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Bleeding Events and Factor VIII levels in Obligate Female Carriers of Haemophilia A

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Abstract

Background: Haemophilia A (also known as classical haemophilia) results from heterogenous mutations in factor VIII gene which is located on long arm of X –chromosome (Xq28). Haemophilia A being X linked disorder affects males while females are mostly carriers of the disease. An obligate carrier is the daughter of an affected male (inheriting the X linked mutation from father) or the one who has more than one affected son or one affected son and one or more affected relatives in the family. A carrier can have low factor levels because of X-inactivation or Lyonization (if the X chromosome with the Haemophilia gene is the active chromosome) Being a carrier with low factor levels many aspects of life of the female get affected including her own health, risk of prolonged bleeding, mennorhagia & postpartum bleeding. It is therefore extremely important for a carrier to know her factor levels.

The aim of study was to measure factor VIII levels and bleeding events of obligate female carriers of Haemophilia A compared with controls.

Objective: To measure factor VIII levels in obligate female carriers of haemophilia A compared with controls.

Materials and Methods: It was a cross sectional comparative study carried out in department of Pathology, Army Medical College, Rawalpindi in collaboration with Armed Forces Institute of Pathology (AFIP), Rawalpindi and Haemophilia Treatment Centre of Pakistan Haemophilia Patients Welfare Society, Rawalpindi-Islamabad. The study was carried in one year from November 2016 to November 2017.Female carriers of all ages were included in study except those who were pregnant or using oral contraceptives or doing aerobic exercises or having chronic inflammation.

Results: A total of 50 subjects were included in the study divided into two groups i.e., obligate female carriers and healthy control females. Out of 50 subjects studied 30 were obligate female carriers and 20 healthy females were included as controls. Mean age of obligate carriers vs controls was statistically insignificant. Factor VIII levels were lower in obligate carriers than in healthy females. Levels observed were statistically significant in obligate carriers vs controls.

Carriers also reported a higher number of bleeding events when compared to controls. Statistically significant difference was observed between carriers and controls.

Conclusion: We studied factor VIII levels and bleeding episodes faced by obligate female carriers. Significantly lower Factor VIII levels and higher bleeding episodes were reported in obligate carriers compared with controls. These carriers with low Factor VIII levels are at risk of increased bleeding events therefore require attention and follow up.

Keywords: Events; Haemophilia A; Factor VIII Levels; Obligate Female Carriers

Introduction

Haemophilia is the commonest coagulation disorder classified as type A and B depending upon absence or deficiency of plasma clotting factors VIII and IX respectively [1]. World-wide incidence of haemophilia A is 1 in 5000 live male births [2]. Prevalence of haemophilia in Pakistan is 1.6/100,000 male population [3].

Haemophilia A is an X-linked recessive bleeding disorder resulting from mutations in factor VIII gene [4]. The disease affects males as they have single X chromosome [5]. Females can be affected if they inherit factor VIII gene mutations from both parents, but mostly females are carriers of the disease [6]. They can be obligate or possible carriers [7]. Plasma concentration of factor VIII levels in female carriers are expected to be half of the normal (50-150% of the normal) [8]. Male hemophiliacs present at an early age with prolonged bleeding after trauma or surgery however female carriers usually remain asymptomatic but can present with menorrhagia, postsurgical bleeding, bleeding after dental extraction, postpartum hemorrhage, haematuria, epistaxis and bruising during their reproductive years [8]. Most serious issue is the risk of having a haemophiliac baby which poses a serious emotional and financial burden on family as well as on the society. These complications raise the need for awareness of a female to know about her carrier status and her Factor levels. This can help prevent complications & can reduce all such burdens.

We planned this study to define extent of bleeding symptoms in obligate female carriers vs controls and measure factor VIII levels of obligate female carriers of haemophilia A compared with controls.

Materials and Methods

It was a cross sectional comparative study carried out in department of Pathology, Army Medical College, Rawalpindi in collaboration with Armed Forces Institute of Pathology (AFIP), Rawalpindi and Haemophilia Treatment Centre of Pakistan Haemophilia Patients Welfare Society, Rawalpindi-Islamabad. The study was carried out in one year from November 2016 to November 2017. Permissions from Ethical Review Committee (ERC), Institutional Review Board (IRB) and hemophilia center were taken.

Operational Definition

For the purpose of this study, obligate female carriers were defined as daughters of hemophiliacs and women who had more than one affected son or one affected son and one or more of affected relatives in family line.

Inclusion and Exclusion Criteria

Female carriers of all ages were included except those who were pregnant or using oral contraceptives or doing aerobic exercises or having chronic inflammation.

Study Subjects were Stratified into Two Groups

Group 1 included obligate female carriers meeting inclusion and exclusion criterias and Group 2 comprised of healthy females, who served as controls.

Sample Size

It was calculated by WHO calculator and was thirty (30). Informed written consent was obtained from all study subjects after explaining the procedural aspects and aim of the study. Demographic data and relevant history were noted on a proforma (Annex A). Presence of clinical signs and symptoms and severity of bleeding episodes were evaluated using ISTH-BAT Scoring system (0, 1, 2 3, 4 as described by ISTH-BAT [9]). Bleeding episodes were classified as trivial, minor and major bleeding [10].

Sample Collection

Two (2) ml of blood was extracted after informed consent from each healthy control and female carrier, and transferred to a vial containing 3.2% sodium citrate for plasma factor VIII analysis. Factor VIII assays were performed at AFIP on Sysmex CA 1500 [11] (automated coagulation analyzer) after running routine quality control procedures. Control and carrier's plasma VIII assays were entered on Annex B.

Data Analysis

The data was analyzed on Statistical Package for the Social Sciences (SPSS) version 22. For qualitative variables, frequency and percentages were calculated. For quantitative variables, mean, standard deviation and 25th, 50th and 75th percentiles were calculated. Qualitative parameters were compared among the groups using the chi-square test. A two-tailed P-value < 0.05 was considered statistically significant. Independent T test was used for comparison between two groups (carriers and controls). One way ANOVA was applied for describing clinical features.

Results

Out of 50 subjects studied, 30 were obligate female carriers and 20 females were controls. Age range of female carriers was 7 to 55 years (mean = 27.2+14.07) and for controls it was 9 to 51 years (mean = 27.00+11.8) showing no statistically significant difference (p=0.95). Among female carriers studied 21 (70%) were married and 9 (30%) were unmarried. Thirteen females (65%) were married and seven (35%) were unmarried among control group. Statistically significant difference was not observed between two groups regarding marital status (p=0.56).

Out of 30 carriers studied, 21 (70%) were mothers of haemophiliac sons, 9 (30%) were daughters of haemophiliac father. Bleeding events were found statistically significant in obligate female carriers (p=0.002). Frequent bleeding episodes faced by carriers were menorrhagia 11 (37%), gum bleeding 3 (10%) and epistaxis 4 (13%), postpartum hemorrhage 3 (10%) and easy bruising 2 (6%). Seven (24%) carriers were asymptomatic.

Bleeders and Bleeding Score

Among carriers, 18 (60%) females were defined as minor bleeders, 5 (16%) females were classified as major bleeders, 7 (24%) females were found to have trivial (no) bleeding. (Table 1).

Bleeders	Carrier females
	(n=30) (n (%))
Major	5 (16%)
Minor	18 (60%)
Trivial (no) bleeding	7 (24%)

Table 1: Showing bleeding status.

Seven (24%) carriers of haemophilia had a bleeding score 0, eight (27%) had bleeding score 1, four (14%) were with a score of 2, three (10%) hemophilia carriers were assigned score 3, three (10%) carriers had a score of 4. Two (6%) carriers had a score of 5, two (6%) had score of 6 and one (3%) had a score of 7.

Factor VIII levels of female obligate carriers were assayed and compared with those of control females. These were lower in carriers than in non-carriers. These values ranged from 26.13% to 140% of the normal (mean= 85.95+33.95). Mothers of hemophiliacs had values ranging from 26.1% to 140% of normal (mean=87.6+33.5). Daughters of hemophiliac fathers had mean level 83+37.8 with a minimum level of 33% of the normal and maximum level of 135% of the normal. Factor VIII levels in mothers and daughters of hemophiliacs were statistically not significant (p=0.09). Out of 30 carriers, only 3 had factor FVIII levels below 50% of the normal. Of these 3 females, 2 carriers were unmarried with FVIII levels 33% & 37% of the normal and gave only history of epistaxis. Third carrier female had FVIII level 26.1% of the normal presented with history of menorrhagia and postpartum hemorrhage. Clinical symptoms observed by carrier females and FVIII levels are shown in table 2.

Bleeding events	Female carriers (n=30)	FVIII levels in obligate female carriers	
		range (%of the normal)	Mean+SD
Menorrhagia	11	26-90	64.24+17.70
Postpartum hemorrhage	3	33-72	52.86+19.80
Gum bleeding	3	37-135	87.30+49.05
Epistaxis	4	91-140	110.5+20.92
Bruising	2	76-130	103+38.1
Asymptomatic	7	69-140	114.77+27.83

Table 2: Bleeding events and FVIII levels in obligate female carriers.

Factor VIII levels of control females had a minimum level 100% of the normal and maximum level of 237% of the normal (mean =157.65+44.3). Statistically significant lower levels were observed in carriers (p=0.000).

Discussion

Carriers of Haemophilia A are expected to be at an increased risk of bleeding and have almost 50% lower levels of factor VIII compared to healthy females, non-carriers of the disease. Bleeding has always been a problematic experience both for patients and health care providers. We did this study in order to evaluate bleeding complications and correlate this with factor VIII levels in carriers of haemophilia A presenting to haemophilia treatment center. Appreciation of bleeding events and complications allows us better understanding of health quality of haemophilia carriers.

In our study, mean age of female carriers was 27 years in contrast to [12] and [13] where mean age was 40 years and 13.2 years respectively. Status of relationship of obligate female carrier in our study was mothers 70% and daughters 30%, compared to [12] where mothers were 52%, daughters 12% and sisters 36%.

Bleeding events were reported and considered significant according to ISTH-BAT. We identified an increase in various bleeding events including menorrhagia, gum bleeds, epistaxis, easy bruising and postpartum bleeding in haemophilia carriers.

Out of female carriers inquired about bleeding events, 76% of females were experiencing increased bleeding episodes whereas 24% females remained asymptomatic. Most common clinical feature observed was menorrhagia, which affected their daily and social activities. Carriers also reported increased severity of bleeding and use of iron, tranexemic acid and other multivitamins to avoid the disturbance caused by menorrhagia. Gum bleeding, epistaxis and easy bruising were also frequent and reported by female carriers. History of postpartum hemorrhage was also given by these female carriers. Study conducted by Hussain et al., 2017, also showed heavy menstrual bleeding, gum bleeding and epistaxis as most common presenting features in females with inherited bleeding disorders [14]. Another study by Burhany et al., 2015 reported that menorrhagia and bruising were most troublesome features in carrier females but contrary to our study majority of carriers were mostly asymptomatic in that study [12]. Paroskie et al., 2015 discussed various clinical features of haemophiliac carriers. Comparable to our study, menorrhagia was also major bleeding event faced by these carrier females [15]. Another study by Plug et al., 2006 reported heavy menstrual bleeding in 57% of the cases[8]. Seven (24%) carriers had a bleeding score 0, 27% had bleeding score 1,14% with score 2, 10% had a score of 3, 10% had a bleeding score of 4, 6% with score of 5, 6% had a score of 6 and 3% had a score of 7 in contrast to [12] where bleeding score was 38%,10%,17%,12%,2.4%,2.4% and 7% respectively, whereas [12] also reported a bleeding score of 8 and 11 in 2.4% haemophilia carriers.

Mean value of FVIII in our female carriers was 85% with controls having mean FVIII level 157%. Factor VIII levels reported by Paroskie et al., 2015 were 82.5%, comparable to our study. FVIII levels reported by [8], [12], [16] and [17] were 60, 74, 90 and 55, in carrier females compared with controls. Surprisingly, in our study, 3 carrier females had FVIII levels below 50% of the normal, 2 were unmarried presented with epistaxis off and on with FVIII levels 33% and 37% but third female was married and reported with increased bleeding events with FVIII level 26.1%. Similarly, a case subject having FVIII:C 37% with increased bleeding symptoms was also reported by [15].

Conclusion

This study showed various problems faced by obligate female carriers of haemophilia A regarding bleeding issues and issues affecting their quality of life. Bleeding events experienced were significantly higher and factor VIII levels were significantly lower in obligate female haemophilia carriers. So, we conclude that obligate female carriers experience varying degrees of bleeding events despite having normal FVIII levels. Only a few of them had low factor VIII levels and majority showed bleeding tendency even with normal factor VIII levels.

Recommendations

A comprehensive study regarding genetic makeup, bleeding phenotypes and functional activity of FVIII in a larger cohort is needed for further evaluation. Our findings underline importance for carriers and health care providers to be aware of complications and problems affecting their life. Obligate female carriers require a special attention. Genetic counseling, access to information about reproductive choices and improvement in quality of life needs to be addressed. Public awareness programs need to be tailored effectively to cater for the needs of these carriers as they constitute a sizeable proportion of our society.

References

1. Keeney S, Mitchell M, Goodeve A (2005) The molecular analysis of haemophilia A: a guideline from the UK haemophilia centre doctors' organization haemophilia genetics laboratory network. Haemophilia 11: 387-397.
2. Rossetti LC, Radic CP, Larripa IB, De Brasi CD (2008) Developing a new generation of tests for genotyping hemophilia□causative rearrangements involving int22h and int1h hotspots in the factor VIII gene. Journal of Thrombosis and Haemostasis 6: 830-836.
3. Stonebraker JS, Bolton□Maggs PH, Soucie JM, Walker I, Brooker M (2010) A study of variations in the reported haemophilia A prevalence around the world. Haemophilia 16: 20-32.
4. Jayandharan G, Shaji RV, Baidya S, Nair SC, Chandy M, et al. (2005) Identification of factor VIII gene mutations in 101 patients with haemophilia A: mutation analysis by inversion screening and multiplex PCR and CSGE and molecular modelling of 10 novel missense substitutions. Haemophilia 11: 481-491.
5. Dobyns WB (2006) The pattern of inheritance of X□linked traits is not dominant or recessive, just X□linked. Acta Paediatrica 95: 11-15.
6. Sami W, Clinical manifestations and complications of haemophilia A in Pakistan. Ann. P
7. Habart D (2005) Molecular diagnosis of haemophilia A in clinical practice. Casopis lekaru ceskych 144: 795-800.
8. Plug I, Mauser-Bunschoten EP, Bröcker-Vriends AH, van Amstel HK, van der Bom JG, et al. (2006) Bleeding in carriers of hemophilia. Blood 108: 52-56.
9. Rodeghiero F, Tosetto A, Abshire T, Arnold DM, Coller B, et al. (2010) ISTH/SSC bleeding assessment tool: a standardized questionnaire and a proposal for a new bleeding score for inherited bleeding disorders. Journal of Thrombosis and Haemostasis 8 :2063-2065.
10. Tosetto A, Castaman G, Rodeghiero F (2013) Bleeders, bleeding rates, and bleeding score. Journal of Thrombosis and Haemostasis 11: 142-150.
11. Lee YW, Chang CW, Lim MS, Lim BJ, Lee YK (2001) Laboratory Evaluation of Automated Coagulation Analyzers Sysmex CA-1500 (TM) and CA-7000 (TM). Journal of Clinical Pathology and Quality Control 23: 253-258.
12. Ay C, Thom K, Abu□Hamdeh F, Horvath B, Quehenberger P, et al. (2010) Determinants of factor VIII plasma levels in carriers of haemophilia A and in control women. Haemophilia 16 :111-117.

13. Borhany M, Shamsi T, Naz A, Khan A, Parveen K, et al. (2011) Congenital bleeding disorders in Karachi, Pakistan. Clinical and Applied Thrombosis/Hemostasis. 17: E131-137.

14. Husain N (2009) Carrier analysis for hemophilia A: ideal versus acceptable. Expert review of molecular diagnostics 9: 203-207.

15. Paroskie A, Gailani D, DeBaun MR, Sidonio RF (2015) A cross□sectional study of bleeding phenotype in haemophilia A carriers. British journal of haematology 170: 223-228.

16. Morange PE, Tregouet DA, Frere C, Saut N, Pellegrina L, et al. (2005) Biological and genetic factors influencing plasma factor VIII levels in a healthy family population: results from the Stanislas cohort. British journal of haematology 128: 91-99.

17. Graham JB, Rizza CR, Chediak J, Mannucci PM, Briet E, et al. (1986) Carrier detection in hemophilia A: a cooperative international study. I. The carrier phenotype. Blood 67: 1554-1559.