ULTRACENTRIFUGE INVESTIGATION OF THE SERUM PROTEINS IN WALDENSTRÖM'S MACROGLOBULINAEMIA

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INTRODUCTION

The clinical and haematological aspects of WALDENSTRÖM's macroglobulinaemia have been discussed in a previous publication[1], with reference to the literature and to personal observations. The various protein-chemical aspects of this disease are the subject of our present investigations. The ultracentrifugal analysis of the serum proteins are presented in this paper.

PEDERSEN[2] was the first to use the ultracentrifuge in exhaustive investigations of human serum. In 1944 WALDENSTRÖM[3] described three patients in whose serum PEDERSEN, with the aid of the ultracentrifuge, demonstrated an abnormal, rapidly sedimentating protein fraction. WALDENSTRÖM referred to these cases as macroglobulinaemia and raised the question as to whether they might involve "incipient myelomatosis or a new syndrome". WALDENSTRÖM'S observations were corroborated by many investigators. Some 200 patients with this form of paraproteinaemia have been described in the literature[4,5] and the condition was found indeed to be a separate clinical entity, which has since become known as WALDENSTRÖM's macroglobulinaemia.

Although ultracentrifuge investigation of serum can deepen our understanding of the composition of the serum proteins and although it is of great significance in many scientific problems, it had (hitherto) been of only subordinate importance in clinical diagnosis. It is only in diagnosing WALDENSTRÖM's macroglobulinaemia that this method of investigation had proved to be indispensable.

NORMAL SERUM

In the ultracentrifuge diagram of serum from normal subjects, obtained under the usual experimental conditions (serum dilution 1 g/100 ml, etc.), three fractions can be distinguished (Fig. 1), *viz.*:

(1) The *A*-fraction*, including the albumins and part of the α- and β-globulins. Sedimentation constant: 4.0 $\pm$ 1.0 *S*. Mol. wt. 60,000–70,000.

(2) The *G*-fraction*, including the major part of the γ-globulins and part of the α- and β-globulins. Sedimentation constant: 7.0 $\pm$ 0.4 *S*. Mol. wt. 150,000–170,000.

* The designation *A*- and *G*-fraction (abbreviation of albumin and globulin fraction) is not entirely correct, because these fractions are not exclusively composed of albumins and globulins respectively. Some investigators denominate the various components in accordance with the number of Svedberg units in which the sedimentation constant of the fraction in question is expressed. They refer to 4 *S*, 7 *S* and 19 *S* components. However, this also is hardly satisfactory because the sedimentation constants of these fractions are subject to individual variations, albeit within relatively narrow limits.

(3) The physiological macroglobulin fraction. In the diagram, this fraction is visible as a small, obtuse peak ($< 5\%$ of the total serum proteins). This fraction includes part of the α-, β- and γ-globulins, *i.e.* the α_2-macroglobulins (α_2M), the β_2-macroglobulins (β_2M) and the macromolecular γ-globulin component. The immunologically important serum protein fractions such as properdin, various agglutinins, etc. proved to be macromolecular globulins, constituting part of the heterogenous physiological macroglobulin component. The sedimentation constant of this fraction is 15–20 *S* (average value found in 200 cases: 17 *S*).

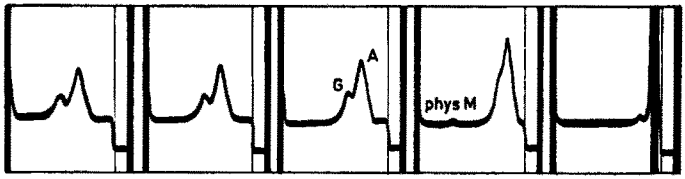
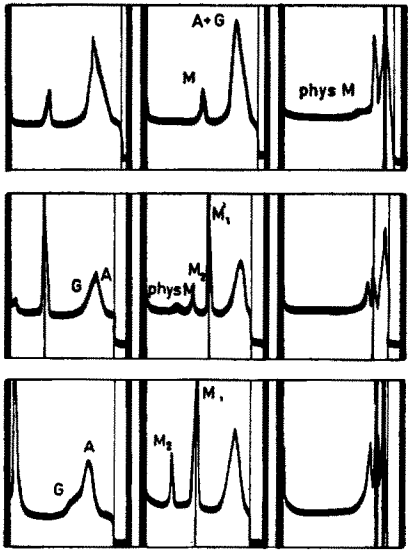


Fig. 1. Ultracentrifuge diagram of normal serum. Direction of sedimentation from right to left. A: $s_{20} = 4.0 \pm 0.15$ *S*; G: $s_{20} = 6.5 \pm 0.3$ *S*; phys. M: $s_{20} = 16.8 \pm 0.2$ *S*.



(a) macroglobulinaemia
A + G: $s_{20} = 3.7 \pm 0.15$ *S*
path. M: $s_{20} = 15.0 \pm 0.5$ *S*
phys. M: $s_{20} = 22.5 \pm 0.5$ *S*

(b) macroglobulinaemia
A + G: $s_{20} = 4.0 \pm 0.1$ *S*
path. M_1: $s_{20} = 15.3 \pm 1.0$ *S*
path. M_2: $s_{20} = 22.1 \pm 1.0$ *S*
phys. M : $s_{20} = 26.5 \pm 1.0$ *S*

(c) macroglobulinaemia
A: $s_{20} = 4.2 \pm 0.25$ *S*
G: $s_{20} = 6.6 \pm 0.4$ *S*
path. M_1: $s_{20} = 15.5 \pm 0.5$ *S*
path. M_2: $s_{20} = 21.0 \pm 1.0$ *S*

Fig. 2. Ultracentrifuge diagrams of the sera of patients with macroglobulinaemia. Direction of sedimentation from right to left.

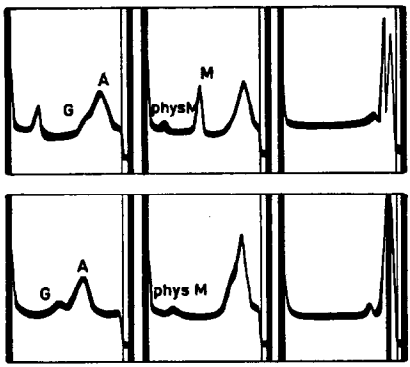
MACROGLOBULINAEMIA

Up to October 1959, 500 ultracentrifuge serum investigations were carried out in the Van 't Hoff Laboratory, Utrecht. In 75 cases, WALDENSTRÖM's macroglobulinaemia was diagnosed on the basis of a pathological macroglobulin fraction thus found. The ultracentrifuge diagram of a patient suffering from this affection is characterized by a small, relatively heterogenous physiological macroglobulin com-

ponent (phys. M), and an abnormal, peaked, rapidly sedimentating fraction (M) — the relatively homogeneous pathological macroglobulin component (Fig. 2a).

From the literature we collected data on 100 patients with macroglobulinaemia. In 90 instances the sedimentation constant of the pathological macroglobulin fraction was reported. We calculated the sedimentation constant of this fraction in our 75 cases (Table I). In fully 95% of cases the sedimentation constant of the pathological macroglobulin fraction was found to exceed 13 *S*. It is questionable whether cases in which an abnormal protein component with a sedimentation constant smaller than 13 *S* was found should be regarded as cases of WALDENSTRÖM's macroglobulinaemia: perhaps they are cases of multiple myeloma with paraproteins of an abnormally high rate of sedimentation.

Occasionally, the ultracentrifuge diagram does not show a single pathological macroglobulin component but several—usually two (Fig. 2b and c) but sometimes three[6] or even four[7].



(a) Total serum
A: $s_{20} = 4.0 \pm 0.2\ S$
G: $s_{20} = 6.0 \pm 0.3\ S$
path. M: $s_{20} = 15.2 \pm 1.0\ S$
phys. M: $s_{20} = 23.5 \pm 1.0\ S$

(b) After removal of the cryoglobulin-fraction.
A: $s_{20} = 4.0 \pm 0.2\ S$
G: $s_{20} = 6.0 \pm 0.3\ S$
path. M: —
phys. M: $s_{20} = 16.5 \pm 1.0\ S$

Fig. 3. Ultracentrifuge diagrams of the serum of a patient with cryo-macroglobulinaemia, before and after removal of the cryoglobulin-fraction. Direction of sedimentation from right to left.

In the presence of a pathological macroglobulin fraction, the sedimentation constant of the physiological macroglobulins was generally higher than that found in the absence of abnormal macroglobulins. The sedimentation constant of the physiological macroglobulin fraction was reported in 43 of the case records found in the literature. In our material, this constant was calculated in 69 cases* (Table II).

The influence of the presence of a pathological macroglobulin fraction on the rate of sedimentation of the physiological macroglobulin component is illustrated by a number of cases of macroglobulinaemia in which the abnormal macroglobulins were at the same time cryoglobulins. In a rotor, pre-heated to 37°, an ultracentrifuge investigation of the total serum was first made. The serum was allowed to cool (precipitation of macro-cryoglobulins), and the macro-cryoglobulins were centrifuged off. The supernatant fluid was subsequently submitted to ultracentrifuge investigation under identical test conditions. In all cases the pathological macroglobulin component was found to be no longer demonstrable (Fig. 3b), while the sedimentation constant of the

* In four of the remaining six cases the physiological macroglobulin fraction—although unmistakably present—was visible only on one photograph; the sedimentation constant consequently could not be calculated. In two cases, no physiological macroglobulin peak was seen.

TABLE I

DATA ON THE SEDIMENTATION CONSTANTS OF THE PATHOLOGICAL MACROGLOBULIN FRACTION IN 90 CASES FROM THE LITERATURE AND 75 PERSONAL OBSERVATIONS

Sedimentation constant in S	*Cases from the literature*	*Personal observations*
11–12	3	—
12–13	3	—
13–14	6	2
14–15	9	15
15–16	33	30
16–17	19	21
17–18	9	3
18–19	7	1
19–20	6	2
20–21	5	1
Total	90	75

TABLE II

DATA ON THE SEDIMENTATION CONSTANT OF PHYSIOLOGICAL MACROGLOBULINS

Sedimentation constant in S	*No Waldenström's macroglobulin*	*Waldenström's macroglobulin*	
		cases from the literature	*personal observations*
10–11	—	—	—
11–12	1	—	—
12–13	2	—	—
13–14	1	—	—
14–15	8	—	—
15–16	40	1	1
16–17	60	1	—
17–18	42	4	—
18–19	24	2	—
19–20	12	—	1
20–21	4	7	8
21–22	2	—	3
22–23	2	9	12
23–24	—	5	15
24–25	—	4	12
25–26	1	2	8
26–27	—	4	5
27–28	—	1	2
28–29	1	2	1
29–30	—	1	1
Total	200	43	69
average	17.0 *S**	22.4 *S*	23.8 *S*
standard deviation separate determ.	1.92 *S*	3.39 *S*	2.25 *S*
standard deviation average	0.14 *S*	0.52 *S*	0.27 *S*

* This is in agreement with the data of KANZOW *et al.*[11], who found an average value of 16.9 ± 0.8 *S*.

physiological macroglobulins had considerably decreased in value (Table III), corresponding with the average established by us for this component in normal serum.

An obvious explanation of these phenomena lies in the association of molecules of the pathological macroglobulins with molecules of the group of physiological macroglobulins.

If many of the macroglobulins may be regarded as smaller units coupled by means of *S-S* bridges[8], then it could be that, say, one or two pathological macroglobulin molecules replace smaller units, as a result of which especially large molecules are formed.

Let one of the molecular types of physiological macroglobulins with molecular weight $M_{phys.}$ consist of three equal parts "$^1/_3\ M_{phys.}$", then, if one or two of these parts are replaced by pathological macroglobulin molecules with molecular weight

TABLE III

DATA ON THE SEDIMENTATION CONSTANTS OF THE VARIOUS SERUM PROTEIN FRACTIONS IN CASES OF MACRO-CRYOGLOBULINAEMIA, BEFORE AND AFTER REMOVAL OF THE MACRO-CRYOGLOBULINS

Case	*Fraction*	*Total serum sed. const. in S*	*After removal of cryoglob. sed. const. in S*
I	A	4.0 ± 0.2	4.0 ± 0.2
	G	6.0 ± 0.3	6.0 ± 0.3
	path. M	15.2 ± 1.0	—
	phys. M	23.5 ± 1.0	16.5 ± 1.0
II	A	4.3 ± 0.2	3.9 ± 0.2
	G	6.6 ± 0.3	6.4 ± 0.3
	path. M	15.0 ± 1.0	—
	phys. M	23.7 ± 1.0	15.9 ± 1.0
III	A	4.0 ± 0.2	4.0 ± 0.2
	G	6.5 ± 0.3	6.5 ± 0.3
	path. M	15.7 ± 1.0	—
	phys. M	24.9 ± 1.0	16.1 ± 1.0

$M_{path.}$, molecules are formed with a molecular weight of $M_I = (M_{path.} + {}^2/_3\ M_{phys.})$ and $M_{II} = (2\ M_{path.} + {}^1/_3\ M_{phys.})$. Postulating $M_{path.} = M_{phys.}$, and assuming all molecular types to be spherical, then it is simple to understand that the sedimentation constants of the molecules $M_{path.}$, M_I and M_{II} must be in the ratio 1 : 1.4 : 1.75.

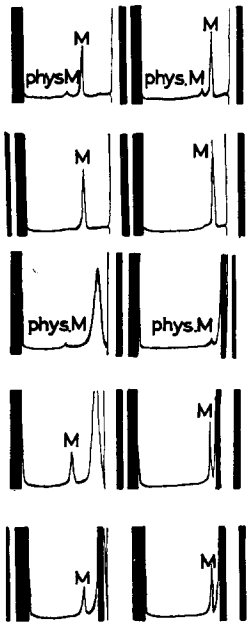
The proportion approximately calculated in this way is reflected in the values which are often obtained for the sedimentation constant of rapidly sedimenting globulin fractions, *viz.* 16, 22 and 28 *S*.

In order to establish experimentally whether an association of pathological macroglobulins with physiologically occurring macroglobulins takes place, an isolated pathological macroglobulin component* was added to normal serum. The

* This pathological macroglobulin component was isolated by means of precipitation at low ionic strength. The precipitate was dissolved in a physiological saline solution at pH 6.5. From this solution, the macroglobulin fraction was again precipitated by dilution with distilled water. This procedure was repeated several times. In this way we obtained a specimen which still contained a "physiological macroglobulin component" ($s_{20} = 25.1 \pm 1.3\ S$) in addition to the pathological macroglobulin fraction ($s_{20} = 17.0 \pm 0.8\ S$). From this specimen, the pure pathological macroglobulin fraction was obtained by preparative ultracentrifugation (Fig. 4).

ultracentrifuge was then used to determine whether the sedimentation constant of the physiologically occurring macroglobulin fraction increased.

This, however, was not demonstrable, no component being found which sedimented more rapidly than the pathological macroglobulin fraction. Repetition under modified test conditions (concentration and time of incubation) yielded identical results.



(a) path. M: $s_{20} = 17.0 \pm 0.8\ S$
phys. M: $s_{20} = 25.1 \pm 1.3\ S$

(b) path. M: $s_{20} = 18.3 \pm 0.9\ S$

(c) phys. M: $s_{20} = 16.7 \pm 0.8\ S$

(d) path. M: $s_{20} = 15.9 \pm 0.8\ S$

(e) path. M: $s_{20} = 16.0 \pm 0.8\ S$

Fig. 4. Ultracentrifugal analysis of the influence of an isolated pathological macroglobulin component on the sedimentation rate of the physiologically occurring macroglobulins. *a.* the isolated pathological macroglobulin fraction, contaminated with the physiological macroglobulin component. *b.* a purified pathological macroglobulin component, after preparative ultracentrifugation of solution (a). *c.* ultracentrifuge diagram of a normal human serum. *d.* ultracentrifuge diagram of a mixture of the isolated pathological macroglobulin fraction (b) and the normal serum (c). The sedimentation constant of the physiological macroglobulin component is not increased. *e. viz.* (d) after prolonged incubation at a high protein concentration. Direction of sedimentation from the right to the left side.

Since it is possible that the "physiological macroglobulin component" in a patient with WALDENSTRÖM's macroglobulinaemia differs from the corresponding component in serum from normal subjects, experiments will have to be made to determine the influence of isolated pathological macroglobulins on the rate of sedimentation of the physiologically occurring macroglobulin component encountered in pathological serum.

DIFFICULTIES IN THE INTERPRETATION

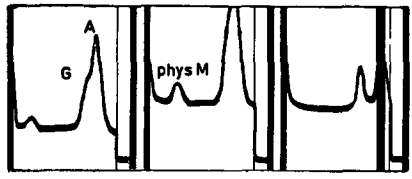
Under the following conditions, the interpretation of an ultracentrifuge diagram may be less simple.

A. Increase in the physiologically occurring macroglobulin fraction

It is generally assumed that the physiological macroglobulin fraction does not exceed 5% of the total serum protein value under normal conditions. Increases in

this fraction have been described in hepatopathies, nephropathies, acute inflammations, syphilis, etc.[9-14]. This increase is as a rule of unimportant quantitative importance. A marked increase in this fraction, however, may impede differentiation from cases of WALDENSTRÖM's macroglobulinaemia in which only one rapidly sedimentating fraction is visible in the ultracentrifuge diagram (as a general rule, the physiological macroglobulin fraction is recognizable beside the pathological macroglobulin fraction; in such cases the diagnosis offers no difficulty).

If only one rapidly sedimentating fraction is visible in the ultracentrifuge diagram, then it is often possible to distinguish between the pathological macroglobulin component and an increased physiological macroglobulin fraction on the basis of the *shape* of the peak. A sharp, symmetrical peak with a narrow base indicates the presence of a relatively homogeneous protein component, *i.e.* pathological macroglobulins. A flat peak with a broad base suggests an increase in the more heterogenous normally occurring macroglobulins (Fig. 5).



A: $s_{20} = 3.8 \pm 0.15\ S$
G: $s_{20} = 6.3 \pm 0.25\ S$
phys. M: $s_{20} = 14.8 \pm 1.0\ S$

Fig. 5. Ultracentrifuge diagram of the serum of a patient with liver cirrhosis. Increase of the physiologically occurring macroglobulin fraction. Direction of sedimentation from right to left.

However, since no exact criteria are available for this distinction in ultracentrifuge investigation, this remains a not uncommon difficulty.

In 16 cases in our material, we found only one rapidly sedimentating fraction and therefore had to differentiate between a pathological and an increased physiological macroglobulin component. In a number of cases the shape of the peak was so typically that of a homogeneous or of a heterogeneous protein component, that it formed a sufficient basis for a decision in this respect. Whenever any doubt remained, we resorted to paper or agar gel electrophoresis. If this revealed *no* abnormal, sharply defined protein fraction, then paraproteinaemia (and therefore WALDENSTRÖM's macroglobulinaemia) could be excluded; the rapidly sedimentating protein fraction in such cases had to be an increased physiological macroglobulin component.

In a few cases of multiple myeloma we observed—in addition to the sharp and usually very high G-peak—a fairly considerable, single, rapidly sedimentating protein fraction with a sedimentation constant of about 17 *S*. Since the clinical diagnosis of multiple myeloma was beyond doubt in these cases and since we have never observed a sharp elevation of the G-peak in macroglobulinaemia, we believe that an increase of the physiological macroglobulin fractions exists in these case of multiple myelomatosis.

When a single fairly considerable, rapidly sedimentating protein fraction is seen in the ultracentrifuge diagram, therefore, it is possible to distinguish between a pathological macroglobulin fraction (WALDENSTRÖM's macroglobulinaemia) and an increased physiological macroglobulin component on the following grounds.

A pathological macroglobulin component is seen in the ultracentrifuge diagram

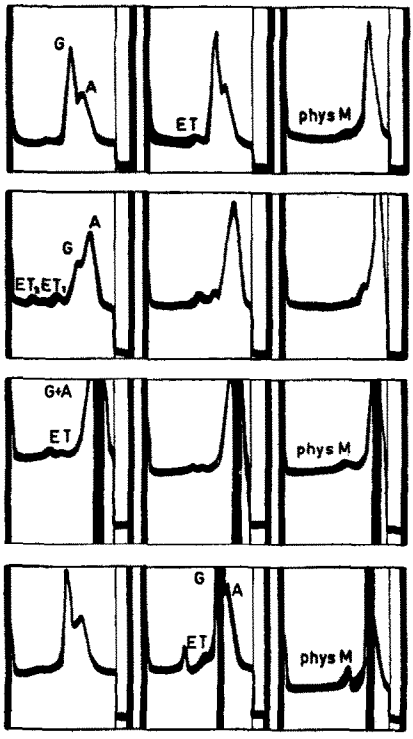
as a sharp symmetrical peak with a narrow base; no sharply raised G-peak is seen in these cases, and electrophoresis reveals a paraprotein fraction in the globulin region.

The physiological macroglobulins are visible in the ultracentrifuge diagram as a more obtuse peak with a broad base, sometimes (in a few cases of multiple myelomatosis) associated with a sharp rise in the G-fraction. In the absence of multiple myelomatosis, electrophoresis reveals *no* paraprotein band.

B. Abnormally rapid sedimentation of protein fractions

(1) Examining sera from patients with multiple myeloma, we rather frequently found relatively *heterogeneous*, abnormally rapidly sedimentating protein components ($s_{20} = 7$–12 *S*), visible in the diagram as *small, flat* additional peaks (E.T.) (Fig. 6).

These additional components are occasionally also encountered in other affec-



(a)
A: $s_{20} = 4.0 \pm 0.15$ *S*
G: $s_{20} = 5.7 \pm 0.3$ *S*
E.T.: $s_{20} = 9.2 \pm 0.4$ *S*
phys. M: $s_{20} = 18.1 \pm 1.0$ *S*

(b)
A: $s_{20} = 4.0 \pm 0.1$ *S*
G: $s_{20} = 6.0 \pm 0.2$ *S*
E.T.$_1$: $s_{20} = 8.5 \pm 0.5$ *S*
E.T.$_2$: $s_{20} = 12.5 \pm 1.0$ *S*

(c)
A + G: $s_{20} = 5.7 \pm 0.3$ *S*
E.T.: $s_{20} = 12.0 \pm 1.5$ *S*
phys. M: $s_{20} = 15.5 \pm 1.5$ *S*

(d)
A: $s_{20} = 3.5 \pm 0.1$ *S*
G: $s_{20} = 4.9 \pm 0.1$ *S*
E.T.: $s_{20} = 6.9 \pm 0.5$ *S*
phys. M: $s_{20} = 16.1 \pm 0.8$ *S*

Fig. 6. Ultracentrifuge diagrams of sera from patients with multiple myeloma. Additional peaks of rapidly sedimentating, rather heterogeneous protein components (E.T.). Direction of sedimentation from right to left.

tions. In the ultracentrifuge diagram, they are readily distinguishable from the pathological macroglobulin fraction as found in WALDENSTRÖM's macroglobulinaemia.

(2) In 10 sera in our material, we found a relatively homogeneous protein component with a sedimentation constant of 7–12 *S* (so-called *atypical macroglobulins*).

(3) Examining serum from a patient with multiple myeloma, we once observed *abnormally rapid sedimentation of part of the G-fraction* (double G-peak) (Fig. 7).

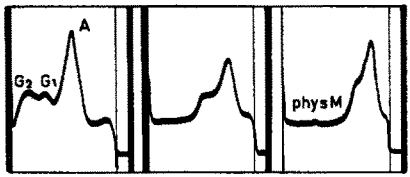
C. Abnormally rapid sedimentation of the entire G-fraction

In some cases of multiple myeloma, the sedimentation constant of the usually high and homogeneous G-component is found to be above the normal (s_{20} = 7–10 *S*).

In the above-mentioned cases, ultracentrifuge investigation reveals abnormally rapidly sedimentating protein fractions. This, however, does not in itself warrant the diagnosis WALDENSTRÖM's macroglobulinaemia. For this diagnosis to be made, the following criteria must be met:

Besides the small, flat peak of the physiological macroglobulins, the ultracentrifuge diagram should show one (sometimes several) abnormal component(s) characterized as follows:

(1) Rapid sedimentation (s_{20} = 13 *S* or more), but less rapid than the associated



A: s_{20} = 4.3 ± 0.4 *S*
G_1: s_{20} = 6.2 ± 0.5 *S*
G_2: s_{20} = 8.0 ± 0.8 *S*
phys. M: s_{20} = 11.0 ± 1.0 *S*

Fig. 7. Ultracentrifuge diagram of the serum of a patient with multiple myeloma. Double G-peak. Direction of sedimentation from right to left.

physiological macroglobulins. *(2)* A recognizable sharp, symmetrical peak with a narrow base in the diagram, as an indication of the relatively homogeneous nature of this protein component (paraprotein aspect). *(3)* Complete separation from the less rapidly sedimentating G-component.

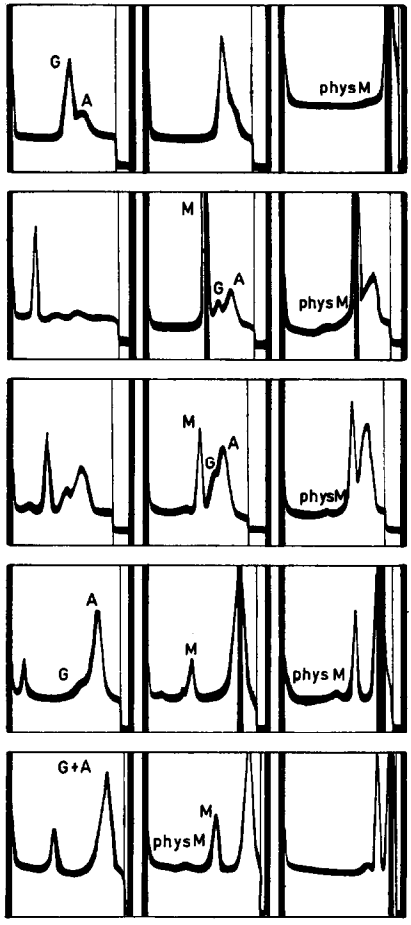
"ATYPICAL MACROGLOBULINS"

In 1955, JAHNKE AND SCHOLTAN[15] described six patients in whose sera they found, by ultracentrifuge investigation, a relatively homogeneous abnormal protein fraction, sedimentating more rapidly than the G-component but with a sedimentation constant lower (s_{20} = 7–10 *S*) than that found in WALDENSTRÖM's macroglobulinaemia. They called these proteins "atypical macroglobulins". Similar findings have also been reported by other investigators[16–18]. JAHNKE AND SCHOLTAN in particular believed that this "atypical macroglobulinaemia" constitutes a transition between the two paraproteinaemias known as multiple myeloma and WALDENSTRÖM's macroglobulinaemia.

An abnormal protein component of this kind was found in ten cases in our material (sedimentation constants: 8.2 *S* in 2 cases; 8.5 *S* in 1; 8.6 *S* in 3; 8.8 *S* in 1; 11.0 *S* in 1 and 11.8 *S* in 1 case).

Fig. 8 presents the ultracentrifuge diagrams of two cases (Fig. 8b and c), which are compared with the diagrams obtained of serum from a patient with multiple myeloma (Fig. 8a) and from two patients with WALDENSTRÖM's macroglobulinaemia (Fig. 8d and e). Indeed it is highly attractive—on the basis of this series—to assume a gradual transition from multiple myeloma via "atypical macroglobulinaemia" to WALDENSTRÖM's macroglobulinaemia.

We hope to present a more detailed discussion of these paraproteins in a later publication which, in addition to ultracentrifuge data, is to include clinical, haematological, biochemical and serological findings. For the time being, there seem to be



(a) Multiple myeloma
A: s_{20} = 4.3 ± 0.3 S
G: s_{20} = 6.5 ± 0.4 S
phys. M: s_{20} = 18.4 ± 1.0 S

(b) Multiple myeloma
A: s_{20} = 4.2 ± 0.2 S
G: s_{20} = 6.5 ± 0.3 S
M: s_{20} = 8.2 ± 0.1 S
phys. M: s_{20} = 15.1 ± 0.8 S

(c) Multiple myeloma
A: s_{20} = 4.0 ± 0.2 S
G: s_{20} = 6.2 ± 0.2 S
M: s_{20} = 8.6 ± 0.1 S
phys. M: s_{20} = 19.8 ± 1.0 S

(d) Macroglobulinaemia
A: s_{20} = 3.8 ± 0.1 S
G: s_{20} = 6.5 ± 0.5 S
M: s_{25} = 14.5 ± 0.5 S
phys. M: s_{25} = 20.5 ± 1.0 S

(e) Macroglobulinaemia
A & G: s_{85} = 4.1 ± 0.1 S
M: s_{25} = 15.0 ± 0.5 S
phys. M: s_{85} = 22.0 ± 1.0 S

Fig. 8. Ultracentrifuge diagrams of sera of patients with multiple myeloma and macroglobulinaemia. Rapidly sedimentating paraproteins. Direction of sedimentation from right to left.

strong arguments in favour of atypical multiple myeloma rather than atypical macroglobulinaemia, because the majority of cases show the typical clinical and other features of multiple myelomatosis[19]. The occurrence of such paraproteins in cases of multiple myeloma has been frequently mentioned in the literature[20-23].

It has been our experience and that of others also, that the sedimentation constant of the abnormal proteins in WALDENSTRÖM's macroglobulinaemia is invariably higher than 13 S. Consequently we believe that it is not justifiable to diagnose WALDENSTRÖM's macroglobulinaemia if ultracentrifuge investigation had revealed an abnormally rapidly sedimentating protein component with a sedimentation constant below 13 S.

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SUMMARY

Data on 90 cases of WALDENSTRÖM's macroglobulinaemia collected from the literature, and personal observations on 75 cases, showed that the sedimentation constant of the pathological macroglobulin component was above 13 *S* in fully 95% of cases. The sedimentation constant of the physiological macroglobulin component —calculated from data on 200 patients not suffering from WALDENSTRÖM's macroglobulinaemia—is about 17 *S*; that in cases of macroglobulinaemia is about 23 *S*. The possible influence of the pathological macroglobulins on the sedimentation constant of the physiological macroglobulin component is discussed and elucidated by results obtained with sera containing cryo-macroglobulins. Difficulties in the interpretation of the ultracentrifuge diagram are discussed, with special reference to the problem of so-called atypical macroglobulins.

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