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Immunoglobulinopathy: Its Diagnosis and Development

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Summary. **A four item screening system was established to detect monoclonal serum proteins: that is, a marked increase in serum total protein over 10 g/100 ml, abnormally low values of TTT and ZTT or their marked dissociation, and dissociation of the results of urinary albustix and sulfosalicylic acid tests.**

A high rate of association of myeloma with other malignant tumors was noted in our series. Twelve malignancies in 11 patients were confirmed in 43 cases with multiple myeloma in 1980. Recently succesive resections of the carcinomas of the sigmoid colon, ascending colon and stomach (double cancers of the stomach) were performed on an 84-year-old male, who is still alive.

Treatment of multiple myeloma should not be too aggressive. Maintenance therapy with low dose anticancer drugs was necessary in most of the cases.

A Method for Detecting Monoclonal Gammopathy from Routine Laboratory Test Results

The experience of a 53-year-old female with multiple myeloma gave me the impetus to undertake an extensive survey on monoclonal gammopathy. The patient underwent a mastectomy because of breast cancer when she was 46 years old. Preoperative laboratory examinations revealed monoclonal protein at the mid gamma region, but thymol turbidity (TTT) and zinc turbidity (ZTT) test, LDH, PSP, erythrocyte sedimentation rate (ESR), blood counts and urinalysis were all within normal limits. A radical mastectomy was done, but no significant reduction in M protein was noted a month after the operation.

She had been free of symptoms for seven years after the operation when she visited our department because of a marked increase in the amount of the serum monoclonal protein. On re-admission the total serum protein was 15.8g/100 ml, the amount of the M protein of IgG lambda type reached 11,392 mg/100 ml, TTT/ZTT 0.4/25.0, ESR 165 mm/h, and many punched out lesions were consistent with multiple myeloma. Repeated bone marrow examinations revealed atypical plasma cells but their percentages were between 3.6 and 10.0 per cent of the total nulceated cells of the bone marrow smears, disproprotionate to the large amount of the serum M protein.

This exprience rendered many problems to be solved, including:

(1) The earliest finding of multiple myeloma;

(2) Correlation of morphological findings of the bone marrow plasma cells and serum M protein;

(3) Whether there exists any chance that so-called benign monoclonal gammopathy develops into multiple myeloma.

Mass survey on monoclonal gammopathy was started in June 1976, in the Department of Hematology, Toranomon Hospital, with assistance of the Departments of Clinical Chemistry, Hematologic Research, Pathology and Immunology.[1)]

Screening Items for the Monoclonal Immunoglobulin(s)[2)]

(1) Total protein of the serum beyond 10 g/100 ml.

(2) M-bow (definite or suspected) on the protein electrophoresis.

(3) Markedly low TTT and ZTT (below 1) or marked dissociation of TTT and ZTT (i.e., TTT under 1 and ZTT over 11),

(4) Dissociation of the results of albustix and sulfosalicylic acid tests for urinary protein (i.e., negative or trace albumin vs. a markedly large quantity of the total protein)—a frequent finding of positive Bence Jones proteinuria.

Seventy-six sera with monoclonal immunoglobulin

were detected through this "four-item screening for the M protein" from a population of about 40,000 out- and inpatients of Toranomon Hospital with various disorders over a period of one and a half years.[1]

Of these, 2/3 were of IgG, and for the light chains, the frequencies of kappa and lambda were almost equal. IgA was next, with the other types being rare.

Premyeloma[3]

Many cases with monoclonal immunoglobulin(s) run a long, uneventful course; they are called "benign" monoclonal gammopathy. Moreover, some cases of definite myeloma do not show any positive symptoms, and remain in latency for many years.

Many terms have been coined to show the diseased state such as "pre-myeloma:" MGUS (monoclonal gammopathy of undetermined significance), smoldering myeloma, or low-percentage myeloma. A diagnosis of the premyelomatous state is easy after the patient develops overt myeloma; it is not so easy to make its diagnosis at the first visit of the patients.

The author made a diagnosis of "premyeloma" for a patient with IgGL monoclonal immunoglobulin during its asymptomatic phase (Table 1). ESR was almost within normal limits, but a dissociation between TTT and ZTT was evident. EgG was 6,347 mg/100 ml. A bone marrow smear from a sternal marrow aspirate in April 1977 showed only 3.6% plasma cells but with an evident atypism. Fig. 1 shows a binuclear plasma cell in a smear from a sternal marrow aspirate with prominent nucleoli. A clot section from the same material revealed no pathological findings suggestive of myeloma. A diagnosis of premyeloma was made. Unfortunately, an overt plasmocytoma —plasma cells numbering more than half of the nucleated cells in a bone marrow smear as show in Fig. 2 were atypical and pathological findings were consistent

Table 1. M.O., Born 1917, male IgGL myeloma

	Tp (Gm/100ml)	Ⓜ%	IgG (mg/100ml)	IgA (mg/100 ml)	IgM (mg/100ml)	CRP / ESR	TTT / ZTT	Plasma cells in the Bone Marrow Aspirates: % (Atypsim)	Histology
March 1977	8.8	26.0%	6347	31 ↓	49 ↓	−/16	0.2/16.9	April 1977 3.6% (+)	Focal, nodular infiltrations (−)
October 1977	8.9	—	5166	27 ↓	34 ↓	−/28	0.2/22.6	.....	.....
May 1978	10.6	—	5450	32 ↓	42 ↓	−/80	0.8/50 ↑	.....	.....
September 1978	12.1	54.9%	9600	30 ↓	32 ↓	−/117	0.2/50 ↑	September 1978 55.2% (++)	(++)

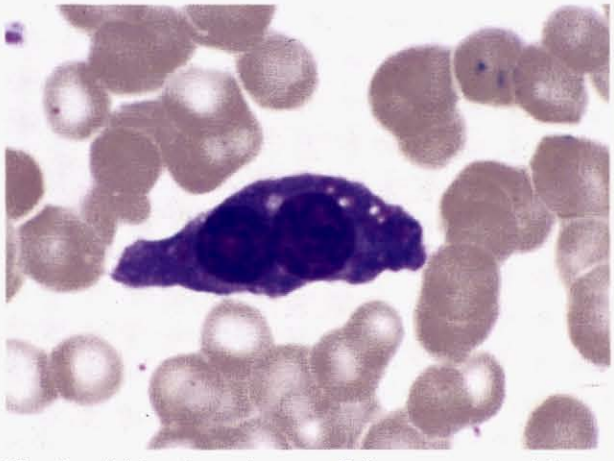

Fig. 1. A binuclear plasma cell from a smear of the sternal marrow punctate on April 28, 1971. (M.O., premyelomatous state)

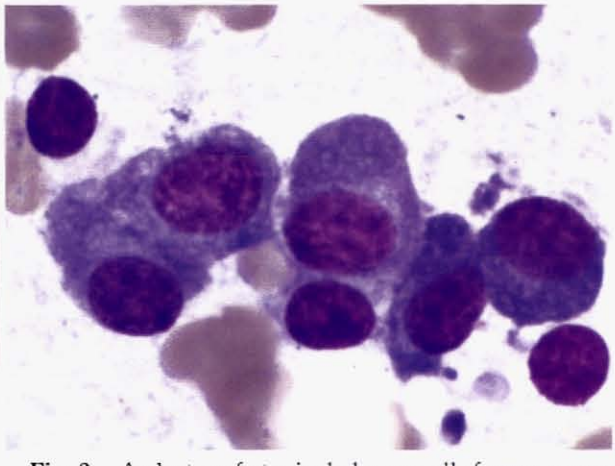

Fig. 2. A cluster of atypical plasma cells from a smear of the sternal marrow punctate on September 26, 1972. (M.O., overt myeloma)

with myeloma— developed one and a half years after his inital visit and he succumbed to an overwhelming infection shortly thereafter.

Complete Remission and Longevity in Multiple Myeloma

The first target in the treatment of acute leukemia is to achieve a complete remission. In the cases of multiple myeloma, it it difficult to achieve this in most cases, partly because the ages of the patients are advanced, and, moreover, there is no definition of "complete" remission which is generally accepted.

A 66-year-old female clerk was first admitted in March 1976 because of lumbago. A sternal marrow aspirate revealed numerous myeloma cells, a marked increase in serum IgA (IgA kappa monoclonal immunoglobulin) was confirmed, and a diagnosis of IgA kappa myleoma was made. Combination chemotherapy with prednisolone, vincristine and melphalan resulted a partial remission, and with two-and-a-half years' trearment with prednisolone and melphalan complete remission was achieved: serum monoclonal protein disappeared in a paper electrophoretic pattern, serum IgA level was 270 mg/100 ml, that is, 1/15 initial level, and a marked increase in IgG was evident, although a faint monoclonal protein was identified in an immunoelectrophoresis; a percentage of the plasma cells in a marrow smear was 1.0% with slight atypism.

She was put on a maintenance chemotherapy with a small dose of prednisolone and melphalan. After seven months a monoclonal protein reappeared in the electrophoretic figures. The patient died of pneumonia five months after the reappearance of the IgA kappa monoclonal protein.

Usefulness of ZTT[4]

Although many laboratories in Western countries have abolished the zinc turbidity test because of its uselessness, it is still efficient as a screening test of multiple myeloma and in the observation of its clinical course.

A male, born in 1914, visited the hospital because of gouty attacks. Monoclonal protein was detected by routine blood examinations: Total protein 7.2 g/100 ml, with monoclonal protein of IgG kappa type 7.3%, ESR 13 mm/h and TTT/ZTT 1.5/4.9.

Regular examinations of serum protein revealed a gradual increase in the amount of the monoclonal protein, and the valeue of ZTT also increased in parallel with the level of serum M protein as shown

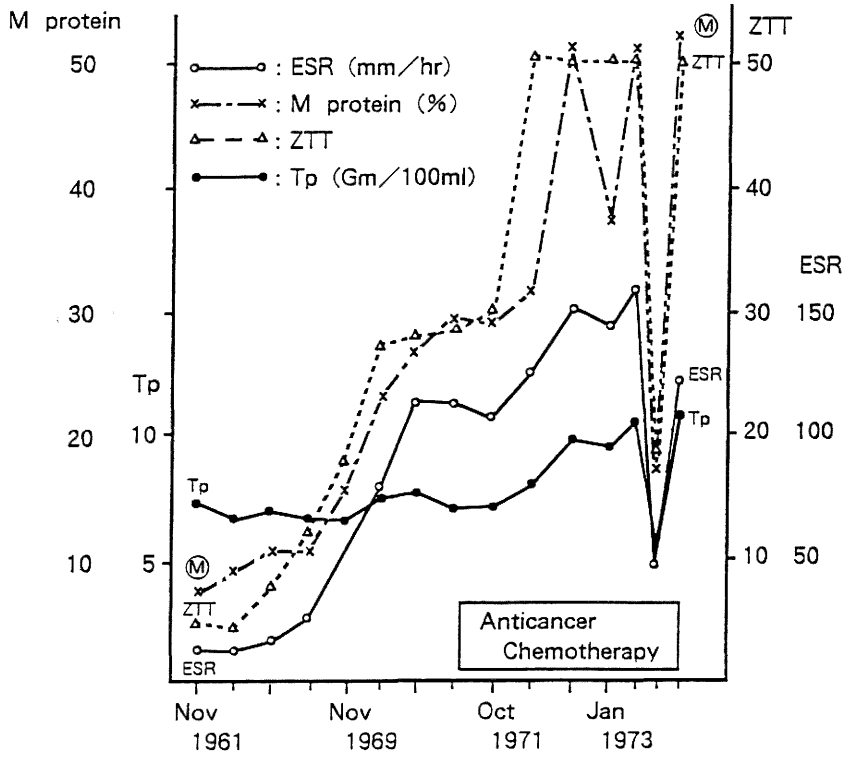

Fig. 3. Male, born 1914. IgGK myeloma.

in Fig. 3. The curves of ESR followed almost the same pattern, but its changes lagged slightly behind.

Ten years after his first visit, monoclonal protein in the slow gamma region (IgG kappa type) increased to 31.3 percent, dissociation of TTT/ZTT was evident (1.1/27.0), and IgG 2,900 mg/100 ml and the levels of the IgA and IgM were very low.

In addition, 25.8 percent of the nucleated cells in a sternal marrow aspirate were atypical plasma cells and kappa type Bence Jones proteinuria was ascertained. A diagnosis of IgG kappa myeloma was made. In 1972 a sudden increase in the monoclonal protein and simultaneous increas e in ZTT were observed; the value of ZTT was off the scale (over 50). Combination chemotherapy with vincristine, cyclophsophamide and prednisolone resulted in simultaneous sharp decreases in monoclonal protein and ZTT levels.

Multiple Myeloma Preceded by Solitary Plasmocytoma[5]

Solitary plasmocytoma may develop into multiple myeloma, but it is, in general, a rare event. We have recently experienced a case of myeloma in which four years after the complete removal of a left lung tumor —at first diagnosed as lung cancer, and the tumor histologically confirmed as plasmocytomal— developed into an overt multiple myeloma with Bence Jones proteinuria.

Serum total protein was 8.1 g/100 ml, including 22.6% M protein (IgG kappa); smears from the bone marrow aspirates revealed from 15.4 to 38 percent atypical plasma cells. After four weeks' VEP therapy, the amount of the M protein in the serum was reduced from 1,831 mg/100 ml, and the patient was asymptomatic.

Treatment of Myeloma in a Patient of Advanced Age[6]

A female, born in 1901, suffering from hypertension, was found to have markedly elevated serum protein and gamma globulin levels and an accelerated red cell sedimentation rate by routine laboratory examination. With a complaint of severe chest pain, she was referred to the author's clinic in February 1979.

Laboratory examinations revelaed anemia, a markedly accelerated red cell sedimentation rate, high serum protein, massive M protein in the slow gamma regions and marked dissociation of TTT and ZTT. Serum M protein was of the IgG kappa type, and a smear from the sternal marrow punctate revealed 9.8 percent of plasma cells with prominent atypism. A clot section from the same material showed a proliferation of plasma cells in small islets.

At first she refused admission because of her age and severe lumbago, but after repeated recommendations she reluctantly entered in March 1979. Under a diagnosis of IgGK myeloma VEP therapy was started. After five weeks' treatment and maintenance chemotherapy with prednisolone and melphalan, the level of the serum M protein fell as low as 2.3 percent initial level, and the level of ZTT declined from 30.3 to 0.8, well paralleling the serum level of the abnormal IgGK globulin (Table 2).

Improvement of her lumbago was noteworthy. When she was first admitted to the hospital, she could

Table 2. R. M., born 1901, female, IgGK myeloma

	Feb 1979	Aug 1979	Jan 1980	Feb 1981	May 1982	Oct 1985	Apr 1988	Jul 1989
Hb (Gm/100 ml)	9.0	9.3	9.9	11.4	11.5	11.7	11.1	10.5
ESR (mm/hr)	119	93	60	33	28	64	44	56
Total protein (Gm/100ml)	10.9	9.5	6.5	6.7	6.4	7.5	7.4	7.3
M protein (%)	58.0	39.8	6.2	5.7	2.3	21.2	18.4	17.2
M protein (mg/100ml)	6,322	3,781	403	382	147	1,590	1,262	1,256
TTT	1.0	3.1	0.6	1.4	1.1	2.8	2.0	1.0
ZTT	30.3	34.7	1.3	1.1	0.8	16.8	13.1	12.1
	← VEP 5 wks →			{ Prednisolone 5-10 mg/day Melphalan 4 mg/day~2mg×2/week →				

not walk, but on discharge was able to walk without a cane. She led a normal life for more than ten years thereafter, dying at the age of 89 of pneumonia on February 17, 1991, twelve years after her first visit.

In this case, too, the ZTT level was a good indicator of the disease activity of IgG kappa myeloma.

Coincidence of Multiple Cancers is a Case of Multiple Myeloma[7)]

A high rate of association of malignancies in cases of multiple myeloma was stressed in 1981, referring to eleven combined malignancies out of 43 cases of multiple myeloma under my care.

An abnormal band was detected by routine laboratory tests in the serum of a male, born in 1907, who visited the Department of Nephrology because of a persistent elevation of BUN (28–34 mg/100 ml). He was referred to the Department of Hematology in 1978. Laboratory examinations revealed serum total protein 8.4 g/100 ml, TTT/ZTT 0.3/2.2, and 141, the percentage of M protein being confirmed at the fast gamma region. CBC was within normal limits and no Bence Jones proteinuria was noted. Immunoglobulin levels were IgG 2,440, IgA 150 and IgM 50 mg/100 ml. Bone marrow aspirations were done on three occasions. Around five percent of the nucleated cells were considered to be atypical plasma cells with multiple muclei and/or prominent nucleoli. A diagnosis of IgG lambda myeloma was made. In September 1970 an emergency admission because of severe abdominal pain resulted in the discovery of villous adenoma of the sigmoid colon with focal cancer. In 1971 a partial sigmoidectomy in a length of 8.4 cm was performed. Further, in July 1974, three moderately differentiated adenocarcinomas were resected from the ascending colon, and in 1988 gastrectomy was done because of two separate cancers (IIc lesion in the corpus, IIa lesion in the prepylorus). The amount of the serum M protein remained unchanged without any specific therapy for the myeloma.

Triple cancer, that is, IgG lambda multiple myeloma, follicular thyroid cancer, and poorly differentiated subcutaneous cancer on the body wall were coincidentally experienced in a 63-year-old female, and many often cases of double cancers have already been reported; however, quadruple malignancy in multiple myeloma is a very rare complication.

Table 3. C.S., born 1929, female, Benign Monoclonal Gammopathy (IgGK)
Course of the results of the laboratory tests

	January 1975	April 1980	February 1986	December 1990
Tp (g/dl)	8.4	8.5	8.3	8.2
Alb (%)	58.7	58.9	57.4	57.1
α_1	3.7	2.4	1.8	1.9
α_2	6.1	6.1	6.2	5.9
β	8.9	8.4	8.8	8.4
γ	5.9	4.5	5.7	7.7
Ⓜ fast γ	16.7 (1403 mg/dl)	19.4 (1649)	20.1 1688)	19.0 (1558)
Hb (g/dl)	10.4	13.3	13.2	13.0
WBC	6.300	5.500	5,400	5,300
Thromb	301,000	217,000	229,000	206,000
Urine: Albus/Sulfo	−/10 ↓	−/10 ↓	−/10 ↓	−/3
CRP	—	—	—	—
ESR (mm/hr)	8	12	16	18
TTT/ZTT	1.5/1.3	1.0/2.7	0.5/10.0	0.4/9.2
Creatinine (mg/100ml)	0.6	0.9	0.7	0.7
LDH	183	154	161	139
Sternal aspirate	NCC 150,500 Plasma cells 1.8% Atypism (−)			
IgG (mg/dl)	2,076	3,400	1,913	2,226
IgA	102	148	100	87
IgM	123	89	49	63

A Case of Benign Monoclonal Gammopathy Followed Regularly for Sixteen Years

In most cases of benign monoclonal gammopathy, the amount of serum M protein remains unchanged for many years, usually between 5 to 20 percent (i.e., 350-11,600 mg/100 ml). An abnormal band was detected in a serum electrophoretic pattern of a woman born in 1929, who complained of nasal bleeding and frequent attacks of "cold."

Results of an examination in January 1975 are shown in the left column of Table 3. M protein was detected at an amount of 16.7 percent at the fast gamma region, with total serum protein 8.4 g/100 ml. Serum IgG increased to 2,760 mg/100 ml, with normal values of IgA and IgM. ESR and TTT/ZTT were within normal limits. Complications of iron deficiency anemia due to hypermenorrhea were successfully overcome by the oral administration of iron. A sternal marrow aspirate revealed a normal percentage of plasma cells without atypism. The patient's condition has been uneventful for sixteen years, and no significant increase in the monoclonal protein has been noted, as shown in Table 3. She underwent an operation for a caruncle of the urethra in 1989 without any problems.

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