Nagoya J. med. Sic. 28: 273-292, 1966.

ON THE PLASMA PROTEIN PATTERN OF PATIENTS WITH MALIGNANCY, USING ELECTRO-PHORESIS OF TISELIUS

KIMIYUKI KATO

1st Department of Surgery, Nagoya University School of Medicine
(Director: Prof. Yoshio Hashimoto)

SUMMARY

Free boundary electrophoresis of plasma protein was performed in accordance with "the Standard Method of Tiselius Electrophoresis". The plasma was obtained, using heparin sodium as an anticoagulant.

Parallel relationship between stages of malignancy and changes of plasma protein pattern was presumed on patients with malignancy, mostly gastric cancer. Changes of plasma protein patterns of patients with malignancy would be increase of fibrinogen in early stage and increase of alpha-globulin and gamma-globulin and decrease of total protein and albumin in the following stage. Absence of increasing gamma-globulin in terminal stage was observed and the same phenomenon was also observed in patients with hematogeneous metastasis.

Plasma protein patterns of professional blood donors with anemia and patients with mal-nutrition resulted from gastroduodenal ulcers with persisting pyloric stenosis were observed and the patterns were both within normal ranges.

A transition of plasma protein patterns of patients with malignancy from preoperative stage to postoperative stage were observed and it was concluded that observation of plasma protein pattern of cancer patient for a short period can not tell prognosis of the patient but the observation for several months of several years would tell it.

PREFACE

From the stand point of clinicians, who deal with pathological phenomena of human organisms, it is important to analyse and grasp a pathological condition of plasma protein. The principal reasons for study of plasma protein in neoplastic diseases are, first, hope of finding useful diagnostic tests; second, necessity for objective criteria for evaluating tumor therapies; and third, necessity for better understanding of systemic effects of tumors on their hosts.

Hitherto, many new techniques, for example, blood sedimentation ratio, turbidity reactions, flocculation reactions, have been eagerly applied to study the plasma protein in neoplastic diseases, with same nonspecific results; abnormal values in patients with advanced cancer as similar abnormal findings

加 藤 王 千
Received for publication, March 5, 1966.

in patients with other chronic wasting diseases or infections[1] and with rare exceptions of negative findings in patients with early cancer[2], where diagnostic tests should be most valuable.

Plasma protein analysis was performed for the first time by Arne Tiselius[3] in 1937 and this method has been employed widely in the medical field. The situations of the plasma protein in many diseases have been investigated by this method and the significance of plasma protein analysis by this method has been gradually recognized. In nephrosis, liver cirrhosis, multiple myeloma and others, which show certain specific plasma protein pattern, the protein analysis by this method presents an important basis for their diagnoses. On the other hand, in those which show no specific plasma protein pattern like tuberculosis, it is useful for observation of pathological condition and clinical progress, and also for judgement of prognosis.

But the evaluation of these plasma protein abnormalities in cancer patients have not yet been established without any objections.

HISTORICAL

In the conclusion by Wuhrmann and Wunderly[4], the blood cheminal picture including the plasma protein picture in a neoplastic disease depends with a large extent on the stage and localization of the tumor, presence or absence of metastases and in certain cases on tissue breakdown.

According to the expression of Riva[5], in patients of malignant tumor with or without local metastases, only mild or moderate dysproteinemia can be found.

From the study of Krystodik[6], the mean values of a group of patieuts with active neoplastic diseases show a decrease of albumin accompanied by an increase of alpha-globulin fraction, and those shifts in the protein pattern of the blood serum are not characteristic or specific for their neoplastic diseases alone.

It was Petermann[7] who described that the quantitative changes in the plasma proteins reflected chiefly the reactions of the host to the presence of tumor and had not yet been proved to be of diagnostic value and that, however, these changes were of great importance in following the clinical state of the patients.

Concerning children with advanced malignant diseases, Stickler[8] reported that protein fractions in such patients disclosed a decrease in serum albumin and its lowest concentration was usually found shortly before the death of the patients.

Sieberth[9] concluded that rising tendency of the alpha-globulin in malignant disease was an unfavorable sign for prognosis and descendig tendency of alpha-globulin was a favorable one.

According to the study of Miller[10], a hyperglobulinemia and hypoalbumi-

nemia are consistently observed in patients with active malignant disease and the changes in both serum proteins are accelerated by progression of the malignant disease, and moreover, 8 subjects free of recurrence for long period after successful surgical removal of breast cancer by radical mastectomy showed normal plasma protein patterns.

METHODS

Approximately 5 ml of the blood was obtained from subjects before breakfast by venipuncture, and was transferred to a dry test tube. Immediately after this procedure 0.1 mg of heparin sodium was mixed. The plasma was obtained by centrifugation. The usage of heparin sodium to this experiments was recommended by Hayase[11]. But heparin sodium showed same mobility as albumin at electrophoresis, it should be used only a little.

The total protein concentration was determined by a refractometer manufactured by Hitachi Co., at room temparature and then temperature correction was done.

The plasma was diluted with phosphate buffer to the protein concentration of 2.0 g per 100 ml. The solution was enclosed in a cellophane bag under slight pressure, leaving about 2 ml of free space in it and dialysed more than 16 hours with continuous rocking of the bag against the buffer solution at 4°C. After dialysis, the cellophane bag was removed, the solution was centrifuged in order to remove all unnecessary particles. The buffer employed was 1/10 M phosphate buffer (pH=7.6, ionic strength=0.144).

Free electrophoresis of the plasma was performed with a Shimazu apparatus. Electrophoresis was carried out for 50 minutes at 10°C, using potential difference of 130 volts and current of 13 milliamperes. The picture was enlarged three times and traced the curve. After measuring the area under each curve with planimeter, the relative percentage of each component was calculated, assuming that the reaction index was the same for each. Only descending side was employed and the area under the epsilon-peak was disregarded[12][13].

The details of techniques which are not described here were the same as "the Standard Method of Tiselius Electrophores"[14] determined at the Japanese Congress of Electrophoresis in 1950.

MATERIALS

1) *Control Group for Normal Ranges*

The materials of this study consist of 8 males and 6 females. All of them were healthy volunteers at the time of drawing the blood specimens and had no history of any illness for more than the last 6 months.

2) *Patients with Malignancy*

Seventy five petients with malignancy admitted consecutively from Sep-

tember 1959 to July 1963 were employed for this study. Seventy one of total 75 cases had abdominal malignancies, mostly gastric cancer, and their diagnoses were confirmed by histology or cytology. None of them had ever received any special treatments for their malignancies such as operation, radiation therapy or antitumor chemotherapy. According to bed side examinations or findings at their operations which were performed later, they were divided into three groups.

The first group; group of radical operation: in this group of patients, radical operations were performed.

The second group; group of exploratory operation: in this one, radical operation could not be performed, but no ascites was observed at the time of operation.

The third group; terminal group: in this one, ascites was observed obviously in bed side examination due to abdominal carcinomatosis.

3) *Patients with Hematogeneous Metastasis*

For the other cases of the terminal stage of neoplastic disease, 5 patients who had multiple blood born metastases were used. These patients were confirmed to have hematogeneous metastatic tumors by X-ray examinations and later autopsies.

4) *Ratients with Gastroduodenal Ulcer*

For the other examples of mal-digestion and mal-absorption, 6 patients with gastroduodenal ulcer were employed, who were proved roentogenologically to have pyloric stenosis for more than 2 months.

5) *Professional Blood Donors*

For the other example of chronic blood loss, 11 males of professional blood donors were used. They came to the Nippon Chubu Blood Bank and were proved to be inadequate for further blood drawing because of low blood counts, resulting from frequently repeated blood drawing. None of these donors' blood counts was above 350×10^4, and hemoglobin 70% by Sahli.

6) *Patients with Malignancy, who Received Radicel Operation*

Twenty one patients of gastric cancer were employed. All of them received radical operation and their postoperative courses were uneventful without any complication and their long-term follow-ups could be observed.

RESULTS

1) *Control Group for Normal Ranges*

The first study was designed to obtain normal ranges of plasma protein pattern in order to compare them with protein pattern of the patients's plasma (Table 1 and Fig. 1).

TABLE 1. Control Group

Age	Sex	Total Protein (g/dl)	Alubumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
20	F	7.5	61.0	6.3	9.4	5.1	18.2
24	F	7.2	67.3	9.8	7.7	4.9	10.4
29	M	7.9	59.1	8.2	12.5	5.5	14.7
26	M	7.1	57.2	10.5	10.4	4.5	17.4
50	M	6.3	62.5	5.4	8.6	7.0	16.5
26	M	7.3	58.0	7.9	12.8	7.8	13.5
27	M	7.8	64.5	1.9	3.1	6.0	18.5
20	F	7.5	58.4	8.1	5.5	3.5	24.5
26	F	7.2	71.5	8.2	6.5	4.3	9.5
36	F	7.9	62.6	9.3	4.6	12.0	7.9
50	F	7.1	63.2	12.6	6.1	3.8	14.9
28	M	7.3	59.3	8.1	7.1	6.9	18.7
47	M	6.3	63.2	10.4	8.3	3.3	14.8
24	M	—	60.6	6.4	18.8	7.4	6.8

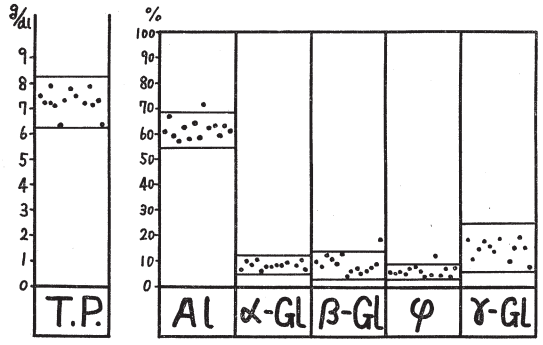

FIG. 1. Control group.

By this study the normal ranges were determined as following (p=0.05); total protein: 7.26±1.00 g/dl, albumin: 62.03±7.54%, alpha-globulin: 8.42±3.64 %, beta-globulin: 7.99±5.56%, fibrinogen (phi): 5.38±2.98%, gamma-globulin: 14.72±9.48%.

These results are used as the standard values for the judgement whether or not the results from following studies are within normal ranges.

2) Patients with Malignancy

The plasma protein patterns of three groups were compared with those of the normal group and were observed whether or not they were within the normal ranges.

(1) The first group; total protein, albumin, beta-globulin and gamma-globulin were grossly within normal ranges. Only fibrinogen deviated obviously to high value and significant difference from normal group was observed statistically (p=0.01) (Table 2 and Fig. 2).

(2) The second group; significant decrease in total protein and albumin and significant increase in alpha-globulin, fibrinogen and gamma-globulin were

TABLE 2. Operative Group

Age	Sex	Diagnosis	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
65	M	Gastric Cancer	6.7	60.7	9.8	7.3	8.9	13 3
55	F	Gastric Cancer	7.2	62.2	9.3	8.5	7.2	12.9
66	M	Gastric Cancer	5.4	51.4	12.6	6.5	10.0	19.5
47	M	Colon Cancer	6.8	66.7	10.5	6.5	6.8	9.5
37	M	Gastric Cancer	6.5	60.2	9.2	6.8	2.6	21.2
51	F	Gastric Cancer	6.6	70.6	4.8	7.0	3.8	13.0
40	M	Gastric Cancer	6.8	57.6	13.9	3.4	6.9	18.2
42	M	Gastric Cancer	5.8	62.0	10.5	14.8	5.9	6.8
28	M	Gastric Cancer	6.4	76.6	9.3	6.0	2.4	5.7
58	F	Gastric Cancer	6.0	56.0	13.0	6.4	8.4	16.2
45	M	Gastric Cancer	6.1	67.7	7.2	7.8	3.8	12.9
56	F	Gastric Cancer	6.3	58.6	12.1	12.9	5.3	11.0
59	M	Grstric Cancer	6.5	58.1	10.1	9.5	2.9	19.4
47	F	Gastric Cancer	4.7	60.5	11.4	2.4	11.9	13.7
55	M	Gastric Cancer	6.8	66.8	11.2	6.7	3.8	11.5
57	M	Gastric Cancer	6.0	70.3	12.3	5.3	6.6	5.5
39	F	Gastric Cancer	6.8	62.6	10.3	7.2	4.4	15.5
58	M	Gastric Cancer	6.4	60.9	7.6	6.3	6.1	19.1
18	F	Gastric Cancer	7.2	85.8	8.2	11.5	9.2	15.3
66	M	Gastric Cancer	6.7	57.2	8.6	10.0	9.5	12.8
64	F	Gastric Cancer	7.2	51.4	13.3	14.5	8.4	12.4
67	M	Gastric Cancer	5.4	49.9	11.6	8.1	11.6	18.8
48	M	Rectal Cancer	6.8	60.6	10.6	11.0	7.8	10.0
39	M	Gastric Cancer	6.5	57.8	7.4	10.1	8.4	16.3
50	F	Gastric Cancer	6.6	63.6	8.9	9.5	5.2	13.4
41	M	Gastric Cancer	6.8	70.5	10.0	9.8	6.4	13.5
43	M	Gastric Cancer	5.8	59.5	8.1	12.9	9.8	3.7
28	M	Gastric Cancer	6.8	67.1	9.4	9.0	5.8	8.7
37	F	Gastric Cancer	6.0	51.6	8.3	14.0	10 3	15.8
46	M	Gastric Cancer	6.1	60.7	9.1	10.1	9.6	10.9
55	F	Gastric Cancer	6.3	52.4	10.1	11.3	10.5	15.7
60	M	Gastric Cancer	6.5	71.1	7.6	8.0	2.8	10.5
48	M	Gastric Cancer	7.0	94 3	5.6	5.4	6.2	18.5
46	F	Gastric Cancer	4.9	61.5	9.0	10.6	8.1	10.8
56	M	Gastric Cancer	6.8	64.2	9.1	10.9	4.9	10.9
56	M	Gastric Cancer	6.9	65.3	7.4	10.9	5.6	10.8
38	F	Gastric Cancer	6.8	61.7	10.0	6.7	8.0	13.6
57	M	Gastric Cancer	6.4	57.2	7.0	12.0	9.2	14.6
19	F	Gastric Cancer	7.2	59.6	10.1	6.1	7.3	16.9

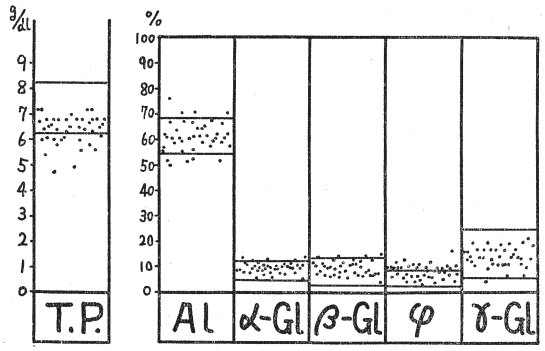

FIG. 2. Operative group.

TABLE 3. Non-operative Group

Age	Sex	Diagnosis	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
48	M	Gastric Cancer	6.0	45.0	11.4	9.6	10.0	24.0
26	M	Gastric Cancer	6.4	45.8	10.3	9.8	13.5	22.0
42	M	Gastric Cancer	7.0	50.6	10.8	4.3	8.7	25.6
61	M	Gastric Cancer	7.7	46.8	11.4	14.1	3.4	24.1
49	F	Gastric Cancer	6.8	53.2	16.7	7.1	8.6	14.4
55	F	Gastric Cancer	7.0	57.6	9.4	7.2	8.4	17.4
34	M	Gastric Cancer	6.5	58.6	13.6	9.4	4.6	14.1
60	M	Gastric Cancer	7.1	61.1	13.0	5.2	5.2	15.0
66	M	Gastric Cancer	6.8	44.9	12.4	6 4	11.0	25.5
42	M	Gastric Cancer	6.9	55.2	15.1	9.9	3.4	16.4
47	M	Gastric Cancer	6.4	47.0	8.6	3.2	7.0	34.0
48	M	Gastric Cancer	6.0	45 0	11.4	9.6	10.0	24.0
48	M	Gastric Cancer	7.2	44.3	10.5	14.3	14.6	10.3
26	M	Gastric Cencer	6.4	54.0	8.6	9.4	8.8	19.4
44	F	Rectal Cancer	6.7	52.2	4.9	3.9	12.2	22.3
54	M	Gastric Cancer	7.0	48.2	10.6	8.5	9.6	23.1
60	F	Gastric Cancer	7.3	54.0	9.9	11.0	5.6	19.2
42	M	Gastric Cancer	7.0	45.6	8.5	11.1	12.3	22.5
61	M	Gastric Cancer	7.7	45.2	12.6	7.0	12.3	22.8
49	F	Gastric Cancer	6.8	55.5	9.8	13.3	8.7	12.7
55	F	Gastric Cancer	7.0	60.4	8.7	9.7	6.9	14.3
34	M	Gastric Cancer	6.5	56.9	10.4	10.6	7.7	14.4
60	M	Gastric Cancer	7.1	58.4	9.6	12.6	6.0	13.4
66	M	Gastric Cancer	6.8	44.7	11.9	12.2	11.0	20.2
42	M	Gastric Gancer	6.9	57.6	9.9	9.6	6.8	16.1

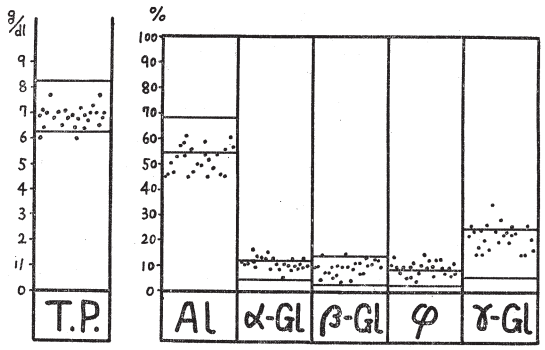

FIG. 3. Non-operative group.

observed (p=0.01). Only beta-globulin was grossly within normal limits (Table 3 and Fig. 3).

(3) The third group. Total protein and albumin decreased significantly, and also alpha-globulin and fibrinogen increased significantly (p=0 01). Beta-globulin and gamma-globulin were grossly within normal ranges (Table 4 and Fig. 4).

3) Patients with Hematogeneous Metastasis

Total protein and albumin decreased and alpha-globulin and fibrinogen in-

TABLE 4. Terminal Group

Age	Sex	Daignosis	Total Protein (g/dl)	Albumin (%)	α-Glbumin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
50	F	Abdominal Carcinomatosis from Gastric Cancer	6.0	49.5	13.5	12.5	11.2	13.8
57	M	Rectal Cancer	4.5	47.4	13.0	14.3	9.9	15.4
46	M	Gastric Cancer	7.0	47.8	13.4	10.6	13.8	14.4
30	F	Gastric Cancer	5.6	54.3	10.1	12.1	10.9	12.6
24	M	Gastric Cancer	5.2	52.7	7.9	16.2	11.3	11.9
42	M	?	6.4	43.6	14.7	10.9	14.4	16.4
51	M	Gastric Cancer	6.0	40.5	16.6	12.8	13.6	16.5
47	M	Gastric Cancer	7.0	47.3	14.8	8.9	12.1	16.6
31	F	Gastric Cancer	3.6	60.9	10.8	6.5	8.0	13.8
25	M	?	5.2	48.7	10.7	10.4	15.1	15.1
41	M	Gastric Cancer	6.4	48.2	19.8	7.2	15.5	9.4

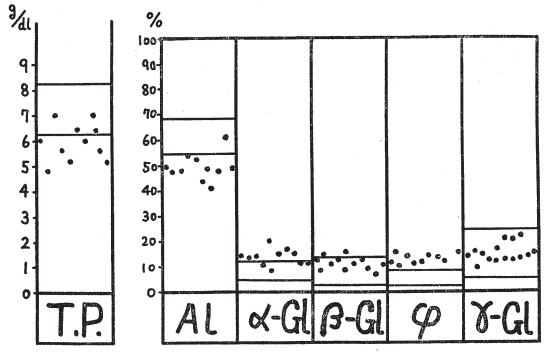

FIG. 4. Terminal group.

creaed significantly (p=0.01). Beta-globulin and gamma-globulin were within normal ranges (Table 5 and Fig. 5).

4) *Patients with Gastroduodenal Ulcer*

In this study, their protein patterns were all within normal ranges and no

TABLE 5. Patients with Hematogeneous Metastases

Age	Sex	Diagnosis	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
35	F	Lung Metastases of Breast Cancer	6.1	50.9	15.6	8.5	11.7	13.0
55	M	Lung Metastases of Fibrosarcoma	6.1	41.1	16.5	11.1	17.3	14.0
63	M	Lnug Metastases of Rhabdomyosarcoma	6.0	52.0	9.4	11.2	6.8	20.6
56	F	Lung Metastases of Skin Cancer	6.0	53.2	9.4	12.4	8.9	13.1
35	M	Brain Metastases of Lung Cancer	6.3	56.4	12.8	6.4	12.2	11.6

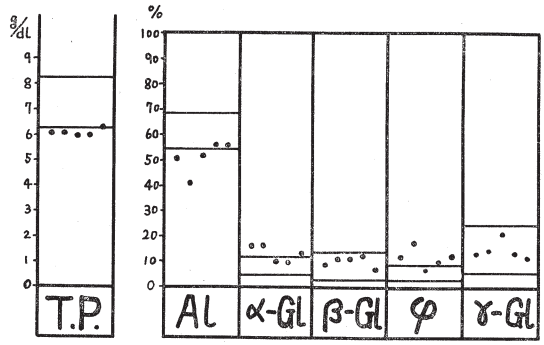

FIG. 5. Patients with hemategeneous metastasis.

TABLE 6. Patients with Gastroduodenal Ulcer

Age	Sex	Periods of Pyloric Stenosis Mons.	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
45	M	2	6.4	65.1	7.6	8.4	6.7	12.2
51	M	1	6.7	58.1	10.1	11.2	6.8	13.8
14	M	6	6.8	62.0	9.4	8.1	7.3	13.2
44	M	2	6.4	70.2	5.2	9.0	2.7	13.9
50	M	4	6.7	59.4	7.5	13.0	3.5	16·6
13	M	10	6.8	64.6	12.7	10.4	3.4	8.9

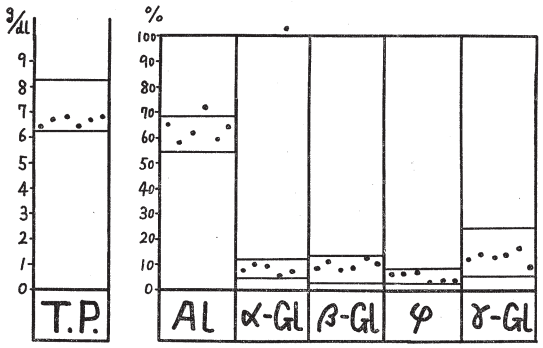

FIG. 6. Patients with gastroduodenal ulcer.

significant difference from normal group was observed (Table 6 and Fig. 6).

5) Professional Blood Donors

Similarly to values of patients with gastroduenal ulcer, protein patterns of the donors were all within normal ranges and no significant difference from normal group was observed (Table 7 and Fig. 7).

6) Patients with Malignancy, Who Received Radical Operation

At first, changes of the plasma protein pattern from proeperative stage to postoperative stage for eight weeks were observed in a patient with gastric

TABLE 7. Inadequet Profesional Blood Donors

Age	Sex	Hemo-globin (%)	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
24	M	66	7.3	61.7	9.6	8.6	5.6	14.2
30	M	66	7.3	68.3	11.5	7.7	5.2	7.3
28	M	66	7.0	67.6	10.7	4.9	9.7	7.4
48	M	64	7.4	55.7	6.2	9.5	6.6	22.0
28	M	51	7.0	67.2	6.9	6.0	5.2	14.7
34	M	57	6 8	64.3	7.4	9.0	6.0	13.2
24	M	54	8.0	56.3	12.6	9.8	8.0	13.1
45	M	51	6.8	57.0	9.9	11.9	7.1	14.1
38	M	58	7.7	53.1	10.9	9.5	7.4	19.1
28	M	52	7.1	66.0	6.4	9.4	8.4	10.0
27	M	55	7.7	65.3	7.9	12.1	4.5	10.2

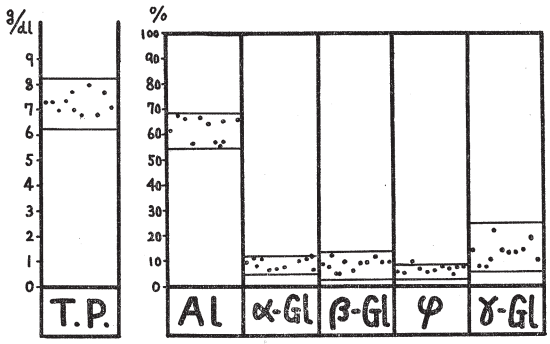

FIG. 7. Inadequate professional blood donors.

TABLE 8. 46-year-old Male with Gastric Cancer, Who Received Radical Operation

	Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)
Before operation	6.9	51.1	8.1	10.8	8.6	21.4
1 week after operation	6.2	42.1	9.6	10.8	19.8	17.7
2 weeks after operation	6.3	43.1	8.1	10.8	8.9	24.0
3 weeks after operation	6.6	55.6	5.5	10.6	6.3	22.0
4 weeks after operation	6.6	60.0	9.5	1.9	8.2	20.4
5 weeks after operation	7.2	54.4	11.0	11.5	4.9	18.2
7 weeks after operation	7.2	65.4	8.2	8.4	3.2	14.8

cancer who had received radical operation (Table 8).

One week after operation, the pattern was varied from that before operation and the pattern in two weeks postoperatively the changes were severer than that in one week. Three weeks after operation, the protein pattern was nearly the same as that before operation. In the following course, the pattern had a tendency to approach the normal pattern.

The pattern on 14th day and 21st day after operation were compared with that of before operation, with respect to the plasma protein of patients with gastric cancer, who received radical operation.

The total protein value decreased on 14th and increased on 21st postoperative day in mean value, but no significant difference was observed (Table 9).

Albumin decreased 2 weeks after and returned to preoperative value by 3 weeks (Table 10). This change was statistically significant.

Alpha-globulin increased significantly 2 weeks after and came back to preoperative value by 3 weeks (Table 11).

Beta-globulin changed not significantly (Table 12).

Gamma-globulin similarly increased 2 weeks after and continued to be in high value for 3 weeks(Table 14).

Now, a comparison between the plasma protein pattern in preoperative stage and that of more than 3 weeks after radical operation was performed in the same patient (Table 15). In order to analyse these data, so called

TABLE 9. Total Protein (g/dl)

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	7.2	6.2	7.0
26	M	6.4	6.4	6.8
28	M	6.8	5.6	6.9
50	F	6.6	6.9	6.5
41	M	6.8	6.5	6.6
46	M	6.1	6.6	6.6
55	F	6.3	5.9	6.8
56	M	6.0	5.8	5.9
50	M	6.9	5.6	6.1
30	M	6.4	6.4	6.8
51	F	6.6	6.9	6.5
42	M	6.8	6.5	6.6
47	M	6.1	6.6	6.6
57	M	6.0	5.8	5.9
Mean		6.50	6.26	6.54

TABLE 10. Albumin (%)

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	44.3	47.6	55.2
26	M	54.0	55.0	55.7
28	M	67.1	57.7	51.1
50	F	63.0	60 2	52.2
41	M	60.5	52.8	57.4
46	M	60.3	53.0	58.2
55	F	52.4	47.8	49.1
56	M	65.3	60.4	63.2
50	M	57.6	56.8	50.2
30	M	45.8	52.3	51.9
51	F	70.6	60.1	57.9
42	M	57.6	50.3	61.1
47	M	67.7	52.1	61.3
57	M	70.3	59.5	70.1
Mean		56.18	54.69	56.11

TABLE 11. Alpha-globulin (%)

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	16.5	11.2	11.6
26	M	8.6	9.4	17.0
28	M	9.4	9.8	8.1
50	F	8.9	8.8	13.5
41	M	10.0	12.1	11.1
46	M	9.1	10.3	8.8
55	F	10.1	12.5	15.0
56	M	7.4	11.8	10.0
50	M	15.1	7.9	11.7
30	M	10.3	13.0	10.6
51	F	4.8	11.3	11.2
42	M	13.9	20.3	12.3
47	M	7.2	12.4	9.6
57	M	12.3	17.4	11.2
Mean		10.22	12.76	11.55

TABLE 12. Beta-globulin (%)

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	14.3	11.5	10.1
26	M	9.4	8.6	10.7
28	M	9.0	8.9	10.8
50	F	9.5	10.1	10.6
41	M	9.8	11.9	9.7
46	M	10.1	9.3	9.8
55	F	11.3	10.4	12.1
56	M	10.9	9.7	9.2
50	M	9.6	12.2	14.0
30	M	9.8	5.6	11.6
51	F	7.8	8.3	13.5
42	M	3.4	13.0	4.5
47	M	7.8	6.5	10.7
57	M	5.3	8.2	4.9
Mean		9.14	9.59	10.87

TABLE 13. Fibrinogen (Phi)

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	14.6	10.0	8.4
26	M	8.8	8.9	10.7
28	M	5.8	7.8	8.6
50	F	5.2	10.5	9.6
41	M	6.3	7.3	5.7
46	M	9.6	12.1	8.5
55	F	10.5	13.1	11.4
56	M	5.6	7.6	6.3
50	M	6.8	7.5	7.0
30	M	13.5	7.2	8.6
51	M	3.8	12.3	5.2
42	M	6.9	4.1	5.5
47	M	3.8	11.1	3.6
57	M	6.6	3.8	4.6
Mean		7.70	8.81	7.41

TABLE 14. Gamma-globulin

Age	Sex	Before operation	2 weeks after operation	3 weeks after operation
48	M	10.3	19.7	14.7
26	M	19.4	18.1	9.9
28	M	8.7	15.8	21.4
50	F	13.4	10.4	14.1
41	M	13.5	15.9	15.9
46	M	10.9	15.3	14.7
55	F	15.7	16.2	21.4
56	M	10.8	10.5	11.3
50	M	16.1	15.6	13.1
30	M	22.0	21.9	17.4
51	F	13.0	8.1	12.2
42	M	18.2	11.8	16.6
47	M	12.9	17.9	14.8
57	M	5.5	11.1	9.2
Mean		13.60	14.88	18.76

"proteinogram" was used. This proteinogram is a graph indicating the variation in percentage of protein fraction of postoperative stage from that of preoperative stage. There are three types of proteinogram (Fig. 8).

Type A; alpha-globulin and fibrinogen tend to increase and albumin tends to decrease, it is the type of aggravation on the stand point of plasma protein patterns of cancer patients who are divided in three groups by author.

Type B; alpha-globulin and fibrinogen tend to decrease and albumin to increase, it is the type of improvement.

Type C; mixed type.

The proteinogram of cancer patient who received radical operation was belonged to one of these three types. The relationship between the type of proteinogram and survival period of the patient was studied but no statistical significance was observed (Fig. 9).

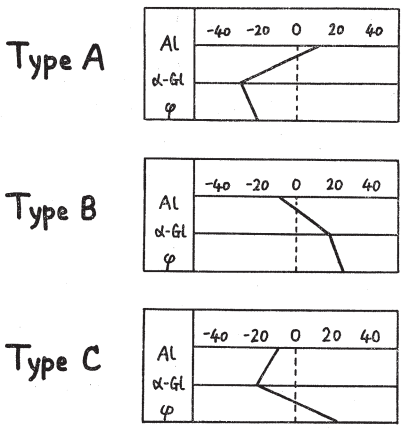

FIG. 8

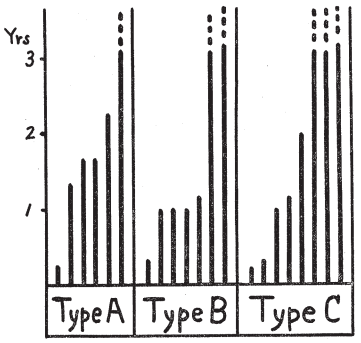

FIG. 9

TABLE 15

Age	Sex		Total Protein (g/dl)	Albumin (%)	α-Globulin (%)	β-Globulin (%)	φ (%)	γ-Globulin (%)	Type of Proteinogram
49	M	preoperative	6.0	45.0	11.4	9.6	10.0	24.0	B
		postoperative	5.8	42.9	8.4	11.7	10.0	27.0	
36	F	preoperative	6.1	41.1	16.5	11.1	17.3	14.0	A
		postoperative	6.7	60.7	11.5	8.7	3.2	15.9	
30	M	preoperative	6.4	45.8	10.8	9.8	13.5	22.0	C
		postoperative	6.8	51.8	10.6	11.6	8.6	17.4	
35	M	preoperative	6.8	76.6	9.3	6.0	2.4	5.7	B
		postoperative	6.9	56.0	7.8	9.1	6.8	20.3	
51	F	preoperative	6.6	70.6	4.8	7.8	3.8	13.0	B
		postoperative	6.5	57.7	11.2	13.5	5.2	12.2	
42	M	preoperative	6.8	57.6	13.9	3.4	6.9	18.2	A
		postoperative	9.6	61.1	12.3	4.5	5.5	16.6	
47	M	preoperative	6.1	67.7	7.2	7.8	3.8	12.9	C
		postoperative	6.6	61.3	9.6	10.7	3.6	14.8	
56	M	preoperative	6.3	58.6	12,1	12.9	5.3	11.1	C
		postoperative	6 0	45.2	13.6	6.4	5.6	29.2	
57	M	preoperative	6.0	70.3	12.3	5.3	9.6	5.5	C
		postoperative	5.9	70.1	11.2	4.9	4.6	9.2	
51	M	preoperative	6.9	55.2	15.1	9.9	3.4	16.4	C
		postoperative	6.1	50.2	11.7	14.0	7.0	13.1	
48	M	pre-operative	7.2	44.3	16.5	14.3	14.6	10.3	A
		post-operative	7.0	55.2	11.6	10.1	8.4	14.7	
48	M	pre-operative	6.0	45.0	11.4	9.6	10.0	24.0	C
		post-operative	5.8	42·9	8.4	11.7	10.0	29.0	
35	F	pre-operative	6.1	50.9	15.9	8.5	11.7	13.0	A
		post-operative	6.7	58.1	8.2	12.9	7.0	13.8	
26	M	preoperative	6.4	54.0	8.6	9.4	8.8	19.4	A
		postoperative	6.8	55.7	17.0	10.7	10.7	9.9	
28	M	preoperative	6.8	67.1	9.4	9.0	5,8	8.7	C
		postoperative	6.9	51.1	8.1	10.8	8.6	21.4	
50	F	preoperative	6.6	63.0	8.9	9.5	5.2	13.4	B
		postoperative	6.5	52.2	13.5	10.6	9.6	14.1	
41	M	preoperative	6.8	60.5	10.0	9.8	6.2	13.5	C
		postoperative	6.6	57.4	11.1	9.7	5.7	15.9	
46	M	preoperative	6.1	60.3	9.1	0.1	9.6	10.9	B
		postoperative	6.6	58.2	8.81	9.8	8.5	14.7	
55	F	preoperative	6.3	52.4	10.1	11.3	10.5	15.7	B
		postoperative	6.0	39.8	15.8	11.8	9.1	23.5	
56	M	preoperative	6.0	65.3	7.4	10.9	5.6	10.8	A
		postoperative	5.9	63.2	10.0	9.2	6.3	11.3	
50	M	preoperative	6.9	57.6	9.9	9.6	6.8	16.1	B
		postoperative	6.1	49.4	15.3	6.8	8.5	20.0	

COMMENTS

Considerable loss of tissue proteins may result minimal decrease in the concentration of plasma proteins, as it is said to be 30 to one[15]. Therefore, a little change of plasma proteins can be raised by a tolerable change of local

tissue proteins. Detailed observations of plasma proteins have revealed that decrease of albumin and increases of globulins are seen in many diseases and, moreover, the changes of plasma protein patterns in one disease can not show any specificity, except for in multiple myeloma, liver cirrhosis and others[16)17)18)].

But observations of plasma protein patterns were said to be useful for the differentiation of cancers from inflammatory diseases (Esser[19)]), for the differentiation of cancers from pulmonary tuberculosis and sarcoidosis (Seibert[20)]) and for the early diagnosis of cancers (Zukschwerdt[21)]). It has been thought that there is no difference in qualities of plasma proteins between patients of cancer and those of other disease but some differences in quantities are there. On the other hand, it was reported by Shimamura[22)] that plasma protein pattern of patient with pulmonary cancer markedly differed from that of pulmonary tuberculosis[23)], pyothorax, mediastinal tumor, tuberculous lymphadenitis of the mediastinum or chronic pneumonia, but had no difference from pulmonary abscess.

These previous works on plasma protein patterns of cancer patients dealt with phenomenalism and revealed no causality without objections.

In this paper, at the first place, the normal values were determined and all these results are in agreement with previous works[12)17)22)24)25)]. The patients with malignancy were divided into three groups according to stages of their malignancies. Differences of plasma protein patterns of patients with malignancy from that of healthy person were observed and obvious differences of the three groups from the normal were observed. The results suggest that the plasma protein patterns of patients who have malignancy of the alimentary tract would advance such as Fig. 10 according to advancing of the malignant tumor.

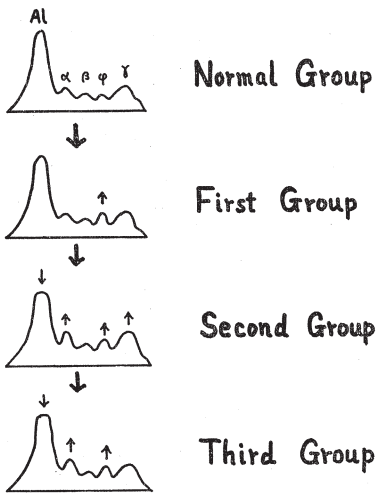

FIG. 10

However, in patients with malignancy of the alimentary tract, obstraction of the alimentary tract can be overcome easily in its early stage in general, so the plasma protein pattern can be easily affected by mal-nutrition. Mereover, in these patients the protein loss caused by continuous bleeding from ulceration of the tumor, which is coexistent almost constantly, could result hypo-albuminemia[26)]. But the plasma protein patterns of patients of gastroduodenal ulcer with pyloric stenosis and professional blood donors who were inadequate for further blood drawing because of anemia resulting from frequently repeated blood drawing were both within

normal ranges. It means that the abnormalities of the plasma protein patterns of patients with malignancy of the alimentary tract are caused by not only mal-nutrition and blood loss, but also the malignant tumor itself.

The changes of plasma protein patterns with malignancy are as following,

1) decrease of total protein
2) decrease of albumin and increase of alpha-globulin
3) increase of fibrinogen
4) gamma-globulin within normal range in terminal stage

1) decrease of total protein

The total protein was said to be decreased in cancer patients by many authors[27)28)29)30)31)32)33)34)], but Locher[35)] reported that the decrease could not always occur. The results of this paper showed that the more advanced the tumors are, the more decreases of total proteins are observed, which indicates the presence of decrease of total protein in cancer patients. The disturbance of liver function by Toxohormon and others are assumed to be main cause and mal-nutrition is not so, because the plasma protein patterns of gastro-duodenal ulcer patients and inadequate blood donors are within normal range.

2) decrease of albumin and increase of alpha-globulin

Decrease of albumin has been reported to be observed in most diseases through their course[4)]. Parallel relationship between severty of a disease and decrease of albumin was presumed by Locher[35)]. Albumin is more easily destructed by proteinase than globulin and albumin in native form is more easily destructed than denatured form[12)]. This evidence makes us to suppose that albumin is most easily affected by change of condition of tissue proteins. On the other hand, resynthesis of albumin is slower than other plasma proteins, so the need of proteins in tumor tissue influences easily and severly on albumin and results decrease of albumin. Moreover, the slower resymthesis of albumin reduces its concentration in the plasma than globulin. But this decrease can not be produced only by a loss of protein, though the loss is chronic, because the plasma protein patterns of inadequate blood donors are within normal ranges. The decrease of albumin can be caused by an increased transportation of plasma proteins to tissue proteins.

Increase of alpha-globulin is accompanied always with decrease of albumin in most diseases[36)] and total amount of alpha-globulin plus albumin varies unremarkably. So it was thought that when albumin decreases, alpha-globulin increases as a compensation. This thought is utterly phenomenalistic. As a fact, in a group of multiple myeloma patients increase of alpha-globulin is not remarkable in spite of remarkable decrease of albumin. Schedlowski and Scudder presumed the relationship between a tissue destruction and an increase of alpha-globulin[37)]. In general, decrease of albumin and increase of alpha-globulin occur by stress in some measure[38)]. In patients with malignancy the

tissue destruction caused by the tumor might result in an increase of alpha-globulin.

3) increase of fibrinogen

Increase of phi-peak in this paper is due to increase of fibrinogen, that was confirmed by measuring fibrinogen by Kijerdal's method. This result is in agreement with earlier works[39]. In general, fibrinogen is said to increase in acute and chronic inflammations. In the report of Schulz[40], increase of fibrinogen was observed in patients with pulmonary cancer also, but no increase in the patients in 3 or 4 weeks before death. But the absence of the increase was not observed in the terminal stage of patients with malignancy in this work.

Fibrinogen decreases markedly in severe liver diseases. This fact indicates fibrinogen to be producted in the liver. Increasing of fibrinogen in early stage of malignacy would be a reaction of the liver, as is the organ of synthesis of fibrinogen, by the humoral stimuli, toxohormon for example, etc.

4) gamma-globulin within normal range in terminal stage

Gamma-globulin fraction is very heterogeneous chemically[4], physiologically[41] and immunologically[42]. The heterogenicity of the gamma-globulin may be demonstrated by immunoserologic methods, ultracentrifugation, measurement of absorption in ultraviolet range and by combinations of electrophoresis and convection[5].

The behavior of the gamma-globulin represents reactive and humoral expression of the reticuloendothelial system, which is in part influenced greatly by preexisting dysproteinemia associated with alpha-globulin elevation.

Increase of gamma-globulin in cancer patients has been believed by many workers[43][44][45][46][47][48]. An apparent increase of gamma-globulin exists when the liver is destroyed by massive intrahepatic replacement by neoplastic tissue. But the result in this papers is that gamma-globulin did not increase but stayed rather within normal limits in patients with abdominal carcinomatosis. Akai[28] reported this phenomenon without comment. A half of gamma-globulin is found in the circulating blood and the other half in the extravascular spaces, especially in the lympnspace of the abdomen and the liver, and on the other hand, the reproduction of gamma-globulin is slow in speed same as albumin. This fact suggests that decrease of gamma-globulin fraction in plasma protein is caused by transudation of gamma-globulin to peritoneal cavity in patient with abdominal carcinomatosis. But protein patterns of ascites of patient with abdominal carcinomatosis is similar to that of plasma of the same patient by electrophoresis[49][50][51].

Basing on these evidence and on the fact that the value of gamma-globulin in patient with hematogeneous metastasis is not high, it seems most reasonable to conclude that not high value of gamma-globulin in terminal stage of patient

with malignancy is not caused by transudation of gamma-globulin to peritoneal cavity as ascites but caused by decreasing of function of the reticuloendothelial system and by disturbed tumor-host relationship[52] and decreased resistence of patient against tumor[53].

In earlier works, changes of plasma protein pattern have been observed for a short period after operation of pulmonary and renal tuberculosis[22], and also on the other report, the postoperative change in gastroduodenal ulcer have been observed for about one week and then afterwards the plasma protein pattern returned to the preoperative level[27][29].

In a short period after operation, plasma protein pattern would be under influences of surgical stress, bleeding, blood transfusion, low intake of diet and others and these influences were said to be continued for one week to 4 weeks by various report[54].

In the result of this study, the influence of operation on plasma protein pattern continued at least 2 weeks. So that, in order to observe the changes of pattern resulted from the radical operation, it is neccesary to pursuate variations of the pattern for more than 3 weeks after the operation. The plasma protein patterns of patients with gastric cancer who received radical operation for more than 3 weeks after the operation did not always return to the preoperative level. Using proteinogram, comparisons between preoperative and postoperative plasma protein pattern were performed and three types of proteinogram were observed but no significant correlation was found between the proteinogram and survival periods of the patients.

From observing the changes of plasma protein patterns of patients with malignancy, possible course and prognosis of the patients have been tried to be inferred. Akai reported that postoperative follow up study showed preoporative high gamma-globulin was a favorable sign for the patient, but in this work no relationship was found between plasma protein patterns of preoperative patient and their survival periods. Recently it was reported that the value of this electrophoretic method for forseeing possible prognosis of a patient was unfounded[55].

CONCLUSION

Free soundary electrophoresis of plasma protein was performed in accordance with "the Standard Method of Tiselius Electrophoresis". The plasma was obtained, using heparin sodium as an anticoagulant.

At first, normal ranges of plasma protein pattern by author's method were determined and all other results were compared with this normal ranges and normal group.

Plasma protein patterns of patients with malignancy, mostly gastric cancer and their operation findings and/or bed side findings were observed and parallel relationship between stages of malignancy and changes of plasma protein

pattern was presumed. Changes of plasma protein patterns of patients with malignancy would be increase of fibrinogen in early stage and increase of alpha-globulin and gamma-globulin and decrease of total protein and albumin in the following stage. Absence of increasing gamma-globulin in terminal stage was observed and the same phenomenon was also observed in patient with hematogeneous metastasis. This absence of increasing gamma-globulin was presumed to be caused by decreasing of function of the reticuloendothelial system and disturbed tumor-host relationship and decreased resistance of the patient against tumor.

In next, plasma protein patterns of professional blood donors with anemia and patients with mal-nutrition from gastroduodenal ulcers with persisting pyloric stenosis were studied and the patterns of both groups were within normal range. By this evidence, it is confirmed that the changes of plasma protein patterns of cancer patients are caused not only by mal-nutrition and chronic blood loss, but also by the malignant tumor itself. A transition of plasma protein patterns of patients with malignancy from preoperative stage to postoperative stage was observed and it was concluded that observation of plasma protein pattern of cancer patient for a short period can not tell prognosis of the patient and the observation for several months or several years would tell it.

ACKNOWLEDGEMENT

The author wishes to express his grateful acknowledgement to Professor Dr. Yoshio Hashimoto for his kind guidance and hearty encouragement.

REFERNCES

1. Doggs, D. R. and Fahey, J. L. Serum protein changes in malignant disease. *J. Nat. Cancer Inst.* **25**: 1381, 1961.
2. Coppi, F. and Gallivanone, A. Electrophoresis in urogenital tumours. *Arch. Ital. Urol.* **33**: 339, 1960.
3. Tiselius, A. *Farad. Soc.* **33**: 524, 1937. Cit. from 4.
4. Wuhrmann, F. and Wunderly, C. Die Bluteiweisskörper des Menschen. Benno Schwabe Verlag, Basel, 1952.
5. Riva, G. Das Serumeiweissbild. Hans Huber Verlag, Bern, 1960.
6. Krystosik, J. and Kwiatkowski, M. Serum protein in various neoplastic disease before and after treatment with X-ray. *Ann. Univ. Curie-Sklodowska.* **13**: 113, 1959.
7. Petermann, M. L. Plasma protein abnormalities in cancer. *Med. Clin. North. Amer.* **45**: 537, 1961.
8. Stickler, G. B. and Pinkel, D. Serum protein fraction in children with malignant tumor. *Cancer* **13**: 446, 1960.
9. Sieberth, E. Über die Bedeutung der Papierelektophorese für die Prognose und für die Therapie Maligner Erkrankungen. *Krebsarzt* **15**: 152, 1960.
10. Miller, B. J. and Ert, L. The serum protein and lipoproteins in patients with carcinoma and in subjects free of recurrence. *Snrg. Gyne. Obstet.* **105**: 487, 1956.
11. Hayase, T. Studies on the preservation of human blood, on the preservation of the

heparinized blood; dextran sulphurized blood and heparinized blood added with mannitol. *J. Jap. Soc. Blood Transfusion* **13**: 1966 (in Japanese).
12. Hirai, H. and Shimao, K. Electrophoresis. Kyoritsu Press, Tokyo, 1955 (in Japaness).
13. Lewis, A. L. Electrophoresis in Physiology. Charles C. Thomas, Springfield, 1960.
14. Hirai, H. *et al.* Method of Electrophoresis. Bunkodo, Tokyo, 1963 (in Japanese).
15. Cantarow, A. and Schepartz, B. Biochemistry. Saundars, Philadelphia, 1957.
16. Berndt, H. and Schwarz, H. The blood-proteins in brochial carcinoma and chronic pneumonia. *Arch. Geschwülstforsch.* **18** 209, 1961.
17. Misawa, N. Paper electrophoretic observation in gynecology and obstetrics. *J. Nagoya Med. Ass.* **82**: 100, 1960 (in Japanese).
18. Nakata, K., Shibata, K., Suzuki, T., Tsuzuki, K. and Ibi, M. On serum protein pattern of various disease using paper electrophoresis. *Saishin-Igaku* **17**: 865, 1962 (in Japanese).
19. Esser, E., Heinzler, F. and Wild, H. Serumeieiweisswerte beim Carcinom. *Klin. Wschr.* **31**: 321, 1953.
20. Sieberth, E. Über Veänderungen der Serum α-Globuline und Leukozyten als Ausdruck der Alarmreaktion bei der Karzinoma Krankheiten. *Krebsarzt* **14**: 11, 1959.
21. Zukschwerdt, L., Knedel, M. and Zettel, H. Eiweissprobleme in der Chirurgie. *Dtsch. Med. Wschr.* **77**: 640, 1952.
22. Shimamura, K. Plasma protein pattern in lung cancer. *J. Jap. Surg. Soci.* **56**: 1205, 1955 (in Japanese).
23. Knedel, M. and Zettel, H. Elektrophoretische Untersuchungen über das Verhalten der Bluteiweisskörper beim Bronchialcarcinom. *Klin. Wschr.* **30**: 596, 1956.
24. Akai, S. Observation of plasma protein and serum protein of patients with malignancy by electrophoresis. *Niigata Med. J.* **69**: 74, 1955 (in Japanese).
25. Yoshida, T. Comparative studies of electrophretic analysis of plasma proteins and pathological aspect in cancer patients, especially gastric cancer pateints. *J. Jap. Surg. Soci.* **61**: 340, 1960 (in Japanese).
26. Nishihara, T. On serum protein in patients with anemia. *Sogo-Igaku* **11**: 421, 1954 (in Japanese).
27. Aga, H. Protein content of blood plasma by Tiselius electrophoresis before and after operations of some surgical diseases measured. *Niigata Med. J.* **67**: 37, 1953 (in Japanese).
28. Akai, S. and Yoshida, T. Comparative studies of electrophoretic analysis of plasma proteins and pathological aspects in gastric cancer. *Jap. J. Cancer. Clin.* **2**: 470, 1956 (in Japanese).
29. Akamatsu, Y., Takao, S. and Kurihara, Y. Statistical observation of plasma protein patterns by electrophoresis in Ginnai Clinic; on gastric cancer and gastric ulcer. *Geka* **17**: 179, 1955 (in Japanese).
30. Berndt, H. and Schwarz, H. The blood-proteins in bronchial carcinoma and carcinoma of the stomach. *Arch. Geschwülstforsch.* **18**: 206, 1961.
31. Bartolomucci, D. and Colella, C. A. Changes of the serum protein pattern in cancer patients subjected to telegamma therapy with Co 60. *Ricerca Sci.* **30**. 1325, 1960.
32. Goswami, P. and Barna, R. K. Paper electrophoretic serum protein pattern in carcinoma. *Indian J. Med. Sci.* **14**: 17, 1960.
33. Marinucci, M. and Pannella, A. The changes in total protein and in the glycoproteins, as determined electrophoretically in cases of cancer. *Riv. Path. Clin.* **14**: 883, 1959.
34. Nettleship, A., Stronther, J. A. and Smith, H. S. Relationship of serum and urinary proteins in neoplastic states; a preliminary survey. *Clin. Chem.* **9**: 609, 1963.
35. Locher, R. Das elektrophoretische Serumeiweissbild bei der krebskrankheit. Bern, 1951.

36. Scheurlen, P. G. Der Einfluss des Albumin die Konzentration der Serumglobuline. *Dtsch. Med. Wschr.* **88**: 260, 1955.
37. Doutenwill, W., Ranz, H. and Moor, U. The significance of necrosis as a cause of blood protein changes in tumors. *München Med. Wschr.* **101**: 1365, 1959.
38. Medvedev. Paper electrophoresis of proteins while preserving the pattern of their localization in tissues. *Nature* **191**: 1021, 1961.
39. Tasaki, Y. Furue, H. and Sakai, K. Cancer and fibrinogen. *Gann* **47**: 487, 1959 (in Japanese).
40. Schulz, E. *Dtsch. Med. Wschr.* **80**: 260, 1955 Cit. from 5.
41. Jahnke, K. and Scholtan, W. Die Bluteiweisskörper in der Ultrazentrifuge. Georg Thieme Verlag, Stuttgart, 1960.
42. Plesnicar, S. and Romes, A. Immunoelectrophoretic analysis of serum proteins of patients with malignant tumours. *Tumori* **48**: 13, 1962.
43. Alexandrov, K. Electrophoretic studies of blood serum proteinin cancer patients. *Vopr. Onkol.* **7**: 64, 1961.
44. Barclay, M., Calathes, D. N., DiLorenzo, J. C. and Helper, A. The relation between plasma lipoproteins and breast carcinoma. *Cancer* **12**: 1163, 1959.
45. Galehr, O. Die Serumeiweisskörper bei malignen Tumoren. *Wien Arch. Inn. Med.* **9**: 378, 1952.
46. Takahashi, S. Variations in the protein fraction of transplantable tumor (Katsumi's Sarcoma) bearing dogs. *J. Nagoya Med. Ass.* **78**: 150, 1959 (in Japanese).
47. Zettel, H. and Endress, M. Das Serumeiweissbild beim Carcinom. *Chirurg.* **24**: 498, 1953.
48. Zographov, D. G. Das Blutbild beim Mammakarcinom. *Zbl. Chir.* **88**: 283, 1963.
49. Burlian, J., Dugandzic, S., Bugarski, O., Rodic, S. and Jevtic, Z. Electrophoretische Untersuchungen des Eiweisses des Magensaftes. *Klin. Wschr.* **40**: 597, 1962.
50. Shimura, H. and Tamazawa, K. Electrophoretic study on plasma protein and protein of ascites in patients with malignancy. *Geka* **16**: 176, 1954 (in Japanese).
51. Tabuchi, N. Studies on the protein patterns of patients with gastric cancer. *Jap. J. Cancer Clin.* **6**: 490, 1960 (in Japanese).
52. Grace, J. T. and Lehoczky, A. Tumor-Host relationship factors in man. *Surgery* **46**: 238, 1957.
53. Dufour, D., Berlingnet, L. and Loiselle, J. M. Antigenic property and electrophoretic behavior of the serum albumin of rats bearing Walker tumor. *Canad. J. Biochem. Physiol.* **37**: 1401, 1959.
54. Kessel, M. and Kessel, J. Elektrophoretische Verlaufsuntersuchungen bei Tumoren. *Zschr. Klin. Med.* **151**: 526, 1954.
55. Fischer, R. Was leistet die Electrophorese der Bulteiweisskörper fur die Diagnose und postopertive Prognose maligner Tumoren? *Langenbeck Arch. Klin. Chir.* **307**: 225, 1964.