

RESPONSE OF SICKLE CELL ANAEMIA PATIENTS TO THERAPEUTIC TRIAL OF AMPLES A&B

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ABSTRACT

Background: Sickle cell anaemia is common in the Niger delta region of Nigeria. This study was conducted to indirectly assess the significance of cobalamin deficiency in sickle cell anaemia using therapeutic trial of Amples A&B containing cobalamin.

Material & Methods: This study was conducted on sickle cell anaemia patients from University of Benin Teaching Hospital and Sickle Cell Centre, Benin, Nigeria. Sixty patients of ≥ 20 years age, with history of ≥ 2 admissions and blood transfusions, were taken as test subjects. While 60 stable, age and sex matched patients were taken as controls. Clinico-haematologic and demographic features of these patients were noted. Test patients were given intramuscular injections of Amples A&B, containing cyanocobalamin 2500 μ g, folic acid 0.9 μ g, niacinamide 12mg and vitamin C 150mg. Blood counts were carried out before therapy, and at 14 days.

Results: The haemoglobin of patients and controls pre-therapy was 6.28g/dl \pm 0.38 & 6.82 \pm 0.40g/dl and at 14 days of therapy 7.7 \pm 0.53g/dl and 6.8 \pm 0.42g/dl respectively. The mean corpuscular volume was 83.8 \pm 20.7fl and 83.2 \pm 21.0 fl pre-therapy and at 14 days 82.1 \pm 15.8fl and 84.1 \pm 70.6fl respectively. The mean reticulocyte count pre-therapy was 2.2 \pm 0.02% & 2.2 \pm 0.03% for test and controls and at 14 days it increased to 2.8 \pm 0.07% and 2.3 \pm 0.05% respectively. The mean white cell count pre-therapy was 11.14 \pm 5.31 $\times 10^3/\mu$ l & 10.48 \pm 5.14 $\times 10^3/\mu$ l for test and controls. At 14 days it showed a decrease to 8.75 \pm 3.49 $\times 10^3/\mu$ l & 9.89 \pm 4.02 $\times 10^3/\mu$ l respectively. The mean platelet count pre-therapy was 480 \pm 195.21 $\times 10^3/\mu$ l & 500 \pm 210.45 $\times 10^3/\mu$ l for test and controls. At 14 days it decreased to 470 \pm 188.34 $\times 10^3/\mu$ l & 480 \pm 193.12 $\times 10^3/\mu$ l respectively.

The p value for the haemoglobin, mean corpuscular volume, white cell count, reticulocyte count and platelet count of the test compared to controls did not show any statistically significant difference ($p > 0.05$). Over 50% of patients during therapy on Amples A&B reported increase in appetite. Less than 1% of the test subjects complained of mild transient bone pains during therapy.

Conclusion: This study revealed a statistically non-significant response in some laboratory parameters and clinical features in sickle cell anaemia patients on therapeutic trial of Amples A&B.

Key words: Sickle cell anaemia, Amples A&B, Cobalamin.

INTRODUCTION

Sickle cell anaemia (SCA) represents about 2% of the population of the Niger delta region of Nigeria and 25% of Nigerians are heterozygous for it.^{1,2} There are four major crises in SCA but vaso-occlusive crisis is the hallmark of the disease. Megaloblastic anaemia in the general population is in the range of 3 to 29%.³ SCA patients are prone to vaso-occlusive episodes of the gut thus reducing the absorption of cobalamin. Folic acid is routinely given to SCA patients because of their continuous state of haemolysis. A study of adult SCA patients showed that more SCA patients had vitamin B₁₂ (cobalamin) deficiency than healthy controls.⁴

Several anti-sickling agents have been tried to ameliorate the sickling of red cells in SCA patient but there is still no such agent. The current use of hydroxyurea in the treatment of SCA has reduced the incidence of crises, anaemia and mortality.^{5,6} The only therapeutic cure for SCA is stem cell transplantation.⁷

The use of folic acid in doses above 0.1 mg daily may obscure pernicious anaemia and worsen the neurological complications due to cobalamin deficiency. The exact dose of folic acid that will worsen cobalamin neuropathy has been reported by some authors as 400 μ g daily or even less.⁸

Several methods including therapeutic trials have been tried for the diagnosis of vitamin B₁₂

deficiency with some drawbacks. However there is still no golden rule in the diagnosis of cobalamin deficiency.⁹

This preliminary study was conducted to access indirectly the significance of cobalamin deficiency in adult sickle cell anaemia patients using therapeutic trial of Amplex A&B, containing cobalamin.

PATIENTS AND METHODS

One hundred & twenty randomly selected cases of SCA were recruited in this study. They comprised of 60 SCA patients of age ≥ 20 years with a previous history of 2 or more admissions for vaso-occlusive crisis and blood transfusions, presenting to out patient department or admitted in University of Benin Teaching Hospital and Sickle Cell Centre, Benin, Nigeria, from February 2003 to July 2003. The ethical committee of the hospital granted approval for the study and informed consent was obtained from all the subjects. Sixty age and sex matched SCA patients in the steady state without any history of crises or blood transfusion in the previous one year were taken as controls.

Clinico-haematologic and demographic features of all the patients were noted. These included

Table 1: Sex and age distribution of SCA patients/control subjects on therapeutic trials

	Age (Yrs)	Male (%) (n=60)	Female (%) (n=60)
Test	20-30	21(35.0)	21(35.0)
Control	20-30	20(33.3)	21(35.0)
test	31-40	9(15.0)	9(15.0)
control	31-40	10(16.7)	9(15.0)
Total		60(100.0)	60(100.0)

Table 2: Response to Amplex A and B and Placebo by SCA patients and control subjects respectively (pre and post therapy)

Parameters	Mean Hb (g/dl)		Mean MCV (fl)		Mean CR (%)		Mean WBC ($\times 10^3$ /ul)		Mean PLT ($\times 10^3$ /ul)	
	Test (n=60)	Control (n=60)	Test (n=60)	Control (n=60)	Test (n=60)	Control (n=60)	Test (n=60)	Control (n=60)	Test (n=60)	Control (n=60)
Before therapy	6.28 \pm 0.38	6.82 \pm 0.40	83.8 \pm 20.7	83.2 \pm 21.0	2.2 \pm 0.02	2.2 \pm 0.03	11.14 \pm 5.31	10.48 \pm 5.14	480 \pm 195.21	500.0 \pm 210.45
72 hr. after therapy	7.3 \pm 0.34	6.76 \pm 0.26	83.1 \pm 19.4	83.0 \pm 19.8	2.3 \pm 0.04	2.2 \pm 0.02	10.80 \pm 4.23	10.21 \pm 3.89	420.0 \pm 173.42	470.0 \pm 178.95
14 days after therapy	7.7 \pm 0.53	6.8 \pm 0.42	82.1 \pm 15.8	84.1 \pm 70.6	2.8 \pm 0.07	2.3 \pm 0.05	8.75 \pm 188.39	9.89 \pm 188.34	470 \pm 188.34	480 \pm 193.72
P-values	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05	P>0.05

age, gender, clinical features including any history of crises or blood transfusion. The homozygous SS genotype was diagnosed based on the typical clinical features, standard morphological blood film and haemoglobin electrophoresis. Full blood count was analyzed by the coulter counter and included haemoglobin (Hb), white cell count (WBC), and Platelet count (PLT). Corrected reticulocyte (CR) count was determined using brilliant cresyl blue stain.

All these patients continued their routine medications (Folic Acid, etc) throughout the study.

The test patients were given intramuscular injections of Amplex A&B, containing cyanocobalamin 2500 μ g, folic acid 0.9 μ g, niacinamide 12mg and vitamin C 150mg, approved by the Federal Drug Control Agency, on alternate day for 6 doses while the controls were given placebo.

Blood counts were carried out before the commencement of study, at 72 hours, and 14 days of the study.

The data was analyzed with SPSS version 11 and the means were compared using student t test. The level of significance was taken as $p < 0.05$.

RESULTS

Male to female ratio for the test and control patients was equally distributed as shown in the age and sex distribution. However the peak age was 23 years with 65% of the Test and 63.8% Control subjects in the age group 20-30 years.

The haemoglobin of patients and controls pre-therapy was 6.28 \pm 0.38 g/dl and 6.82 \pm 0.40 g/dl. At 14 days of therapy, it was 7.7 \pm 0.53 g/dl & 6.8 \pm 0.42 g/dl respectively.

The MCV pre-therapy was 83.8 \pm 20.7 fl & 83.2 \pm 21.0 fl for test and controls. While at 14 days of commencing therapy it was 82.1 \pm 15.8 fl & 84.1 \pm 70.6 fl respectively.

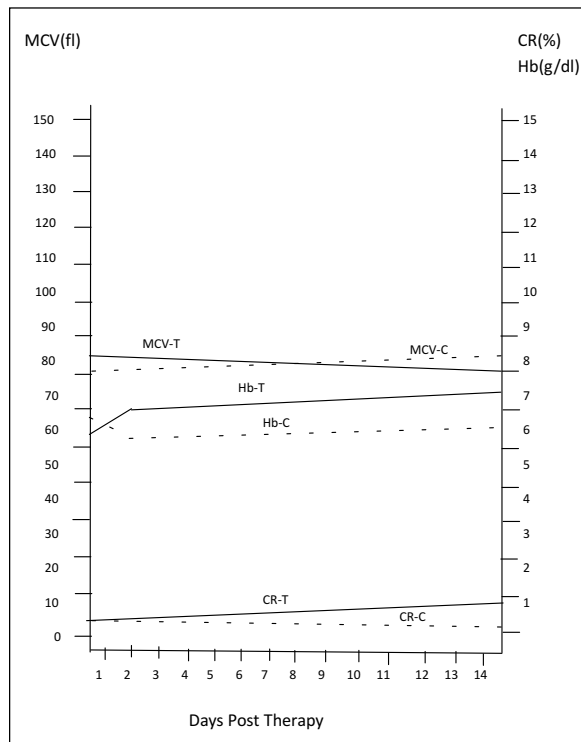
The mean CR pre-therapy was $2.2 \pm 0.02\%$ & $2.2 \pm 0.03\%$ for test and control patients. At 14 days of therapy it increased to $2.8 \pm 0.07\%$ & $2.3 \pm 0.05\%$ respectively.

The mean WBC pre-therapy was $11.14 \pm 5.31 \times 10^3/\mu\text{l}$ and $10.48 \pm 5.14 \times 10^3/\mu\text{l}$ for test and controls. At 14 days of therapy the WBC showed a decrease to $8.75 \pm 3.49 \times 10^3/\mu\text{l}$ and $9.89 \pm 4.02 \times 10^3/\mu\text{l}$ respectively.

The mean platelet count pre-therapy was $480 \pm 195.21 \times 10^3/\mu\text{l}$ & $500 \pm 210.45 \times 10^3/\mu\text{l}$ for test and controls. However at 14 days of therapy the platelet count decreased to $470 \pm 188.34 \times 10^3/\mu\text{l}$ and $480 \pm 193.12 \times 10^3/\mu\text{l}$ respectively.

The p value for the Hb, MCV, WBC, CR and platelet count of test compared to controls did not show any statistically significant difference ($p > 0.05$).

Over 50% of patients during therapy on Amplex A&B reported increase in appetite. Less than 1% of the test subjects complained of mild transient bone pains during therapy.



MCV = Mean corpuscular Volume
Hb = Haemoglobin CR = Corrected Retics
T = Test patients C = Control patients

Fig. 1: Monitoring of response to Amplex A and B and Placebo by SCA patients and control subjects.

Figure 1 is a graphical representation of the monitoring of test and control subjects.

DISCUSSION

Amplex A&B (cyanocobalamin 2500 μg , folic acid 0.9 μg , niacinamide 12mg and vitamin C 150mg) is used as parenteral therapy for sickle cell anaemia patients with megaloblastic anaemia especially when accompanied with malabsorption. It has been shown that the complementary roles of vitamin C include the prevention of bleeding associated with thrombocytopenia, and small sized platelets¹⁰ and functionally abnormal platelets in some megaloblastic anaemia patients.¹¹ Niacin has been shown to be more effective in neurological functions when combined with vitamin B and C groups. Limitations to the use of cyanocobalamin is its short half life when compared to hydroxocobalamin but this is improved by increasing the dose to 2500 μg . However cyanocobalamin has the advantage of being cheap.¹²

The result of response of SCA patients to Amplex A&B compared to control patients showed no statistical difference ($p > 0.05$) in all the parameters including Hb, MCV, CR, WBC and Platelet Count. Table 11 and Figure 1 showed a slight increase in the level of the mean Hb and CR from pre-therapy when compared to 14 days after commencement of therapy. Also when compared to control subjects there was a slight decrease in the mean WBC count of test patients from pre-therapy to 14 days post-therapy.

Despite this increase in Hb, CR and a reduction of MCV there was no statistical significance when compared to control subjects. This could be explained by the fact that sickle cell anaemia is a genetic disorder with impaired Haemopoietic production, therefore response may not be as significant as expected in other patients (HbAA) with cobalamin deficiency.

Over 50% of the test patients reported an increase in appetite as a positive side effect while less than 1% complained of mild bone pains. The increase in appetite may be the general effect of vitamin B complex. The increase in Hb leads to an increase in blood viscosity which may be responsible for cellular dehydration and VOC. These mild bone pains has also demonstrated that Amplex A&B have no anti-sickling properties.

Clinicians may never consider cobalamin deficiency as a possible cause of anaemia in adults and children with SCA.¹³ Although diagnosis is difficult but it has been documented to be safe even at 10,000 times the normal dosage of cobalamin

and therefore safe to give cobalamin to patients suspected to have deficiency without facilities for diagnosis.¹⁴

Unnecessary folate supplements for SCA patients could hamper the recognition of neuropsychiatric complications of cobalamin deficiency and some authors have reported SCA patients with unsuspected pernicious anaemia developing neuropsychiatric symptoms while receiving folic acid prophylaxis.¹⁵

CONCLUSION

The result of this preliminary therapeutic trial with Ample A&B has revealed a non-statistically significant increase in some laboratory parameters in sickle cell anaemia patients such as haemoglobin, and reticulocyte count. While there was a reduction in the post-therapy values of MCV and white cell count.

We recommend the routine estimation full blood count with red cell indices in sickle cell anaemia patients, especially in those with neuropsychiatric symptoms.

There is also need to ascertain the long term benefits of cobalamin and combination therapy in sickle cell anaemia.

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