

Plasma Protein of Sickle Cell Anaemia Patients in Enugu

¹Ugonabo, M. C., ²Okafor, E. N., ²Ezeoke, A. C. J. and ²Aduba, O.

¹Department of Microbiology, University of Nigeria, Nsukka Campus, Enugu State.

²Department of Chemical Pathology, University of Nigeria, Enugu Campus, Enugu State.

Corresponding Author: Ezeoke, A. C. J. Department of Chemical Pathology, University of Nigeria, Enugu Campus, Enugu State.

Abstract

The plasma total protein, albumin and globulin levels from 93 sickle cell anaemia (HbSS) patients from UNTH, Enugu, and aged between 15–30 years were determined. The Albumin/Globulin ratio was equally calculated. For the control, ninety-six (96) healthy volunteers with normal adult haemoglobin (HbAA) matched for age and sex were recruited for the study. A comparison of the results obtained from the HbSS patients with that of the controls, showed that HbSS patients had significantly lower albumin levels and higher globulin levels than controls, irrespective of sex. The albumin/globulin ratio of patients was significantly lower than that for controls whereas there was no significant change in the total protein levels. The significantly higher globulin levels seen in patients do not seem to confer any immunity on the patients. (P<0.05)

Introduction

The gene for sickle hemoglobin seems to have prospered as a result of slower but ultimately more deadly environmental cataclysm. The most virulent of the human malarial *falciparum* malaria, may have been the exclusive agent of positive selection of Hemoglobin S¹

Since first described by Herrick², a number of advances in the understanding of sickle cell anaemia have constituted important milestones in human genetics. The sickling phenomenon is expressed in both those with sickle cell anaemia and those with sickle cell trait, though more severe in the former.

Sickle cell anaemia in the first two decades of life is marked by periods of clinical quiescence and relative well-being interspersed with intermittent episodes of acute illness. It is remarkable to note that the steady state is characterized varied biochemical abnormalities^{3,4}. A number of plasma factors are believed to play vital roles in the initiation and modulation of the sickling phenomenon. In so far as erythrocyte adhesivity is mediated by plasma proteins, fluctuating levels of those that respond to acute phase reactants during concurrent illness portend adverse results^{5,6}.

Patients with sickle cell anaemia are usually prone to infections probably due to reduced immune status. Since proteins form the bedrocks of the immune status of any individual, the author felt that estimating plasma protein levels would be a worthwhile venture. This, it is hoped may help throw more light into the repeated episodes of infections to which such patients are prone.

Materials and Methods

For this study, a total of 93 patients between the ages of 15 and 30 years, whose hemoglobin genotype was HbSS, were selected from the sickle cell clinic, at UNTH, Enugu. These comprised 44 males (47.3%) and 49 female (52.68%). All patients

were in the steady state, and none had any underlying renal, liver or cardiac problems.

For the control, 96 healthy volunteers with HbAA genotype matched for age and sex were drawn from among hospital staff and students. This number comprised 44 males (45.83%) and 52 (54.17%) females.

Following informed consent, 5 ml of fasted venous blood was collected from the patients into EDTA bottles. Plasma was obtained after centrifugation at 5000 x g for 10 minutes and was then stored in appropriately labeled tubes, ready for further analysis. Plasma total protein level was determined by Biuret method⁷. The plasma albumin was equally determined after ether extraction of globulin by the use of Biuret method⁷. The globulin level was estimated subtracting the albumin value from total protein value.

Data Presentation and Analysis

The student t-test was used as a test of significance. P<0.05 means significant; P>0.05 means not significant.

Results

Table 1: Shows mean and standard deviation of total protein in patients and control. There is no significant variation between the patient and control in total protein (P>0.05). Table 2 Shows Mean and standard deviation of plasma Albumin G/L. There is statistical significant variation in Albumin level in patient and control (P<0.05). Table 3: Shows mean Globulin and standard deviation of patients and control. There is statistical significant difference between the patient and control (P<0.05). Table 4: Shows the mean and standard deviation of plasma albumin. Globulin Ratio (G/L). There is statistical significant difference between the ratio (P<0.05).

Table 1: Mean total protein. By age and sex for patients and control

Age Group (years)	Male			Female		P value
	Patient	Control	P Value	Patients	Control	
15-19	73.0±38 (n = 15)	71.0±3 (n = 15)	>0.05	73.6±5 (n = 15)	72.8±1.32 (n = 17)	>0.05
20-24	72.9±51 (n = 14)	72.3±22 (n = 15)	>0.05	74.3±5 (n = 17)	72.6±31 (n = 19)	>0.05
25-29	72.9±43 (n = 15)	72.1±34 (n = 14)	>0.05	74.2±44 (n = 17)	72.2±34 (n = 16)	>0.05

Table 2: Mean plasma albumin. By age and sex for patients and control

Age Group (years)	Male			Female		P value
	Patient	Control	P Value	Patients	Control	
15-19	29.3±16 (n = 15)	71.8±32 (n = 15)	<0.05	28.9±27 (n = 15)	42.4±0.25 (n = 17)	<0.05
20-24	28.9±15 (n = 14)	41.9±07 (n = 15)	<0.05	28.7±17 (n = 17)	42.0±23 (n = 19)	<0.05
25-29	29.3±19 (n = 15)	42.2±26 (n = 14)	<0.05	29.1±1 (n = 17)	42.1±22 (n = 16)	<0.05

Table 3: Mean plasma albumin. By age and sex for patients and control

Age Group (years)	Male			Female		P value
	Patient	Control	P Value	Patients	Control	
15-19	44.5±43 (n = 15)	30.0±13 (n = 15)	<0.05	45.5±48 (n = 15)	30.4±0.48 (n = 17)	<0.05
20-24	44.0±43 (n = 14)	30.4±2 (n = 15)	<0.05	44.9±0.48 (n = 17)	30.9±24 (n = 19)	<0.05
25-29	43.7±41 (n = 15)	29.9±14 (n = 14)	<0.05	45.0±0.36 (n = 17)	30.7±22 (n = 16)	<0.05

Table 4: Mean plasma albumin/globulin ratio. By age and sex for patients and control

Age Group (years)	Male			Female		P value
	Patient	Control	P Value	Patients	Control	
15-19	0.649±0.096 (n = 15)	1.40±0.12 (n = 15)	<0.05	0.652±0.099 (n = 15)	1.42±0.12 (n = 17)	<0.05
20-24	0.659±0.004 (n = 14)	1.38±0.11 (n = 15)	<0.05	0.645±0.077 (n = 17)	1.37±0.13 (n = 19)	<0.05
25-29	0.675±0.08 (n = 15)	1.42±0.09 (n = 14)	<0.05	0.659±0.054 (n = 17)	1.34±0.12 (n = 16)	<0.05

Discussion

Generally, very few patients with sickle cell anaemia attend sickle cell clinic. The probable reasons for this poor attendance range from poverty lack of awareness to psychological embarrassment suffered by these patients at the hand of the public. It is common knowledge that sickle cell anaemia patients are prone to infection which usually precipitate hemolytic and/or painful crisis.

A comparison of the results in many respects shows that patient with sickle cell anaemia have significantly lower plasma albumin than normal controls for the same age group and sex (Table 2). However, plasma globulin level in patients is significantly higher than in control (Table 3). Interestingly, the albumin / globulin (A/G) ratio in patients is also significantly lower than in controls for all age groups in both sexes (Table 4). Genetic disease cause considerable human suffering and place an enormous financial burden on the health care system. Such genetic disorders affecting red cells are frequently encountered in the Nigerian population. In our area with a history of malaria

endemicity, the frequency of the abnormal genes is high and is believed to be a consequence of the natural in-born resistance provided to the carriers of these genes against malaria^{8,9}.

The low plasma albumin seen in patients with sickle cell anaemia as compared with controls is in contrast with the findings of recent study¹⁰, but agrees with the result of an earlier study¹¹. The difference in the findings may be due to the fact these workers carried out their work on the patients in varying severities of the disease, while the present work was done on patients in the steady state. The low plasma albumin seen in these patients may be due to increased utilization, reduced hepatic synthesis or low protein intake. It may be argued that during the acute phase of the disease, there is a rapid elaboration of these protein in response to the crisis situation.

Since sickle cell anaemia is usually associated with increased reticulocyte count and hence, protein mobilization for hemoglobin synthesis, it is to be expected that plasma albumin level should be low. The low plasma albumin seen in sickle cell anaemia patients may explain partly

the low opsonisation of pathogenic microorganisms in these patients and hence the higher incidence of infection^{12,13,14,15}. Conversely, the significantly higher plasma globulins in the patients though relative, may be due to repeated and chronic infections to which these patients are usually prone, as well as attempts by the immune system to come to terms with these noxious agents. The relative hyperglobulinemia seen in patients, however, does not seem to confer any measure of immunity on these patients, as borne out by repeated infections to which they are usually subjected^{16,17,18}.

In so far as erythrocytes adhesivity is mediated by plasma proteins, fluctuating levels of those that react as acute phase reactants during concurrent illness portend adverse results^{5, 6}. Further work relating to the protein sub-fractions both in steady states and during crisis is recommended.

Acknowledgement

Our appreciation goes to Dr. S.C. Harrada of Dept. of Haematology UNTH for permission to use some of her patients. We are also grateful to Mrs. C.O. Ilo of Chemical Pathology Department for Secretarial assistance and Mr. E. Ele of Chemical Pathology Dept. for technical assistance.

References

- Allison, A.C. (1954) protection afforded by sickle cell trait against subtertian malaria. *Brit. Med. J.* 1:290 – 294.
- Herrick, J. R. (1910): Peculiar elongated and sickle – shaped red blood corpuscles in a case of severe anaemia. *Arch. Intern. Med.* 6:517
- Fleming, A.F. (1989): The presentation, management and prevention of crisis in sickle cell disease in Africa. *Blood Rev.* 3 (1): 18-28
- Person, H.A. (1987): Sickle cell disease: Diagnosis and Management in infancy and childhood. *Pediatrics in Review* 9(4): 121-130.
- Warrier, R.P., Kuvibidila, S., Gordon, L. and Humber, J. (1984): Transport proteins and acute phase reactant proteins in children with sickle cell anaemia. *J. Nat. Med. Assoc. Jan.* 86(1): 33-39.
- Pasvol, C., Weatherale, D.J., Wilson, R.J.M. (1978): Cellular mechanism for the protective effect of Hemoglobin S against *Plasmodium falciparum* malaria. *Nature* 274: 701-703.
- Gornall, A.G. Baidawil, C.J. and David, M.M. (1949): Biuret method of determination of serum proteins. *Mj. Biol. Chem.* 177: 751
- Friedman, M.J. (1978): Erythrocyte mechanism of sickle cell resistance to malaria. *Proc. Natl. Acad. Sci. (USA)* 75: 1994.
- Fleming, A.G., Storey, J., Molineaux, L., Iroko, E.A. and Attai, E.O. (1979): Abnormal Haemoglobin in the Sudan Savanna of Nigeria, 1. Prevalence of haemoglobins and relationships between sickle cell trait, malaria and survival. *Annals of Tropical Medicine and Parasitology* 73: 161-172.
- Hedo, C.C. Aken'ova, Y.S., Okpala, I.E., Durojaiye, A.O., Salimonu, L.S. (1993): Acute phase reactants and severity to homozygous sickle cell disease. *J. Intern. Med.* 233 (6): 467-70.
- Isichei, U.P. (1979): Serum protein profile in sickle cell disease. *Journal of Clinical Pathology* 32: 117-121
- Kaine, W.N. (1983): Morbidity of homozygous sickle cell anaemia in Nigerian children. *Journal of Tropical Pediatrics* 29: 104-111.
- Barrett – Connor, E. 1971 Bacterial infection and sickle cell anaemia. An analysis of 2550 infectious in 166 percents a review of the literature. *Medicine* 50 98-112.
- Zarkowsky, H.S.D. Caccagier F.M. Gill W.C. Wang J.M. Faletta W.M. Londe P.S. Levy J.L. Verter and D. Wethers (1986), Bacteraemia in sickle. Hemoglobinopathies. *J. Pediatr.* 106-579, 585.
- Anna Nowak Wegrzyn, Jerry A. Winkelst E.N., Andrea J. Swift, Howard M. Ledexman and the Pneumococcal conjugate vaccine study group (2000) *Clinical and diagnostic laboratory immunology.* 7 (5): 788-793.
- Akinyanju, O. and Johnson, A.O.I. (1987): Acute illness in Nigerian children with sickle cell anaemia. *Annals of Tropical Pediatrics* 7: 181-186
- Adewuji, J.O. (1988): morbidity in sickle cell disease in early childhood. *J. Trop. Pediatr.* 34: 93.
- Rogers, D.W. Vaidya, S. and Serjeant, G.R. (1978): Early splenomegaly in homozygous sickle cell disease: An indicator of susceptibility to infection *Lancet.* 2: 963-965.