

## Thromboembolic events in $\beta$ -Thalassemia major patients

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### Summary:

**Background:** Seventy eight patients with beta - thalassemia major were Collected randomly from the thalassemia center in Ibn-Al-Baldy Hospital (all of them were transfusion dependent), together with fifty six age and sex matched healthy children were collected as a control.

**Aim of the study:** To determine the incidence of thromboembolism among them and to evaluate the precipitating factors.

**Patients and Methods:** History was taken and physical examination was done . EDTA Anticoagulated blood samples were taken prior to the next transfusion , platelet parameters were estimated for both groups using the MS – 9 coulter counter .

**Results:** Thromboembolic events was detected in five patients (6.41%).The main site was the central nervous system in 4 of them ( 5.2%) followed by the lung ,1 case ( 1.2%). Associated predisposing factors were found in 3 of them (60%).The patients had a statistically significant higher platelet counts, platelet crit, mean platelet volume and platelet distribution width than the control group. The splenectomized had higher platelet counts ,platelet crit than the non splenectomized with nearly equal mean platelet volume and platelet distribution width , a results that is in favour of thrombosis.

**Conclusions:** This results suggest that beta- thalassemia patients had a hypercoagulable state which makes them susceptible to thrombosis and pulmonary embolism even in the young age group and even before splenectomy is performed.

**Key words:** Thalassemia , Thromboembolism

### Introduction:

Thalassemia is a congenital hemolytic disorder caused by partial or complete deficiency of  $\alpha$  or  $\beta$  globin chain synthesis<sup>(1)</sup>. Homozygous suffer from severe anaemia and other serious complications due to iron overload from early childhood<sup>(1,2)</sup>. Vascular complications such as pulmonary embolism , transient ischemic attack (TIA) and strokes have been reported among them<sup>(3)</sup>. The platelet counts are slightly elevated in thalassemia unless there is secondary hypersplenism<sup>(4)</sup>. The mean platelet volume (MPV) measures platelet size ,is increased with increased platelet function and activation<sup>(5)</sup>. It is increased in patients with thalassemia major , thalassemia minor and sickle cell anaemia<sup>(6)</sup>, it has been proposed as a clinically risk factor for thromboembolic diseases<sup>(7)</sup>. platelet distribution width (PDW) represent platelet anisocytosis<sup>(8)</sup>. It indicates platelet heterogeneity<sup>(9)</sup>, this platelet heterogeneity may be involved in pulmonary thrombosis<sup>(1,10,11)</sup>.

### **Patients and Methods:**

Seventy-eight patients (37 females, 41 males) with thalassemia major were collected randomly from the thalassemia center in Ibn-Al-Baldy hospital. All of them were transfusion dependent, admitted to the center for blood transfusion. History was taken and physical examination was performed, blood Samples were collected by veni-puncture just prior to the next transfusion using EDTA anticoagulant. Complete blood picture was obtained within 2 -4 hours using automated full digital cell counter (MS-9, Melet, schloesing laboratory/ Cergy Pontoise, France) with standard calibration and quality control), the control of this instrument was available at the time of samples collection, in addition internal quality control was done for the machine every two months)/ double checking method. The electronic counter measures the platelet counts, platelet crit, mean platelet volume and platelet distribution width with a normal range value given by the manufacturer<sup>(12)</sup>.

Fifty-six healthy children, age and sex matched were also collected as a control and for them the same platelet parameters were estimated and compared with reference range developed for normal subject<sup>(13,14)</sup>.

Mean, standard deviation were estimated , statistical analysis was done using the student t-test through Microsoft Excel program , p value < 0.05 is considered as statistically significant.

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**Results:**

The age of the patients collected range from (5-21years), with a mean and standard deviation of (13.93  $\pm$  4.71) all of them were on chronic transfusion regimen, the age of the control range from (7-17years) with a mean and standard deviation of (11.91 $\pm$ 2.8).

Of the studied patients, five of them (6.41%) developed thromboembolic events, the main site was the central nervous system in four of them (5.2%) with a variation in the clinical presentation as it is shown in ( table- 1) . The remaining one patient, (1.2%) got sudden chest pain, shortness of breath, death within few hours, suggestive of pulmonary embolism which happened after the use of contraceptive pills ( table -1). All of the affected patients are irregularly transfused either because of large family size with more than one member of the family being affected with thalassemia major and poor financial support or because of non availability of the blood when needed .

The platelet count was higher in the patients then the control with a range of ( 62 – 959  $\times 10^9/L$ ) ( 154 – 400  $\times 10^9/L$ ) respectively and a mean of (306.808 $\pm$ 201.231 $\times 10^9/L$ ), (241.19  $\pm$  66.38  $\times 10^9/L$ ) respectively,being statistically highly significant ,p <0.01 as shown in (table-2).

The platelet crit is significantly higher in the patients then the control, with a range of( 0.1 -1%),( 0.1 – 0.3%) respectively and a mean of (0.34  $\pm$  0.45%),(0.2 $\pm$ 0.08%) respectively, p< 0.01.(table-2).

The patients had a significantly higher mean platelet volume than the control, with a range of (7.4 -12.7 fl) , ( 7.9 – 10 fl) and a mean of (10.05 $\pm$ 1.75 fl),(8.15 $\pm$ 0.65 fl) respectively which is statistically highly significant, p.<0.01, (table-2).

The patients had a significantly higher platelet distribution width then the control with a rang of ( 8.1 – 11.2 ) ,( 7.5 – 9.9 ) respectively and a mean of (9.81  $\pm$  0.83),(8.51  $\pm$  0.78) respectively which is statistically highly significant, p.<0.01 (table-2).

Of the 78 thalassemic patients, 56 ( 71.80%) Cases were non splenectomized, and 22( 28.20%) underwent splenectomy.

The mean age of the splenectomized was (14. 27 $\pm$ 5.67),and the mean age of the non splenectomized was (13.80 $\pm$  4.33) .

The splenectomized had significantly higher platelet count then the non splenectomized with a mean of (475.86  $\pm$  273.59),(240.39  $\pm$  110.39) respectively, p. <0.01 as shown in (table-3).

The splenectomized had a higher platelet crit then the non splenectomized with a mean of (0.55  $\pm$  0.79),(0.26  $\pm$  0.14) respectively which is statistically significant, p<0.05, (table-3).

The MPV is nearly equal , in the splenectomized group with a mean of (9.90  $\pm$  2.80), and (10.10 $\pm$ 1.14) for the non splenectomized, statistically not significant p>0.05 (table-3).

The PDW is nearly equal in the splenectomized and the non splenectomized, with a mean of (9.6  $\pm$  0.90), (9.89  $\pm$  0.80) respectively, the result is statistically not significant, p>0.05. (Table-3).

Table – 1- demographic characters and clinical presentation in patients with central nervous system and pulmonary thromboembolism

Clinical presentation	Age/ year	Sex	Associated predisposing factors
TIA followed after 2 months by Headache , Seizure , Hemiplegia	5	male	/
Sudden Hemiplegia	7	male	/
Sudden Hemiplegia	12	female	Splenectomy
Hemiplegia , Seizure	15	male	Diabetes Mellitis
Sudden chest pain shortness of breath ,death	16	female	Contraceptive pills

Table –2- mean , standard deviation of platelet parameters in the patients and the control

Platelet parameters	Control	Patients	P -value
Platelet count $\times 10^9/L$	241.19 $\pm$ 66.38	306.808 $\pm$ 201.231	*** 0.004272
Platelet crit %	0.2 $\pm$ 0.0809	0.34872 $\pm$ 0.4509	*** 0.002752
MPV fl	8.15 $\pm$ 0.65	10.0513 $\pm$ 1.75741	*** 0.0000

PDW	8.51 ± 0.78	9.81282 ± 0.8378	*** 0.0000
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\* p-value > 0.05 not significant

\*\* p-value 0.01 - 0.05 significant

\*\*\* p-value < 0.01 highly significant

Table-3-

mean, standard deviations of Platelet parameters in the splenectomized and non splenectomized patients

Platelet parameters	Splenectomized	Non splenectomized	p-value
Platelet count x 10 <sup>9</sup> / L	475.86 ± 273.59	240.39 ± 110.39	***0.00033
Platelet crit %	0.55 ± 0.79	0.26 ± 0.14	**0.0496
MPV fl	9.90 ± 2.80	10.10 ± 1.14	*0.37168
PDW	9.6 ± 0.90	9.89 ± 0.80	*0.09421

\* p-value > 0.05 not significant

\*\* p-value 0.01- 0.05 significant

\*\*\* p-value < 0.01 highly Significant

### Discussion:

The overall incidence of thromboembolic events in this study was ( 6.41%) which is slightly higher than Italian studies which showed an incidence of ( 1.1%) (Borgna – Pignatti et al)<sup>(15)</sup>, ( 3.95%) (Moratelli S et al)<sup>(16)</sup>, ( 5.3%) (Borgna – Pignatti et al)<sup>(17)</sup>.

The central nervous system events was reported in ( 5.2%) , in comparison with a study done in Greece , stroke syndrome was reported in ( 2) cases out of (138) and a neurological deficit comparable with transient ischemic attack was reported in ( 20%) of cases (Logothetis J et al)<sup>(18)</sup> . An Italian study reported cerebral thromboembolic events accompanied by a clinical picture of headache , seizure and hemiparesis in (16) individuals with  $\beta$  - thalassemia major out of (735) patients and pulmonary embolism in ( 3) out of (735) patients(Borgna – Pignatti et al)<sup>(15)</sup>.

The abnormalities in the platelet parameters noticed in this study is in agreement of Abbas study<sup>(19)</sup> as well as Bunyaratvej A. study<sup>(10)</sup> and Hathirat P. et al study<sup>(20)</sup>. This high platelet counts is in favour of thrombosis and may lead to pulmonary artery disease due to platelet aggregation in the pulmonary circulation<sup>(4,21)</sup>. In addition the increased mean platelet volume is considered as a risk factor for thrombosis because it's level is related in a complex way to thrombopoietic stimulation<sup>(22)</sup> . High platelet distribution width indicates platelet heterogeneity , it's value is linked to platelet count and mean platelet volume<sup>(23)</sup> , this heterogeneity may be involved in pulmonary thrombosis<sup>(1,10,11)</sup> . These finding suggest platelet

activation and indicates that a hypercoagulable state already exist in  $\beta$  - thalassemia major since early childhood even in the absence of overt thromboembolism<sup>(21)</sup> .

Associated predisposing risk factors was found in ( 3) cases ( 60%) compared with ( 15.3%) (Moratelli S et al)<sup>(16)</sup> up to ( 50%)(Borgna – Pignatti et al)<sup>(15)</sup>.

Of the risk factors encountered was diabetes mellitus , it is known that platelet from diabetic individual are hyperactive, and as a consequence of platelet activation the outer layer of it's phospholipid membrane is more procoagulant stimulating thrombin formation<sup>(24)</sup> .In addition the mean platelet volume is increased in diabetes mellitus<sup>(25)</sup> . Contraceptive pills as a hormonal therapy was other risk factor, it was proved that an increase in the platelet counts and mean platelet volume occur within 6 weeks of hormonal therapy , this increase in the mean platelet volume indicates platelet reactivity<sup>(26)</sup> . Splenectomy as a risk factor is followed by increase in the platelet counts and mean platelet volume and it leads to morphological platelet abnormalities that may contribute to enhanced risk of vascular complications<sup>(27)</sup> .

Affected patients with thromboembolism were irregularly transfused , this irregular transfusion enhanced thromboembolism because the number of circulating damaged red blood cells and the number of platelets will be more<sup>(28)</sup> , these damaged red blood cells were demonstrated to facilitate thrombin formation due to abnormalities involving the membrane phospholipid<sup>(29)</sup> a fact

that make thromboembolic manifestations more frequently recorded in less developed countries with limited transfusion resource<sup>(28)</sup>, so regular transfusion can reduce thromboembolic events by eliminating the abnormal aggregate observed with thalassaemic red cells<sup>(28)</sup>. In addition many of those patients may have abnormal liver function and possibly reduced protien C and protien S which are considered as a contributing factors for thrombosis<sup>(16,17,23,30,31)</sup>.

Close monitoring of thalassaemic patients who are at risk of developing thromboembolic events is recommended as well as frequent measurements of platelet parameters are indicated since early childhood and even before doing splenectomy. Aspirin prophylaxis is recommended specially for those with splenectomy to prevent thrombosis.

### Conclusion:

Increased platelet numbers, mean platelet volume, platelet heterogeneity (PDW) are involved in thrombotic events.

Recurrent thromboembolism may occur involving mainly the central nervous system followed by the lung.

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