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Abstract

Human brain turnors removed from 126 patients were histochemically examined and following results were obtained. 1. In general, alkaline phosphatase activity is decreased in poorly differentiated gliomas, but is not related to the tumor cell infiltration. 2. All the cases of alkaline phosphatase negative gliomas have poor reconvalescent course and most of the positive cases show good reconvalescence. 3. Alkaline phosphatase, leucine aminopeptidase and acid phosphatase activities are remarkable in fibroblastic meningioma, moderate or feeble in meningocytic meningioma, and negative in malignant meningioma. 4. The activities of alkaline phosphatase, β-esterase, leucine aminopeptidase and acid phosphatase are decreased in most of meningocytic meningiomas when the duration of symptoms and signs is short. 5. Succinic dehydrogenase, malic dehydrogenase, isocitric dehydrogenase and β-glucuronidase are strongly reactive in malignant meningioma; from strong to moderate in meningocytic meningioma and from moderate to feeble in fibroblastic meningioma. 6. There is a slight increasing tendency of the activities of succinic dehydrogenase, malic dehydrogenase, isocitric dehydrogenase in fibroblastic meningioma and p·glucuronidase for a short duration of symtoms and signs. 7. In the case of acoustic neurinomas the higher the alkaline phosphatase activity, the longer is the duration of symptoms and signs.

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THE RELATIONSHIP BETWEEN HISTOCHEMICAL ENZYME ACTIVITIES OF BRAIN TUMORS AND CLINICAL FEATURES OF THE PATIENTS

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There are many reports of the histochemical studies on brain tissues and tumors concerning to their enzyme activities but the relation between the clinical feature and the enzyme activity of brain tumors has not yet been systematically studied. In order to make this point clear, the authors made the histochemical observations on human brain tumors referring to the clinical symptoms. In this paper it is reported that there are some relation between the appearance and duration of clinical symptoms and the enzyme activities of tumor tissues.

MATERIALS AND METHODS

The materials used were human brain tumors removed from 126 patients admitted to the attached hospital of Okayama University Medical School, Surgery and Neurosurgery Department, from June 1965. Two-thirds of these specimens were kept at −20°C immediately after the removal, and the rest were frozen quickly by ice aceton mixture. With these two groups of tumor specimens frozen sections of 20 microns in thickness were prepared in a −20°C cryostat for histochemical observations. The rest part of tumors was fixed in 10 per cent formol for the routine histologic observation, paraffin sections and hematoxylin eosin stain. For the histochemical demonstration of hydrolytic enzymes, the tissue sections were fixed in 10 per cent cold formalin and rinsed in distilled water, and then incubated with the specific medium prepared for each enzyme as follows. Alkaline phosphatase: 10 mg of sodium α-naphthyl phosphate were dissolved in 20 ml of CLARK and LUB's buffer at pH 9.2 and 20 mg of fast blue B were added. The sections were incubated at 20°C for 30 minutes, dehydrated and mounted in balsam. Acid phosphatase: 10 mg of sodium α-naphthyl phosphate were dissolved in 20 ml of acetate buffer at pH 5.8, to which 20 mg of fast blue B were added. The sections were

incubated for one hour at 20°C, dehydrated and mounted in balsam. β-esterase: 5～10 mg sodium β-naphthyl acetate were dissolved in 1 ml of aceton, and 20 ml of MICHAELIS buffer at pH 7.2 and 20 mg of fast blue B were added. Incubated at 20°C, for 30 minutes and mounted in glycerin. β-glucuronidase: The method of SELIGMAN *et al.* (1954) was applied, in which 6-bromo-2-naphthyl-β-D-glucuronide was used as the substrate. Leucine aminopeptidase: The method of NACHLAS *et al.* (1957) was used.

For the demonstration of oxidative enzymes, the sections dried at room temperature were incubated with the following media. Succinic dehydrogenase: Incubation mixture was composed of 5 ml of 0.2 M sodium succinate, 5 ml of 0.2 M phosphate buffer at pH 7.6, to which 10 ml of nitro-BT aqueous solution (1 mg/1 ml) were added. The sections were incubated in the mixture at 37°C for 30 minutes, fixed in 10 per cent formalin and mounted in glycerin without dehydration. Lactic, glutamic, α-glycerophosphate and β-hydroxybutyric dehydrogenase: Incubating solutions were consisted of 4 ml of 1 M substrate solution, 3 ml of nitro-BT solution (5mg/3ml), 11 ml of 0.1 M phosphate buffer at pH 7.6, 2.5 mg of NAD (100 per cent), 2 ml of 0.1 M KCN and adjusted to pH 7.6 with 0.5 M HCl. Malic dehydrogenase: Incubating solution was composed of 5 ml of 1 M sodium malate, 3 ml of nitro-BT solution (5 mg/3 ml), 10 ml of phosphate buffer (0.1 M) at pH 7.4, 2.5 mg of NAD, 2 ml of 0.1 M KCN and adjusted to pH 7.4 with 0.5 M HCl. Glucose-6-phosphate dehydrogenase: Incubation mixture was consisted of 4 ml of 0.002 M disodium glucose-6-phosphate, 3 ml of nitro-BT solution (5 mg/3 ml), 11 ml of 0.1 M Veronal buffer at pH 7.6, 3 ml each of 0.01 M $MgCl_2$ and 0.5 M $MnCl_2$ solutions and with 7 mg of NADP. Isocitric dehydrogenase: Incubation mixture was consisted of 4 ml of 0.1 M sodium isocitrate, 3 ml of nitro-BT solution (5 mg/3 ml), 11 ml of 0.1 M Veronal buffer at pH 7.4, 2 ml each of 0.01 M $MgCl_2$ and 0.5 M $MnCl_2$ solutions and with 2.5 mg of NADP. For the lactic and malic dehydrogenases, incubation was carried out at 37°C for 30 minutes, and for the other NAD and NADP linked dehydrogenases one hour.

RESULTS

Enzyme activities of tumors:

Gliomas are classified morphologically according to the classification by BAILEY and CUSHING and Armed Forces Institute of Pathology (Table 1). Normal glial cells showed no alkaline phosphatase activity but those in gliosis faint activity. The cells of astrocytoma grade 1 showed a marked reaction of alkaline phosphatase in their cytoplasm (Fig. 1), particularly strong in protoplasmic astrocytoma, and moderate in oligodendroglioma (Fig. 2). Spongioblastoma

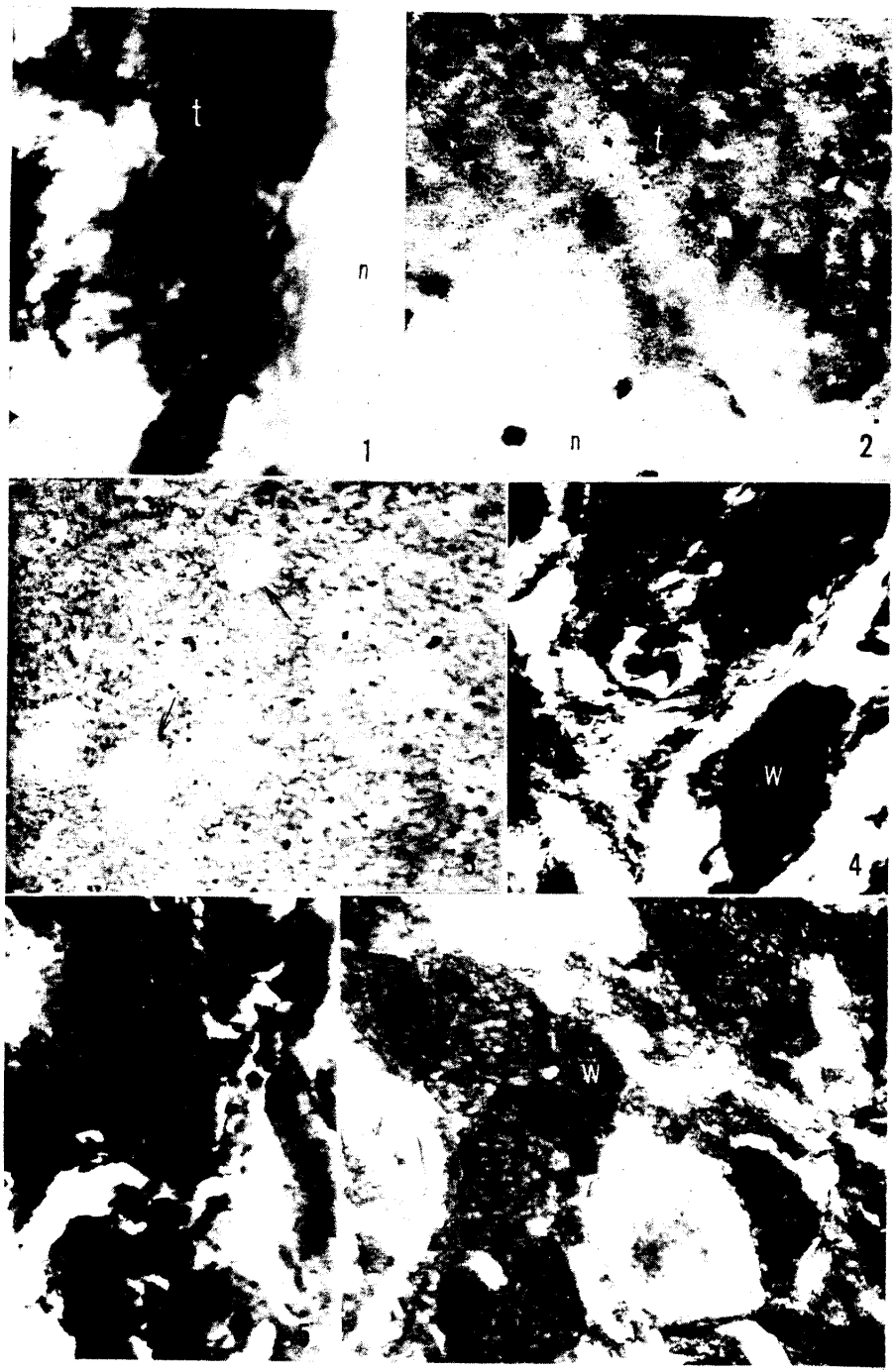
Table 1 Enzymatic Activities in Humar. Brain Tumors

	Astro. 1	Oligo.	Spong.	Epend.	Astro. 2—3	Medull.	Gliobl. Mult.	Fib. Mening.	Men. Mening.	Mal. Mening.	Neurin.	Pineal.	Cranioph.	Pituit. Ad.	Papill. Ep.
Case numbers	6	5	6	6	10	3	10	8	18	3	12	2	5	4	3
ALP	###~##	##	+~−	±~−	±~−	—	±~−	###~##	##~±	—	##~−	###~##	###~+	—	###~##
ACP	##	+	##~±	##~±	##~±	##~+	+	###~±	##~±	±~−	##~−	##~±	##~+	##	###~##
β-EST	+~±	+~−	+~−	+~±	+~±	—	+~−	##~±	###~−	##~−	±~−	±~−	+~±	±	##~+
LAP	—	—	—	—	—	—	—	##~−	+~−	±~−	—	—	—	—	+~±
β-GL	###~##	##~+	##	###~+	##~+	##~+	##	##~±	###~+	###~##	###−±	+~±	+~±	##	##~+
LDH	####~##	###	###	####~##	###~##	###~##	###~##	####~##	####~##	###~##	####~##	##~+	###~##	###~##	###~##
G6PDH	##~±	##~+	##~±	##~+	##~±	##~+	##~±	###~+	###~+	##~+	##~+	##~+	##~±	###	###~##
SDH	###~##	##	+~±	##	###~+	##~+	##~±	##~±	###~##	###~##	##~+	###~±	###~+	##~+	+
MDH	###~##	###~##	##	###~##	###~+	###~##	###~##	###~+	###~##	###~##	###~##	+~±	###~+	###~##	###~##
ICDH	##~±	+~±	+~±	##~±	+~±	##~+	##~−	##~±	###~±	####~±	##~+	##~+	##~±	##	##
GDH	###~+	##	##	###~+	##	##	###~+	###~+	###~+	###~+	###~##	+~±	###~+	###~##	###
α-GDH	##	###=+	##~±	##~+	##~+	##	##~+	###~+	###~+	###~+	##~+	##~+	###~##	###~##	###~##
β-HDH	+	+	+~±	##~±	##~±	+~±	##~±	##~±	##~±	##~±	##~±	##~±	##~±	##	##~+

The Enzymatic Activities of Specimens Stained Histochemically were Graded Microscopfoally on the Basis of Color Relation in − to ### by Inspection of All Cells. −: Complete negative, ±: Faint, +: Slight, ##: Moderate, and ###: Intense Staining

Astro.: Astrocytoma
Oligo.: Oligodendroglioma
Spong.: Spongioblastoma polare
Medull.: Medulloblastoma
Gliobl. Mult.: Glioblastoma multiforme
Fib Mening.: Fibroblastic Meningioma
Men. Mening.: Meningocytic meningioma
Mal. Mening.: Malignant meningioma
Neurin.: Neurinoma
Pineal.: Pinealoma
Cranioph.: Craniopharyngioma
Pituit. Ad.: Pituitary adenoma
Papill. Ep: Papillary ependymoma

ALP: Alkaline phosphatase
ACP: Acid phosphatase
β-EST: β-Esterase
LAP: Leucine aminopeptidase
β-GL: β-Glucuronidase
LDH: Lactic dehydrogenase
G6PDH: Glucose-6-phosphate dehydrogenase
SDH: Succinic dehydrogenase
MDH: Malic dehydrogenase
ICDH: Isocitric dehydrogenase
GDH: Glutamic dehyrogenase
α-GDH: α-Glycerophosphate dehydrogenase
β-HDH: β-Hydroxybutyric dehydrogenase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presented a weak activity in its process. The less differentiated gliomas displayed nearly negative reaction. In general, alkaline phosphatase reaction was increased in the surrounding tissues of the blood vessels, negetive in the parts of calcium salt deposition. Capillaries and corium of the vessels of well differentiated gliomas revealed very high activities, but the reduced activity of alkaline phosphatase was observed in those less differentiated cells. The acid phosphatase activity of normal glial cells was slight or negative and moderate in gliosis. Most of gliomas showed from moderate to negative reaction, that were not related to cell differentiation and located closer to nuclei. Smaller glioma cells had less activity than larger ones having rich protoplasm. In the β-esterase reaction, dye deposition was usually absent in glial cells both in normal and gliotic tissues, excepting the microglia located at the regions surrounding the vessels which gave sometimes a positive reaction. In gliosis over half of tumors displayed various grade of the cytoplasmic activity which was not correlated with morphological malignancy. One outstanding feature to be mentioned is that in most cases of glioma consisted of small tumor cells the activity was negative, and also the cases of oligodendroglioma and medulloblastoma. No leucine aminopeptidase activity was seen in normal brain and glioma tissues. β-glucuronidase reaction was rather intense in the brain cortex than in the medulla, probably due to weak or slight activity in astrocyte and moderate in oligodendroglia. The tissues of gliosis gave a moderate to weak activity and moderate in glioma. Usually dehydrogenase activities were more remarkable in glioma cells than in nomal glia, but no appreciable differences between gliosis and glioma. In glioma cells the strongest reaction was lactic dehydrogenase, moderate reaction malic dehydrogenase, glutamic dehydrogenase, α-glycerophosphate dehydrogenase and succinic dehydrogenase, and rather weak glucose-6-phosphate dehydrogenase, isocitric dehydrogenase and β-hydroxybutyric dehydrogenase. However, no correlationship was fouud between the degree of anaplasia and the enzyme activities. Sometimes large tumor cells showed stronger activities. Necrotic area hardly showed any positive reaction but the tissue adjacent to

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Fig. 1 ALP stain ×100: Astrocytoma grade 1 (t) shows a strong activity and normal cortex (n) negative.

Fig. 2 ACP stain ×100: Showing a moderate activity in oligodendroglioma (t), and no activity in normal medulla.

Fig. 3 SDH stain ×100: Ependymoma is moderately reactive but sucker of rosette formations (↓) is not active.

Fig. 4 LDH stain ×100: The strongest activity is noticed in the areas of whorl formation (w) in meningocytic meningioma.

Fig. 5 ALP stain ×100: Fibroblastic meningioma reveals a remarkable activity.

Fig. 6 ACP satin ×100: The parts of whorl formation in meningocytic meningioma (w) are strongly reactive.

necrosis and hemorrhage gave a slight activity. The vessel walls found in the tumor tissues gave a positive reaction, rather strong one on the thick vessel walls. Concerning the positive reaction, the activities showed a tendency to increase slightly in the part of rosette and cartwheel formation and to reduce in the part of sucker feet (Fig. 3), and these enzyme activities appeared diffusely in the tumor tissue, though slightly stronger at marginal area of tumor in some cases.

Meningioma was classified into three types according to the concept introduced by ZIMMERMAN *et al.*; meningocytic meningioma, fibroblastic meningioma and malignant meningioma (Table 1). Those of mixed type are also found but they were classified to the one whose characteristics appeared more predominant. Normal arachnoidal villi cells served as control for meningocytic meningioma, and leptmeninx cells for fibroblastic meningioma. As a whole enzymatic activities of control tissues were similar to or slightly stronger than those of meningioma excepting malignant ones. Alkaline phosphatase activity was remarkable in fibroblastic type (Fig. 5), from moderate to very weak in meningocytic one (Fig. 6), and negative in malignant meningioma. At the area of whorl formation or concentric arrangement of the cells in the tumor, the highest activity was noticed. The psammoma and its abjacent zone showed no activity and found only in the tumor with positive activities. Alkaline phosphatase activity of the wall of vessels proliferated apparently in meningioma were similar to or slightly weaker than that found in the control. The acid phosphatase activity was moderate to weak in benign meningioma but feeble or negative in malignant ones. In the part of whorl formation the reaction of acid phosphatase showed a similar tendency to that of alkaline phosphatase. β-esterase and leucine aminopeptidase reactions were negative in controls but positive in many cases of the meningiomas, more than one half of the cases. Generally, these enzyme activities appeared weaker in malignant meningioma than in other types of tumor. The activities of β-glucuronidase and dehydrogenases belonging to TCA cyle such as succinic dehydrogenase, malic dehydrogenase and isocitric dehydrogenase appeared inversely to the activity of alkaline phosphatase, i. e. strong to faint activities in benign tumors and rather strong in malignant meningiomas. Among the other dehydrogenases without any relation to the three types of meningiomas, lactic dehydrogenase reaction was strongest (Fig. 4), glucose-6-phosphate dehydrogenase, glutamic dehydrogenase and α-glycerophosphate dehydrogenase stronger and β-hydroxybutyric dehydrogenase weakest. At the part of whorl and concentric arrangement of cells, all of dehydrogenases and β-glucuronidase were intensely active. Generally, benign meningiomas with calcium salt deposition gave a relatively weak dehydrogenase activity than that without calcification, though the calcified area was negative in all the enzymic

activities.

Since neurinomas did not show any histochemical specificity as illustrated in Table 1, it was difficult to identify fibroblastic meningioma, spongioblastoma and neurinoma by means of enzyme histochemistry. In neurinomas the lactic dehydrogenase activity was highest among all the enzymes studied, and the others reacted from strong to weak except for alkaline phosphatase, β-esterase and leucine aminopeptidase. Alkaline phosphatase was active in varying degrees, β-esterase very weak or negative, and leucine aminopeptidase not active.

Miscellaneous tumors: In craniopharyngiomas the alkaline phosphatase activity was feeble or negative in the single layers of the columnar cells from the lining of the cysts, and strong to weak in squamous epithelial cell layers. Acid phosphatase and β-glucuronidase showed moderate to strong reactions in epithelium. However, β-esterase activity was very weak and no leucine aminopeptidase activity. Dehydrogenase activities were marked in the single cell layer and in the outer side of squamous epithelium layer, and low in the inner part of epithelium. All pituitary adenomas investigated were of chromophobe type in which dehydrogenase activities were marked except for isocitric dehydrogenase, glutamic dehydrogenase and β-hydroxybutyric dehydrogenase. Acid phosphatase, β-glucuronidase, isocitric dehydrogenase, glutamic dehydrogenase and β-hydroxybutyric dehydrogenase were moderate, and alkaline phosphatase, β-esterase and leucine aminopeptidase were almost negative. In pinealomas heavy alkaline phosphatase, succinic dehydrogenase and β-glycerophosphate dehydrogenase were noted. Lactic dehydrogenase, glucose-6-phosphate dehydrogenase, isocitric dehydrogenase, β-hydroxybutyric dehydrogenase, acid phosphatase, β-esterase, β-glucuronidase, glutamic dehydrogenase and malic dehydrogenase were moderately active, and leucine aminopeptidase not active. Large epitheloid cells in this tumor had more predominant enzymatic activities than lymphoid cells. Capillary ependymomas were remarkable in the activities of alkaline phosphatase, acid phosphatase, lactic dehydrogenas, glucose-6-phosphate dehydrogenase, malic dehydrogenase, α-glycerophosphate dehydrogenase and glutamic dehydrogenase. β-esterase, β-glucuronidase, succinic dehydrogenase, isocitric dehydrogenase and β-hydroxybutyric dehydrogenase were of moderate activity and leucine aminopeptidase weak. Capillary wall showed the strongest activity of alkaline phosphatase in these miscellaneous tumors.

Relationship between histochemical enzyme activities and clinical features:

The records of histochemical observations were studied referring to the clinical symptoms of the limited patients survived through the surgical operation whose reconvalescent courses were traced in 92 per cent of the cases. Fatal cases due to operation and those given irradiation treatment were discarded from the statistical observation.

Glioma: Reconvalescent course was poor in many cases of histologically malignant gliomas showing weak alkaline phosphatase activity, especially in all 12 cases with no alkaline phosphatase activity (Table 2). Exceptional cases

Table 2 Prognosis of Cases Operated on Gliomas

Type of tumors		Astro. I	Oligo.	Spong.	Epend.	Astro. II/III	Medull.	Gliobl.
Prognosis	good	3	2	1	2	0	1	0
	poor	4	1	2	2	4	0	4
ALP activity		--	±	+	++	+++	++++	
Prognosis	good	1	2	2	2	2	0	
	poor	12	1	2	1	0	0	
G6PDH activity		−	±	+	++	+++	++++	
Prognosis	good	0	1	6	1	0	0	
	poor	0	2	4	7	0	0	
SDH activity		−	±	+	++	+++	++++	
Prognosis	good	0	1	1	7	0	0	
	poor	0	2	4	8	2	0	

Numerals iudicate the case number observed.
Abbreviation and symbols: Refer to Table 1.

with positive alkaline phosphatase activity, in spite of poor prognosis, had infiltrating tumors (Table 3). Other enzyme activities showed not any specific

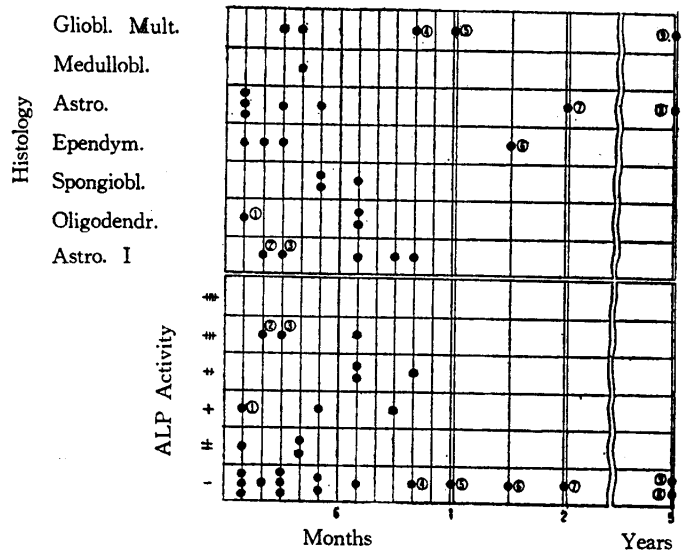
Table 3 ALP Positive Cases in Poor Reconvalescent Group on Glioma

ALP activity	No. of cases	Type	Weight of excised tumor	Growing pattern
+++	1	Astro. I	70g	infiltrative
++	1	Oligo.	150g	relatively circumscribed
+	2	Astro. I	25g	infiltrative
		Astro. I	100g	infiltrative

Abbreviations and symbols: Refer to Table 1.

relation with the prognosis (Table 2). In two-thirds of the cases, the longer the duration of clinical symptoms and signs, the more benign was the histological pictures and the stronger in alkaline phsphatase activity. However, in the others from 1 to 9 as shown in Table 4, no specific relation was found between the

Table 4 Duration of Symptoms and Signs of Cases Operated on Gliomas



Duration of symptoms and signs means the period from the first complaint of glioma to the operation.
Abbreviations and symbols: See Table 1.

duration of symtoms and the enzyme activity and to the extent of anaplasia. Figure 1 in the circle denotes a case of oligodendroblastoma, 2 and 3 are those found in early stage, and figures 4 to 9 are truely exceptional ones. Other enzyme activities were not correlated with the duration of symptoms and signs. Chemical analyses of urine, stool, blood, blood serum and cerebrospinal fluid were likewise not related with histochemical results.

Meningiomas: Prognosis of meningiomas was dependent on its localization (Table 5), and it was not correlated with histological and enzymatic histochemical findings in the present study (Table 6). The cases of a short duration

Table 5 Relation between Enzymatic Activities of Meningioma of Poor Reconvalescent Cases and the Localization

Localization of Tumors	ALP	β-EST	LAP	SDH
Frontal Basis	++	++	−	++
Sphenoidal Ridge	+++, ±	−, −	−	±, ++
Supra Sellar	±	+	±	++
Parasagittal	−, +	++, −	±, ±	+++, ++
Convexity	±	±	−	++
Falx	±	−	−	++

Abbreveations and symbols: See Table 1.

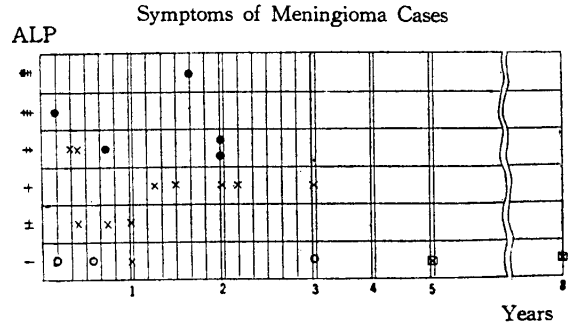
Table 6 Prognosis and Enzyme Activities on Meningiomas

		Fibrobl.		Meningo.		Malignant	
Prognosis	good	3		5		2	
	poor	1		6		1	
ALP activity		−	±	+	++	+++	++++
Prognosis	good	3	1	2	4	0	0
	poor	1	4	1	1	0	1
β-EST activity		−	±	+	++	+++	++++
Prognosis	good	1	1	3	4	1	0
	poor	4	1	1	2	0	0
LAP activity		−	±	+	++	+++	++++
Prognosis	good	6	1	0	3	0	0
	poor	4	3	0	0	0	0
SDH activity		−	±	+	++	+++	++++
Prognosis	good	0	0	5	2	3	0
	poor	0	1	0	6	2	0

Abbreviation and symbols: See Table 1.
Numerals indicate the case numbers observed.

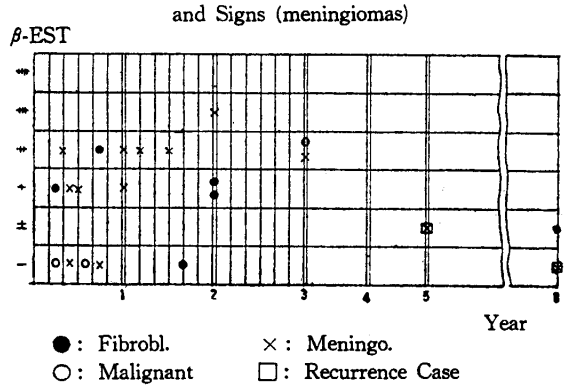
of symtoms and signs were suggestive of benign meningioma. However, there was such a tendency that the stronger the alkaline phsphatase, acid phosphatase, β-esterase and leucine aminopeptidase activities, the longer was the duration of symptoms and signs in meningocytic meningioma (Tables 7, 8), and the weaker

Table 7 ALP Activity and Duration of Signs and Symptoms of Meningioma Cases



●: Fibrobl., ○: Meningo., ×: Malignant, □: Recurrence Case (period from the lst operation to the second op.)

Table 8 β-EST Activity and Duration of Symptoms and Signs (meningiomas)



the succinic dehydrogenase, malic dehydrogenase, isocitric dehydrogenase and β-glucuronidase activities the longer was the duration in symptoms and signs in fibroblastic meningioma (Table 9). Other clinical features as described of gliomas did not show any relationship with the enzyme activities of tumor.

Table 9 SDH Activity and Duration of Signs and Symptoms of Meningioma Cases

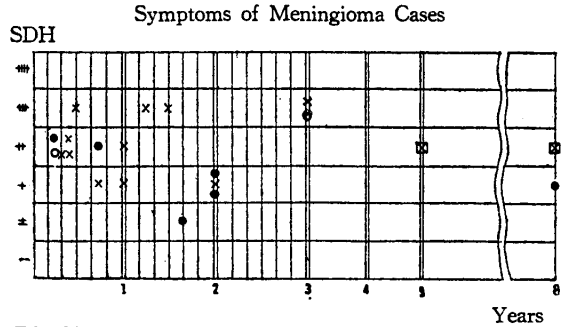
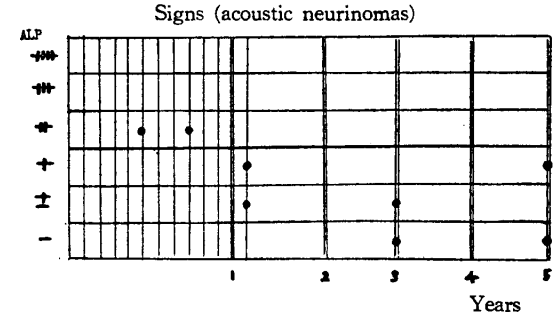


Table 10 ALP Activity and Duration of Symptoms and Signs (acoustic neurinomas)



Acoustic neurinomas: All the cases showed a good reconvalescent course. There was an increasing tendency of the alkaline phosphatase activity for a long duration of symptoms and signs (Tablo 10), but other enzymes revealed no correlation with the clinical features.

DISCUSSION

LANDOW *et al.* and OCONNOR *et al.* observed histochemically a similar tendency of alkaline phosphatase activity in gliomas as obtained in our present study, and NASU *et al.* have observed 66 cases of gliomas and reported that alkaline phosphatase activity is rather strong in astrocytic tumor cells, weak in oligodendroglioma, and inconstant in glioma, consisting of round shape cells. FEIGIN *et al.* have presented the opinion that there is no correlation between alkaline phosphatase activity and the degree of anaplasia in 21 cases of gliomas. In the present study the extent of anaplasia was correlated with alkaline phosphatase activity of gliomas and all cases having no activity showed a poor reconvalescent course. Therefore, alkaline phosphatase activity offers an excellent clue to predict the prognosis. However, because the fact that the enzyme activity is not related to the tumor cell infiltration, it is important to conduct microscopic observation of gliomas sufficiently before the removal of tumors and with hematoxylin-eosin stain specimens of the tumors. Concerning the acid phosphatase activity there are works by NAS *et al.* and WOLF *et al.* They have described that acid phosphatase reaction in spongioblastoma and oligodendroglioma is weaker than that in another gliomas. These results are similar to the present results. As for the β-esterase activity in gliomas, virtually the same results were obtained as did NASU *et al.* According to ALLEN about the activity of β-glucuronidase in glioblastoma multiforme reveals a strong activity, ependymoma moderate, astrocytoma and oligodendroglioma weak. In the present study, however, the activity proved to be not uniform; from strong to weak in every type of gliomas. Dehydrogenase activities of malignant gliomas are remarkable according to MOSSAKOWSKI and PAXON, while NASU *et al.* have stated that every glioma had an unpredictable dehydrogenase activity as in the present survey. As mentioned in the present communication, there can be recognized no correlation between the activity of the enzymes except alkaline phosphatase and histology of gliomas on one hand, and between the enzyme activity and clinical features of gliomas. NASU studied histochemically alkaline phosphatase of 43 meningiomas and stated that the activity of meningocytic meningioma was strong to moderate, fibroblastic strongest and rapid growing, negative. BÜTTGER *et al.* also reported the same tendency. Moreover, LANDOW *et al.* and NASU have found that the meningiomas with calcifications show a considerable activity of

alkaline phosphatase. The results of the present study agree well with theirs. It is interesting to note that alkaline phosphatase reaction is strongest in the part of whorl formation but negative in calcium salt itself, its adjacent zone and in its central part probably presaging calcification in near future. Perhaps alkaline phosphatase has not only a certain connection with the tumor growth but also with calcification in meningiomas. Clinical findings of symptoms and signs are mutually associated. The duration of symptoms is so long that alkaline phosphatase, acid phosphatase and leucine aminopeptidase responses are predominant in meningocytic type. According NASU rapid growing meningiomas have more abundant β-esterase than the rest type of tumors in his histochemical observations, but in the present survey malignant ones exhibited varying degrees of activity of β-esterase, and the longer the duration of symptoms and signs, the stronger β-esterase activity in meningocytic type as with the case of alkaline phosphatase. β-glucuronidase, succinic dehydrogenase, malic dehydrogenase and isocitric dehydrogenase activities show an opposite inclination against the alkaline phosphatase activity, and the malignant ones show a remarkable response in agreement with the report of the oxidative enzyme activities by NASU. Succinic dehydrogenase activity in meningocytic meningiomas is not related to the duration of symptoms and signs, but the fibroblastic ones display a tendency to increase succinic dehydrogenase activity for a long duration of symtoms and signs. From these facts, it is assumed that alkaline phosphatase, β-esterase and leucine aminopeptidase play an important role in the growth of meningocytic meningioma. About acoustic neurinomas, there is no enzyme histochemical characteristic feature, however, the duration of symptoms and signs is long when alkaline phosphatase activity is weak, which exactly reverses to the alkaline phosphatase activities in both glioma and meningocytic meningioma. As for miscellaneous tumors, similar clinico-histochemical investigations are yet to be conducted.

SUMMARY

Human brain tumors removed from 126 patients were histochemically examined and following results were obtained.

1. In general, alkaline phosphatase activity is decreased in poorly differentiated gliomas, but is not related to the tumor cell infiltration.

2. All the cases of alkaline phosphatase negative gliomas have poor reconvalescent course and most of the positive cases show good reconvalescence.

3. Alkaline phosphatase, leucine aminopeptidase and acid phosphatase activities are remarkable in fibroblastic meningioma, moderate or feeble in meningocytic meningioma, and negative in malignant meningioma.

4. The activities of alkaline phosphatase, β-esterase, leucine aminopeptidase

and acid phosphatase are decreased in most of meningocytic meningiomas when the duration of symptoms and signs is short.

5. Succinic dehydrogenase, malic dehydrogenase, isocitric dehydrogenase and β-glucuronidase are strongly reactive in malignant meningioma; from strong to moderate in meningocytic meningioma and from moderate to feeble in fibroblastic meningioma.

6. There is a slight increasing tendency of the activities of succinic dehydrogenase, malic dehydrogenase, isocitric dehydrogenase in fibroblastic meningioma and β-glucuronidase for a short duration of symtoms and signs.

7. In the case of acoustic neurinomas the higher the alkaline phosphatase activity, the longer is the duration of symptoms and signs.

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