

Immunological Studies on Esophageal Cancer --Cellular Immunocompetence and Histological Responses in Main Tumor and Regional Lymph Nodes in Esophageal Cancer Patients--

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Introduction

Recently cancer therapy has made remarkable progress in not only operation therapy, but also radiation therapy, chemotherapy, immunotherapy, etc., so that the five year survival rate has remarkably improved. In esophageal cancer, the remote results and prognosis have been improved through the development of the operation method and anesthesia, but in comparison with gastric cancer the five year survival rate of esophageal cancer is very low and only twenty percent¹⁹⁾. The cancer is difficult to diagnose in the early stage. Another reason for the low five year survival rate is the advanced age of most patients⁸⁾. Furthermore it metastasizes extensively and the cell-mediated immunity is remarkably suppressed as compared with other cancers²⁴⁾. Then, as the influential factors of the prognosis of esophageal cancer, in accordance with guide lines for the clinical and pathologic studies on carcinoma of the esophagus, the invasion to the adventitia ("a" factor), the degree of lymph node metastasis ("n" factor), the stage, etc., are all investigated. But these factors do not refer to the host's responses to the cancer.

In the past the cellular infiltration around the tumor (lymphocytes and plasma cells infiltration), and the response of the regional lymph node (sinus histiocytosis and follicular hyperplasia) have been studied. Also the relation between these responses and the prognosis have been investigated. Because of this the author examined the cell-mediated immunity of the esophageal cancer patients using the skin tests (SK-SD, Candida, PHA, PPD) and phytohemagglutinin-stimulated lymphocyte transformation, and observed the histological findings of the main tumor and the regional lymph nodes in the resected specimens and investigated the relation between these reactions and the prognosis.

Case selection

One hundred and eleven cases of primary esophageal cancer were resected at the 2nd Surgical Clinic, Yamaguchi University Hospital, from 1970 to June, 1978. Of these 111 cases, thirty-

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three cases which had preoperative radiotherapy and thirty-three cases which hadn't preoperative therapy were investigated histologically on the resected main tumors and regional lymph nodes, with the exception of the absolute-non-curative cases. From 1978 to 1979 skin tests were performed in 19 cases of esophageal cancer. Also PHA-stimulated lymphocyte transformation was studied in 42 cases.

Items of examination & Method of evaluation

1. Skin Tests

- a) SK-SD
Streptokinase-streptodornase (Torii): 10 unit/0.1 ml
- b) Candida
Protein of Candida (Torii): 500 dil./0.1 ml
- c) PHA
Phytohemagglutinin (Wellcom): 5 μ g/0.1 ml
- d) PPD
Purified protein derivatives (Jpn. BCG): 0.05 μ g/0.1 ml

The above four antigens were injected into the skin of the flexion side of the forearm in the doses of 0.1 ml. After 24 hours the length of the erythema was measured. The value of measurement was half of the short diameter plus the long diameter of the erythema, and according to the criteria decided in Table 1, the values were evaluated.

2. Measurement of the lymphocyte reactivity

The lymphocyte reactivity was studied according to the method by SUZUKI, et al²⁸⁾, which was partly improved by the author. Peripheral blood, collected in preservative-free heparin (10 u/ml), was obtained aseptically from the patient. The blood was diluted with 16 volumes of RPMI-1640 culture medium (Gibco) containing 20% fetal calf serum (FCS, Gibco). Aliquots of the cell suspension (0.1 ml), were distributed to each well of the microplate (Falcon). Each of the triplicate cultures received 30 μ g/ml PHA (Difco). The microplate was cultured in 37°C. CO₂-incubator for ninety-six hours, after which each of the wells received 5 μ C/ml ³H-thymidin and the microplate was cultured for an additional twenty-four hours. Before the cultures were harvested with distilled water, each of the wells

Table 1. The criteria for evaluating the results of the skin tests.

	(-)	(+)
SK-SD	0~9 mm	10~
Candida	0~9 mm	10~
PHA	0~24 mm	25~
PPD	0~9 mm	10~

Table 2. Scores of histological responses.

Lymphocyte infiltration		Follicular hyperplasia	
Score	Detail	Score	Detail
0	Absent	0	Germinal centers rare
1	Present (+)	1	Germinal centers are recognisable in cortex
2	Present (++)	2	Germinal centers are recognisable in cortex medulla
3	Present (###)	3	Germinal centers are recognisable in all lymphnode areas

Plasma cell infiltration (number per High Power Field)		Sinus histiocytosis	
Score	Detail	Score	Detail
0	0	0	Sinus 0 in the lymphnode area
1	0.5~2 per HPF	1	Sinus below 1/4 in the lymphnode area
2	3~10 per HPF	2	Sinus 1/4~1/2
3	More than 10 per HPF	3	Sinus more than 1/2

had added 400 dil. waihem-7 (Toa), were lightly vibrated on the micromixer for 3 minutes, and sat still for 2 minutes. Samples were counted in a liquid scintillation counter.

3. Structural response

The histological specimens of the main tumor and regional lymph nodes of esophageal cancer (Hematoxylin-eosin stain) were observed. These structural responses were evaluated as shown in Table 2, according to the reports of IRIS, et al.¹⁷⁾ and HASHIMOTO, et al.¹¹⁾ Namely the lymphocyte and plasma cell infiltration (lymphocyte infiltration scores+plasma cell infiltration scores=cellular infiltration scores [C.I.]) were studied in the main tumor, and follicular hyperplasia (F.H.), which is the degree of the prominence of germinal centers in

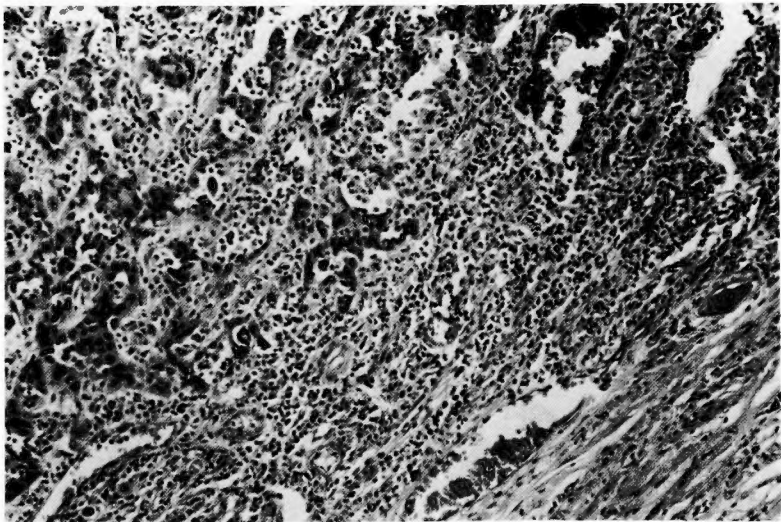


Photo. 1 Marginal portion of the tumor with extensive infiltration of lymphocytes and plasma cells (C.I. 3) $\times 80$

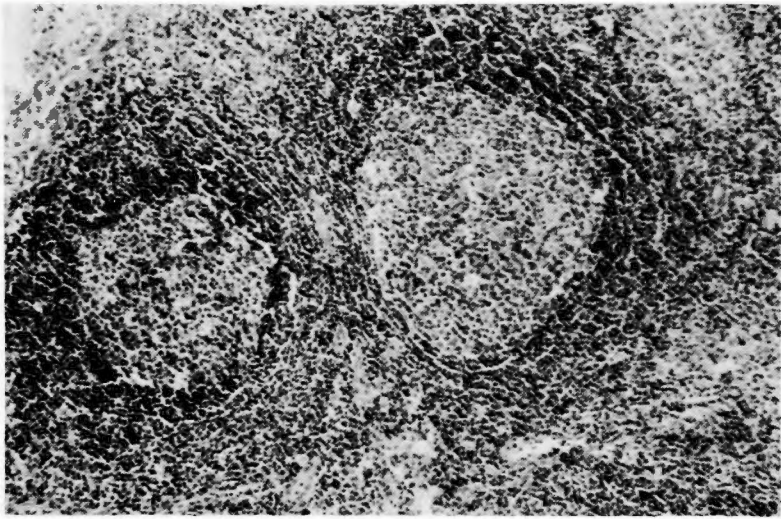


Photo. 2 Portion of the lymph node in which germinal centers were found (F.H. 3) $\times 80$

lymph node, and sinus histiocytosis (S.H.), which is the degree of the prominence of sinus in lymph node, were studied in the regional lymph nodes. The resected lymph nodes were evaluated in all specimens, and the most dominant scores were taken. The highest scores of C.I., F.H., and S.H. are shown in Photo. 1, 2 and 3.

Results

1. Skin test

The four type skin tests were evaluated according to the above criteria, and the esophageal cancer patients were compared with healthy subjects in their twenties.

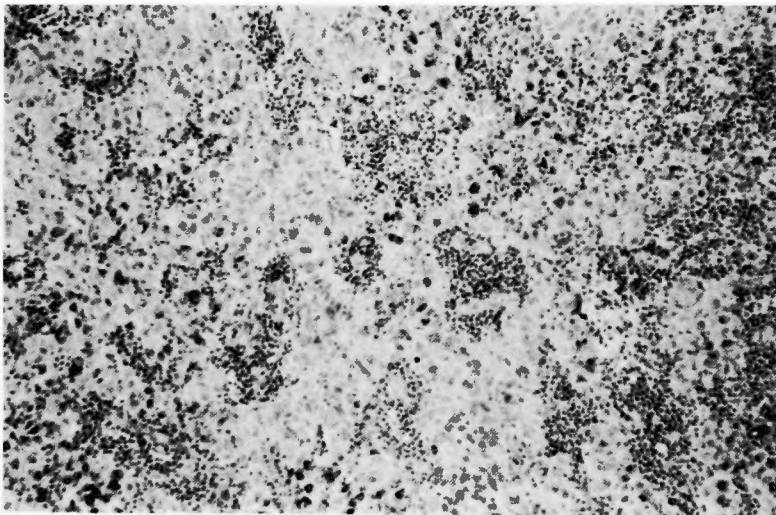


Photo. 3 Portion of the lymph node in which histiocytes proliferated (S.H. 3) $\times 80$

Table 3. Positive percentage of the skin tests in the healthy subjects and the esophageal cancer patients.

Groups \ Skin test	SK-SD*	Candida*	PHA**	PPD
Control	7/14 (50%)	12/14 (86%)	14/14 (100%)	11/14 (78%)
Esophageal cancer patients	4/19 (21%)	6/19 (32%)	9/19 (47%)	6/19 (32%)

* $P < 0.05$, ** $P < 0.01$

- a) Comparison between the esophageal cancer patients and healthy subjects in the skin tests (Table 3).

A positive reaction of SK-SD was observed in 50% of the healthy subjects, as against 21% of the esophageal cancer patients. The significant difference from the sampling point of view was ($P < 0.05$). A positive reaction of Candida was observed in 86% of the healthy subjects, as against 32% of the esophageal cancer patients. The significant difference was ($P < 0.05$). A positive reaction of PHA was observed in 100% of the healthy subjects, as against 47% of the esophageal cancer patients. The significant difference was ($P < 0.01$). The positive reaction of PPD was observed in 78% of the healthy subjects, as against 32% of the esophageal cancer patients. No significant difference between the two was found. As the esophageal cancer patients showed obvious decrease in skin reactivity to SK-SD, Candida and PHA, it was considered that cell-mediated immunity was depressed in the esophageal cancer patients.

- b) The summarized judgment of the four-type skin tests (Table 4).

More than 3 positive reactions in four-type skin tests were shown in 100% of the healthy subjects while they were evident in only 15% of the esophageal cancer patients. The remarkable significant difference from the sampling point of view was ($P < 0.01$). The use of the 4 types of skin tests showed the obvious difference more than any single test detected.

- c) Comparative study of the skin tests and lymphocyte count in preoperative and postoperative periods (Fig. 1).

In preoperative and immediate postoperative periods, lymphocyte count and skin

Table 4. The summarized judgment of the four types of skin tests in the esophageal cancer patients.

$P < 0.01$		
	Number	%
0	4	21
+	6	32
++	6	32
+++	2	10
++++	1	5

* In healthy persons, the percentage of more than +++ was 100%.

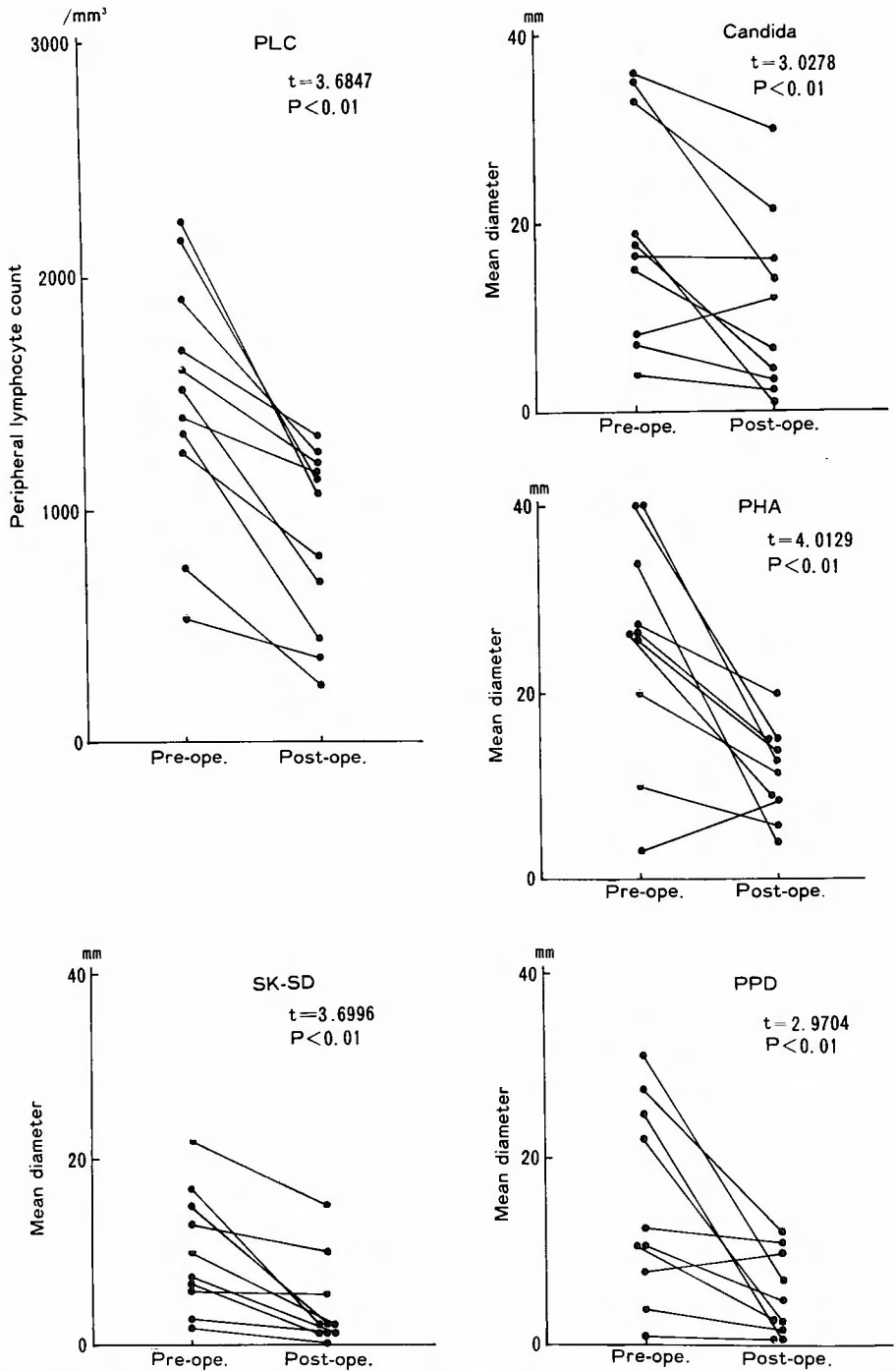


Fig. 1. Comparative study of the skin tests and lymphocyte count in preoperative and postoperative periods.

reactions of SK-SD, Candida, PHA and PPD were measured in the esophageal cancer patients. In the postoperative period, all values significantly decreased, as compared with values in the preoperative period. As a result, in the postoperative period it was observed that the cell-mediated immunity of the patients were remarkably suppressed.

Evaluation of lymphocyte reactivity using whole blood (Fig. 2).

In PHA-stimulated lymphocyte reactivity, more than 10000 cpm values were observed in 18 among 23 (78%) of the healthy subjects, while on the other hand, in only 4 among 42 (10%) of the esophageal cancer patients. The significant difference was ($P < 0.01$). The result showed the remarkable depression of cell-mediated immunity in the patients, as shown in the skin tests.

Histological reactions

The author studied each of the preoperative radiotherapeutic group and the preoperative non-therapeutic group.

a) Histological reactions of the preoperative therapeutic group

Thirty-three cases irradiated before operation were studied. Twelve cases were confirmed in the prognosis except in the direct operative dead cases. Six cases among

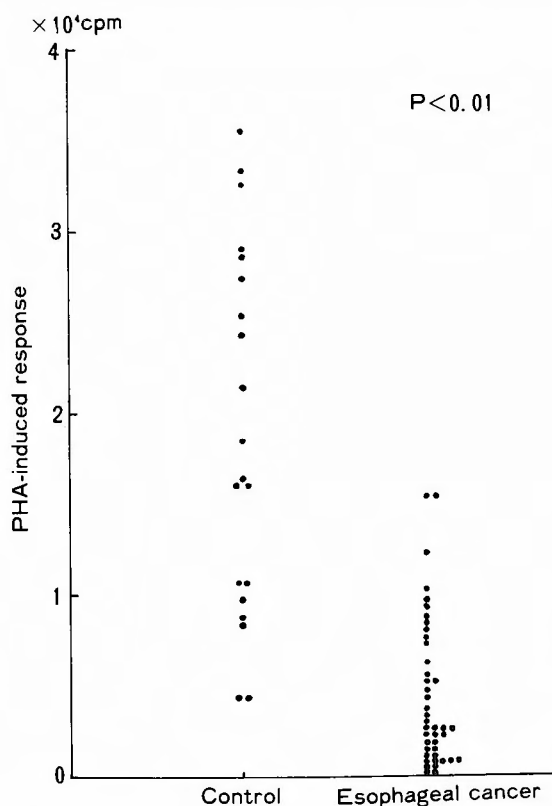


Fig. 2. Comparison between the healthy subjects and the esophageal cancer patients in evaluating lymphocyte reactivity using whole blood.

twelve survived more than one year.

- 1) Comparison between scores of cellular infiltration and the interval between the end of the preoperative radiotherapy and the operation (Fig. 3).

The author studied how cellular infiltration around the tumor changed during the period from the end of the preoperative radiotherapy to the operation. It was noted that cellular infiltration did not correlate with the length of this interval. Fourteen among thirty-three cases (42%) showed 0~2 scores of C.I., seven (21%) showed 3~4 scores, eleven (36%) showed 5~6 scores. The depression of cellular infiltration was observed in a high percentage after radiation.

- 2) Relation between prognosis and "a" factor, "n" factor and stage (Table 5).

In the preoperative radiotherapy group, the author studied the relation between prognosis of the esophageal cancer patients and "a" factor, "n" factor and stage. In less-than-one year survival cases, those who died of a recurrence of the cancer were two cases in "a" 0~1, two in "a" 2 and two in "a" 3. Also in these cases there were three cases in "n" 0~1, one in "n" 2 and two in "n" 3, and there were no cases in stage I~II, two in stage III, four in stage IV. On the other hand, in more-than-

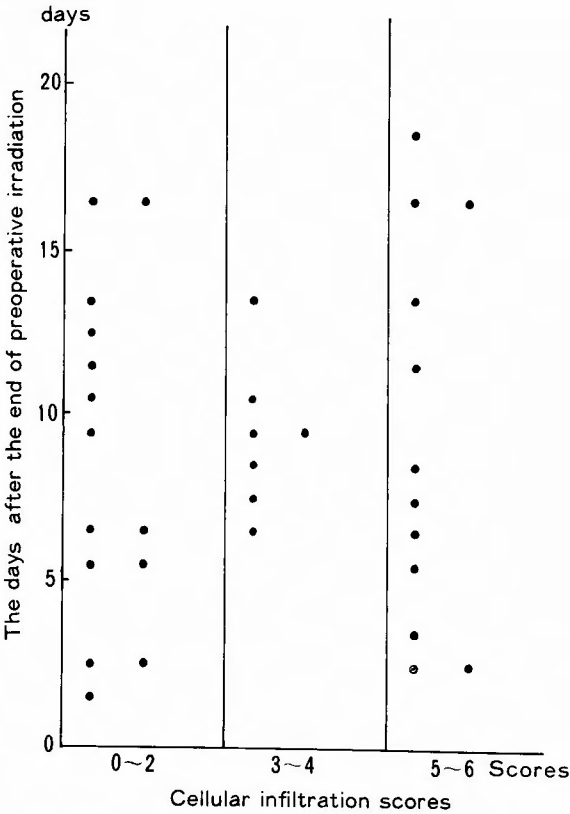


Fig. 3. Comparison between the scores of cellular infiltration and the interval between the end of the preoperative radiotherapy and the operation.

Table 5. The relations between "a" factor, "n" factor, stage and the prognosis in the preoperative radiotherapeutic group.

A factor & prognosis (N.S.)			n factor & prognosis (N.S.)			Stage & prognosis (N.S.)		
Factor \ Survival year	1 <	1 ≥	Factor \ Survival year	1 <	1 ≥	Stage \ Survival year	1 <	1 ≥
a ₀	2	1	n ₀	1	3	Stage I		
a ₁	0	1	n ₁	2	0	Stage II	0	1
a ₂	2	2	n ₂	1	1	Stage III	2	1
a ₃	2	2	n ₃	2	2	Stage IV	4	4

one year survival cases, there were two cases in "a" 0~1, two in "a" 2, and two in "a" 3. Also in these cases there were 3 cases in "n" 0~1, one in "n" 2 and two in "n" 3, and one case in stage I~II, one in stage III and four in stage IV. These factors did not correlate with prognosis.

3) Relation between histological responses and prognosis (Table 6).

Cellular infiltration equal to 0~4 scores were observed in five among six patients who died of recurrence of the cancer in less than one year. On the other hand, 5~6 scores were observed in all six patients who survived more than one year. The significant difference was ($P < 0.01$). Sinus histiocytosis equal to 0~1 score was observed in all six patients who died of a recurrence of the cancer in less than one year. But it equaled 2~3 scores in all six patients who survived more than one year. Significant difference was ($P < 0.01$).

Follicular hyperplasia equal to 0~1 score was observed in three among six patients who died of a recurrence of the cancer in less than one year. But it equaled 2~3 scores in five of six patients who survived more than one year. The obvious difference was ($P < 0.25$). From the above, cellular infiltration and sinus histiocytosis correlated well with prognosis.

b) Histological reaction of the preoperative non-therapeutic group

Thirty-three cases, who were not treated before operation and whose prognosis could be confirmed were studied. Fourteen of the thirty-three survived more than one year.

1) Relation between stage and prognosis (Table 7).

In observed frequencies of each stage of thirty-three cases, stage 0~1 was 2%,

Table 6. The relation between the structural responses and prognosis in the preoperative radiotherapeutic group.

C.I.			S.H.			F.H.		
P < 0.01			P < 0.01			P < 0.25		
Scores \ Survival year	1 <	1 ≥	Scores \ Survival year	1 <	1 ≥	Scores \ Survival year	1 <	1 ≥
0~2	3	0	0~1	6	0	0~1	3	0
3~4	2	0	2	0	2	2	2	5
5~6	1	6	3	0	4	3	1	1

Table 7. The relation between stage and prognosis in the preoperative non-therapeutic group.

Survival year	Stage	0~I	II	III	IV
0~1		1	1	5	12
1~2		2	1	2	0
2~3		0	0	3	0
3~4		2	1	2	0
4~5		0	1	0	0

stage II was 12.1%, stage III was 36.4% and stage IV was 36.4%. So, many cases were in the advanced stage. Of cases surviving more than one year, four (80%) were stage 0~1, three (75%) were stage II, seven (58%) were stage III and zero (0%) were stage IV. These figures show that as the stage advanced, the prognosis became poor ($P<0.01$).

- 2) Relation between histological reactions in stage II and stage III and prognosis (Table 8).

Even in the cases of stage II and stage III, who were expected to survive more than one year, some died of recurrence of the cancer. To study its causes the author observed the relation between prognosis and "a" factor, "n" factor, C.I., S.H., and F.H. in sixteen cases of stage II and stage III. Concerning "a" factor, of the cases who died of recurrence of the cancer in less than one year, two were "a" 0,

Table 8. "a" factor, "n" factor, C.I., S.H., and F.H. in the stage II and stage III patients.

Stage	Survival term	a	n	Ly. + Pl.	F. H.	S. H.
III	4M	a ₂	n ₀	4	1	1
III	6M	a ₀	n ₂	2	1	1
III	9M	a ₂	n ₂	3	1	1
III	11M	a ₂	n ₀	2	1	1
II	11M	a ₁	n ₀	3	1	1
III	11M	a ₀	n ₂	4	1	2
III	1Y	a ₁	n ₂	4	1	1
III	1Y2M	a ₂	n ₀	4~6	2	3
II	1Y4M	a ₁	n ₀	2	3	3
III	2Y	a ₁	n ₂	2	3	3
III*	2Y	a ₂	n ₂	4	1	3
III*	2Y	a ₀	n ₂	4	1	2
III*	3Y	a ₂	n ₀	2	1	2~3
III*	3Y	a ₂	n ₀	4	2	3
II*	3Y	a ₁	n ₀	6	1	2~3
II*	4Y	a ₁	n ₁	6	2	2

(*; alive)

Table 9. The relation between the histological responses and the prognosis in the preoperative non-therapeutic group.

C.I.				S.H.				F.H.			
		N.S.				P<0.05				N.S.	
Scores	Survival year	1<	1≥	Scores	Survival year	1<	1≥	Scores	Survival year	1<	1≥
0~2		9	3	0~1		14	4	0~1		12	6
3~4		8	6	2		4	3	2		4	5
5~6		2	5	3		1	7	3		3	3

one was "a" 1, and three were "a" 2. On the other hand, of the cases who survived more than one year, one was "a" 0, five were "a" 1 and four were "a" 2. A significant difference was not found. Concerning "a" factor, of those who died of recurrence in less than one year, three were "n" 0, three were "n" 2. On the other hand, of those who lived more than one year, five were "n" 0, one was "n" 1 and four were "n" 2. Again there was no significant difference between the two. Concerning C.I., the cases who survived more than three or four years showed high scores, but C.I. did not correlate with prognosis. Concerning F.H., all six cases who died of a recurrence in less than one year were score 1, and five among ten cases who survived more than one year were score 2~3. No significant difference was found. But it seemed if the score of F.H. was low, that the patient might die in less than one year. Concerning S.H., five among six cases died in less than one year and were score 1, nine among ten cases who survived more than one year were score 2~3. As a result S.H. significantly correlated with prognosis ($P<0.05$).

- 3) Histological reactions of the preoperative non-therapeutic group and prognosis (Table 9).

The relation between prognosis and histological reactions (C.I., F.H., and S.H.) were studied in thirty-three cases of preoperative non-therapeutic group. In the case of S.H. fourteen (73.7%) among nineteen cases who died of a recurrence of esophageal cancer in less than one year were score 1, while ten among fourteen cases who survived more than one year were score 2~3. S.H. significantly correlated with prognosis. But there was no statistical correlation between C.I., F.H. and prognosis.

Table 10. The relation between the histological responses and the skin tests.

C.I.				S.H.					F.H.					
Skin test	Scores	0~2	3~4	5~6	Skin test	Scores	0~1	2	3	Skin test	Scores	0~1	2	3
0~+		3	4	2	0~+		3	6	1	0~+		5	4	0
++		3	0	1	++		2	0	1	++		2	2	0
+++~###		2	1	0	+++~###		3	0	0	+++~###		2	1	0

4. Relation between histological reactions and skin test (Table 10)

Each histological reaction was compared with positive percentage of the four type skin tests in sixteen cases of esophageal cancer. But as in Table 10, C.I., F.H., and S.H., which are considered host response to cancer, were not correlated with skin test which seems to reflect cell-mediated immunity.

Discussion

It is now supported that the tumor-bearing host has immunoresponses against its own neoplasma and its responses depend chiefly upon the mechanism of cell-mediated immunity⁷⁾. Therefore, it is important to assess the cell-mediated immunocompetence of the tumor-bearing host and there are various methods³⁴⁾ of measurement of the immunocompetence. On the other hand, it is not always true that these immunoparameters reflect the stage and prognosis of the cancer.

It was reported that immunoresponse of the esophageal cancer patients was suppressed in the early stage²⁴⁾. The author selected and measured the four-type skin tests (SK-SD, Candida, PHA and PPD) and lymphocyte blastogenic response as the index of cell-mediated immunocompetence. There are various antigens¹³⁾ of skin tests which have been assessed in the cancer patient. Generally, it has been reported that the positive percentage of skin tests in the cancer patient was less than in the healthy subjects,^{2,13)} and it was reported that PHA skin tests correlated with the prognosis²⁸⁾.

The author also observed significant depression of SK-SD, Candida and PHA skin tests in the esophageal cancer patients. But, it is difficult to evaluate the immunocompetence of the cancer patient only by the use of skin tests^{13,34)}. Furthermore it is impossible to assess the immunocompetence of the cancer patient by using only one skin test. As the author studied the immunosuppression of cancer patients by the total judgment of four-type skin tests, he found that it was useful to practise the skin test of various antigens^{21,23)}. The measurement of lymphocyte blastoid response depends upon the kind of antigen. PHA was reported to cause chiefly the blastogenesis and mitosis of T-cells⁹⁾. So PHA is qualified to measure the cellular immunocompetence. There are two methods of the measurement of lymphocyte blastogenesis, one is the use of separated lymphocytes³⁶⁾ and the other is the use of whole blood^{10,30)}. The author selected the whole blood method. Because there is no change of lymphocyte subpopulation which may change when lymphocytes are separated²⁵⁾, it is not necessary to draw a large volume of blood, and the lymphocyte response can be measured without removing the immunosuppressive substances which were reported to be contained in the serum of the cancer patients³⁵⁾. It was reported that lymphocyte blastogenic reaction was significantly suppressed in patients with unresectable lung cancer¹⁶⁾ and that its reaction was suppressed in patients with stage III and stage IV gastric cancer²⁷⁾. The author also observed that PHA-stimulated lymphocyte response was remarkably suppressed in patients with esophageal cancer. The above results of four-type skin tests and PHA-stimulated lymphocyte response show cell-mediated immunity is suppressed in almost all patients of esophageal cancer compared with healthy subjects.

In postoperative cases the importance of the operative intervention on such immunosuppressive patients was investigated. It was confirmed experimentally that operative intervention such as thoracotomy and/or laparotomy enhanced tumor growth¹²⁾. It is widely practiced that esophageal cancer patients receive thoracotomy and or laparotomy. These operations are the cause of severe stress. The author observed that all values of four-type skin tests and lymphocyte count clearly decreased after operation, compared with these values in the preoperative period. Therefore, it seems that immunocompetence of patients, which was already suppressed in the preoperative period, was more suppressed by operative intervention, so the postoperative patients were under the influence of strong immunosuppression. As a result, it is necessary that immunocompetence of patients should be recovered by administration of immunopotentiators before operation and in the immediate postoperative period.

On the other hand, the existence of responses of tumor-bearing hosts against their own neoplasma can be estimated from the fact that there is spontaneous regression⁶⁾ and some good prognostic cases among same stage cases of cancer. BLACK, et al. reported that in gastric cancer and breast cancer cases, most of the cases with high grade of lymphoid infiltrate had a good prognosis regardless of the histological type, and the cases with high grade of F.H. and S.H. had also a good prognosis^{3,4,5)}.

INOKUCHI, et al.¹⁵⁾ reported the importance of stromal reaction, and HASHIMOTO, et al.¹¹⁾ found that lymphoid reaction and S.H. correlated well with prognosis of gastric cancer, but there was no correlation with F.H. The plasma cell infiltration is nearly always found to accompany lymphocyte infiltration. It was reported that the infiltration correlated well with prognosis of breast cancer¹⁾. The author decided that the scores of lymphocyte infiltration plus plasma cell infiltration were cellular infiltration (C.I.) scores, and investigated the relation between C.I. and prognosis. Concerning esophageal cancer, one reported that lymphoid infiltrate correlated well with the prognosis³¹⁾, while another reported that F.H. correlated well with the prognosis²⁰⁾. Recently, KIKUCHI reported²²⁾ that almost all lymphocytes around the tumor were proved to be T-cells by the fluorescent antibody method. And that it seemed to be a possibility of T-cell identification, and also that the stage of breast cancer and T-cell infiltration were in inverse proportion. OBOSHI²⁶⁾ regarded the lymphocyte invasion around the tumor which was observed after therapeutic irradiation as a kind of rejection phenomena against cancer.

As described, there are various evaluations concerning histological reactions. The following became clear after the investigation in the relationship between the value of C.I., F.H., and S.H. as against prognosis of esophageal cancer patients. In the preoperative non-therapeutic group, almost all cases whose scores of S.H. were 2~3 survived more than one year and almost all cases whose scores of F.H. were 0~1 died of recurrence in less than one year, but C.I. did not correlate with prognosis. In the preoperative radiotherapeutic group, cases with high scores of C.I. and S.H. had good prognosis but F.H. did not correlate with prognosis. As a result, it seems that these histological reactions are host defense reactions against cancer and have a relation to prognosis.

As the influential factors of the prognosis of esophageal cancer, various factors have been

investigated. ISHIGAMI, et al.²⁸⁾ reported the importance of "n" factor while others reported the importance of "a" factor¹⁴⁾ and the length of the tumor³³⁾. But besides these factors, it seems that histological reaction and regional lymph node reaction is useful to judge the prognosis of esophageal cancer. Then the author observed the relation between the skin tests which were reported to reflect immunocompetence and C.I., S.H. and F.H. which were reported as host response against cancer. STEWART²⁹⁾ reported the existence of a definite correlation between skin reactions in patients toward cellular extracts of their malignant tumors and stromal reaction.

It was also reported that PPD skin test correlated well with the degree of lymphocyte infiltration³²⁾. But the author did not observe the correlation between skin test and histological reaction. So, it seems that the degree of skin reaction is not always parallel with their antitumor competence.

Conclusion

1. In the four-type skin tests of SK-SD, Candida, PHA and PPD, the esophageal cancer patients were compared with healthy subjects. There was significant difference in skin reactivity to SK-SD, Candida and PHA. It seems that the immunocompetence of esophageal cancer patients was suppressed.
2. In the summarized judgment of four-type skin tests, in healthy subjects a positive percentage in more than three of the four skin tests was 100%. But in the esophageal cancer patients the positive percentage was only 15%. So the summarized judgment clearly reflected the immunosuppression of esophageal cancer.
3. In lymphocyte blastoid reaction using whole blood method, 78% of the healthy subjects were more than 10000 cpm in PHA stimulated value, on the other hand, only 10% of the esophageal cancer patients were more than 10000 cpm. It seems that the immunocompetence of the patients was remarkably suppressed.
4. Compared with skin tests and lymphocyte percentages in preoperative and postoperative periods, these values were clearly depressed in the postoperative period. It seems that the operative stress remarkably suppressed the immunocompetence.
5. In the preoperative radiotherapeutic group, almost all cases with high scores of C.I. and S.H. survived more than one year. But "a" factor, "n" factor and stage did not correlate with prognosis.
6. In the preoperative non-therapeutic group, the prognosis became worse as the stage of cancer advanced. On the other hand, almost all cases with low scores of S.H. had a bad prognosis.
7. There did not exist a definite correlation between skin tests and histological responses.

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Reference

- 1) Berg JW: Inflammation and prognosis in breast cancer. *Cancer* **9**: 935-939, 1959.
- 2) Betsuyaku T: Skin test reactions to various antigens in each stage of gastric cancer patients. *Cancer & Chemotherapy* **4**: 753-759, 1977. (Jpn)
- 3) Black MM: Microscopic structure of gastric carcinomas and their regional lymph nodes in relation to survival. *Surg Gynec Obstet* **98**: 725-734, 1954.
- 4) Black MM: Survival in breast cancer cases in relation to the structure of the primary tumor and regional lymph nodes. *Surg Gynec Obstet* **100**: 543-551, 1955.
- 5) Black MM: Prognostic significance of microscopic structure of gastric carcinomas and their regional lymph nodes. *Cancer* **27**: 703-711, 1971.
- 6) Everson TC and Cole WH: Spontaneous regression of cancer. WB Saunders, Philadelphia, 1966.
- 7) Friedman H and Southam CM: Area of relationship between immunology and clinical oncology. *AJCP* **62**: 224-242, 1976.
- 8) Fujimaki M: Preoperative combined treatment for esophageal cancer surgery. *J Clin Surg* **33**: 1271-1278, 1978. (Jpn)
- 9) Greaves MF and Janossy G: Lymphocyte activation i. Response of T and B lymphocytes to phytomitogens. *Clin Exp Immunol* **9**: 483-498, 1971.
- 10) Han T and Pauly J: Simplified whole blood method for evaluating in vitro lymphocyte reactivity of laboratory animals. *Clin Exp Immunol* **11**: 137-142, 1972.
- 11) Hashimoto I: Studies concerning the anti-cancer reaction in the lymph node, I. Prognosis in gastric cancer in relation to the structural representations in lymph node. *J Kyoto Pref Univ Med* **83**: 553-560, 1974. (Jpn)
- 12) Hattori T: Enhancing effect of thoracotomy on the growth of ascitic tumor or lung metastases in rats. *JJATS* **26**: 836-845, 1978. (Jpn)
- 13) Hosokawa M: Skin reaction test. *J Clin Surg* **33**: 1397-1402, 1978. (Jpn)
- 14) Iizuka T: Factors influencing the prognosis of the esophageal cancer. *JJATS* **25**: 953-959, 1977. (Jpn)
- 15) Inokuchi K: Prognosis in gastric cancer patients performed radical operation in relation to stromal reaction. *Surgery* **27**: 559-570, 1965. (Jpn)
- 16) Inoue H: Evaluation of lymphocyte transformation using whole blood culture in patients with primary lung cancer: Correlation with DNCB reactivity. *JJSS* **79**: 1518-1525, 1978. (Jpn)
- 17) Iris ME: Possible host resistance in carcinoma of the breast: A histological study. *Brit J Cancer* **21**: 383-401, 1968.
- 18) Ishigami K: Treatment of carcinoma of the thoracic esophagus judging from the local findings. *Gastroenterological Surg* **3**: 617-627, 1980. (Jpn)
- 19) Isono K: Prognosis of the esophageal cancer. *Surg Diagn Treat* **20**: 1223-1227, 1978. (Jpn)
- 20) Kakuta H: Prognostic significance of histological structure of the regional lymph nodes and activity of peripheral lymphocytes on esophageal cancer patients. *Recent Med* **30**: 681-686, 1975. (Jpn)
- 21) Kamei H: Immunological parameters in cancer bearing patients after treatment with immunostimulants. *Jap J Cancer Clin* **24**: 948-955, 1978. (Jpn)
- 22) Kikuchi K: Surgical immunology and lymphatic system. Lymphatic system and cancer regression. *Surg Diagn Treat* **23**: 1187-1192, 1979. (Jpn)
- 23) Kobayashi H: Skin reaction test in cancer patients. *Jpn Med J* **2805**: 8-12, 1978. (Jpn)
- 24) Kumashiro R: Studies on immunological responsiveness of cancer patient. II. Phytohemagglutinin stimulated lymphocyte transformation in patients with gastric or esophageal cancer. *Fukuoka Acta Med* **69**: 339-349, 1976. (Jpn)
- 25) Nakamura Y: Lymphocyte transformation by ultra-whole blood culture method. *Jpn J Nucl Med* **14**: 871-875, 1977. (Jpn)
- 26) Oboshi S: Tumor-host-relationship in cancer radiotherapy. *Jap J Cancer Clin* **16**: 651-657, 1970. (Jpn)
- 27) Orita K: Blastogenesis. *Jap J Cancer Clin* **24**: 941-947, 1978. (Jpn)
- 28) Sone S: Phytohemagglutinin (PHA) skin test and prognosis in cancer patients. *Igaku No Ayumi* **101**: 715-717, 1977. (Jpn)
- 29) Stewart IHM: The presence of delayed hypersensitivity reaction in patients toward cellular extracts of their malignant tumors. *Cancer* **23**: 1380-1387, 1967.

- 30) Suzuki H: Micromethod using whole blood culture for evaluating lymphocyte response to phytohemagglutinin. *Igaku No Ayuki* **105**: 882-884, 1978. (Jpn)
- 31) Takahashi K: Squamous cell carcinoma of the esophagus. Stromal inflammatory cell infiltration as a prognostic factor. *Cancer* **14**: 921-933, 1961. (Jpn)
- 32) Tanaka K: Relationship between postoperative survival rate and stromal reaction in relation to PHA and PPD reaction in gastric cancer. *Jap J Cancer Clin* **25**: 1389-1395, 1979. (Jpn)
- 33) Tasei Y: A evaluation of prognosis for esophageal cancer following radiation therapy. *Jap J Cancer Clin* **21**: 159-163, 1975. (Jpn)
- 34) Tsubura E: Cancer immunotherapy and its diagnostic parameter. *J Clin Surg* **33**: 1365-1371, 1978. (Jpn)
- 35) Urushizaki I: Cell-mediated immunity in cancer-patients. *Jap J Cancer Clin* **24**: 378-385, 1978. (Jpn)
- 36) Yamamura M: Standardization of lymphocyte transformation to phytohemagglutinin. *Clin Exp Immunol* **14**: 457-467, 1973.

和文抄録

食道癌の免疫学的研究 —食道癌患者における細胞性免疫能と主病巣 および所属リンパ節の動態—

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食道癌患者の免疫能を4種の皮内反応(SK-SD, Candida, PHA, PPD) および全血によるリンパ球幼若化能にて測定し、同時に食道癌患者における宿主側の反応として、組織学的に主病巣におけるリンパ球および形質細胞の浸潤(細胞浸潤)と所属リンパ節における洞組織球増生症および濾胞増生症について、それぞれ score をつけて、予後との関係を検討した。その結果は以下のとおりであった。

- 1) SK-SD, Candida, PHA および PPD の4種皮内反応に関して、健康人と食道癌患者を比較すると、SK-SI), Candida および PHA について有意の差を認め、食道癌患者の細胞性免疫能が低下していることを予想せしめた。
- 2) 4種皮内反応を総合判定すると、健康人は3種以上陽性の者が100%であったのに対して、食道癌患者ではわずか15%であり、総合判定がより確かに患者の免疫能を反映することを知った。
- 3) 全血によるリンパ球幼若化能の判定において、PHA による刺激値は10000 cpm 以上のものが正常人では78%であったのに対して、食道癌患者では10%に

すぎず、明らかな低下を認め、細胞性免疫能の低下を予想せしめた。

- 4) 術前および術直後における皮内反応とリンパ球の%を比較すると、明らかに術後には低下しており、手術によって免疫能が著しく障害されたと考えられた。
- 5) 術前照射群において、細胞浸潤度、洞組織球増生度および濾胞増生度を1年未満再発死亡群と1年以上生存群にわけて比較すると、細胞浸潤度および洞組織球増生度の scores の高いものほど予後が良好であったが、a 因子、n 因子および stage と予後のあいだには相関を認めなかった。
- 6) 非術前治療群では stage が進むほど予後が不良であったが、同時に洞組織球増生度の score の悪いものは明らかに予後が不良であるという結果を得た。
- 7) 術前の皮内反応と組織反応のあいだには相関を認めなかった。

以上の成績より、食道癌患者は術前および術後において細胞性免疫能が低下しており、予後の指標としては洞組織球増生度の検討が有用であると考えられた。