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A study in respect of transfusion related complications in hemophiliacs with special reference to developing factor inhibitors

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ABSTRACT

Prophylaxis and treatment with factor replacement have greatly improved the quality of care for patients with hemophilia.However, development of factor inhibitors is the most serious and challenging complication of therapy. Other complications are viral infections like Hepatitis B, Hepatitis C, and HIV, and the other infective diseases, which can be transmitted by the blood and blood products transfusion.

The aims of this study is to study the complications in the hemophiliacs who have been treated prophylactically or 'on demand' with fresh frozen plasma, cryoprecipitate and concentrated products of FVIII and

During the study period, all patients (100) with Hemophilia attending Gandhi Medical College are taken under consideration. The presence of an inhibitor was determined by a simple mixing experiment using the test plasma and normal pooled plasma and 3rd generation enzyme linked immunosorbent assay (ELISA) method& anti-HBsAg.statistical

The inhibitor study showed that in 7%(3.2-13.98 at 95% confidence interval) patients, APTT was not corrected after mixing patients plasma with pooled normal plasma (PNP) and applying the test immediately and after one hour of incubation. serological tests showed antibodies for HCV were positive in 4% of cases (1.1%-9.93% at 95% CI), whereas antibodies against HBsAg was positive in 1% of cases(0.3%-5.45% at 95% CI), which was less then anti HCV. Transfusion associated complications were higher in severe form of Haemophilia as compared to moderate and mild form of Hemophilia.

Developing antibodies to infused factor concentrates (inhibitors) remains a major source of morbidity and mortality in the treatment of patients with hemophilia. Novel treatment approaches for these patients are in developmental stage, which include therapeutic agents that mimic factor VIII or augment thrombin production by bypassing the inhibitors, as well as agents that act by inhibiting the natural anticoagulants.

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1. Introduction

Prophylaxis and treatment with factor replacement have greatly improved the quality of life in patients with hemophilia.Though, development of factor inhibitors is the most serious and challenging complication of therapy. Other complications are viral infections like Hepatitis B, Hepatitis C, HIV and other infective diseases, which can be transmitted by the blood and blood products transfusion.

10%-20% of patients with hemophilia A will develop inhibitors and 3% of patients with hemophilia B.[1–3] Development of inhibitors increase the cost of care significantly, increase the financial burden and psychosocial stress on patients and their families as well as cause negative impact on disease morbidity and mortality by making therapy less effective.[4,5] In spite of development of various new generation therapeutic products that treat bleeding episodes patients without inhibitors, bypassing agents (BPAs) are the advanced treatment modalities for patients with inhibitors. Therefore, novel approaches that address bleeding in the setting of antibodies against clotting factors are an active area of investigation. Other serious complications related to treatment of haemophilia are transfusion related infections like viral hepatitis B (HBV),[6] viral hepatitis C (HCV),[7]

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HIV etc. The cause of transmission of these infections can be understood by the fact that concentrating coagulation factors are prepared of plasma from thousands of blood donors that didn't undergo viral inactivation.[8–10] Majority of infected patients do not suffer from acute illness and clear the infection spontaneously, though some patients (<50%) become chronic carriers of virus[11] and latter may develop into chronic active hepatitis and finally progress to liver cirrhosis; especially in HBV and HCV infections.[12–14] Because hamophilics received multiple transfusions of the factor VIII and IX concentrates prepared from pool plasma of thousands of donations as well as FFP and cryoprecipitate so there are risk of acquiring Transfusion Transmitted Infection. Therefore, this paper is focused on the above mentioned complications of the patients with hemophilia associated with transfusion of blood components or derivatives.

2. Materials and Methods

This hospital based observational study was conducted on 100 patients with Hemophilia in Gandhi Medical College and associated Hamidia hospital Bhopal since 1^{st} of March 2017 to 30^{th} of June 2018. During the study period, all patients with Hemophilia attending Gandhi Medical College & associated Hamidia hospital are taken under consideration. Patients having other bleeding disorders or on medication affecting coagulation profile were excluded from study. After taking written consent, Patients with hemophilia were subjected for a thorough clinical history including family history and history regarding prophylaxis or therapy of Hemophilia. Blood sample were collected in EDTA as well as in citrated tubes. Complete blood count was done with Mindray BC-5300 hematology analyserand peripheral smear examination was also done. Sample containing Sodium citrate anticoagulant was subjected for coagulation profile done manually (Manual Tilt- tube technique)infectious agents.

The presence of an inhibitor can be determined by a simple mixing experiment using the test plasma and normal pooled plasma. Inhibitors against coagulation factors which influence the APTT are of two type; immediate acting and time dependent. Test plasma which contain an immediate acting inhibitor, when mixed with normal plasma, reveal very small or no correction of the clotting time while time dependent inhibitors require a period of incubation with normal plasma before they can be detected. Both, normal plasma as well as test plasma were incubated at 37°C for 1 hour, separately and as a mixture of 50:50. Then APTT is measured on the normal plasma, test plasma, incubated mixture and a mixture prepared from equal volumes of test and normal plasma after separate incubation. The value of correction of the APTT of each mixture is compared. Poor correction in: (a) The mixture prepared after separate incubation is suggestive of an immediate acting inhibitor. (b) In one hour incubation mixture is suggestive of a time dependent inhibitor.

3. Observations

Most common type was Hemophilia A i.e. 89% (82.21%-93.91% at 95% confidence interval). Most common age group of patients was between 6-15 years which was 49% (39.42%-58.65% at 95% confidence interval) and mean age of patients was 19.02 ± 12.58 years with a age range of 1-65 years.

Regarding age of onset of first clinical manifestation, majority of patients was below 1 year. (41.35%-60.58% at 95% confidence interval) and a mean was 2.44 ± 2.77 years with a range of 1 month to 14 years. 57% patients had positive family history of hemophilia (47.21%-62.27% at 95% confidence interval) showing correlation of its X- linked recessive nature. Most common clinical manifestation was hemarthrosis which was 72% (62.48%-79.90% at 95% confidence interval). 52% patients presented with knee joint heamarthrosis (56.78%-76.61% at 95% confidence interval)

Involvement of various joint in the study population was:
Knee > Elbow > Ankle > Hip = Shoulder > Wrist

All the patients showed raised APTT and 57% had APTT more than 80 seconds (47.21%-66.27% at 95% confidence interval).

Patients with severe hemophilia usually had APTT above 80 seconds and

96.2% patients with spontaneous bleeding episodes have APTT range >80 secs.

Severe haemophilia was the most common type of hemophilia i.e 52% (42.32%-61.54% at 95% confidence interval).

Inhibitors were present in 7% of haemophilia patients (3.2% - 13.98% at 95% confidence interval).

Screening for TTI is done in all haemophiliac patients as a part of routine practice because of high risk of these infection.

Hepatitis C was more prevalent than Hepatitis B in Hemophiliac patients.

4. Discussion

In present study in 7 out of 100 (7%) patients, APTT was not corrected after incubating with 1:1 pooled normal plasma (PNP) These patients had history of recurrent bleeding and had received multiple factor VIII transfusions. Prevalence of factor inhibitor is seen in study done by R.K. Nigam[15] et al (9.09%) and Payvandi[16] et al (30.3%). Several hypothesis have been put forward to explain these difference in prevalence rate. These include:

1. The distinction between controlled clinical studied and pharmacovigilance studies. In these pharmacovigilance studies the inhibitor assay were performed at

Table 1: Agewise distribution of hemophiliac patients

Age(In years)	Hemophilia A	Hemophilia B	Total	Percentage
0-5	4	0	4	4%
6-15	45	4	49	49%
16-30	26	4	30	30%
>30	14	3	17	17%
Total	89	11	100	100%

Table 2: Distribution of presence of factor inhibitors

Type of haemophilia	Inhibitor present	Inhibitor absent	Total	Percentage
Hemophilia A	07	82	89	89%
Hemophilia B	00	11	11	11%
Total	07	93	100	100%

Table 3: Hepatitis B and Hepatitis C Positivity in Hemophilia

Hepatitis type	Number of patients
Hepatitis B	01(1%)
Hepatitis C	04 (4%)
HIV	00(00%)
Total	**05(05%)**

Fig. 1: Hemophilia patients with multiple joints Heamarthrosis

6-12 month interval and hence transient or low factor inhibitors can be missed.

2. The inhibitors were known to disappear spontaneously.
3. The residual and contaminating proteins in the intermediate purity may have immune modulatory effect on immune system of the recipient thus reducing the likelihood of inhibitor formation.
4. The intermediate purity products contain well preserved Von Willebrand factor multimers that may block epitopes on the light chain of the factor VIII molecule to which the most of the allo-antibodies to factor VIII react.
5. The possible difference between type of mutation between various patient populations. Severe hamophilia caused by mutations associated with a major loss of coding information such as large

deletions, inversions and mutation which results in premature translation stops have been shown to have an increased risk of inhibitor formation (tuddenham EGD et al).[17]

6. The possibility that repeated switching from one factor VIII product to another facilitates an immune response.
7. The possibility that some products are more immunogenic than others.

In the present study only 7 out of 100 patients had evidence of inhibitors. This could be because of.

1. Inhibitors most commonly arise following an intensive episode of replacement therapy in mild or moderate hemophiliacs. (Hay CRM et al 1998)[18] None of the patients included in this study had history of intensive episode of replacement therapy.
2. Factor VIII or IX inhibitors are often transient and may disappear spontaneously or following immune tolerance induction.

In present study 1% patient show seropositivity for Hepatitis A and 4% patients show seropositivity for Hepatitis C. Hepatitis C was more prevalent than Hepatitis B in hemophiliac patients. Percentage of hepatitis B and Hepatitis C was 6.5% and 9.1% (Saurabh Mishra et al[19]) and 2.5% and 5% (R.K. Nigam et[15]) Very low seropositivity for Hepatitis B and Hepatitis C in present study may be due to:

1. Patients received screened blood and blood products.
2. Use of Recombinant factors and factor concentrate pre-treated for viral inactivation.
3. Development of factor VIII for the management of bleeding episodes causes remarkable lowering in the morbidity as well as mortality due to repeated and prolonged bleeding episodes in patients with haemophilia. Nevertheless, the application of replacement therapy is associated with remarkable complications, such as generation of antibodies against their respective clotting factors i.e factor VIII and factor IX. Also there are significant risk of spread of transfusion transmitted infections.[20] Risk of development of transfusion transmitted infections can be understood by the fact that production of factor concentrate at a large scale for the replacement therapy manufactured from a large pool of plasma that did not undergo viral attenuation increases the risk of transfusion related infections[13] such as HBV, HCV along with successive increase in morbidity and mortality rate up to five fold.[21] Continuous prolonged bleeding was the most common cause of death in haemophiliac patients in early nineteen whereas infections by HIV, HBV and HCV were the most common cause of death during 8th decade of nineteen century.[18] The results for HCV seropositivity were in accordance with study done by Goedert et al., which was conducted during the period 2001-2003 in a large number of Hemophilia centres showed seroprevalence of HCV in 30% of haemophiliacs.[20] Results of other studies are consistent with present study.[22–27] The prevalence of HCV seropositivity was at its peak before the implementation of HCV screening, which was as high as in a range of 92% -100%.[28,29] Similarly, prevalence of HBV infection was also high before the establishment of donor blood screening for HBV seropositivity.[30,31]

5. Conclusion

Generation of inhibitors against factors continue to exist as a provoking complication related to treatment in hemophilia. Treatment of factor Inhibitors by the application of immune tolerance therapy exists as a standard of care and resides as a longterm approach for the augmentation of factor therapy. Though, immune tolerance therapy is a time dependent and high cost procedure, therefore can cause significant impact on psychological and financial factors on patients and their families. Regarding other treatment modalities in patients with inhibitors who have bleeding episodes, besides bypassing agents very few options are there. Several researches are going through with the aim of provision of optimal prophylactic and therapeutic modalities for better care of patients with having risk of therapy related complications.[32–34] Beside this there are also many novel therapy approaches are in the developing stage by mean of several active clinical trials, which can lead to significant decrease bleeding risk and burden of disease in patients with haemophilia.[35–38]

Improvements in transfusion-transmitted diseases risk through nucleic acid amplification assays (PCR), administration of efficient viral inactivation techniques and the use of recombinant factor concentrates would significantly reduce the infectious complications in patients with hemophilia facing today.

6. Source of Funding

None.

7. Conflict of Interest

None.

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