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(From Med. Clin. Akad. Hospital, Upsala (Sweden). Chief: Prof. G. Bergmark).

Incipient myelomatosis or «essential» hyperglobulinemia with fibrinogenopenia — a new syndrome?

By

JAN WALDENSTRÖM.

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The real nature of myelomatosis.

The title of this paper may at first seem somewhat surprising. The myeloma has of old had a reputation as a well defined clinical entity. With the aid of the typical changes on the X-ray film and guided by the examination of the cells from a sternal puncture the diagnosis should therefore be easy and there ought not to be found any serious diagnostical troubles. In the following I am going to give a description of two cases, who have several symptoms suggesting myelomatosis but also show decided differences. They are very much alike even as regards details in the chemistry of the blood proteins and it seems probable according to my opinion, that they suffer from the same malady. A third case very much resembles these two patients but also shows other signs, that do not fit in so well with the picture.

It might be possible to regard these patients as myeloma without myeloma i. e. the hypothesis may be propounded, that we have to do with a premyelomatous stage of the malady with only increase in serum globulins but not yet with the secondary deposition of myeloma tissue, plasma cells, in the bone marrow (Waldenström 1942). I have recently pointed out, that many facts in the

clinical picture of myeloma would be explained, if we assume that the malady were really a thesaurismosis. The primary change should be a deranged protein metabolism with secondary deposition of these pathological products, above all globulins, in the bone marrow. If this were true the disease ought not to be regarded a sa true neoplasm but rather as an analogon of the socalled lipoidoses e. g. Mb. Gaucher or Schüller-Christian (Also the skeletal xanthomata were until quite recently regarded as tumours). The real cause of the disturbed protein metabolism is as yet unknown but some possible explanations will be discussed later.

Before I proceed to a discussion of the diagnosis etc. I shall give the case histories.

Case histories.

Case I. Farm labourer born in 1881. Nr. 794/42. Hereditarily nothing known (mother dead, father unknown). Previous maladies of no interest. In *Aug. 1935* pains in the shoulders and left wrist. Admitted to the Medical Clinic on Aug. 19th (for hematological data se table 1).

From internal organs nothing of interest. Blood pressure 145/110. Left-sided hydrocele, the size of cocoanut. Left shoulder no swelling or redness but diffuse painfulness on pressure chiefly from the axilla. All movements painful. Other joints show nothing abnormal. No anemia, W. R. neg., no albuminuria. X-ray: Left shoulder: no skeletal changes, no calcified bursa. Heart: aortic configuration. Lungs: normal. No fever. Later several severe nose bleedings which caused some anemia. Dismissed on Nov. 22d »no cause of the increased S. R. has been found». — Ordered iron pills and active movements. After springtime 1936 no troubles from the joints but several, severe, nose bleedings. In the springtime *1941* 3 weeks in the Otiatric Clinic for these complaints. On *March* 16th *1942* »influenza». Admitted on March 21st with typical signs of lobar pneumonia. Has had no severe bleedings lately. From status: Dyspnoic, slight edema of the feet, some cyanosis. Rather severe eczematous changes on the trunk. Slight on arms and legs. Typical signs of pneumonia sin. (X-ray) Heart nothing abnormal. Bloodpressure 180/115. High fever. Blood values: se table. The condition of the patient improved rapidly. Fractional test meal: Free HCl 55 after 0.2 g caffeine. Serumiron 56 γ %; non protein nitrogen 43 mg %. Meulengracht 2. Electrocardiogram: nothing pathological. From the hydrocele were drawn 2 litres of fluid. X-ray of cranium and pelvis: no signs of myeloma. W. R. neg. (slight auto-inhibition). The status of the patient was controlled several times. He usually showed some albuminuria, but no sediment or Bence-Jones proteinuria. Blood examination: see table. Has been able to work during the autumn but has had some severe nasal and gingival bleedings. No pains of any

kind. The patient was treated several times at the Medical Clinic with venesections, retransfusions of his own erythrocytes and transfusions. No real change in the status during the year but there were found a few enlarged lymph glands. One of these was excised. After this operation *bleeding from the wound for more than a fortnight.* Micr. picture: catarrhal changes in the sinuses with localised fibrous and hyaline induration. Some increase of plasma cells. No signs of tumor. Decidedly no myeloma (Gellerstedt).

In *Jan. 1943* new transfusion after severe epistaxis. Coagulation-time tested with glass-bead in a small test tube 11.5 min. (normal person 3.25 min.). Bleeding time 8.5 min. (normal person 2.5 min.). Meulengracht 4. The values for citric acid and phosphatase were normal. The patient was treated with transfusions of in all 900 cm^3 and large venesections of a litre; the red corpuscles were allowed to sediment and were reinfused. After this treatment no bleedings.

Readmitted to the Surgical Clin. on *March* 23rd *1943* for severe abdominal pains on the right side. From the status: severe meteorism and painfulnes in the right upper abdominal quadrant. Leucocytosis of 14000 cells. In the sediment there were found some erythrocytes. The pains disappeared after a few days. The patient was treated with large transfusions of in all 1300 ml blood. In spite of this not more than 1.5 mill. Ery. Bloodpr. 115/75. Persistent slight albuminuria. No casts Bence Jones neg. Water tolerance test (1000 ml). In 2 hours 280 ml; lowest spec. grav. 1,012. 4 hours 405 ml, 1370 ml in 24 hours, with highest spec. grav. 1,021. Decreased from 65—63.3 kg in weight. Blodpressure 115/75. Sternal puncture: no marrow could be aspired. On April 3rd suddenly pains in the right knee joint (probably bleeding). Ocular fundi show multiple bleedings round the papillae. X-ray of the skeleton only showed a periosteal thickening on the left femur.

A new water tolerance test was performed with 1500 ml instead of one liter. During the first 2 hours excretion of 310 ml in 4 hours 515 in 24 hours 1400. Spec. grav. 1,017—1,020. After several transfusions his status was considerably improved. He had no bleedings and felt much better. Dismissed with Hgb 30. Red. 1.6, White 7000 on May 27th.

The serum and blood of the patient showed many peculiarities which will be discussed and described in the text.

Case II. Farmer born in 1878. No 2394/1942. Hereditarily nothing of importance. In 1928 trauma against the left eye, with luxatio lentis et ablatio retinae. Since Aug. 1941 decreasing vision on the right eye. Treated in the Ophthalmiatric Clinic Aug. 9th—Aug. 16th 1941 for thrombosis venae centralis retinae dxt. Since then status quo. Is not able to read. Oct. 1941 bleedings from nose and gingiva. Treated with injections without any certain effect.

Admitted to the Med. Clin. on April 29th 1942. No dyspnoea, no fever, some bleedings from the gingiva. Some enlarged lymph glands in the fossae supraclaviculares and below the mandibula. Heart: nothing abnormal.

Blood pressure 120/85. No albuminuria. Urobilin: traces. Urobilinogen neg. X-ray: skeleton no signs of myeloma. W. R. neg. Meulengrach 2. Citric acid 31.4 γ/ml. Phosphatase 3.6 Ph. units. Readmitted several times during the following months for bleedings. Blood values see table. Treated with large transfusions; bleedings very much improved. Feels better than for several years. In Oct. and Dec. 1942 and Febr. 1943 treadmitted for transfusions. X-ray skeleton several times, no signs of myeloma. Serum calcium normal, citric acid very high (79.7 γ/ml.) Phosphatase low. Meulengracht 4.

Readmitted on April 7th 1943 for transfusions. Has been quite well for about a month after the last transfusions. During this time no bleedings from the nose or mouth but these have relapsed of late. Has felt very tired and dyspnoic. No edema, no pains of any kind. On admittance gingival and nasal bleedings. Lymph glands as before. Heart nothing abnormal, blood-pressure 125/95. No albuminuria. Hgb 36 %, Ery 2.1 mill. Leucocytes 9500. Platelets 120000. Prothrombin index 103. F-gel 10 sec. Retic. 12‰ *Coagulation time (Howell-Gram) 10 min. A sternalpuncture* was attempted but it was impossible to aspire any fluid through the needle. There was continous bleeding from the wound for more than 12 hours. Water tolerance test. 1000 ml. After 2 hours 545 ml, 4 hours 790 ml., 24 hours 1800. Spec. grav. varied between 1.009 and 1.020. Citric acid in the serum 76.8 γ/ml. Phosphatase 2 Ph. un.

Readmitted on May 27th 1943. Has felt better but never been free from bleedings which have increased considerably during the last days. The mouth has been filled with blood. Status as before. Blood pressure 105/75. Hgb 20 %, Ery 1.3, No albuminuria. Platelets 115000. After a transfusion considerable improvement. Hgb 30 Ery 1.6. White 6700. Dismissed on June 10th.

Case III. Farmer born in 1880. No. 1095/1941. Hereditarily nothing of importance, no previous maladies. In 1940 some coughing and anemia. Treated with injections. Admitted on April 16:th 1941 for fatigue and dyspnoea. No pains, no loss of weight, no fever. Some lymph glands the size of a bean on the neck, in the axillae and inguina. Internal organs otherwise normal. Blood pressure 120/70. No albuminuria or urobilinuria. Free HCl 75 after 0.2 caff. A lymph gland was excised. Mitroscopical diagnosis: localized proliferation of the reticulum but without any signs of granuloma or tumor. Partial reticulosis (Gellerstedt). W. R. neg. No autoinhibition. Blood values see table.

The status of the patient has been followed several (7) times in the Outpat. Dep. The Hgb was varied between 56 and 38 %. The Ery 3.8 to 2.0 mill. Serum iron low 25 γ %. Persistent slight albuminuria. No Bence *Jones protein. Sed: nothing* pathological. The skeleton was x-rayed several times but nothing pathological was found except a spondylitis deformans.

Readmitted on March 18th 1943 for increasing fatigue. Otherwise no special symptoms, no pains. Moderately enlarged lymph glands as before. Blood pressure 120/70. X-ray: Skull, vertebral column, no signs of mye-

Table 1.

Chemical and hematological data.

Date	S. R. mm/hr.	Hgb %	Ery mill.	Leucocytes 1000	Platelets 1000	Formolgel	Total prot. %	Alb. %	Glob. %	F-gen %	Albuminuria
Case I.											
1935											
5/10	135	80	3.5	5							
20/11	132	85	3.2	8							
1936											
30/1	117										
1941											
10/6	145										
1942											
23/2	130	55	2.9	9		1'					
9/4	145	50	2.9	6			8.4	2.3	6.1		(+)
19/10	150	40	2.0	7		10"					
18/11	150	45	2.5	8	115	15"	10.8	2.5	8.2	0.09	(+)
1943											
28/1	165	35	1.4	7	120	15"	10.2	2.2	8.0	0.25	
23/3	176	25	1.1	14	73		10.0	2.3	7.7	0.39	+
25/5	157	30	1.6	7		50"	6.6	2.0	4.6	0.67	—
Case II.											
1942											
30/4	150	45	2.1	10	85	14"					—
5/5	140	40	2.0			10"	12.4	2.2	10.2		—
9/6	140	40	1.8		150	15"					
16/7	153	40	2.1			7"	11.4	2.1	9.4	0.14	—
13/8	152	43	2.2		85	14"					—
19/10	156	40	2.1	8	82	11"					
11/12	160	40	2.0	7	91		12.8	1.9	10.8	0.07	—
1943											
9/2	160	30	1.4	4	150	9"					—
9/4		35	2.1	9.5	120		11.5	2.0	9.5	0.2	—
25/5	160	26	1.9	0		10"	12.4	2.2	10.3		—
Case III.											
1941											
16/4	140	56	3.1	9	440						—
23/4	120	56	3.5	11		15"					—
3/6	140	56	3.5			7'	10.6	3.3	7.3		
25/8	140	51	2.7	10							
19/12		43	2.7	13							+
1942											
5/2		43	2.2	13							+
1/4	156	38	2.1	10		2'					
6/6	151	47	2.7	10	457	10'					
1943											
19/3	160	25	1.3	10	353	24'	7.7	1.8	5.9	0.6	+
25/5		22	1.7	7	230	20'	6.4	0.75	5.7	0.7	++

loma. Bleeding time 3.5' Coagulation time (Howell-Gram) 7' Albuminuria 1—2.5 ‰. Non protein N. 30 mg %. Kongo red test: after one hour very slight blue colour of plasma after addition of HCl. No coloration in the urine with HCl.

Readmitted on May 27th. Very feeble. Bad appetite. Edema of the legs, abdominal wall and hands. No bleedings. Heart normal, blood pressure 80/35. Liver enlarged. Albuminuria 2.5—4 ‰, but no Bence Jones protein. Capillary resistance: no bleedings after 50 min. pressure for 15 min. or 100 mm for 15 min. X-ray of vertebral column, pelvis and cranium no signs of myeloma.

The general status was very poor. The patient had no appetite at all. His edema increased considerably (very severe hypo-albuminemia!) in spite of blood transfusions. After a period of diarrhoea the patient died on June 18th.

Differential counts from blood and sternal punctures from Cases I—III.

Case I.

Blood.

	Neutroph.		Eo	Baso	Lymphoc.	Monoc.	Normobl.	Myeloc.	Plasmacells
21/4	42	50.6	6.5	—	37	6			
19/10	»	55	1	—	36	8	2/200		
17/11	»	51.5	0.5	—	45.5	2.5	3/200		
27/3	43	56	2	—	30	8.5		2 %	1.5 %

Bone marrow.

17/11 42. Myeloblasts 0.6 % Neutrophilic myelocytes 2.0 Neutroph. leucoc. 23.2 Monocytes 2.6 »Lymphocytes» 70.2 Plasma cells 0.4 Normoblasts 1.0.

27/3 43. N. Myelocytes 2.0 % Eosinophilic m. c. 0.2. Neutroph. leucoc. 28. Monocytes 2.0. »Lymphocytes» 64. Normoblasts 3.2. Plasmacells 0.4. Rare erythrocytes show polychromasia and basophilic stippling. Slight anisocytosis. Very few typical plasma cells. No cells suspect for myelomatosis.

Case II.

Blood.

	Neutro.		Eo.	Baso.	Lympho.	Mono.	Normoblasts.
10/6	42	40	2	—	54	8	
23/7		44	1	—	50	3.5	2/200
19/10		60	0.5	0.5	35	4	1/200
15/2	43	49	—	0.5	44.5	4	

Bone Marrow.

10/6 42. Neutrophilic myelocyte 0.6. Neutrophilic leucocytes 1.8 Normoblasts 0.4 »Lymphocytes» 93.2. Plasma cells 4 %.

The lymphocytoid cells were mostly spindle shaped with protoplasmatic bodies often separated from the mother cells, sometimes vacuolized.

Case III.

Blood.

Neutro.	Eo	Baso.	Lympho.	Mono.	Normoblasts.
3/6 41 54	1	0.5	34	10.5	
19/12 41 39	1.5	0.5	31.5	8	
1/2 42 40	2.5	—	51	6.5	
6/6 42 58	0.5	—	35	6.5	1/200
20/3 43 57	1.5	—	30.5	9	2/200

Bone marrow.

16/4 41. Myeloblasts 0.6. Promyelocytes 0.8 Neutrophilic m. c. 13.8 Eo. m. c. 0.4. Neutrophilic meta m. c. 0.6 N.phil. leucoc. 33.8. Lymphocytes 14.4 Monocytes 2.0. Plasmacells 3.4. Normoblasts 20.6.

1/2 42. Very cellular marrow. Plasma cells in groups very rare. Spindle-shaped »lymphocytes» are to be seen.

M. bl. 0.2 %. N. ph. m. c. 16.2. Eo. m. c. 0.6. N.meta m. c. 20.2. N. l. c. 29.0. Eo l. c. 0.6. Monoc. 0.6. Lymphoc. 13.6. Plasma cells 1.6. Normoblasts 16.8.

20/3 43 M. bl. 0.2 % N. m. c. 2.4. Eo m. c. 0.2. N. Metam. 3.0. Neutrophils 17.0 Eo 1. 0.6. Baso 1. 0.2. Monocytes 1.4. »Lymphocytes» 69.4. Plasma cells 1.6. Normoblasts 3.4.

Very frequent darkly basophilic protoplasmatic bodies, partly vascuolized, partly resembling polychromatic erythrocytes. It is sometimes clearly to be seen, that they originate from the lymphocytoid cells. Such bodies are not to be found in the blood. Very numerous naked nuclei.

Discussion of the diagnosis.

The *clinical picture* in the first two cases is very similar and this seems to indicate that they suffer from the same malady. An elderly man comes to the doctor because of symptoms, that are probably caused by an *anemia* with a tendency to bleedings. At the ordinary clinical examination we find a generalized slight enlargement of the lymph glands with considerable anemia. This is normochromic or slightly hyperchromic, without signs of hemolysis (bilirubinemia or urobilinuria) or increased regeneration (reticulocytosis). A certain leucocytosis was found in Case II. His blood also showed mononuclear cells of a type that is difficult to determine. The *bone marrow* has the same appearance in both cases with *chiefly lymphocytoid* cells. On repeated examinations plasma cells were only found in

normal percentages or at the upper limit of the normal. The white blood cells show nothing, that indicates a leukemia. The patients have been followed for a long time (one for more than seven years, the other for two years without any rapid changes in the status). Among the clinical features the *excessive sedimentation of the erythrocytes* is one of the most striking. A detailed analyses shows, that it is caused by a very marked *increase* in the *globulin* content of the *serum. The content of fibrinogen* however *is* pathologically *low.* These facts make it possible to characterize the cases and place them in a special group. Is it possible to correlate them with other wellknown maladies?

A diagnosis that has always been the cause of much discussion is chronic aleukemic lymphadenosis. As far as this condition later develops into a manifest lymphatic leukemia the diagnosis aleukemic leukemia seems to be legitimate. Sternal puncture may leed us to suspect this malady in cases without visible enlargement of the glands or typical blood picture. Such cases with a primary proliferation of lymphoid cells in the bone marrow later followed by a typical leukemia are of very great importance. They are very rare (cf the work of Hynes 1940 where no case developed a real leukemia). Very often there is really an aplastic anemia present which later develops into a final acute leukemia.

This diagnosis: panhemocytophtisis with or without myeloblastic proliferation (so-called myeloblastic leukemia) might also be discussed because of the atypical bone marrow picture.

The longdrawn clinical course however speaks against it, nor was it possible to find any typical pathological myeloblasts among the small mononuclear cells found at sternal punctures.

Is marked increase in serum globulins a sign, that has been described in leukemia? Snapper publishes a rather extensive study concerning this problem and he points out, that hyperproteinemia speaks against the diagnosis uncomplicated leukemia. In rare instances S. found border — line values. Out of 15 cases one showed a globulin value of 3.3 % (with 1.0 % euglobulin). It was an instance of myelosis chronica. In a case of chronic lymphatic leukemia the value was 3.0 %. Low values for the fibrinogen have not been published as far as I know.

Isolated cases with marked hyperglobulinemia in the literature are reported to have shown signs of leukemia at the postmortem.

Gross has published a patient, whose malady was interpreted as aleukemic myelosis with a protein content of 14.8 %. Keilhack in a similar case but without post mortem found 8.6 % globulin of which 75.6 % was euglobulin. The patient had 9000 leucocytes in the blood; of these some were unripe. Thrombocytopenia. No X-ray examination of the skeleton. Bürkel places his case, that is often quoted, among the atypical myelomata and it is evident that also the anatomical diagnosis may be very difficult and uncertain. Some of the above-mentioned patients may possibly have suffered from the same malady as my two patients.

Snapper discusses the possibility of a simple combination of the two independent maladies: myeloma and leukemia in a patient he was able to observe. In my cases the long-drawn course (up to 7 years without the appearance of signs of real leukemia), the histological picture of the bone marrow, the severe fibrinogenopenia, the remarkable increase in serum globulins together with the absence of the common clinical signs of leukemia speak against this latter diagnosis.

In none of my patients was there any pains indicating a skeletal affection but in some cases recently published, who showed a widespread myelomatosis at the section, there were never any pains (Cases 1 and XII, Waldenström, 1942). X-ray pictures have been taken of the skeleton at different stages of the malady but no signs of myeloma have been found. As a matter of fact this does not prove that there is not present the form of the malady, that we call general myelomatosis. The result of the sternal punctures seems more important and the bone marrow has therefore been examined on several occasions. Both in Case 1 and Case II there was found a picture that is definitely pathological. According to the experiences put down in literature and also with regard to my own of 10 cases with myeloma, the picture does not in any way resemble what we find in this malady. There is not any decided increase in plasma = myeloma cells; the pathological cells are all very small with no characteristic histological details.

The third patient differs from the others in certain respects. His platelet counts for instance are constantly elevated not decreased as with the other patients, the formol-gel reaction is not so rapid, even if the value for the globulin is very high, he does not suffer from any bleedings and shows no fibrinogenopenia, he has a

more marked leucocytosis and lately he has developed a massive albuminuria indicating a »nephrosis». At the present stage it is not easy to judge if these differences are more important than the resemblances. At the last sternal puncture plasma cells in small clusters have been observed. This may possibly speak in favour of the diagnosis multiple myeloma (cf. Waldenström, 1942).

The question is near at hand if there are no exactly similar cases published in the literature. I have tried to go through all papers, that may possibly have some bearing upon this question and I have found a few instances with a very similar clinical picture. One was published by Bing and Plum (later also by Gormsen 1942). This patient was also an elderly man (65 years). For 3 years troubles with nosebleedings and melaena. After 2 years also impaired vision. He was examined in a hospital where the diagnosis retinitis haemorrhagica was made. Hyperchromic anemia (45 % 2.1 mill.) 6900 leucocytes. The diagnosis lymphatic leucemia was made and the patient has since been regarded as an instance of this malady. He had severe hyperglobulinemia (8—8.9 %) strong formol-gel reaction (2') and a S. R. of 160/hr. A later differential count showed 50 % lymphocytes; 43 % neutrophiles and 7 % monocytes with a total count of 5000. Serum calcium 11.4 mg%. Bleeding time 10 min. A year after the first status the patient showed no enlargement of the lymph glands. Liver and spleen normal. X-ray of the lungs showed stasis? Bence Jones in the urine positive. Free HCl in the gastric juice. Blood count as before. On X-ray no signs of myeloma. Sternal puncture: 58 % lymphocytes but only 1 % plasma cells. The possibility of a myeloma was therefore discarded. It was impossible to follow the patient any further. The diagnosis lymphatic leucemia as a matter of fact seems very uncertain and it is at present impossible to ascertain if the malady was rather an incipient myeloma or the same as in my patients. The resemblances in the clinical picture are marked but there were no determinations of the fibrinogen (of the bleedings) or any more detailed investigation of the serum proteins.

Among Bing's cases there is another history (1940 Case 69) of a man 75 years old. He showed extensive moderate enlargement of the lymphatic glands. The leucocytes terminally rose to 55000 with about 50 % atypical lymphoblasts. Otherwise neutrophilic leucocytes. Formol-gel 2,5. Globulins 6.9 % High fever. The post

mortem gave no certain diagnosis but showed generalized enlargement of the lymph glands. A para-aortal gland was examined microscopically. It showed complete loss of structure with a generalized proliferation of rather large mononuclear cells somewhat resembling plasma cells. In the red bone marrow there were found numerous large and small non-granulated mononuclear cells not resembling lymphocytes. Some plasma cells. The case was interpreted as a septicemia with a leucemoid reaction, also the diagnosis leucemia was discussed. The resemblance with my patients seems obvious. The label reticulosis may also be used but it hardly furthers the understanding of the process.

In 1938 Rohr discusses the problem hyperglobulinemia and reticulum cells.

von Marsovszky (1940) has published a case of a similar type. A man of 50 showed severe bleedings from the nose and gums in 1939. He suffered from some anemia (58 % 3.2 mill.) Platelets 150000 S. R. 135 mm. The erythrocytes could not be counted in Hayems solution but only in normal saline. He had large retinal hemorrhages. Skeleton: X-ray normal, sternal puncture nothing pathological. Bence Jones neg. Total protein in the serum 12.2—16 %. Albumin 1.4—0.7! Fibrinogen 0.6—0.7 %.

The post mortem showed nothing definitely pathological. v. M. published the observation under the title: Hyperproteinemia causa incerta. It is obvious, that the clinical picture in all essential respects is identical with my cases, only the value for fibrinogen was increased. It is to be noted however, that the fibrinogenopenia was no constant finding in my cases either.

In 1940 Malmros published three cases of hyperglobulinemia, where it was impossible to find any cause of this symptom. We have been able to observe a number of similar patients in the Medical Clinic of Upsala these last years. Some of them are now published in Nordisk Medicin (1943). They are decidedly not instances of the same pathological process as was present in Case I and II.

Fanconi in 1941 publishes an instance of »dysproteinemia» in a luetic infant. The blood was highly viscous, did not coagulate but there was coagulation of the plasma at 56°.

Some clinical and chemical observations.

One of the most striking properties of the sera from Cases I and II was their very *high viscosity* already at room temperature. I have therefore tried to investigate this property more closely especially as the viscosity of the serum in hyperglobulinemia has been very little studied. From later years there are only a few determinations in cases of myeloma by Albers and Magnus Levy but no attempts at a systematic analysis have been made.

For the measurements I have used a slightly modified Ostwald's viscosimeter, an instrument whose simplicity makes it well suited also for clinical work. The only real draw-back with the original instrument is the difficulty to tell beforehand, what will be the correct time to read the instrument and this leads to a series of unnecessary observations in order not to miss the right moment when the meniscus passes the lower limit. I have therefore constructed a modification of the original instrument, where it is possible first to determine 1/10 of the total time. This is usually done quickly. A clock is then started that gives a signal when the instrument ought to be observed again. With the aid of this modified instrument it seems, as if viscosimetry would be a simple and useful clinical method.

All determinations are performed in a waterbath and the temperature is corrected with automatic thermoregulator, when the difference against room temperature is great. The fall-time for a number of sera with increased globulin content was determined at different temperatures and with different instruments. The determinations with viscosimeters having varying dimensions of the capillaries gave no real differences even in sera with very high viscosity and the results will not be discussed here.

Determinations at different temperatures were made in order to find out, if the very marked temperature dependance of viscosity found in the serum from Case I might be observed also in the sera from other patients. Measurements were therefore made at every 4th grade from 9°—39° C on a number of sera (see fig. 1). In others determinations were only made at a few definite temperatures (usually 13° and 37°). The temperature 17° has also been used in a number of instances, as it corresponds well with ordinary room temperature. Under such circumstances there are certain impor-

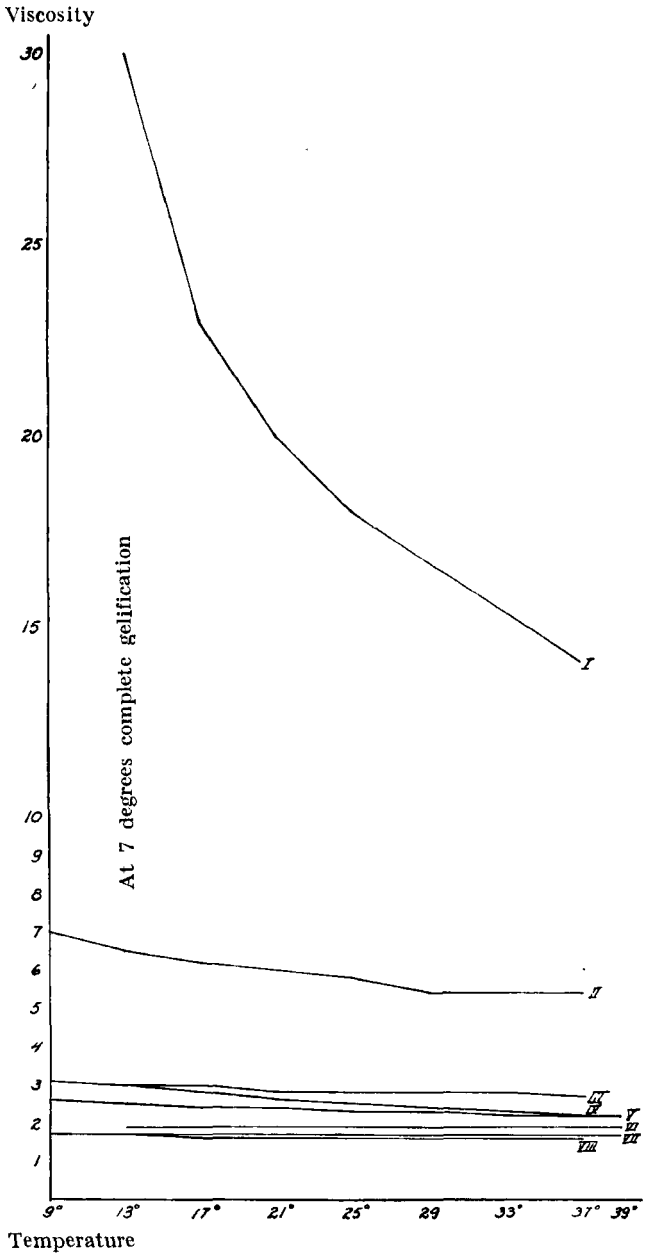

Fig. 1. The curves show the very great difference in viscosity (η) for different sera. I represents the serum from case II. It is seen that the viscosity is very high at all temperatures but also that the influence of the temperature in this case is much more marked than in any other. Curve II shows the very gradual increase in the viscosity of the serum from case I with falling temperature. At 7° complete gelification however. Curve III shows the serum from a case of myeloma with high globulin content but no very marked viscosity or increase with falling temperature. Curve IV shows that the serum from case I has still a very marked increasing viscosity with falling temperature even after dilution 1: 1 with saline Curves V and VI show the very slight increasing viscosity with falling temperature in two other instances of hyperglobulinemia. A normal serum VII has absolutely the same form as a curve for water. Curve VIII gives the subnormal values for a serum dilution 1: 3 from case II but with still marked increase at low temperature.

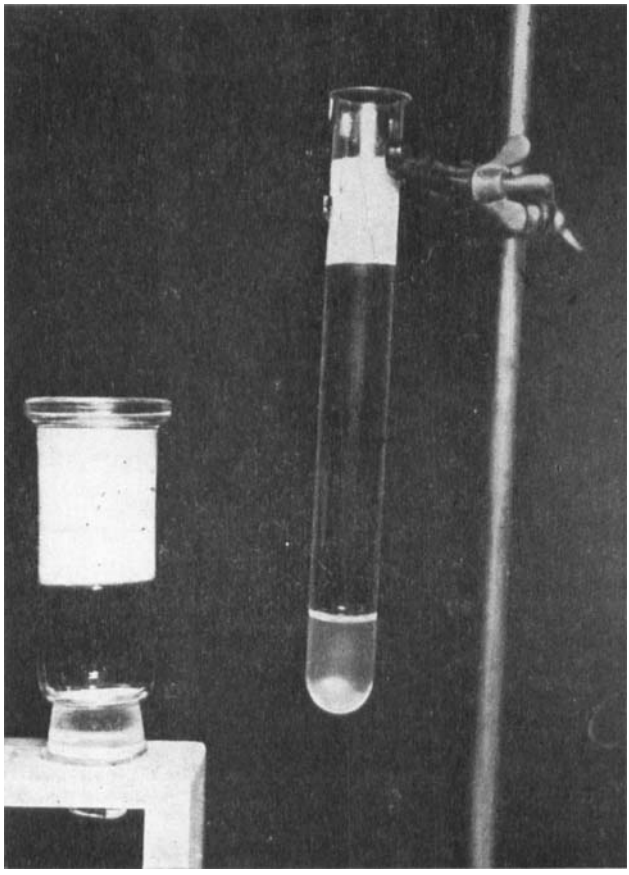

Fig. 2. The stoppered flask that is put upside down contains serum from case I that has been kept in a refrigerator at + 5° C. The serum is gelified and opaque. In the test tube a sample of the same serum kept at room temperature. It is quite limpid (but viscous) and translucent.

tant facts to be observed. The term relative viscosity (η) is used to denote the relation of the fall-time to that of water or normal saline; the very slight influence of specific grav. is disregarded For many sera it is quite constant in the temperature range 9°—37°. They behave in other words just like water and there is only a slight increase in viscosity with sinking temperature. Some of the sera with high globulin content behave in quite another manner. In one patient with a serum globulin value of 5.7 % the

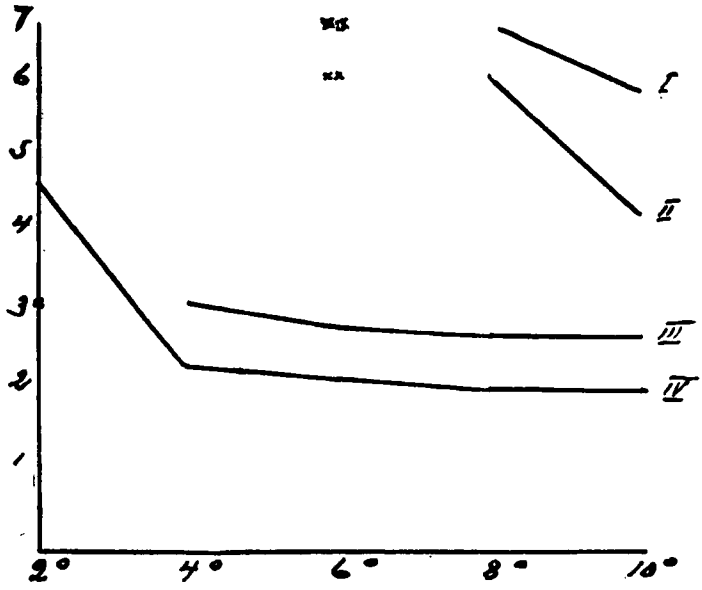

Fig. 3. *The influence of dilution on gelification of the serum from case II at low temperature.*

I Undiluted serum + + Gelification at 6°
II 2.5 ml serum + 0.5 ml saline } + + » » »
III 2 ml serum + 1 ml saline + Gel at 2°
IV 1.5 » 1.5 » + » » »
In dilution III there is no gelification above 2°. In IV there is no gelification whatever.

η at 13° was 2.7, at 17° = 2.6, at 37° = 2.4. In cases I and II the differences were still larger.

A patient, who suffers from myeloma and shows a serum globulin of 10.2 % showed a relative viscosity at 17° of 3.7. In another patient with 8.7 % globulin it was as high as 6.2 but in a case with 10.8 % globulin it was only 2.3. Also a dilution of the most viscous sera to half the initial globulin value gives a much higher viscosity than was found in other sera with the same original globulin content.

These relatively large differences are not merely a result of a very high globulin content as another patient with a still higher globulin value (5.9 %) and the same viscosity at 13° (2.7) only showed 2.6 at 37°. The relation of viscosity to temperature is

thus different in different patients and is probably to be regarded as a result of certain qualitative differences in the globulin and not as a function of the globulin content.

An extremely high viscosity in relation to the globulin content seems to be characteristic for patients with what I call essential hyperglobulinemia but it has also been observed in two instances with myeloma and in one case of polyarthritis with sepsis. In order to get an expression for the temperature dependance of the viscosity I have calculated the relation between relative viscosity at 13° and at 37° and multiplied the value by 100. This index seems to give a rather good expression of the temperature variability of viscosity. In normal sera it lies at 100 or very near this value. The highest value I have found is 211 i. e. the viscosity is more than doubled from 37°—13°.

The variability of the *fibrinogen viscosity* with temperature was also determined in 12 plasmata from different patients with a high sedimentation rate. It is well known, that this protein has a high viscosity, and it is rather surprising, that the easily determined specific viscosity has not been used for qualitative or quantitative studies. The investigations of later years above all with the ultracentrifuge have shown, that fibrinogen after precipitation and resolution is easily denatured. I have therefore made some experiments with plasma and serum from the same patient. A man with a practically normal serum viscosity, who suffered from chronic polyarthritis, had a very high plasma viscosity and a short plasma formol-gel time. Formol-gel neg. in the serum. The viscosity of the plasma showed a very marked dependance on the temperature with a value of 120 comparable to the serum globulins in case I.

In 11 cases the plasma viscosity was practically the same at 13° and at 37°, when the serum viscosity was normal, and the serum-formol-gel was neg. It may therefore be assumed, that fibrinogen usually does not show a very marked influence from falling temperature on its viscosity.

The *serum* from one of the here published patients (Case I) showed a very curious phenomenon. His serum and plasma were at room temperature highly viscous but not to such a degree as the serum from Case II (see fig. 1). If a sample is left *in the ice chest at + 4—5° it develops into a jelly*, that gets white and opaque (see fig. 2). At higher temperatures it is possible to »thaw» it up again.

The sample then »melts» peripherically leaving a solid centre. None of the many sera with hyperglobulinemia that I have been able to examine showed anything similar and I have not found the phenomenon mentioned in literature. With the aid of the viscosimeter it has been possible to examine the problem more in detail. It was found, that the sample had not become solid at 9° but at 7° it did not pass through the capillary. The serum was left in the apparatus over night at a temperature of 17° but this was not enough to lower the viscosity to such an extent, that more than a few drops passed the capillary. When warmed to 37° the serum again became liquid.

The influence of dilution was tested in the following way. (see Fig. 3). The nondiluted serum gelified at 6°. Also slightly diluted it behaved in the same way. In a dilution 1 part saline + 2 parts serum there was a measurable viscosity at 6° and 4° but at 2° there was complete gelification. Dilution 1.5 + 1.5 gave a high viscosity but no jelly at 2°. An increase in the number of water molecules present lowers the temperature for gelification. It is thus obvious, that the temperature limit is not dependent on any sudden change in the composition of the protein at a definite temperature but rather on its concentration. According to my opinion gelification is seen, when all water molecules of the solution are bound to the protein. I. e. there is no more free solvent present.

As it might possibly be suspected, that the high viscosity were caused by some mucin substance, even if this was not very probable, the serum was treated with a highly concentrated mucinase, that I obtained through the courtesy of Doctors L. Hahn and B. Skanse. The viscosity was not altered by the influence of this enzyme.

What is the real *cause of a high viscosity?* If it is allowed to make a generalization it would perhaps be suitable to say that it is caused by a thronging of the molecules in a fluid either because they are very long-drawn, thread-like or because there is such a high concentration of the dissolved molecules or such a tendency for them to bind water molecules that much of the water is fixed. Threadlike molecules and molecules with a strong tendency to hydratization are biologically the most important causes of a high viscosity. There are e. g. sera containing large amounts of globulins with a very high hydratization. If the concentration of these

molecules is high enough there develops a gel — all the water molecules are bound and there is no longer a free solution (cf. Case I).

In the two cases published here (I and II) there was a very high content of euglobulin in the serum that may be precipitated by a very simple procedure (dilution with H_2O or dialysis against water). The precipitate that is formed in this manner does not resemble an ordinary dry protein. It is a slimy translucent jelly, insoluble in water but easily soluble in for instance normal saline. It is obvious that the high viscosity is caused by this substance which in itself gives highly viscous solutions. The serum after precipitation of the euglobulin with dialysis is no longer very thick.

It is seen from Table I that there is no real parallellism between the values obtained from chemical or electrophoretical analysis of the globulin and e. g. viscosity or formol-gel time. Also the discrepancy between the formol-gel time and the viscosity is obvious. The five sera that showed the highest viscosity had formol-gel values varying between 24', 3'7", 50", 5'55" and 10". Among the sera with the shortest formol-gel time the viscosity was 3.7, 2.3, 6.2 and 20. What may be the cause of this?

Partly it may be explained by assuming a different chemical structure in the different globulins. It is obvious that the chemical structure of the very hydrophilic euglobulins in case I and II ought to differ greatly from other globulins. On the other hand the viscosity in Case D.-B. is also very high and lies between the values for Cases I—III even if he has no euglobulin increase and shows a rather long formol-gel time. It is probable that this last mentioned fact is caused by a low concentration of the dialyzable factor necessary for gelification with formaline. (Waldenström 1943). It is also possible, that the surprizingly low time in Case W. depends upon a milieu that is optimal for the reaction. When we know about the importance of low-molecular factors for the rapid formation of a gel such atypical instances, and also Jersild's patient with severe hyperglobulinemia and neg. formol-gel are less difficult to understand.

Another question seems to be of importance. Is it possible with simple means and without ultracentrifugation to get an idea about the molecular weight of a special pathological protein? This is obviously not yet the case. I have tried to find out if it were possible

Table 2.

The relation of relative viscosity and temperature for some sera with high globulin content.

Name Diagn.[1]	K. J. J. 1:3 Ess.	Normal	A. 1: 3 Ess.	S. E. Ess.	A. P. Ess.	J. W. Prem.	E. K. Cirrh.	G. H. Sarcoid	L. E. Lupus eryt.	T. A. Endocard.	I. B. M.	K. J. J. 1: 1 Ess.	A. A. Case III Ess?	B. A M.	O. Chron arthr.	A. 1: 1 Ess.	K. J. J. Case I Ess.	D. B. M.	A. Case II Ess.
Glob. %				4.5	4.3	6.1	5.9	5.3	5.9	5.7	7.5	4.3	5.9	10.2	7.3	5.4	8.7	7.8	10.8
Curve		VII	VIII	VI		V					III	IV					II		I
9°		1.7				2.6				2.8	3.1			4.1			7.0[2]		
13°	1.6	1.7	1.7	1.9		2.5	2.6		2.7	2.7	3.0	3.0	3.0	3.8	4.5	3.1	6.5		30.0
17°		1.7	1.6	1.9	2.1	2.4		2.4		2.6	3.0	2.8	3.4	3.7			6.2	6.7	23.0
21°	1.6	1.7	1.6	1.9		2.4				2.6	2.8	2.6		3.5			6.0		20.0
25°		1.7	1.6	1.9		2.3				2.5	2.8	2.5		3.4			5.8		18.0
29°	1.6	1.7	1.6	1.9		2.3				2.5	2.8	2.4		3.4			5.4		16.7
33°		1.7	1.6	1.9		2.2				2.4	2.8	2.3		3.3			5.4		15.4
37°	1.6	1.7	1.6	1.9		2.2	2.4		2.6	2.4	2.7	2.2	2.5	3.2	3.0	2.5	5.4		14.1
39°		1.7		1.9		2.2													
$\frac{100 \times \eta_{13}}{\eta_{37}}$			106			113	108		104	112	110	136	120	118	150	124	120		211

[1] Ess. = Essential hyperglobulinemia
Prem. = »Premyeloma»
M. = Myeloma
[2] = gelification at 7°

from determinations of the relative viscosity of the sera from Cases I and II to find out the shape of the molecules. This was not possible. The globulin component with a very high molecular weight that is present in these cases certainly gives the serum its high viscosity but in another highly viscous serum from a patient with myeloma (D.-B.) there was no component with such big molecules and on the other hand the serum of Case III. contained a large molecule but showed no very high viscosity.

At present it is necessary to collect more data about cases with abnormal serum globulins before we are allowed to state anything definite about the relative importance of different methods for the investigation of these substances.

Knowing the importance of the serum proteins for the *water transport* in the blood we may assume that a water tolerance test on these patients should give an abnormal result. In case III there was present a severe hypalbuminemia and consequent edema. No water tolerance test was therefore performed. In case II the result of the test was not extremely abnormal (2 hours 545 ml, 4 hours 790, 24 hours 1800, spec. grav. 1.009—1.020). In case I however there was a severe disturbance. After 1500 ml an excretion of only 310 ml in 2 hours, 515 in 4 hours, 1400 in 24 hours, spec. grav. varied 1.017—1.020. No hypertension, no casts but slight albuminuria.

It seems very striking that an increase of the specific viscosity of the serum to more that 10 times the normal does not cause any serious disturbance of the circulation and edema does not appear, when the albumin value is not very low (as in Case III). Determinations of the colloidal osmotic pressure have not been performed.

As the previous experiments have shown, that viscosity and total protein or globulin content of the serum do not run parallell it would seem possible, that some special globulin fraction caused the high viscosity. The globulin has of old been divided into two fractions: pseudoglobulin and *euglobulin*. They have different solubility in high electrolyte concentrations and the euglobulin is most easily salted out. According to Howe euglobulins are those proteins, that are precipitated by a concentration of 13.5 % Na_2SO_4 at 37°. The next fraction, precipitated at 17.4 % he called pseudoglobulin I and the last, needing 21.5 % Na_2SO_4 for complete precipitation, pseudoglobulin II. What then remains in solution is

regarded as albumin. It seems clear, that such a division, built upon the molar concentration of the electrolyte (the mentioned concentrations correspond to 1.0, 1.25 and 1.5 molar solutions) must give absolutely artificial limits between the »protein fractions». On the other hand the method seems to give reproducible results, when tested on large clinical materials representing different maladies.

I have used another and more specific of the old definitions for euglobulin i. e. the protein fraction, that is precipitated on dilution of the serum with water (or after dialysis against water). An old test for hyperglobulinemia has been based on this fact. A drop of blood is allowed to fall into water and it is stated if a gray haze develops. This test does not give any valuable information and I have tried another technique, that may give a chance of stating also qualitative differences in the euglobulin.

A simple method that may be used in any laboratory as an orientation for detecting pathological euglobulin is the following. One ml serum is mixed with 16 ml aq. dest. In the presence of euglobulin there forms a more or less massive precipitate, which is centrifuged down and washed with distilled water. The amount may be judged from the size of the precipitate, which is a rather approximate method, or by weighing. Some sera give a precipitate of the ordinary floccular type usually seen in proteins. Others show a practically limpid, highly viscous sediment that is obviously very hydrophilic. This sediment redissolves quantitatively in the presence of electrolytes. It is also possible to make a fractionated precipitation, as the euglobulins in some sera need higher electrolyte concentrations to keep in solution than others do. In Case I the euglobulin began to fall out already after the mixture of one ml serum with 6 ml water and the precipitation was complete in 1: 10, with the formation of a very voluminous sediment and no further precipitation on dilution. In Case II there was beginning opacity also in 1: 6 but the precipitation was not complete until 1: 16. It is possible, that this means a real difference in the composition of the euglobulin and is not only a result of concentration. In some other cases of hyperglobulinemia there was found a slight precipitation in the dilution 1: 2—3. Only in one case was there no decided precipitation before the dilution 1: 9—10. The vast majority of cases with hyperglobulinemia did not show any precipitate

at all. Also in sera from a varying material of 80 cases with mixed diagnoses was there sometimes found traces of euglobulin but this was also present in the blood from quite healthy individuals and a slight opalescence with the formation of some precipitate is certainly not pathological.

The more chemical common methods for the determination of serum protein after *fractional precipitation* with Na_2SO_4 according to Howe and Kjeldalization of the precipitate are much to complicated to become really popular as routine methods in a hospital laboratory. For theoretical questions however they will probably be indispensable also in future. From many points of view the electrophoretical method, as it has been worked out by Tiselius, and the ultracentrifugation according to Svedberg are those methods that make the least damage to the protein molecule. In the three cases published here it has been possible to make such investigations on the sera. The results that seem to be of great interest will be published later on in greater detail by Doctor K. O. Pedersen.

Beside the methods, that have already been mentioned for the qualitative and quantitative examination of proteins there are a few phenomena that may lead to a diagnosis if they are correctly interpreted. The curious fact has been known for some time, that it may be impossible to count the erythrocytes as there appears a fine precipitate of globulins certainly caused by the mercury in Hayems solution. This has been wrongly mixed up with the autoagglutination of the erythrocytes, that may sometimes be seen at temperatures below 37° (see Waldenström, 1942). This is quite another process caused by the action of an agglutinin at lower temperatures. It is not affected by the use of normal saline but only by warming.

Gros and Brockmann have used the flocculation of certain globulins in Hayems solution as a test for myeloma. I have tried the reaction in a number of cases with hyperglobulinemia but only once (in a patient with myeloma) found it positive. Its value therefore seems to be limited but a positive reaction probably indicates the increase of some special globulin fraction and may perhaps be used in future, when more is known about the chemical side of the question.

In the literature (cf for instance the excellent work by von Bonsdorff 1937) there are discussed a few other abnormitities in sera

from patients with myeloma. The deficient retraction of the coagulum may be very striking but as it is very difficult to interpret also in other conditions it may be hard to judge its real importance for the diagnosis. Another curious anomaly, that was first described by Wintrobe and Buell in 1933 and later by von Bonsdorff in 1937 and by Bing in 1940 is the division of the serum into two different strata, after standing in the ice chest. The lower is highly viscous and contains much euglobulin which may even be crystallized. The upper is more liquid and has not such a high globulin content. Also the coagulation of serum at 56° (on inactivation of the complement) has been discussed e. g. by Cantarow. Auto-inhibition for the Wassermann reaction (this anticomplementary effect has been studied in Sweden especially by Holmberg and Grönvall and by Olhagen) is another such phenomena.

Case II shows a clinical symptom that ought not at first to be regarded as a consequence of the hyperglobulinemia namely a *thrombosis v. centralis retinae.* The patient published by Wintrobe and Buell as well as one of Bing and Plums cases suffered from the same or a similar disease of the eyes and W. and B. regarded it as caused by the enormous agglomeration of the erythrocytes intravascularly. It is not easy to tell if the interpretation given by the American authors is correct but is does not seem impossible. Fåhræus has treated this question of capillary emboli from agglomerated erythrocytes already in 1921. The tendency to bleedings may also be a cause of intraocular changes (Bing and Plums case) as in case I, where extensive perivascular bleedings were found.

Beside the enormous increase in sedimentation rate the *anemia* is at first sight the most striking clinical symptom. In case III it was slightly hypochromic and the value for serum iron was somewhat lowered. The anemia was decidedly not caused by iron deficiency as a consequent iron therapy had no effect on the blood values. It is to be regretted, that the determination of serum iron is very uncertain in these cases as the protein precipitate is so massive, that it is difficult to obtain enough filtrate for a determination. The filtrate is also slightly opalescent. These complications may be suspected to work both increasing and decreasing the serum iron value and it is therefore best to state, that all determinations of the serum iron are uncertain in presence of considerable hyperglobulinemia.

The anemia is most decidedly not pernicious: hydrochloric acid in normal amounts was present in the gastric juice and the bone marrow did not show any megaloblasts in spite of severe anemia. In cases of myeloma a normochromic or slightly hyperchromic anemia is regarded as typical. Its explanation in this malady has simply been the crowding out of the normal marrow by the foreign myeloma cells. But also other explanations are possible. An essay to analyze the factors that might cooperate at the regulation of the normal erythrocyte level is not simple. A priori it seems probable, that the viscosity should play a part. When this is very much increased by the high globulin content it is possible that a decreased erythrocyte production may be an attempt at compensation. This is a mere conjecture but its validity should perhaps be tested in a suitable case.

The two patients case I and II have both shown a tendency to sickering *bleedings* from the gums and the nasal mucosa. These disturbances were even the reason, why the patients have come to hospital on their last visits. A determination of the coagulation time has shown, that it is decidedly increased. The quantitative determination of fibrinogen shows a considerable decrease and this ought to explain the retarded coagulation at least to a great part. This low fibrinogen content of the blood is probably the explanation of an observation that dr. Melander has made on the blood of this patient namely that a coagulum may dissolve spontaneously. I have myself once seen this in a patient with myeloma, who also showed protein precipitation when the blood was diluted with Hayems solution. The content of fibrinogen was not determined in this case. It does not seem probable, that the rather slight decrease of the platelet count should be of any influence on the coagulation if it is not assumed, that their quality is also changed. If it is the low content of fibrinogen or a low »quality» of the fibrinogen present or a deficiency in some other protein component necessary for hemostasis, that causes the haemorrhages in these patients the future will show. As it is it seems evident that large transfusions had a very favourable effect on the bleedings.

A third very striking change in the blood from these patients, when one has once observed it, is *the quality of blood smears made on slides (see fig.* 4). The first time when one of the nurses in the laboratory complained of the bad quality of the smears from case II we

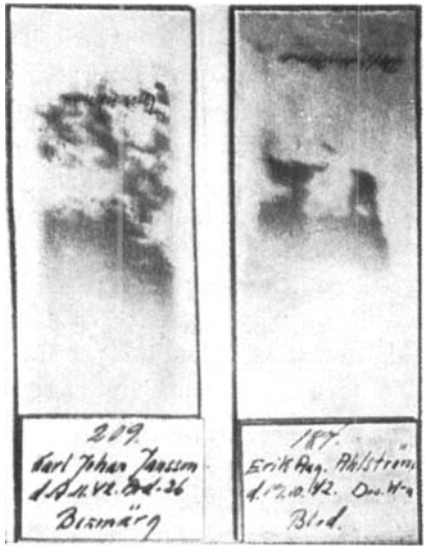

Fig. 4. Two slides with smears from blood (case II) and bone marrow (case I). Stained with May-Grünewald-Giemsa. Also slides from case III show similar changes but less marked, otherwise no such preparations were found among a large collection of sternal puncture specimens.

thought that it might possibly be a mere chance and depend upon a deficient cleaning of the slides. Several controls however showed that there was no technical fault. When it was later found, that also slides from Case I showed exactly the same picture, it was obvious that this change might have some diagnostic importance. Also in Case III older slides showed the same type. The change must be caused by the lowered stability of the protein film, that is spread and then dried and fixed on the slide. It is impossible to tell the cause of this as it has not as yet been possible to reproduce these changes experimentally.

This phenomenon has nothing to do however with a condition, that has been thoroughly studied in this Clinic, namely the autoagglutination of red blood corpuscles on cooling. Also in such cases the blood smears are of poor quality but this is caused by a clumping together of the erythrocytes through autoagglutination at temperatures below 37° C. This clumping and consequent streakiness of the slides is seen at once, not as is the case with the fragmentation of the blood smears from case I and II only after dyeing, fixation and staining.

Pathological protein fractions in the blood.

In 1848 Bence-Jones published the discovery of a new urinary protein. Since that time the study of definitely pathological proteins in different maladies seems to have made rather insignificant progress. The chief interest has been devoted to quantitative determinations of protein fractions and the work of Tiselius, Blix, Shedlovsky, Gutman and others on the electrophoresis of normal and pathological sera has much increased our knowledge in these matters. Of late the interest in the protein pattern of the serum has been very intense also in Scandinavia. This is probably partly explained by the important work on the physical chemistry of proteins, that has been done in the Institute of physical Chemistry in Upsala. (Prof. The Svedberg and Prof. Arne Tiselius).

In 1937 von Bonsdorff, Groth and Packalén published a very interesting case of myeloma in a man 37 years old. The patient had a marked hyperglobulinemia (7.5 %). When the serum was left standing at room temperature nothing happened but in the refrigerator there was seen a stratification with a highly viscous lower layer and an upper stratum with lower viscosity. The content of globulin was 10.7, viz. 5.3 %. In the lower stratum, where the euglobulin but not the pseudoglobulin had increased very much there was after some time found crystals. This crystallizing protein was examined very carefully from many points of view. It was precipitated as a globulin. The molecular weight was determined by K. O. Pedersen in the Institute for Physical Chemistry in Upsala and was found to be 200000 at ultracentrifugation i. e. somewhat higher than is found in the usual serum globulin fractions. Many facts favour the assumption, that this is not a normal component of the serum globulins. Nor is it a Bence Jones' protein, as the molecular weight of the latter is low (35—40000).

Packalén has later (1940) published one more case of myeloma with spontaneously crystallizing globulin.

Bing (1940) has described a case of myeloma with the same curious division of the serum on standing in the ice chest and he also believed that there were formed crystals in the lower layer.

Recently Holmberg and Grönvall have described a protein, that crystallizes readily from the serum under special conditions. The patient showed no signs of myeloma but she suffers from a

chronic arthritis. This protein is also a globulin with normal molecular weight (ultracentrifugation by K. O. Pedersen) and an electrophoretic migration that corresponds to the behaviour of a serum globulin. The γ-fraction was considerably increased but the pathological globulin (1.3 %) did not fit in with any of the previously described electrophoretic fractions. Also after electrophoretic purification did it give a positive Wassermann reaction. It is perhaps somehow related to the anticomplementary substance described by Olhagen, which seems to be increased not only in some instances of myeloma but also in chronic arthritis.

The three cases that have been published in this paper all show pathological proteins in the serum. Doctor K. O. Pedersen, who is at present working on the purification of these substances from the serum of Cases I—III has been kind enough to give a summary of his work. In the serum from case I there is found a fraction that seems homogenous on ultracentrifugation and gives a sed. constant at 20° of 19.2×10^{-13} corresponding to a molecular weight of over 1,000,000. In case II the sedimentation constant was of the same magnitude 20.6×10^{-13}. It is not easy to say, if this very high molecular weight is caused by an aggregation of globulin molecules or if there is really present a giant molecule. The decidedly monodisperse appearance on centrifugation rather speaks in favour of a preformed giant molecule.

Biologically of prime importance is the obscure question: What is the cause of these changes in the structure of the serum proteins?. It must be stressed from the beginning that there is not only found a hyperglobulinemia but also at the same time a very marked lowering of the albumin to values at or below 2 %. In a large material of protein analysis in 102 cases with disturbances of the protein metabolism Gormsen only found 4 patients with a serum albumin value $\leqq 2$ %. None of these were instans of myeloma. The A : G quotient in my 3 cases thus changed not only through the absolute increase in globulin. Lowered albumin values are not uncommon in myeloma but I have never been able to find any explanation of this decrease. In case III there were certainly losses of albumin through the kidney during the last months as he had a picture resembling nephrosis but from the beginning he only showed slight albuminuria and this was absent or very slight in case I and II. Any really serious disturbance of liver function, that is otherwise

regarded as an important cause of hypalbuminemia, would be hard to reconcile with the clinical picture in general even if the high values for citric acid in Case II (but not in Cases I and III) might point to that organ. One explanation would be the assumption that there is some genetical connection between the albumin and globulin fractions of the serum.

I have not been able to find any description in the literature of a fibrinogenopenia associated with hyperglobulinemia as was found in Cases I and II. Probably this was the most important cause of the severe bleedings. The analyses of fibrinogen for instance in cases of myeloma, a malady, where the other globulin fractions have been investigated very closely in later years seem to be very few. Some authors have found an increase (v. Bonsdorff, Magnus Levy). If a decrease of fibrinogen may speak against the diagnosis myeloma, is not easy to tell at present judging from the very sparse material, that has been collected.

It might perhaps be expected, that this very rare but interesting and important symptom, a lowering of the fibrinogen content of the blood (inopenia) should be described also in other related or perhaps identical conditions. As a matter of fact such a finding is regarded as extremely rare in adults. Very low values for fibrinogen have in rare instances been noted as a congenital, hereditary error, experimentally after severe damage to the liver through chloroform or phosphorus and thirdly (by Jürgens and Trautwein) in a case where an extensive growth of cancer cells in the bone marrow was regarded as the cause of the inopenia. Perhaps somewhat rashly this last observation has been taken as a sign, that the bone marrow plays a part in the formation of fibrinogen.

Most characteristic for the first two cases published here is the very large amount of pathological globulin with the solubility of a euglobulin. The electrophoretic protarties in case II corresponded with a β-globulin (in case I electrophoresis was impossible because of the firm jelly, that formed at low temperature) but were not identical with this normal fraction. In case III there was also found a very high globulin content. On eiectrophoresis the fraction mostly resembled a β-globulin. In this case there was also present a component with very high molecular weight.

Discussion.

What may be regarded as the explanation of all these curious data? Why are these persons carriers of hitherto not described globulin fractions with giant molecules? If these are compared with other known proteins with a very high molecular weight we find, that their size most closely resembles some of the human antibodies e. g. against pneumonia (the specific pneumococcal antidies in rabbits are considerably smaller, of the same size as ordinary globulin). It should be remembered, that a marked hyperglobulinemia is found (as an immune reaction?) in some probably infectious diseases of unknown origin e. g. in certain cases of lymphogranuloma benignum and in the virus disease lymphogranuloma venereum besides myelomatosis, that might be regarded as a more or less primary disturbance in protein metabolism. In lymphogranuloma venereum the protein fractions have been thoroughly studied by Gutman, Wise et al. Also in another chronic infectious disease namely Kala-azar a considerable increase in globulin has been observed.

May it be allowed to put forward another hypothesis as a possible explanation. We know from the experiments with plant viruses of different kinds, that the inoculation of an organism with a special virus may change a great part of the protein in the host plant into virus protein. This is e. g. a well established fact as regards tobacco mosaic virus. If a patient is »infected» with some virus protein it may be possible, that his own serum protein is transformed to virus protein instead. The hypothesis may possibly be tested experimentally on lymphogranuloma venereum, where it does not seen wholly improbable, that the pathological serum globulin is virus itself, which then ought to be contagious. It is also possible, that several conditions with a marked hyperglobulinemia are really chronic virus infections. In cases I and II the formation of virus protein might interfere with the synthesis of both albumin and fibrinogen. This seems more probable than the assumption, that the formation of some hypothetical antibody against an unknown infection should interfere with the production of such an indispensable substance as fibrinogen.

The danger of such a hypothetical virus infection would there-

fore not consist in any toxic action but rather in the virus forming a pathological matrix or inductor. The protein synthesis of the body thus runs along a pathological pathway and necessary building material is taken from the normal synthesis of e. g. albumin and fibrinogen.

In order to get a decided answer to all these questions it would be best to treat the problem experimentally e. g. with the infection of apes or monkeys with blood from these patients. It is to be observed however that only positive findings have any real value as proof, when we know how difficult the inoculation even with known virus diseases may be.

Summary.

The possible nature of myelomatosis as a disease of protein metabolism with secondary deposits in the bone marrow is discussed (cf the lipoidoses with high blood cholesterol and deposits in the tissues).

The author then gives a thorough clinical description of two patients probably suffering from a hitherto unknown disease in the bone marrow. A case that may possibly be interpreted as an instance of premyeloma is described (Case III). He shows long-standing hyperglobulinemia and severe anemia. In the bone marrow sternal puncture only revealed some plasma cells clustered together and never any roentgenological signs of myeloma. The course was progressive as regards the anemia. There was found a pathological globulin, but no inopenia and no hemorrhagic diathesis. Even at the post mortem the case remained obscure. There were not found any typical signs of myeloma.

Cases I and II showed signs of severe derangement of protein metabolism with very high globulin and low albumin values. There was also found a tendency to hemorrhages and low fibrinogen values. A severe normochromic anemia was noted. In both patients pathological euglobulins with very high viscosity, in one patient with a tendency to gelification of the serum at temperatures below +7°, were found. Obviously the molecule of this globulin must be a very large one. Studies in the ultracentrifuge and with the aid of electrophoresis by Doctor K. O. Pedersen in the Institute for

Physical Chemistry showed the presence of a globulin fraction with a very large molecule (mol. weight about 1,000000) and a migration that did not correspond to any of the known globulin fractions. It lay next to the β-globulin.

The possible explanation of different anomalies in these patients: gelification of serum below + 7°, character of blood smears (see fig. 4), the high viscosity, the bleedings, the low albumin and the fibrinogenopenia, are discussed and related to each other. The causes of increased globulin content of the blood are treated and the hypothesis is put forward, that a mechanism analogous to the predominant formation of virus protein in a plant infected with e. g. tobacco mosaic virus may be present in these cases. A large amount of the protein in the blood would then be formed after the image of some abnormal »virus» protein, thus explaining the low values for the different types of normal protein (albumin and fibrinogen) and the very high molecular weight, that is of the same magnitude as that for virus proteins.

Literature.

Waldenström: Acta chir. scand. 1942 (Festskrift for H. Waldenström. Suppl.). — Hynes: Quart. J. med. *9*, 1940, 177. — Piney: Fol. hämat. *46*, 1932. — Bing: Acta med. scand. *88*, 1936, 478. — Gross & Brockmann: D. Arch. klin. Med. *177*, 1935, 470. — Snapper: Ziekten van het Skelet. Haarlem. 1937. — Keilhack: Fol. hämat. *55*, 1936, 406. — Bürkel: Zschr. klin. Med. *127*, 1935, 552. — Bing: Acta med. scand. 103, 1940, 547. — Bing & Plum: Acta med. scand., *92*, 1937, 415. — Gormsen: Knoglemarvsundersögelser. Diss. Köpenhamn 1942. — Rohr: Helvetica medica acta. *5*, 1938, 544. — von Marsovszky: Klin. Wschr. 1940, 1190. — Malmros, Nordisk Medicin 1941. — Fanconi: Die Störungen der Blutgerinnung beim Kinde, Leipzig 1941. — Fåhraeus: The suspension stability of the blood, Stockholm 1921. — Albers: Zschr. klin. Med. *132*, 1940, 807. — Magnus, Levy: Acta med. scand., *95*, 1938, 217. — Svedberg & K. O. Pederser: The Ultracentrifuge. Oxford 1940. — Jersild: Zschr. klin. Med. *130*, 1936, 670. — Bing: 1936 see above and ibid. *91*, 1937, 336. — Waldenström: Nordisk Medicin 1943. — Longsworth, Shedlovsky and Mac Innes: J. exp. Med. *70*, 1939, 399. — Tiselius: Biochem. J. *31*, *1937*, 1464. — Stenhagen: Biochem. J. *38*, 1938, 714. — Blix: Zschr. f. d. ges exper. Med. *105*, 1939, 575. — Gutmann and Wise: Proc. Soc. exper. Biol. and Med., *35*, 1936, 124. — von Bonsdorff, Groth, Packalén: Finska läk. förh. *80*, 1937, 486. — von Bonsdorff, Groth, Packalén: Fol. hämat. *59*, 1938,

184. — von Bonsdorff, Groth, Packalén: Acta med. scand. *91*, 1937, 55. — Wintrobe & Buell: Bull. John Hopkins Hosp., *52*, 1933, 156. — Packalén: Acta med. scand. *100*, 1939, 1. — Cantarow, Am.: I. med. sc., *189*, 1935, 425. — Holmberg & Grönvall: Hoppe Seylers Zschr. *273*, 1942, 199. — Olhagen: Acta med. scand. — Magnus Levy: Acta med. scand. *95*, 1938, 217. — Risak: Zschr. klin. Med. *128*, 1935, 605. — Gutmann, Gutmann, Moore et al.