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Measles Antibodies, Anti-Proteinase and Plasminogen Distribution in Serum and Plasma from Patients Affected with Multiple Sclerosis and Patients Affected with Non-Neurological Diseases

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Summary: Total protein content, α_1-antitrypsin, α_2-macroglobulin and plasminogen levels and measles antibody titers were determined in serum and plasma from patients affected with multiple sclerosis and patients affected with non-neurological diseases.

The results were compared with those from a control group of healthy donors. Both multiple sclerosis patients and patients affected with non-neurological diseases differed from controls for the following parameters: total protein, plasminogen and measles antibody activity. However, when studied longitudinally the different parameters were not altered to the same degree in multiple sclerosis and non-neurological diseases, a fact which is translated in the difference of significance levels. Individual plasminogen values were very often higher in non-neurological diseases than in multiple sclerosis, whereas for increased measles antibody titers it was the reverse. Also, there were no notable changes in α_1-antitrypsin and α_2-macroglobulin values in multiple sclerosis, whereas in some non-neurological disease patients particularly high α_1-antitrypsin and α_2-macroglobulin values were observed.

In the multiple sclerosis patients, no correlations existed between the duration of the disease and disturbed biochemical parameters, or between the disturbed parameters themselves.

Masern-Antikörper-, Proteaseinhibitoren- und Plasminogen-Verteilung in Serum und Plasma bei Patienten mit Multipler Sklerose und nicht-neurologischen Erkrankungen

Zusammenfassung: Die Konzentrationen von Gesamtprotein, α_1-Antitrypsin, α_2-Makroglobulin und Plasminogen sowie die Titer der Masern-Antikörper wurden in Serum und Plasma von Patienten mit Multipler Sklerose und nicht-neurologischen Erkrankungen bestimmt.

Die Ergebnisse wurden mit den bei Gesunden erhaltenen verglichen. Beide Gruppen von Patienten unterschieden sich von den Gesunden aufgrund folgender Kenngrößen: Gesamtprotein, Plasminogen und Masern-Antikörper. Wurden die unterschiedlichen Kenngrößen jedoch longitudinal verfolgt, waren sie bei Multipler Sklerose und nicht-neurologischen Erkrankungen nicht im selben Umfang verändert, was sich in der Differenz der Signifikanzwerte ausdrückt. Die individuellen Plasminogenkonzentrationen waren bei nicht-neurologischen Erkrankungen oft höher als bei Multipler Sklerose; das Gegenteil traf für die Titer der Masern-An-

tikörper zu. Bei Multipler Sklerose wurden auch keine bemerkenswerten Änderungen der α_1-Antitrypsin- und α_2-Makroglobulinkonzentration gegenüber Gesunden beobachtet, während bei nicht-neurologischen Erkrankungen besonders hohe Werte gefunden wurden.

Bei den Patienten mit Multipler Sklerose bestand keine Korrelation zwischen der Dauer der Erkrankung und den gegenüber Gesunden veränderten Kenngrößen sowie zwischen den veränderten Kenngrößen selbst.

Introduction

The importance of fibrin formation is not limited to its role in extravascular thrombotic occlusion, but the deposition of fibrin is an integral part of tissue inflammatory response to any injury, be it traumatic, thermal, microbial or immunological; conversely the fibrinolysis mechanism not only plays a role in the removal of fibrin from the vascular bed, but also in several other biological phenomena such as a malignant transformation (1), macrophage function (1) etc.; moreover fibrinolysis may be considered as a fundamental mechanism of repair of tissue injury (2).

Activation of the fibrinolysis enzyme system depends on the presence in plasma and other body fluids of proteolytic enzyme precursor in large quantities: i.e. plasminogen or profibrinolysin. The activated protease is termed plasmin or fibrinolysin. Plasminogen can be converted to plasmin under the influence of a number of physiological plasminogen activators. It has been proposed that demyelination may be initiated by plasminogen activator released from activated macrophages; and, on the other hand, that plasmin itself causes demyelination (3).

Neurochemical studies have also shown that there is an increase of acid proteinase in multiple sclerosis plaque and that the activity of neutral proteinase can be increased in the acute phase of plaque formation (4). Acid proteinase is one of the major intracellular enzymes involved in the breakdown of proteins. Its activity is elevated in a number of degenerative conditions including muscular dystrophy, inflammatory and allergic conditions (5), and experimental allergic encephalomyelitis (6).

Demyelination, for example, represents breakdown of myelin by proteolytic and lipolytic enzymes, although the mechanisms responsible for their release and their mode of action are unknown. In serum, proteinases are normally inactivated by α_2-macroglobulin and α_1-antitrypsin, which are broad spectrum inhibitors. It is therefore of interest to establish whether multiple sclerosis patients present a particular profile of these parameters. Total protein, α_1-antitrypsin, α_2-macroglobulin, plasminogen contents and measles antibody titers were determined in serum and plasma from patients affected with multiple sclerosis, and from patients affected with non-neurological diseases, and the results were compared with those from a control group consisting of healthy adults.

Materials and Methods

Sample donors and patients

- Control sera and plasma were obtained from 20 healthy adult donors whose ages ranged from 22 to 55 years.
- 25 non-neurological patients were selected on the exclusion of any neurological symptoms, and cancer patients were also rejected. This disease group is very heterogeneous and includes pancreatitis, gastric ulcers, gall bladder lithiasis, cardiovascular syndromes, diabetes, respiratory tract infections, spondylarthrosis and cervico-arthrosis. The ages of the patients varied from 22 to 86 years.
- 29 patients affected with clinically confirmed multiple sclerosis were considered; their ages ranged from 22 to 65 years and the evolution period of the disease from 2 to 27 years. 20 Patients displayed the classical remission-exacerbation pattern, 5 patients presented a slowly progressive and 4 an uncertain evolution.

No sex differentiation was made.

Assay procedures

Total protein was measured according to the Biuret reaction; α_1-antitrypsin, α_2-macroglobulin were determined in single radial immunodiffusion using NOR–Partigen plates (NOR Partigen α_1-antitrypsin SLA 03 and NOR Partigen α_2-macroglobulin SLR 03 Behringwerke AG – Marburg Lahn – Hoechst AG) and plasminogen using M-Partigen plates (M-Partigen – Plasminogen TBV 03, also from Behringwerke).

All Partigen plates were calibrated with Behringwerke standard serum OSMH 07 for the determination of α_1-antitrypsin and α_2-macroglobulin. Behringwerke standard plasma TFI was used for the determination of plasminogen. Haemagglutination inhibition and complement fixation tests were carried out as previously described (7, 8).

Quality control

To guarantee the accuracy and validity of the assays 4 controls were used in haemagglutination inhibition:

a) antigen control: must agglutinate at the chosen dilution
b) measles negative serum: must be void of agglutinins and non-specific inhibitors.
c) measles positive serum: serum pool from children in the acute phase of measles infection.
d) quality control of the red blood cells: they must settle correctly within 1 1/2 h after gentle deposition.

In complement fixation 2 more controls were added:

a) control of the possible anticomplementarity activity of all samples and the antigen.

b) validity control of the complement at the chosen dilution.

Since a normal distribution cannot be assumed in pathological states, the statistical method used to compare the different populations was the non parametric U-test of *Wilcoxon*[1]).

The existence or non existence of correlations between the chosen variables was determined qualitatively by scatter diagrams as reported by *Spiegel* (9).

Results

Table 1 groups the results we obtained from the multiple sclerosis patients. Total protein was generally lower than control values, α_1-antitrypsin and α_2-macroglobulin levels presented a distribution profile similar to that of the control, but plasminogen displayed increased levels.

[1]) Documenta Geigy – Tables scientifiques – Basel.

The distribution of measles antibody activity measured by haemagglutination inhibition and complement fixation was completely disturbed, and in 7 patients (pat. 1, 2, 14, 17, 20, 22, 29) particularly high titers were recorded. Analysis of the results showed no association between particularly increased plasminogen values and measles antibody titers (fig. 1). Moreover, no correlation between the duration of the disease and the disturbed plasminogen and measles antibody parameters could be detected (fig. 1). Indeed the scatter diagrams show that the distribution of the variables is random: the points do not lie near a line with either a positive or a negative slope; they are uncorrelated.

Table 2 presents the parameters from the patients affected with non-neurological diseases.

Here also total protein was generally lower than control values and α_1-antitrypsin and α_2-macroglobulin distribution mainly showed profiles similar to that of the control, with the exception of 5 especially high

Tab. 1. Plasma protein parameters and measles antibodies titers in multiple sclerosis.

Patients n = 29	Duration of disease[1]) (years)	Evolution type[1])	Total protein[2]) (g/l)	α_1-Antitrypsin[2]) (g/l)	α_2-Macroglobulin[2]) (g/l)	Plasminogen (mg/l)	Measles antibody titers Complement fixation	Haemagglutination inhibition
1	27	e-r	67	4.40	4.25	172	1/128	1/512
2	26	s-p	61	2.48	1.98	178	1/256	1/512
3	25	e-r	65	3.38	2.23	178	1/128	1/256
4	24	e-r	63	3.66	2.11	172	1/64	1/128
5	24	e-r	59	2.60	2.63	200	1/32	1/64
6	22	e-r	61	3.38	2.23	214	1/32	1/64
7	20	e-r	61	2.35	1.86	172	1/32	1/64
8	18	e-r	59	2.23	2.36	172	–	1/16
9	18	e-r	67	3.25	1.98	174	1/256	1/64
10	18	s-p	nd	nd	nd	206	1/16	1/32
11	15/20*	s-p	56	2.98	1.40	146	1/16	1/32
12	16	e-r	64	2.72	2.11	178	1/32	1/64
13	14	e-r	68	3.25	2.11	214	1/32	1/64
14	13	e-r	72	2.98	3.18	172	1/256	1/1024
15	12	e-r	58	2.00	1.40	214	1/128	1/256
16	11	u	75	2.48	1.98	206	1/128	1/256
17	10	e-r	59	3.25	1.74	200	1/128	1/512
18	8	e-r	70	2.35	2.11	178	1/64	1/64
19	7	e-r	59	2.85	2.76	174	1/64	1/128
20	6	s-p	67	3.81	2.36	236	1/256	1/1024
21	6	s-p	61	2.85	2.23	152	1/16	1/32
22	6	e-r	67	5.02	2.70	116	1/256	1/2048
23	6	e-r	66	3.52	2.76	206	–	–
24	5	e	74	3.11	2.36	172	1/128	1/256
25	4	e-r	61	3.95	1.98	178	1/16	1/64
26	2	e-r	73	2.98	2.36	178	1/8	1/16
27	u	u	66	3.38	2.23	166	1/64	1/128
28	u	u	68	3.52	2.76	222	1/32	1/128
29	u	u	67	1.89	2.11	172	1/128	1/512

[1]) e-r = exacerbation remission; s-p = slowly progressive; e = exacerbation; u = unknown
[2]) nd = not determined
*: uncertain

α_1-antitrypsin values (pat. 17, 19, 22, 23, 24); however only patients 17 and 24 combined those high values with particularly high plasminogen levels; one patient (pat. 2) also had high α_2-macroglobulin levels.

Plasminogen values were generally much increased when compared to control values and, very surprisingly, measles antibody activity was increased, especially in haemagglutination inhibition.

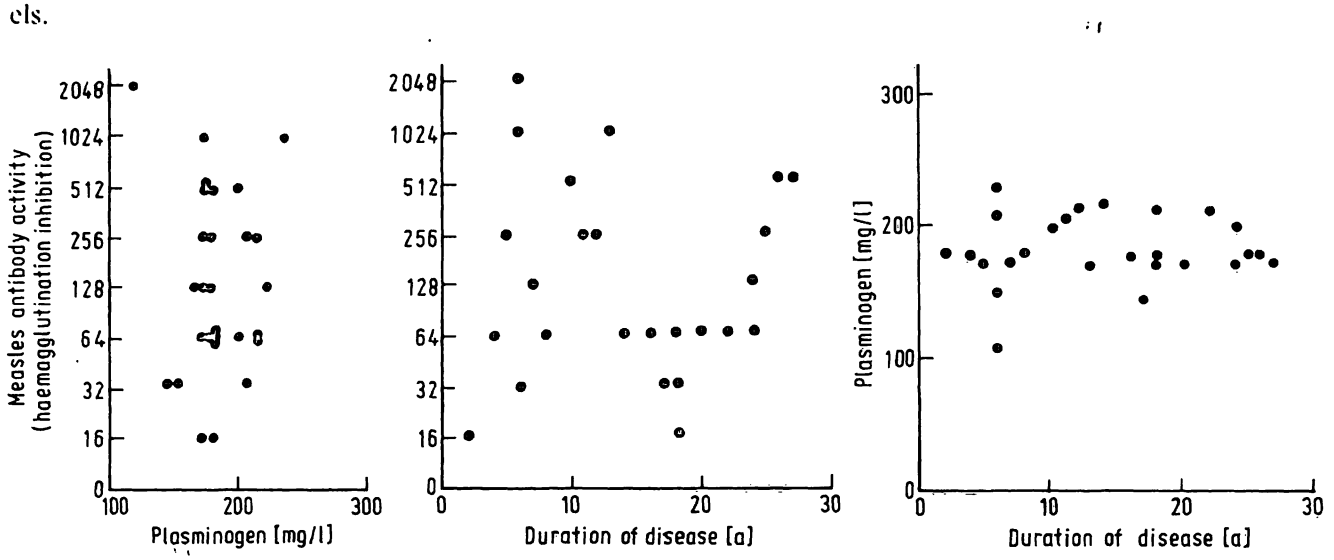

Fig. 1. Scatter diagrams of the variables, measles antibody activity, plasminogen levels and duration of disease in multiple sclerosis patients.

Tab. 2. Plasma protein parameters and measles antibody titers in non-neurological patients.

Patients n = 25	Total protein (g/l)	α_1-Antitrypsin (g/l)	α_2-Macroglobulin (g/l)	Plasminogen (mg/l)	Measles antibody titers[1]) Complement fixation	Haemagglutination inhibition
1	62	4.40	2.49	236	–	1/16
2	67	1.78	4.41	206	1/64	1/512
3	70	3.66	3.48	186	1/128	1/256
4	54	3.11	2.70	228	–	1/16
5	55	2.12	1.51	200	1/32	1/128
6	72	2.98	2.11	172	–	–
7	77	2.85	2.49	200	–	1/8
8	69	2.98	3.00	232	–	–
9	70	2.98	1.98	200	1/64	1/1024
10	65	3.81	2.11	100	1/32	1/32
11	68	3.11	1.51	150	1/8	1/128
12	75	3.95	3.63	208	–	–
13	74	2.85	1.86	232	–	1/16
14	72	4.40	2.11	160	nd	nd
15	72	3.11	1.74	194	1/16	1/128
16	66	2.98	2.90	282	1/64	1/256
17	58	5.85	3.63	194	1/16	1/64
18	56	4.55	2.36	156	1/128	1/256
19	61	5.02	1.98	162	1/16	1/64
20	76	2.72	2.23	194	1/64	1/256
21	61	3.81	3.48	208	1/16	1/32
22	65	6.54	1.74	162	1/64	1/128
23	68	5.77	2.11	156	–	1/64
24	73	7.09	2.11	282	1/16	1/64
25	77	3.66	3.63	208	–	1/16

[1]) nd = not determined

Finally the median values and significant differences between the control group and pathological groups are reported in table 3. The non-neurological disease group significantly differed from controls for total protein, plasminogen values and haemagglutination inhibition measles antibody titers, whereas the multiple sclerosis group significantly differed for total protein, plasminogen values and measles antibody titers measured by both, complement fixation and haemagglutination inhibition.

Discussion

A general increase in plasminogen was observed in all pathological cases, and there was no correlation of increased plasminogen values with any particular neurological or non-neurological disorder.

In both pathological groups the plasminogen increase is most probably due to an equilibrating feedback system compensating for a higher demand of turnover of plasminogen into plasmin. It is an acknowledged fact that plasmin and plasminogen activators are implicated in a variety of biological processes (3, 10, 11, 12). In vivo the main target of plasmin is fibrin, which forms the fabric for cellular proliferation and migration (8, 13). Several authors have suggested that plasminogen is adsorbed to polymerizing fibrin and converted to active enzyme by activators which diffuse into the locus of injury (1, 14).

Tab. 3. Median values and significant differences between controls and pathological samples for $2\alpha = 0.01$, $2\alpha = 0.02$.

	N	Total protein (g/l) median	α_1-Antitrypsin (g/l) median	α_2-Macroglobulin (g/l) median	Plasminogen (mg/l) median	Measles antibody titers: Complement fixation median	Measles antibody titers: Haemagglutination inhibition median
Controls	20	74	3.32	2.36	139	1/16	1/16
Multiple sclerosis	29	66°	3.10	2.23	178°	1/64°	1/128°
Non-neurological patients	25	68*	3.66	2.23	200*	1/16	1/64**

° Multiple sclerosis values differ from controls for $2\alpha = 0.01$
* Non-neurological patients differ from controls for $2\alpha = 0.01$
** Non-neurological patients differ from controls for $2\alpha = 0.02$

There is also most probably a link with the complement cascade: the complement plays an important role in inflammatory responses; the splitting of C_3 in vivo by thrombin and plasmin and the conversion by plasmin of C_1 to C_1 active esterase is of importance in antibody-independent inflammatory reaction.

However as far as the non-neurological group is concerned the plasminogen increase may also be explained by the fact that the mean age is considerably higher than the mean age of the multiple sclerosis group (15).

For multiple sclerosis, the increased plasminogen serum levels agree with earlier findings in CSF, where higher levels of plasminogen were found in CSF from patients with various neurological disorders (13, 16). The authors observed a good correlation between CSF plasminogen values and protein concentrations, from which it was concluded that the CSF plasminogen is probably derived from the blood, rather than produced in the nervous system.

Presumably in multiple sclerosis, the immunological disturbances can activate the coagulation cascade, with the consumption of fibrinogen. Consequently, as already stated, there will be a higher demand for the turnover of plasminogen into plasmin.

When looking at the distribution profile of α_1-antitrypsin and α_2-macroglobulin, a puzzling observation is that in spite of the increased proteolytic proenzyme plasminogen activity, serum levels of these proteinase inhibitors are not significantly altered in the majority of patients.

Normally the end result of a fibrinolytic episode at the time when all circulating plasmin has been inhibited, will be a fall of plasminogen as well as plasminogen inhibitor levels in plasma. The reverse is also true, so one should expect an increase of anti-proteinase levels when plasminogen levels are increased.

α_1-Antitrypsin is the major broad spectrum serum protease inhibitor in plasma; it is an important acute-phase protein, binding and neutralizing a range of proteases which are then cleared by the RES; α_2-macroglobulin, a macrophage product, participates in this clearance, but α_2-macroglobulin plays a more specific role in the inactivation of plasmin. However it reacts more slowly with plasmin than does α_2-antiplasmin and acts as a second line inhibitor (17, 18).

Since α_1-antitrypsin plays an important role in the modulation of inflammatory processes it is the more surprizing that in multiple sclerosis, where the CNS is involved in an important inflammatory reaction, the anti-proteinase distribution was even more regular than in the non-neurological disease group, where in a few patients particularly high α_1-antitrypsin and α_2-macroglobulin values were observed. However, one can argue that what happens at the peripheral blood level is no true reflection of what happens at the cellular level. At the cellular level there might be a local production of anti-proteinases, but the activity might be very different from that found in the serum.

As far as the measles antibody activity is concerned, even if the general increase in measles antibody titers is more elevated in multiple sclerosis, the finding that elevated titers do not solely appear in multiple sclerosis, but in non-neurological diseases as well, adds more evidence in favour of the hypothesis that increase of viral antibody titers may be the result of non specific stimulation (19), related in some unknown way to the disease process in the CNS; as far as multiple sclerosis is concerned, it also confirms our earlier results (20, 21).

Poskitt et al. (22) demonstrated that immunization of single lymphnodes with various antigens led to the appearance of cells in the afferent lymph that secreted antibody specific for antigen and for a number of

unrelated, non-crossreacting antigens. From their observations, they concluded that the micro-environment within the lymphnode, responding to an antigen, induces not only the maturation of specific antigen reactive lymphocytes, but also the maturation of lymphocytes of unrelated specificities.

Finally the longitudinal differences observed between multiple sclerosis and non-neurological patients which are translated by different significance limits, indicate that in multiple sclerosis, which is primarily a demyelinating disease, the homeostatic functions of the organism are differently altered. Indeed, we do not know how the disturbances of the general metabolism may influence the biochemical function of the immune organ, but it seems that the outstanding features of the disease in multiple sclerosis patients, i.e. the frequent occurrence of relapses and remissions, and the liability of patients to these relapses throughout their life, are a manifestation of particular alterations of homeostatic function.

Conclusions

1. There was no trend for the observed general plasminogen increase in pathological cases to be associated with any disorder in particular, be it neurological or not. Consequently, although the activated plasminogen, plasmin, causes or may cause demyelination, the plasminogen increase in multiple sclerosis patients is still to be seen as part of a general and fundamental mechanism of tissue repair versus tissue injury: but which for unknown reasons, has its privileged area in the CNS, and is translated by the formation of plaques.

2. The quasi-normal distribution of anti-proteinases, and more particularly of α_1-antitrypsin, since it plays an important role in the modulation of inflammatory processes, is the more surprising in multiple sclerosis, where the CNS is involved in an important inflammatory reaction.

   Recent studies (23, 24) have suggested that α_1-antitrypsin may have a role in the regulation of immune responses. Studies with human lymphocytes implied that α_1-antitrypsin inhibited the proliferative responses to mitogenic stimulation and regulatory activity appears to be mediated via the macrophages. According to the high measles antibody activity, found by complement fixation and haemagglutination inhibition, inhibition of the proliferative response to mitogenic stimulation seems not be the case in multiple sclerosis. Moreover in multiple sclerosis there is also evidence for aberrant immune responses that could play a primary or secondary role in the pathogenesis of the disease (25, 26).

   Breit et al. (27) postulated that α_1-antitrypsin might modulate the activation of T-cells through its effect on monocytes, leading to abnormality in immunoregulation and hence a predisposition to the development of a variety of immunologic disorders in α_1-antitrypsin deficient subjects.

   Such a postulate could be accepted for multiple sclerosis but does not account for the very heterogeneous non-neurological group; the probability that all non-neurological patients should present a deficient peripheral α_1-antitrypsin synthesis is practically nil. The answer most probably lies at the cellular level, where local deficient anti-proteinase production might induce a more or less pronounced impaired capacity for protease regulation and increased tissue destruction could occur due to proteolytic damage.

3. Our finding that elevated measles antibody titers, when they are observed, are not associated exclusively with multiple sclerosis, adds more evidence in favour of the non-specific mitogenic stimulation hypothesis and confirms our earlier findings:
   a) in a previous paper (28) we reported the failure to detect measles antibodies in brain specimens from several multiple sclerosis patients.
   b) in a subsequent paper (20), isolated serum immunoglobulins from several multiple sclerosis patients were investigated for the presence of measles antibodies. The results did not provide convincing arguments for the measles virus etiology of the disease, since more than half the patients did not display particular haemagglutination inhibition measles antibody titers, or were even frankly negative.
      We wondered whether measles virus etiology can be generally accepted for multiple sclerosis, with certain patients reacting more strongly to the causative agent, or whether, in certain patients, there is, next to and independent of the disease, a particular immunological response to measles virus.
   c) finally, in a paper on measles antibodies, $\kappa-\lambda$ light chain distribution and immunoglobulins in serum, CSF and brain of a patient affected with multiple sclerosis (21), the data assembled did not add up to convincing arguments for a measles virus etiology. We concluded that most probably the etiology of

multiple sclerosis is not specifically related to measles antigen and agreed with *Nordal* (19), that if there is local antibody production it could well be a result of non-specific mitogenic stimulation associated in some unknown way with the disease process in the CNS.

4. In multiple sclerosis patients, correlations between the duration of the disease and disturbed parameters, and between the perturbed variables themselves were non existent. In addition, the disturbed parameters were not specific for multiple sclerosis; only the significance limits were specific.

The abnormalities observed in multiple sclerosis patients may perhaps be considered as a consequence of a non-specific activation of the coagulation system in a chronic immunological disease. It becomes more and more clear that if one looks for specificity in multiple sclerosis, it must be looked for in the mechanisms by which the CNS specifically becomes the "privileged" area of the disease. The evidence for participation of immune mechanisms in the pathogenesis of this neurological disease is undisputable, but how the mechanisms work is still an enigma.

References

1. Reich, E. (1975) Proteinases and biological control, pp. 333–341, Cold Spring Harbor Laboratory.
2. Astrup, T. (1978) Progress in chemical fibrinolysis, Vol. 3, pp. 1–57, Raven Press.
3. Norton, W. T., Cammer, W., Brosnan, C. F. & Bloom, B. R. (1981) Abstracts, p. 265, 8th Meeting of International Society for Neurochemistry, Nottingham, U.K.
4. Kipelainen, H. O., Halonen, T., Kekoni, J., Molnar, G. K. & Riekkinen, P. J. (1980) Acta Neurol. Scand. *62*, 39.
5. Dingle, J. T. & Fell, H. B. (1969) Liposomes in Biology and Pathology, 2, North Holland Publishers.
6. Riekkinen, P. J., Clausen, J., Frey, H. J., Fog, T. & Rinne, U. K. (1970) Acta Neurologica Scand. *46*, 349.
7. Thiry, L., Dachy, A. & Lowenthal, A. (1969) Arch. Ges. Virusforsch. *28*, 278–284.
8. Zissis, G. & Clinet, G. (1974) Lancet *II*, 754–755.
9. Spiegel, M. R. (1972) Schaum's outline of theory and problems of statistics, pp. 241–242., The Graw-Hill International Book Company, New York.
10. Becker, D., Ossowski, L. & Reich, E. (1981) J. Exp. Med. *154*, 385–395.
11. Skriver, L., Nielsen, L. S., Stephens, R. & Dano, K. (1982) Eur. J. Biochem. *12*, 409–414.
12. Wasi, S., Vadas, P., Gregory, W. C., Movat, H. Z. & Hay, J. B. (1982) J. Immunol. *128*, 1076–1077.
13. Collen, D. (1980) Thromb. Haemost. *43*, 77–89.
14. Alkjaersig, N., Fletcher, A. P. & Sherry, S. (1959) J. Clin. Invest. *38*, 1086–1095.
15. Meade, T. W. & North, W. R. S. (1977) Br. Med. Bull. *33*, 283–288.
16. Kenneth, Kun-Yu, W., Jacobsen, C. O. & Hoak, J. C. (1973) Arch. Neurol. *28*, 64–66.
17. Collen, D. (1976) Eur. J. Biochem. *6*, 209–216.
18. Müllertz, S. & Clemmensen, I. (1976) Biochem. J. *159*, 545–553.
19. Nordal, H. J., Vandvik, B. & Norrby, E. (1978) Scand. J. Immunol. *7*, 473–479.
20. Bollengier, F. & Mahler, A. (1979) J. Neurol. *220*, 105–112.
21. Bollengier, F., Mahler, A. & Clinet, G. (1981) J. Neurol. *225*, 135–143.
22. Poskitt, D. C., Frost, H., Cahill, R. N. P. & Trnka, Z. (1977) Immunology *33*, 81–89.
23. Breit, S. N., Robinson, J. P., Luckhurst, E., Clark, P. & Renny, R. (1982) J. Lab. Clin. Immunol. *7*, 127–135.
24. Van Furth, R., Kramps, J. A. & Diesselhof-den Dulk, M. M. C. (1983) Clin. Exp. Immunol. *51*, 551–557.
25. Waksman, B. H. (1981) Immunology today *2*, 87–93.
26. Bloom, B. R. (1980) Nature *287*, 275–276.
27. Breit, S. N., Luckhurst, E. & Penny, R. (1983) J. Immunol. *130*, 681–686.
28. Bollengier, F., Mahler, A., Clinet, G. & Lowenthal, A. (1978) Brain Res. *152*, 133–144.

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