mesenchymal scar with a few remaining atrophic nerve cells in h2. Several foci of loss and atrophy of neurons, especially of Purkinje cells and diminution of nerve cells in the granular layer were found in the cerebellum.

Case 2. S.S. (Matsuzawa No. 1456). 28 years old male. The patient afflicted by JBE with high fever and coma, which was verified serologically, at 6 years of age (1947). Since then he was mentally retarded. At 14 years of age, generalized grand mal seizures and absence began to occur. Because of epileptic personality changes the patient was admitted to the Matsuzawa Hospital on Dec. 3, 1958. Neurologically, elevation of deep tendon reflexes of extremities, dysdiadochokinesis, atactic gait, tremor of fingers and perioral muscles, loss of associated movements of the arms and legs, were observed. On August 8, 1970, the patient died of the generalized convulsions. The total course of the illness was 22 years.

Autopsy Findings. The brain weighed 1150 g. Slight herniation of the uncus hippocampi and tonsil of the cerebellum, small cystic lesions in both occipital poles, left inferior temporal gyrus, left inferior parietal gyrus, atrophy of both Ammon's horns, dilatation of lateral and IIIrd ventricles, were found. Both thalami were atrophic and in the left, a linear scar of 0.8 cm length ran along the LMM. Slight depigmentation in the SN, atrophy of the left red nucleus, locus coeruleus, and inferior olivary nucleus were also noted. *Microscopically*, the lesions in the thalamus were the most characteristic. Linear scars ran along the left LMM thalami, and it showed several small round or oval patches of "LCF" inside the dense glio-mesenchymal scar (Fig. 2). Many of the remaining nerve cells in the foci were calcified and binucleated (Fig. 5). The "LCF" were also found in the SN (Fig. 6). Small cystic scars were scattered in parietal, occipital, temporal and insular cortices. Both Ammon's horns showed atrophy and gliosis. Loss and atrophy of neurons with mild astrogliosis were observed in the other cerebral and cerebellar cortices, periaquaductal regions, the inferior olivary nucleus, and the spinal cord.

Case 3. K.F. (Matsuzawa No. 1406). 78 years old male. The patient afflicted by "Yoshihara Flu" (influenza in Yoshihara—a famous prostitute town in Tokyo at the time, August, 1901) which was thought as JBE (Hayashi, 1960), at the age of 11 with high fever and delirous state which lasted several days, and showed marked personality changes and mental retardation thereafter. Because of derailed conduct, the patient was admitted to the Matsuzawa Hospital in August, 1910. Neurologically, mild tremor in the arms and legs, elevated deep tendon reflexes, especially of lower extremities, were found. During his 57 years of hospitalization he became gradually milder and engaged in a cleaning work around hospital premises. He died on 24th of March 1968 due to general prostration. The total course of the illness was 67 years.

Autopsy Findings. The brain weighed 1300 g. Marked atherosclerosis of the basilar arteries, enlargement of lateral and IIIrd ventricles, circumscribed cystic scars in the left frontal gyrus and in the right caudate nucleus, atrophy of both Ammon's horns, were noted. Half transparent, brownish scars of 10 cm length ran along with the LMM in the left thalamus, similar ones, in the ventral part of the right thalamus and of the right pulvinar thalami, were found. The SN of both sides, the left dentate nucleus, and the tegmentum of the pons were atrophic. *Microscopically*, symmetrical foci were observed in the thalamus, SN, and Ammon's horn. Atrophy of the left thalamus was more marked than the right. Linear scars with characteristic "LCF" (Figs. 3 and 4), ran along with the LMM thalami and the adjacent portions of the lateral nucleus including centre median on both sides. Many calcified and binucleated nerve cells were found nearby. The similar lesions occupied most of the zona compacta of both SN. The boundaries of the "LCF" here were less clear than those in the thalamus. Though loss of nerve cells was marked, calcified neurons were not, but 1 or 2 binucleated ones were found nearby. Selective loss of neurons with gliosis in h1 and h3 in both Ammon's horns. Scattered atrophy and irregular arrangement of neurons, small rarefaction necrosis, thickening and hyalinization of small arteries associated with a few round cells, were found in the cerebral cortices, basal ganglia and brain stem regions. Focal necrosis covering the whole cortex and a spongy state in the white matter running along the U-fibers were observed in the left precentral gyrus. The cerebellum showed little change except a few foci of absence or atrophy of Purkinje cells and of nerve cells in the dentate nucleus.

Case 4. A.Y. (Matsuzawa No. 1103). 19 years old male. The patient contracted JBE at the age of 3 years and 3 months (August, 1935) which began with sudden outbreak of high fever and comatose state and occasional convulsions. A year later epileptic grand mal seizures reappeared. The patient showed marked mental retardation, though he attended school until the 3rd grade. Because of the epileptic personality changes he was admitted to the Matsuzawa Hospital in July, 1948 (16 years old). He was infantile in his behavior with dull mimic, slow movements, and hostility. Neurologically, deformation of the right pupil, elevated deep tendon reflexes on both sides, ankle clonus on both sides, rigidity of the exremities, dysarthria, dysdiadochokinesis, were observed. The patient gradually weakened and died in Nov. 1951 at the age of 19. The total course was 16 years.

Autopsy Findings. The brain weighed 1350 g. The poles of the temporal lobes, gyrus rectus and a part of the orbital gyri of both sides were atrophic due to scars with small cysts. Both thalami showed foci of softening of 2 mm size near the pulvinar thalami, the SN was slightly depigmented. *Microscopically* the thalamus showed several small to moderate sized necrotic scars in the medial, lateral, and anterior nuclei. Inside the scars illdefined spongy-like rarefaction foci similar to "LCF", but less characteristic were discernible. Gliosis was accompanied by focal loss of nerve cells, calcified and binucleated neurons, and pseudocalcium concrements. In the SN, dense cellular and fibrous gliosis with 1 or 2 glial nodules contained small "LCF", and occupied the rostral, medio-ventral parts. Here nerve cell loss was rather mild though depigmented. Small cystic necrosis with "LCF" and pseudocalcium concrements were found in several parts of the gyrus rectus and orbital gyri, but the margin of "LCF" to the scar was vague. Small rarefaction necroses in the temporal cortex and moderated gliosis in the adjacent white matter were found. No change was observed in the cerebellum.

Summary of Neuropathological Findings

The most characteristic and common neuropathological findings in the CNS of these verified and suspected old JBE cases were as follows: 1. A special pattern of distribution of the lesions. Main lesions in the brain of old cases of JBE were uniformly localized in the thalamus, SN, and Ammon's horn, in addition to relatively mild ones in the cerebral cortices. The main lesions were symmetrical with minor differences in extent (Fig. 1). 2. Focal lesions in the thalamus (Figs. 1–4) were characterized by their localization and nature. The lesions in the thalamus consistently involved linearly the LMM with the nucleus intralaminalis and adjacent portions of the lateral nucleus, though the lesions in case 2 were somewhat irregularly located. The localization of the lesions in the thalamus was similar to those of vascular origin. However, the lesions were characterized by "LCF" (Figs. 2–4), and abundance of calcified and binucleated neurons (Fig. 5). The former consisted of several light, small, round or oval areas which were poor in cellular and fibrous elements compared to the adjacent dense glio-mesenchymal scars. The boundary between "LCF" and surrounding scar tissue became gradually less marked as the patient survived longer. 3. The lesions in the SN were almost identical in nature with those in the thalamus, i.e. "LCF" surrounded by the dense glio-mesenchymal scars (Figs. 6 and 7), though there were fewer calcified and binucleated neurons. The lesions usually occupied the middle parts of the zona compacta and ran columnwise rostral to caudalwards. 4. The lesions in the Ammon's horn (Fig. 1c) were presumably those of vascular origin associated with inflammatory processes during the acute stage. They consisted of dense glio-mesenchymal scar of nonspecific nature. 5. Focal lesions in the cerebral cortices were less constant and less severe than those in the thalamus and SN, except in case 2 where a large cyst was found in the parietal lobe. The cortical lesions were usually devoid of characteristic "LCF" except in a focus in case 1. Changes found in the other parts of the brain consisted of atrophy and loss of neurons accompanied by mild to moderate glial proliferation and cystic changes. Alzheimer's neurofibrillary change was not found. Minimal perivascular cuffings around the few vessels and a glial nodule or two were found in the brain stem tegmentum in case 1 and 2.

Discussion

The pattern of distribution of the lesions in the 4 old cases of JBE was surprisingly uniform, i.e. a combination of focal lesions in the thalamus, SN, and Ammon's horn, in addition to mild ones in the cerebral cortex. Neuropathological changes in the brains of acute cases of JBE (Hayashi, 1931; Zimmerman, 1946; Haymaker, 1947; Suwa, 1950; Kanno, 1959; Takeya, 1962; Shiraki, 1963) are characterized by panencephalitis with abundant glial nodules and perivascular cuffing and necrosis with or without characteristic "circumscribed necrotic foci" (focal acellular plaque, Zimmerman, 1946). These findings are more widely and variably distributed than those in our chronic cases, though the severest ones were likewise located in the thalamus, SN, and Ammon's horn. Protracted cases (Kunimoto, 1960, 88 days; Shiraki et al., 1969, 151 days) also show wider localization of the lesions than do chronic cases. Thus the lesions in chronic cases tended to localize in narrower sites. Among them, only severe necrotic scars with characteristic "LCF" are prominent. This means either that the lesions in the acute stage converge to the limited sites as the patient survives longer (Takeya, 1962), or that only those with milder lesions survive.

The lesions of the thalami in the present cases showed surprisingly uniform localization. They consistently involved linearly the LMM with the nucleus intralaminalis and adjacent portions of the lateral nucleus, and occasionally also the nucleus dorsomedialis and nucleus medialis. Shinohara's chronic case (1973) as well as other protracted cases (Kunimoto, 1960; Shiraki et al., 1969) also showed similar localization of the lesions in the thalami. The lesions in the SN usually localized in the middle part of the zona compacta (Fig. 6).

The nature of the residual lesions in the present cases of JBE also showed characteristic features compared with those in the acute cases, i.e. the presence

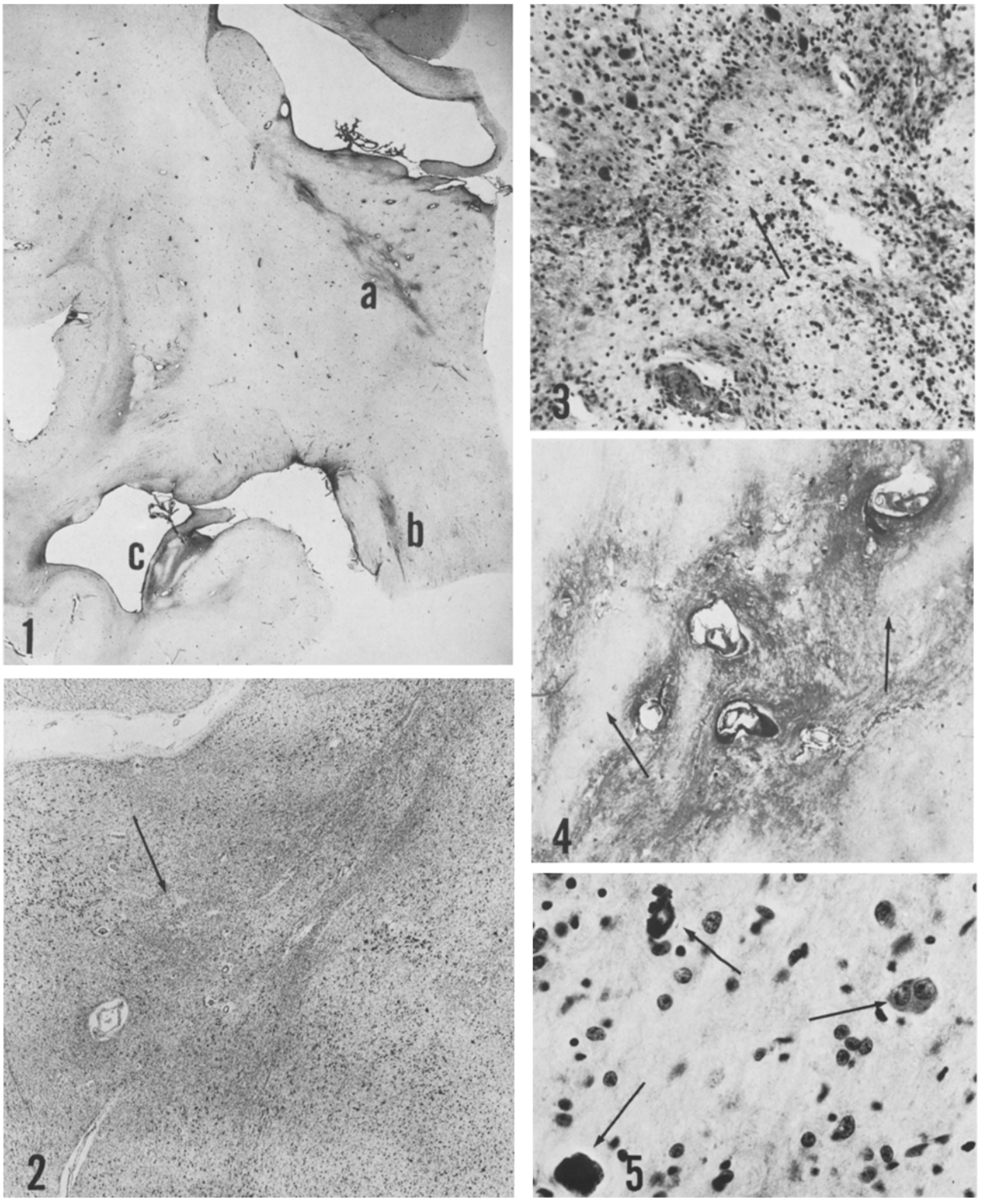

Fig. 1a – c. Localization of main residual pathological lesions of JBE. Case 1. Holzer preparation. **a** Scar in thalamus, **b** scar in substantia nigra, **c** scar in Ammon's horn

Fig. 2. Lesion in thalamus. Case 2. Lesion localized in lamina medullaris medialis thalami and adjacent portions. Arrow: characteristic "LCF". Nissl stain

Fig. 3. Higher magnification of scars in thalamus. Case 3. Characteristic "LCF" (arrow) surrounded by dense glio-mesenchymal scar. H.-E. stain

Fig. 4. Dense gliosis in scar of thalamus. Case 3. Holzer preparation. Arrows: the poor gliosis in "LCF"

Fig. 5. Binucleated and calcified neurons in scar of thalamus. Case 1. H.-E. stain

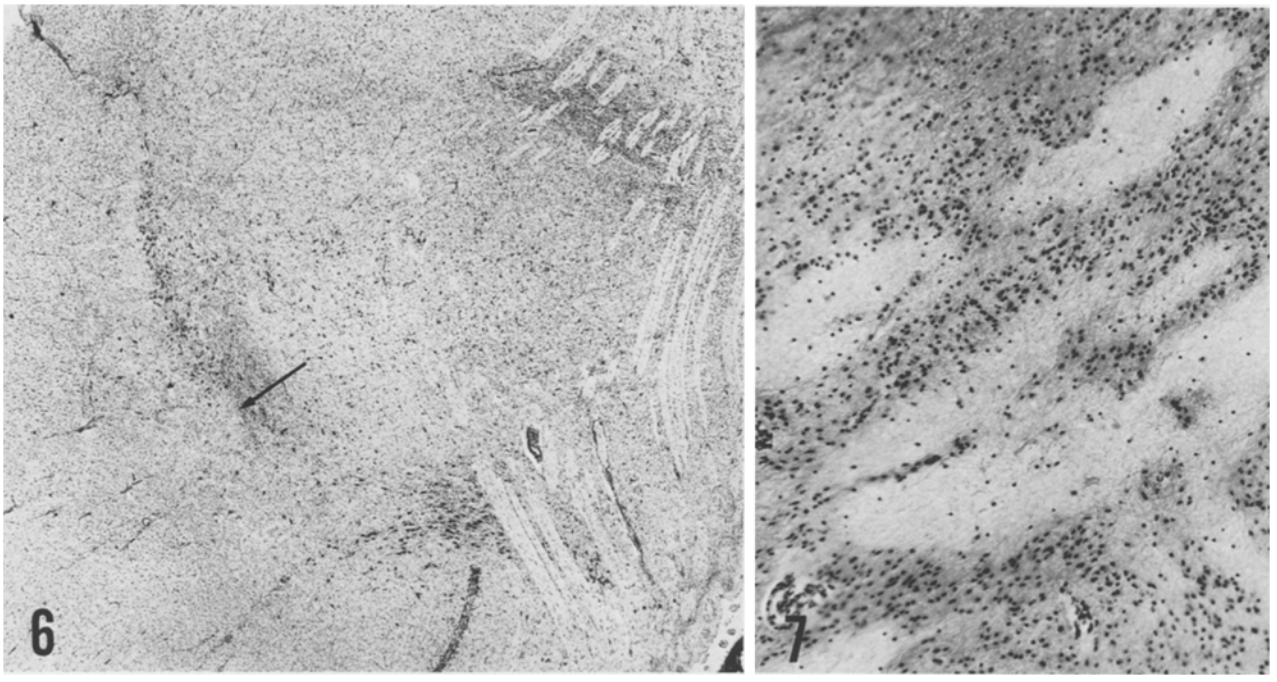

Fig. 6. Lesion in substantia nigra. Lesion occupied middle part of SN. Arrow: "LCF". Case 2. Nissl stain

Fig. 7. Higher magnification of lesion in SN. Cell-deficient, rarefied areas (LCF) were surrounded by dense glio-mesenchymal scar. Case 1. H.-E. stain

of characteristic "LCF" in the thalamus, SN, and occasionally in the cerebral cortex, appearance of calcified and binucleated nerve cells in the thalamic lesions and less frequently in those of the SN, in addition to the absence of acute inflammatory changes such as glial nodules and perivascular cuffing except for a glial nodule or two. "LCF" in the old cases of JBE resembled the "circumscribed necrotic foci" in the brains of acute JBE (Hayashi, 1931; Zimmerman, 1946; Takeya, 1962; Shiraki, 1963). We consider that the "LCF" were actually vestiges of the latter. "LCF" seems to undergo a gradual transformation as the patients survive longer. The boundary between the "LCF" and adjacent tissue in case 1 (12 years survival) was much clearer than that in case 3 (67 years survival). Nevertheless, survival of the "circumscribed necrotic foci" in the brain of an acute JBE for 67 years is surprising, and may mean that the glio-mesenchymal reaction in "LCF" is extremely slow. This slow process which continued for many years, may be a reparative or degenerative one. But a few glial nodules in the brain stem in case 1 and 2 might be interpreted otherwise, i.e. as evidence of slow virus activity as those in the acute stage (Matsuyama, 1962). With regard to the slowly on-going process, we are reminded of postencephalitic parkinsonism of the Economo type, the brains of which also showed continuous decomposition of melanin pigments and the abundant appearance of Alzheimer's neurofibrillary changes. The last mentioned change is one of the most characteristic features of the latter condition, as well as of senile brains and of Parkinsonism-Dementia Complex on Guam. However, we could not find Alzheimer's change in any of the present chronic residual cases of JBE.

Calcified and binucleated nerve cells in the foci (Fig. 5) add another characteristic feature to the lesions in the thalami. Calcified nerve cells and calcium concretions are often found in and around necrotic scars in other conditions, but these are usually few in number. Here they were profuse. Zimmerman (1946) likewise reported widespread calcium concretions in the cerebral cortex and basal ganglia in the brain of acute cases with JBE. Binucleated nerve cells are rare in the CNS. Here many of them were found. What kind of agent or stimulation could induce nuclear divisions or cellular fusion in remaining nerve cells is not clear, but some links between JBE virus and the nuclei of nerve cells might be a matter of speculation (Hamazaki, 1942; Oyanagi, 1966).

We consider that the above combination of features is of diagnostic value. Pette-Doering's encephalitis differs in localization of the lesions and the absence of

"LCF", and of calcified and binucleated nerve cells from the findings in our cases. Economo-type post-encephalitic parkinsonism can be distinguished by the presence of Alzheimer's neurofibrillary changes. Whether old cases of other types of encephalitis such as St. Louis encephalitis or Russian spring-summer encephalitis show similar changes to those of JBE, is an interesting but unresolved problem.

References

Hamada, S., Ishii, T.: Neuropathology of chronic residues of Japanese B encephalitis. Psychiat. Neurol. jap. **68**, 60–78 (1966)

Hamazaki, Y., Watanabe, T.: Intranuclear inclusion body in mice infected by virulences of epidemic encephalitis in Japan and the United States. Okayama Igakkaishi **54**, 555 (1942)

Hayashi, M.: Encephalitis epidemica Japonica. Allg. Z. Psychiat. **95**, 55–58 (1931)

Hayashi, M.: On Japanese B encephalitis. Recent Advanc. Res. Nerv. Syst. **5**, 49–52 (1960)

Haymaker, W., Sabin, A. B.: Topographic distribution of lesions in central nervous system in Japanese B encephalitis, nature of the lesions, with report of a case on Okinawa. Arch. Neurol. Psychiat. (Chic.) **57**, 673–692 (1947)

Kanno, H.: Neuropathology of epidemic Japanese encephalitis. Report of Hayashi Psychiatric Institute, vol. 1, pp. 1–143 (1959) (Japanese edition)

Kunimoto, G.: Histopathologic examinations of central nervous system in protracted cases of encephalitis Japonica. Kyoshu Neuropsychiat. **8**, 63–79 (1960)

Matsuyama, H.: Pathology of Japanese encephalitis. Problem of viral multiplication within cells. Recent Advanc. Res. Nerv. Syst. **6**, 87–89 (1962) (English-Japanese edition)

Oyanagi, S.: An electron microscopic study on the fine structure of cerebral cortex in the mous infected with Japanese encephalitis virus. Neuropath. pol. **4**, 457 (1966)

Shiraki, H., Goto, A., Narabayashi, H.: État passé et présent de l'encéphalite Japonaise au Japon. Rev. neurol. **108**, 633–699 (1963)

Shiraki, H., Fujisawa, K.: A case of protracted (151 days) Japanese B encephalitis. Recent Advanc. Res. Nerv. Syst. **13**, 309–317 (1969)

Shinohara, T., Morimatsu, Y., Hayashi, N., Koya, G., Murofushi, K., Shiraki, H.: An autopsy case of postencephalitis Japonica at infant on 4th month. Recent Advanc. Nerv. Syst. **17**, 337–345 (1973)

Suwa, N.: Patho-anatomy of encephalitis Japonica. Encephalitis Japonica in 1948–1949, problem commission for encephalitis Japonica, pp. 25–30 (1950) (Japanese edition)

Takeya, S.: Histopathology of Japanese summer encephalitis. Recent Advanc. Res. Nerv. Syst. **6**, 75–87 (1962) (Japanese edition)

Zimmerman, H. M.: The pathology of Japanese B encephalitis. Amer. J. Path. **22**, 965 (1946)

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