Original Article

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Pharmacology Section

High dose Intravenous Anti-D Immune Globulin is More Effective and Safe in Indian Paediatric Patients of Immune Thrombocytopenic Purpura

TRUPTI REKHA SWAIN[1], RABINDRA KUMAR JENA[2], KALI PRASANNA SWAIN[3]

ABSTRACT

Introduction: Immune Thrombocytopenia (ITP) is characterised by an autoimmune antibody-mediated destruction of platelets and impaired platelet production. Few controlled trials exist to guide management of patients with ITP in Indian scenario for which patients require an individualized approach. Anti-D (Rho (D) immune globulin) at a higher dose can prove to be a cost effective and safe alternative for Indian patients with ITP.

Aim: To compare the safety and efficacy of higher dose (75µg/kg) intravenous Anti-D immune globulin against the standard does of 50µg/kg for the management of ITP in Indian patients.

Materials and Methods: One hundred 64 children with newly diagnosed ITP between 4-14 years were randomly selected for inclusion and were treated with 50µg/kg (standard dose) or 75µg /kg (higher dose) of Anti-D to compare the efficacy and safety of higher dose intravenous anti-D immune globulin. Efficacy of Anti-D was measured in terms of rate of response and median time to response for increase in platelet counts. Any adverse event was noted. A decrease in haemoglobin concentration suggested accompanying haemolysis.

Results: Seventy one out of 84 patients treated with Anti-D at 75µg/kg produced complete response (85%) with median time of response being 2.5 days. On the contrary, 45 patients (70%) patients treated with 50µg/kg had complete response. However there was no significant increase in haemolysis with higher dose. A significant correlation was found between dose and peak increase in platelet count measured at 7th day following administration. However, there was no relationship between the decrease in haemoglobin and the dose given, or between the increase in platelet count and fall in haemoglobin.

Conclusion: A 75µg/kg dose of Anti-D is more effective with acceptable side effect in comparison to 50µg dose for treatment of newly diagnosed Indian patients of ITP.

Keywords: Autoimmune, Children, Platelet count

INTRODUCTION

Immune Thrombocytopenic Purpura (ITP) has an incidence of up to 6.4 per 100000 children per year [1]. This disorder is believed to differ biologically in children compared to adults. In the former, the disease is mostly self limited and often recovers spontaneously and in later the disease is characterised by remission and relapses with chronicity in significant number of cases. The primary objective of treatment is to take care of the patient as a whole considering his age, severity of bleeding and platelet count.

American society of Hematology guideline 2011 has outlined the indication of treatment of newly diagnosed patients of ITP and has approved corticosteroids, Intravenous Immunoglobulin (IVIG) and Anti-D Immunoglobulin as first line drugs to be used in newly diagnosed ITP patients requiring therapy. Though IVIG is the most potent agent among these for raising platelet count, it is expensive and time-consuming for administration for which it is not favored in Indian set up [2]. Corticosteroids are used mostly because of low cost, easy oral administration but at the cost of a long list of side effects including growth retardation in children, hyperglycemia and weight gain [3]. Also, the median time to raise the platelet count beyond haemostatic level (>50 x 10^9/L) is prolonged i.e., 10-14 days, thus not suitable for patients with serious bleeding who need rapid rise of platelet count within a short span. Thus, there is a need of agents that are safe, effective to raise the platelet count fast with acceptable safety profile and cost, particularly in the Indian scenario.

Intravenous Anti-D immune globulin was licensed by the US Food and Drug Administration in 1995 for the treatment of ITP. The cost of treating a patient with intravenous anti-D immune globulin is roughly less than half the cost of IVIG and takes approximately 5 minutes to administer. An expected side effect of anti-D is a self-limited haemolytic anaemia. However, use of anti-D as frontline therapy in ITP decreased significantly after a black box warning by Food and Drug Administration (FDA) to immune globulin (Anti-D) for ITP warning possible complications related to severe haemolysis [4,5].

A thorough literature scan did not reveal adequate studies in India regarding the use of Anti-D in newly diagnosed ITP patients except one study by R Naithani and Rajat kumar who have observed that efficacy of 50µg/kg of Anti-D can produce overall response of 70% in children with median time to response of 3 days in Indian children [6]. However, no study has proved the role of higher dose anti-D in Indian scenario. Hence, present study aimed at exploring the safety and efficacy of higher dose (75µg/kg Anti-D) for the management of ITP in Indian patients.

MATERIALS AND METHODS

This open label, prospective two dose comparison study was conducted in the Dept. of Clinical Haematology in collaboration with Dept of Pharmacology of S.C.B Medical College & hospital, Cuttack, Odisha.

Newly diagnosed patients of ITP were enrolled from July 2012 to June 2014. Patients were defined as newly diagnosed ITP as per recent ITP international working group consensus report,

that has divided the disorder into three phases: newly diagnosed ITP (≤ 3 months), persistent ITP (3 months-1 year) and chronic ITP (> 1 year) [7]. Only typical cases of thrombocytopenia were included for the study for which no special investigation was done. Recommendation of American society of Haematology evidence base practice guideline (ASH- 2011) was followed for the diagnosis of patients for inclusion in this study [8]. This includes a careful history taking, physical examination, review of the complete blood count, peripheral blood smear examination and exclusion of other secondary causes of thrombocytopenia like Anti- Phospholipid syndrome, autoimmune thrombocytopenia, common variable immunodeficiency, drug in-duced thrombocytopenia, infection with cytomegalovirus, HIV, *H. Pylori*, SLE and other vaccine side effects. The principles of indication of Anti D treatment were strictly followed as per the ASH -2011 guidelines. The protocol was approved by institutional ethics committee before starting the study and informed consent was obtained in all cases.

Inclusion Criteria

Newly diagnosed case of ITP with:

- Total platelet count < 50 x 10^9 /L with features of significant bleeding. (skin manifestations like bruising and petichae were considered as minor bleeding).
- Rh positive blood group.
- Direct Anti globulin test- negative.
- Not undergone splenectomy.
- Absence of any other causes of thrombocytopenia.

Exclusion Criteria

- Absence of any one criteria mentioned in the inclusion criteria was qualified for exclusion from the study.

Efficacy and Evaluation

A total 150 patients meeting the inclusion criteria were enrolled in the present study. The patients were divided into two groups randomly considering male female proportion, age range and pretreatment platelet count (<10x10^9/L). Eighty-four patients were treated with Anti-D at the dose of 75µg/kg while 64 patients received the conventional dose of Anti-D (50µg/kg). Physical examination, assessment of severity of bleeding and complete blood count was evaluated in all cases every day during first week and in every alternate day next week. Response was evaluated at the end of 7^{th} day and was categorized into Complete Response(CR), Response (R) and No Response (NR) as per the criteria of international working group [8].

CR: TPC ≥ 100 × 10^9 / Liter measured in two occasions > seven days apart and absence of bleeding.

R: TPC > 30 × 10^9 / Liter and greater than two fold increase from base line measured in two occasions > seven days apart and absence of bleeding

NR: TPC < 30 × 10^9 / Liter or less than two fold increase in TPC from base line seven days apart or the presence of bleeding

Time to response was calculated as time taken from starting anti-D to achievement of CR or R.

Primary End Point: The quality of response (CR, R, and NR) at the end of one week of administration of Anti D was considered as primary end point of the study.

Secondary End Point: Incidence of haemolysis and other side effects during the study period.

Administration of Anti-D

Patients were divided into two groups with compared demographic and other parameters. Anti-D (INDKDIV001, Synergy Diagnostics Pvt. Ltd. Thane India) was provided to all enrolled participants of this study. These hospitalised patients were included randomly to each of the two groups who received either 50µg/Kg or 75µg/kg of Anti–D as a slow IV push over 3–5 min [9]. All patients were pre-medicated with paracetamol and diphenhydramine. Steroids were not routinely given prior to treatment. Two patients in each group were lost to long-term follow-up and were not included in the analysis of long-term treatment response.

Adverse Events

Any haematological or other adverse events were actively searched in the study participants. The observed adverse events were noted in a standard CDSEO form. Special importance was given to search for incidence of haemolysis that is manifested in the form of decrease in haemoglobin concentration or any other side effects.

STATISTICAL ANALYSIS

All subjects who fulfilled the criteria were included in the analysis as long as adequate data were obtained to measure the rise in platelet count and fall in haemoglobin within 14 days of the infusion. The highest count was chosen, reflecting the best response. Dose of 75µg/kg body weight as single bolus intravenous dose was given to all subjects while the second group received the usual dose of 50µg/kg body weight. Data on rise in platelet count were analysed after logarithmic transformation in order to meet distributional assumptions of the analysis. All analyses were performed using SPSS software version 2014 for analysis. All the patients were enrolled randomly to both groups from indoor.

RESULTS

Male patients predominated in both the groups (45/39 and 38/26 in two groups). The age range of the admitted patients was found out to be between 4-14 years with median age of presentation between 6-7 years [Table/Fig-1]. Median haemoglobin (Hb) was 10.8 g/dL (8.2-12.2) Although haemolysis was seen in nearly all infusions, with a median haemoglobin fall of 1.2 g/dl (range, 0.2 to 2.4 g/dl), the decrease in haemoglobin was greater than 1.5 for only three infusions and maximum fall in haemoglobin (2.4) was noted in a child with an underlying anaemia. When safety of Anti-D was evaluated in terms of decrease in haemoglobin level (due to haemolysis) higher dose (75µg/kg) dose produced 1.2gm decrease in haemoglobin level (range: 0.2-2.4). Standard dose of Anti-D (50µg/kg) produced one gm decrease (0.2-2) in haemoglobin concentration due to haemolysis. Relapse rate was slightly less (in 18 patients) with higher dose Anti-D while 22 number of patients had relapse with 50 µg/kg dose of Anti-D. However, the difference was not statistically significant. No mortality was observed in both the groups till the end of study period [Table/Fig-1].

Complete Response (CR) was observed in 85% of patients treated with higher dose of Anti-D while usual dose of Anti-D produced positive response in 70% of patients. Partial Response (R) was found in 16% and 14% of patients treated with high dose and usual dose of Anti-D, respectively. Importantly, incidence of NR was totally absent in patients treated with 75µg/kg anti D. Nine patients were landed to No Response (NR) category treated with 50µg/kg Anti-D therapy. Thus overall response (OR) was significantly high (99%) in patients treated with 75µg/kg dose of Anti-D compared to 86% with 50µg/kg dose [Table /Fig-2].

DISCUSSION

Although ITP in children is usually of the acute, self-limiting type, the chronic form can occur even in young children; some cases continue for longer than 6 months, yet still remit entirely. In children, in whom the risk of post-splenectomy sepsis is higher, a goal of therapy is to delay or avoid splenectomy. Our data demonstrate that the effect of Anti-D on platelet count in children treated for acute ITP is dose-related. Haemolysis is not dose-limiting, at least in the dose used in this study; majority of patients showed a fall in haemoglobin of less than 2.4 g/dL.

Dose of Anti-D	No. of Patients	Male female ratio	Median age	Range (in years)	Pretreatment platelet count	Post administration decrease in Haemoglobin (g/dL)	Range	Relapse	Mortality
Single IV dose of 75 µg/kg	84	45/39	6	4-14	< 10 x 10^9/ L	1.2	0.2-2.4	18	0
Single IV dose of 50 µg/kg	64	38/26	7	5-14	< 10 x 10^9/ L	01	0.2-2	22	0

[Table/Fig-1]: Characteristics of newly diagnosed ITP patients treated with two doses of Anti-D.

Dose of Anti-D	Post treatment Response				
	Median Time to response	Complete Response (CR)	Response (R)	No response (NR)	Overall Response (OR)
Single IV dose of 75 µg/kg (n 84)	2 .5 days (1-12 days)	71*(85%)	12 (14%)	01 (1%)	83 * (99%)
Single IV dose of 50 µg/kg (n 64)	3 days (2-14 days)	45 (70%)	10 (16%)	09 (14%)	55 (86%)

[Table/Fig-2]: Rate of Response to Anti D in immune thrombocytopenic Purpura
* p-value < 0.05 compared to 50 microgram /kg dose.

Our hypothesis for dose dependence of increase in platelet count without a similar effect on haemolysis can be described as: with administered amounts of Anti-D, red blood cells are bound, but can be removed from the circulation only at a rate determined by the number of Fc receptors in the spleen. As red blood cells are removed, Fc receptors become available, allowing additional cells to be bound and destroyed. However, the rate of haemolysis will reach maximum constant rate, easily compensated by a modest increase in the reticulocyte count. Low doses of Anti-D may manifest a dose effect on haemolysis because these doses may be insufficient to saturate Fc receptors, as the fall in haemoglobin using lower doses is less than that seen in our study. For platelets, in contrast, marrow production is already accelerated in ITP due to feedback from the thrombocytopenia. Thus, as soon as Fc receptors are blocked by antibody-coated red blood cells, the platelet count is poised for a rapid increase. Furthermore, we hypothesize that the platelet count should remain relatively high for as long as Fc receptors are blocked. Therefore, a higher dose of Anti-D will elevate the reticulocyte count and platelet count for a longer period of time without creating a greater degree of anaemia.

Although acute ITP is usually more responsive to a given therapy than is chronic ITP, our data show in addition that the acute form has a steeper dose response.

Acute ITP is a dynamic disease, with resolution in a few months, so subsequent therapy will always seem more effective than initial therapy. Chronic ITP is a more static disease, so the effect of the order of therapies (or dose levels) will be more clearly manifest.

Historical data using only low doses of IV Anti-D (25 to 60µg/kg/d) found no evidence of increase in efficacy with increasing doses of Anti-D; however, it did suggest that haemolysis increases with dose. To determine if a dose response was indeed present and its effect on either platelets or haemoglobin level we analysed data using a larger dose range in 84 children treated with intravenous Anti-D immune globulin for acute ITP at single institution [9]. Multiple clinical trials in both adults and children have shown that anti-D effectively raises the platelet count in majority of the treated patients and is associated with an acceptable safety profile. Duration of effect appears to be comparable to IVIG, with some reports of longer duration of effect in anti-D–treated patients [10]. Multiple clinical trials in both adults and children have shown that Anti-D effectively raises the platelet count in the majority of treated patients and is associated with an acceptable safety profile. Duration of effect appears to be comparable to IVIG, with some reports of longer duration of effect in Anti-D–treated patients [11].

Salama et al., in 1983 reported an increase in platelet counts in ITP patients who were also positive for rhesus D antigen after the infusion of IV Anti-D [11]. Current wisdom favours the notion that IV anti-D coats the RBCs that are positive for D antigen, and these opsonised RBCs in turn compete with opsonized platelets in the spleen for sequestration [12,13]. Reports suggest that a dose of 75 µg/kg over 3-5 minutes is more efficacious than a 50µg/kg dose, although the side effects are more common with the higher dose. Some of the commonly encountered infusion-related side effects are fever, chills, nausea, and headache. Another important adverse effect is a fall in haemoglobin secondary to haemolysis. Usually the fall in haemoglobin is not more than 2g/Dl [14,15], but in rare cases, haemolysis can be severe and can lead to renal failure and disseminated intravascular coagulation [16,17].

One randomized study found no significant difference in efficacy between IVIG, Anti-D and oral prednisone, but the dose of Anti-D used (25ug/kg/d X 2) was suboptimal. Our study was not designed to compare the efficacy of Anti-D with that of IVIG or corticosteroids. However, the overall efficacy of Anti-D at the doses used in this study is comparable to historical data for these therapies for acute ITP in this population. This supports differing mechanisms of treatment effect. We have demonstrated that Anti-D is safe and effective for acute ITP in children, even at the relatively high doses used in our patients. Because of its low cost (compared with IVIG), favorable safety profile and ease of administration, the presence of a dose effect suggests that higher doses may offer a therapeutic advantage over IVIG or corticosteroids.

Our study showed that, haemolytic anaemia, thought previously to be dose-limiting, was not clinically significant at these higher doses in the subjects, although caution should be used in those with pre-existing anaemia or haemolysis. Our study indicates that the optimum dose of Anti-D in the treatment of acute ITP has not been found, demonstrating the need for a prospective study to determine the speed, duration and degree of platelet increase and haemolysis for different doses of Anti-D. Although, a dose of 75µg/kg body weight demonstrated positive results in majority of our subjects, rapidity and duration of response are clinically at least as important as the absolute or relative increase in the platelet count. Since rapidity, duration and degree of rise in the platelet count may not directly correlate, they should be assessed specifically in future studies. It is believed that intravenous Anti-D immune globulin functions by binding to Rh-positive red blood cells in the circulation. The antibody is then bound to Fc receptor in the reticuloendothelial system, mostly in the spleen, thus blocking the system's effect on platelets and increasing their survivals. This mechanism requires the patient to be both Rh-positive and to have a functioning spleen.

LIMITATION

Accurate measurement of rapidity and duration of response are not readily available from this protocol. Platelet counts after study duration (14days) were not observed on a regular basis and many patients were followed for only 3months with monthly platelet counts, making duration of response difficult to measure accurately

and to analyse objectively. Another study limitation was the fact that patients did not have a 12-hour platelet count. One of the goals of a planned study will be to obtain a 12-hour count on all patients so the rate of rise can be quantified.

CONCLUSION

Intravenous Anti-D immunoglobulin at the dose of 75µg/kg dose produced higher rate of complete response and overall response compared to 50µg/ kg dose in newly diagnosed patients of Immune Thrombocytopenic Purpura. However there was no significant difference in median time of response. Incidence of haemolysis was not significantly higher in higher dose group.

REFERENCES

[1] Terrell DR, Beebe LA, Vesely SK, Neas BR, Segal JB, George JN. The incidence of immune thrombocytopenic purpura in children and adults: a critical review of published reports. *Am J Haematol.* 2010;85:174-80.

[2] George JN, El-Harake MA, Aster RH: Thrombocytopenia due to enhanced destruction by immunologic mechanisms. In Beutler E, Lichtman MA, Coller BS, et al (eds): Williams haematology (ed 5). New York. NY, McGraw-Hill.1996, pp 1315-1355

[3] George JN, Woolf SH, Raskob GE. Idiopathic thrombocytopenia purpura: A practice guideline developed by explicit methods for the American Society of Haematology. *Blood*. 1996;88:3-40.

[4] Long M, Kalish L, Neufeld E, and Grace R. Trends in Anti-D immune globulin for childhood immune thrombocytopenia: usage, response rates, and adverse effects. *Am J Haematol*. 2012;87(3):315–17.

[5] Klima J, Despotovic J, O'Brien S. Anti-D immunoglobulin therapy for pediatric ITP: before and after the FDA's black box warning. *Pediatr Blood Cancer*. 2013;60(11):149–51.

[6] Naithani R, Kumar R, Mahapatra M, Tyagi S, Mishra P. Efficacy and safety of Anti-D for immune thrombocytopenic purpura in children. *Indian Pediatrics*. 2010;47:517-19.

[7] Rodeghiero F, Stasi R, Gernsheimer T. Standardization of terminology, definitions and outcome criteria in immune thrombocytopenic purpura of adults and children: report from an international working group. *Blood*. 2009;113:2386–93.

[8] Neunert C, Lim W, Crowther M, Cohen A, Solberg L, Crowther MA. The American Society of Haematology 2011 evidence-based practice guideline for immune thrombocytopenia. *Blood*. 2011;117:4190-207.

[9] Tarantino MD, Young G, Bertolone SJ, Kalinyak KA, Shafer FE, Kulkarni R, et al. Acute ITP Study Group. Single dose of anti-D immune globulin at 75 micro g/kg is as effective as intravenous immune globulin at rapidly raising the platelet count in newly diagnosed immune thrombocytopenic purpura in children. *J Pediatr*. 2006;148(4):489-94.

[10] Saurez CR, Rademaker D, Hasson A. High-dose steroids in childhood acute idiopathic thrombocytopenia purpura. *Am J Pediatr Haematol Oncol*. 1986;8:111-15.

[11] Salama A, Mueller-Eckhardt C, Kiefel V. Effect of intravenous immunoglobulin in immune thrombocytopenia. *Lancet*. 1983;2(8343):193–95.

[12] Stasi R, Provan D. Management of immune thrombocytopenic purpura in adults. *Mayo Clin Proc*. 2004;79(4):504–22.

[13] Cines DB, Blanchette VS. Immune thrombocytopenic purpura. *N Engl J Med*. 2002; 346(13):995–1008.

[14] Scaradavou A, Woo B, Woloski BM. Intravenous Anti-D treatment of immune thrombocytopenic purpura: experience in 272 patients. *Blood*. 1997;89(8):2689–700.

[15] Gaines AR. Disseminated intravascular coagulation associated with acute haemoglobinemia or haemoglobinuria following Rh(0)(D) immune globulin intravenous administration for immune thrombocytopenic purpura. *Blood*. 2005;106(5):1532–37.

[16] Gaines AR. Acute onset haemoglobinemia and/or haemoglobinuria and sequelae following Rh(o)(D) immune globulin intravenous administration in immune thrombocytopenic purpura patients. *Blood*. 2000;95(8):2523–29.

[17] Bussel JB, Graziano JN, Kimberly RP. Intravenous anti-D treatment of immune thrombocytopenia purpura: Analysis of efficacy, toxicity, and mechanism of effect. *Blood*. 1991;77:1884-93.

PARTICULARS OF CONTRIBUTORS:

1. Associate Professor and In charge, Clinical Pharmacology Unit, Department of Pharmacology, SCB Medical College and Hospital, Cuttack, Odisha, India.
2. Head, Department of Clinical Haematology, SCB Medical College, Cuttack, Odisha, India.
3. Assistant Professor, Department of Neurology, SCB Medical College, Cuttack, Odisha, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:

Dr. Trupti Rekha Swain,
Associate Professor, In charge Clinical Pharmacology Unit, Department of Pharmacology,
SCB Medical College and Hospital, Cuttack-755007 Odisha, India.
E-mail: drtruptiswain@gmail.com

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