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[1] Adams, C. W. M., *Nature*, **192**, 331 (1961).
[2] Adams, C. W. M., and Tugan, N. A., *J. Path. Bact.*, **82**, 131 (1961).
[3] Hartsough, G. R., and Gorham, J. R., *Vet. Med.*, **44**, 345 (1947).
[4] Munson, T. O., Holzworth, J., Small, E., Witzel, S., Jones, T. C., and Luginbühl, H., *J. Amer. Vet. Med. Assoc.*, **133**, 563 (1958).
[5] Nishio, S., *Bull. Nat. Inst. Agric. Sci.* (Ser. G), **21**, 89 (1962).
[6] Mason, K. E., Dam, H., and Granados, H., *Anat. Rec.*, **94**, 265 (1946).
[7] Saito, F., Yano, S., and Ishii, T., *Bull. Nat. Inst. Agric. Sci.* (Ser. G), **21**, 105 (1962).

## High Antihaemophilic Factor in Multiple Myeloma

An investigation of antihaemophilic activity in 18 patients with multiple myeloma showed a high antihaemophilic factor (AHF) in 10 cases. The elevated AHF values were not related to hyperglobulinaemia or any other factor which could be determined.

AHF assay was performed by the method of Bergna[1]. Assays on 52 normal plasmas ranged from 50 to 200 per cent of the group mean. Assays were performed on 18 patients with multiple myeloma in various stages of the disease. The blood was drawn at 10.00 a.m. and at bed rest except for one ambulant patient. Symptoms of fever and pain and medications were noted. No patients were studied directly after transportation to the hospital, and none had acute fractures.

The AHF assays of the 18 patients were examined in relation to the age and sex of the patient; total serum protein and albumin and globulin fractions; serum and urine electrophoresis with quantitation of serum proteins by electrophoresis using the Beckman/Spinco 'Analytrol'; immuno-electrophoresis[2] using anti-gamma 2 and anti-gamma$_1$ *A* sera; content of plasma cells in particle smears of aspirated marrow; therapies employed in treatment; and the clinical state of the patient.

Twelve patients had elevated AHF values. Nine were over 300 per cent of the normal group mean, the highest value recorded being 630 per cent. Eleven of these 12 cases with elevated AHF had hyperglobulinaemia, and one had a normal serum globulin level with gamma globulin in the urine. The patient with the highest AHF assay was untreated and had normal total protein and a normal serum globulin fraction by chemical determination, but had an elevated beta fraction by electrophoresis, and gamma$_1$ *A* by immuno-electrophoresis. Of the remaining six cases, five had normal AHF levels and marked hyperglobulinaemia. One patient, untreated, aged 74, was lowest in AHF (30 and 60 per cent) and had a very high gamma of 77·2 per cent with a total protein of 10·1 g per cent.

The plasma and serum of one patient, whose plasma contained 390 per cent AHF, were studied in the thromboplastin generation test[3]. His plasma, diluted with 7 parts of haemophilic plasma, produced a normal TGT, while normal plasma diluted with 3 parts of the same haemophilic plasma give an abnormal TGT. The patient's serum showed no thromboplastic effect. Cohn 'Fraction I' prepared from this patient assayed at 85 per cent of the original plasma AHF. Later, 160 ml. of the same patient's plasma containing 2·92 units of AHF in 1 ml. (1 unit = 1 ml. of normal plasma assaying 100 per cent AHF activity) was fractionated by the method of Cohn[4]. 'Fraction I', dissolved in 15·5 ml., had an assay of 22·7 units per ml., representing 75 per cent of the original activity.

Pitney and Elliott[5] demonstrated that Australian Aborigines, who have hyperglobulinaemia, have a higher plasma AHF than white Australians. They also found elevations of AHF in white Australians who had hyperglobulinaemia associated with lymphosarcoma, autoimmune disease, hepatic cirrhosis, nephrosis and multiple myelomatosis. They thought the rise in AHF might be related to the increased globulin production. Pitney *et al.*[6] studied AHF in normal families and concluded that there was no relation to sex, but there was an increase with age. They found no relation between concentrations of AHF and plasma proteins, particularly gamma globulin, in their normal individuals. Amundsen *et al.*[7] reported elevated AHF levels in patients with carcinoma. Ingram[8] found an increase in AHF following infusion of adrenaline, and Iatrides[9] demonstrated elevated AHF after exercise. Increase in AHF after trauma and childbirth has been shown by Davidson and Tomlinson[10]. Increase in AHF in pregnancy has been reported by Strauss and Diamond[11].

In the present study there was no consistent correlation of the AHF activity with age, sex, therapy, hyperglobulinaemia, type of abnormal globulin (gamma 2 or gamma$_1$ *A*), degree of plasma cell hyperplasia in the bone marrow, or clinical state of the patient. No patients had any recognized thrombotic complications.

The site of origin of AHF is not known. One wonders if the abnormal protein production by the plasma cells in multiple myeloma could include AHF or AHF activator[12] in some cases.

The high AHF values in patients with multiple myeloma are a good source of this factor for experimental purposes. The highly concentrated AHF from such plasma is being used in an attempt to produce anti-AHF antibody in the experimental animal.

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[1] Bergna, L. J., *Blood*, **15**, 637 (1960).
[2] Potter, M., and Kuff, E. L., *J. Nat. Canc. Inst.*, **26**, 1109 (1961).
[3] Biggs, R., and Douglass, A. S., *J. Clin. Path.*, **6**, 23 (1953).
[4] Cohn, E. J., Strong, L. E., Hughes, W. L., jun., Mulford, D. J., Ashworth, J. N., Melin, M., and Taylor, W. L., *J. Amer. Chem. Soc.*, **68**, 459 (1946).
[5] Pitney, W. R., and Elliott, M. H., *Nature*, **185**, 397 (1960).
[6] Pitney, W. R., Kirk, P. L., Arnold, B. J., and Stemhouse, N. S., *Brit. J. Haemat.*, **8**, 421 (1962).
[7] Amundsen, M. A., Spittell, J. A., Thompson, J. H., sen., and Owen, C. A., jun., *Ann. Int. Med.*, **58**, 608 (1963).
[8] Ingram, G. I. C., *J. Physiol.*, **156**, 217 (1961).
[9] Iatrides, S. G., *Fed. Proc.*, **21**, A, 58 (1962).
[10] Davidson, E., and Tomlinson, S., *J. Clin. Path.*, **16**, 112 (1963).
[11] Strauss, H. S., and Diamond, L. K., *New England J. Med.*, **23**, 1251 (1963).
[12] McLester, W. D., and Graham, S. B., *Nature*, **197**, 708 (1963).

## Gel-filtration of Isotopically labelled Ferritin from HeLa Cells

HeLa human cancer cells have been shown to produce ferritin when grown in the presence of iron[1]. HeLa ferritin is electrophoretically heterogeneous; its properties have been compared with those of ferritin isolated from other cells grown *in vitro* and from human and animal tissues[2]. On DEAE chromatography of homogenates of HeLa cells grown in the presence of $FeSO_4$ ($^{59}Fe$), the distribution of the radioactivity was practically the same as the distribution of ultra-violet-absorbing components in the case of purified horse ferritin; the assignment of the activity to ferritin has been confirmed by paper electrophoresis[3]. The use of radio-iron in such experiments results in great sensitivity and specificity. We now report the behaviour of HeLa ferritin on gel-filtration.