ORIGINAL ARTICLE

The Effect of Colchicine on Thrombocytopenia in Dengue Fever

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ABSTRACT

Objective: To study the effect of colchicine on platelet counts in patients with Dengue fever and to compare the effects of colchicine with placebo.

Material & methods: A randomized-control trial was conducted during a dengue fever out break at Services Hospital, Lahore. The total study duration was from November 3rd, 2010 till November 24th 2010. 212 Volunteers aged 14-70 participated according to inclusion criteria. The institutional review board approval was obtained. Group A received oral soda bicarbonate every twelve hours for 72 hours; Group B received oral colchicine, 0.5 mg every twelve hours for 72 hours. Platelet counts were checked on day 0, after 24 hours (day 1), and then for 3 consecutive days. End points were a platelet count rise above 150,000 or fall below 40,000

Results: The rise in mean platelet counts from the baseline at day 0 and at days 1, 2, 3 and 4 were statistically insignificant between the placebo and colchicine groups: p value; 0.679, 0.734, 0.502 and 0.502 respectively. The endpoints of platelet rise above 150,000 and fall below 40,000 were statistically insignificant between the groups: p value; 0.182 and 0.478 respectively.

Conclusion: Colchicine does not increase platelet counts in dengue fever.

Keywords: Colchicine, Dengue, Thrombocytopenia, Treatment

INTRODUCTION

Dengue is the most common mosquito-borne viral disease of humans that in recent years has become a major international public health concern. Globally, 2.5 billion people live in areas where dengue viruses can be transmitted[2]. Antiviral agents have been ineffective but early detection and access to proper medical care lowers fatality rates below 1%[3]. The exact etiology of thrombocytopenia is unclear in dengue fever. There is evidence of direct bone marrow suppression by the virus[4]. In addition, there is evidence to suggest peripheral immune mediated platelet destruction[5,6]. Mucosal bleeding occurs in patients in dengue fever. Up to 15% of patients with dengue can present with mucosal bleeding[7]. Colchicine has been used for the treatment of immune thrombocytopenic purpura (ITP). A possible mechanism of action for colchicine in ITP is decreased clearance of opsonized platelets secondary to inhibition of microtubule-dependent events in macrophages[8,9].

MATERIAL AND METHODS

The study was designed to determine the response of colchicine on thrombocytopenia in patients with dengue fever during a dengue outbreak. It was a randomized-control trial. Approval from the hospital institutional review board was obtained. The total study duration was 21 days from November 3rd, 2010 till November 24th, 2010. 212 volunteers aged 14-70 participated according to inclusion criteria. It was conducted at medical out-patient department, and the medical emergency of Services Hospital, Lahore.

Non-probability, convenience sampling of a population of normal, non pregnant, healthy volunteers age 14-70 was done to recruit a sample of 212 subjects according to the inclusion criteria. After approval from hospital ethics committee, written informed consent was taken from the volunteers and information was filled in Annex 1. Subjects were assigned randomly in two different groups based upon registration numbers. Group A, assigned even registration numbers, received standard conventional therapy with tablet Paracetamol® 60 mg/Kg body weight, adequate oral hydration, thrice daily skin application of standard market mosquito repellent (MOSPEL by Abbott ®) and oral soda bicarbonate, Soda Bicarb® every twelve hours for 72 hours. Group B, assigned odd registration numbers, received tablet Paracetamol® 60 mg/Kg body weight, adequate oral hydration, thrice daily skin application of standard market mosquito repellent (MOSPEL by Abbott ®) and oral colchicine, "Colchicine®" 0.5 mg every twelve hours for 72 hours. Platelet counts were obtained at the time of recruitment i.e. day 0, and then after 24 hours, i.e. day 1, and subsequently on days 2, 3 and 4. A total of 5 platelet count readings were obtained. End points were defined as a rise in

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platelet counts of more than 150,000 or a fall below 40,000.

Inclusion Criteria:

- A probable diagnosis of dengue fever during an outbreak suggested by WHO. Annex 1
- Age between 14 to 70 years of both genders.
- Platelet counts of below 100,000 and above 20,000

Exclusion Criteria:

- Evidence of dengue hemorrhagic fever or warning signs as suggested by WHO. Annex 1.
- Serious mucosal bleeding.
- Need for urgent platelet transfusion
- Hypotension (systolic BP <90 and Diastolic BP< 60)
- Presence of co-morbid illnesses affecting platelet counts e.g. Chronic liver disease, chronic renal failure, immune thrombocytopenia etc. as determined by history, renal function tests and liver function tests.
- History of intake of drugs affecting platelet function in past 7 days, e.g. Aspirin and non steroidal anti-inflammatory drugs, etc.
- A history of intake of drugs affecting platelet quantity in past 7 days, e.g. ranitidine, rifampicin

Statistical analysis: The data was collected and compiled in the computer and analyzed using SPSS version 17.

RESULTS

Two patients were recruited, 150 male and 62 females all of whom belonged to Lahore, Pakistan (Figure 4). Age range of subjects was between 14 to 70 years (Figure 3)

Patients were randomized to placebo (Group A) and colchicine (Group B). The age and sex characteristics were the same in both groups (Figures 3 and 4).

As mentioned earlier, platelet counts were obtained at the time of recruitment i.e. day 0, and then after 24 hours, i.e. day 1, and subsequently on days 2, 3 and 4. A total of 5 platelets count readings were obtained. The mean rise platelet counts from the baseline levels (day 0) were calculated at days 1, 2, 3 and 4. The results were compared in both the groups (Table 1). The significance of difference in both groups was calculated by applying the two independent samples t-test at 5% level of significance. The rise at days 1, 2, 3 and 4 between both the groups was statistically insignificant: p values: 0.679, 0.734, 0.502 and 0.502 respectively.

There was no difference in both the groups who reached the end point of a platelet count of more than 150,000; p-value 0.182 (Table 2, Figure 1). Similarly, the number of patients whose platelet counts remained above 40,000 did not differ between the groups; p-value 0.478 (Table 3, Figure 2).

Table 1: Comparison of mean platelet counts at baseline and subsequent days between Colchicine and Placebo groups

	Intervention given	=n	Mean	Standard Deviation	P-Value
Day1 Change from baseline	Colchicine	76	+ 6.2632	55.03802	0.679
	Placebo	76	+9.7105	47.22678	
Day2 Change from baseline	Colchicine	53	+22.0000	61.70120	0.734
	Placebo	55	+26.0727	62.62110	
Day3 Change from baseline	Colchicine	39	+29.0256	57.42523	0.502
	Placebo	35	+40.8857	91.41849	
Day4 Change from baseline	Colchicine	27	+58.5185	99.74212	0.502
	Placebo	20	+40.4000	76.75621	

Table 2: Endpoint, Platelets above 150,000 compared in both groups

Platelet Counts above 150,000	Intervention given		Total	P value
	Colchicine	Placebo		
Not reached	8	5	13	0.187
Reached	38	49	87	
Total	46	54	100	

Table 3: Endpoint, Platelets below 40,000 compared in both groups

Platelet Counts below 40,000	Intervention given		Total	P value
	Colchicine	Placebo		
Not reached	16	10	26	0.478
Reached	14	7	21	
Total	30	17	47	

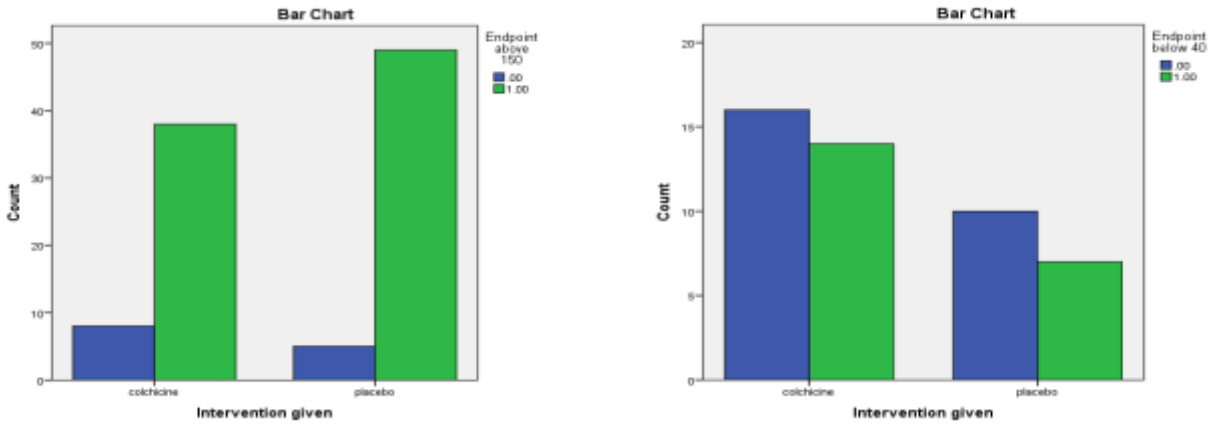

Figure 1:

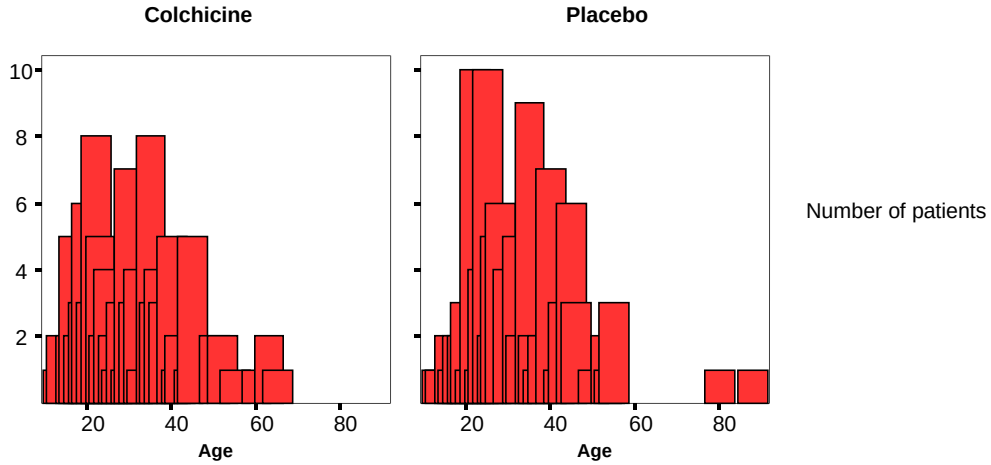

Figure 2**:**

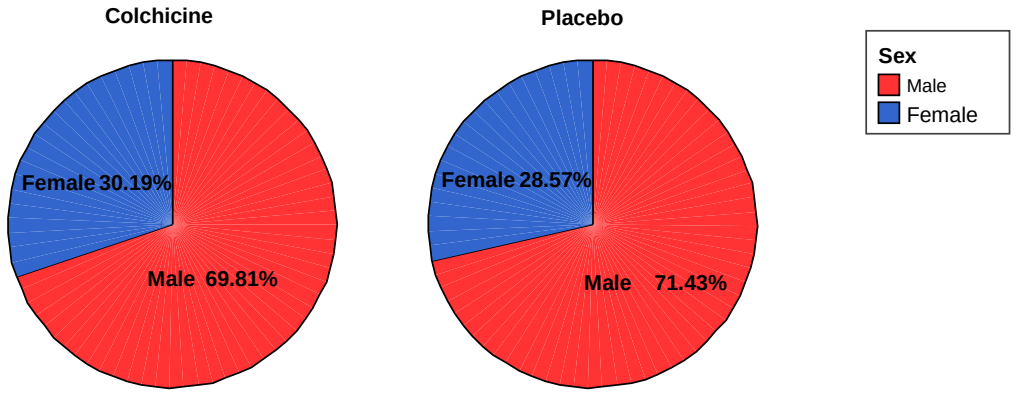

Figure 3:

DISCUSSION

Since 2010, Lahore has witnessed two out breaks of dengue. Medical data suggests that anti-viral agents and steroids have not been beneficial in the treatment of the disease. To make matters worse, the presence of four viral sero-types has impeded the development of a commercially available vaccine. Under these circumstances, treatment is purely supportive. Thrombocytopenia in dengue is associated with an increased risk of mucosal bleeding. Prophylactic platelet transfusions in dengue fever have not shown any benefit[10]. Resource poor settings are also faced with the cost constraints of single donor platelets.

The rationale for using colchicine is its unique anti-inflammatory properties. Colchicine was the first drug known to bind tubulin[11]. By doing so, it may inhibit tubulin assembly thereby destabilizing the microtubules[12]. Microtubules are part of the cytoskeleton in almost every eukaryotic cell. Disruption of microtubules in neutrophils inhibits their migration towards the chemotactic factors. It has been documented that colchicine causes significant inhibition of interaction between white blood cells and endothelial cells by interfering with their transmigration[13]. This might explain the increase in platelet counts observed with colchicine in immune thrombocytopenic purpura.

The data presented shows that the mean sequential rise in the platelet counts from the initial presentation till the fourth day is the same in both colchicine and placebo groups. This shows the lack of efficacy of colchicine on increasing platelet counts in dengue fever. Out of the 147 patients who reached the end points, 100 patients (68%) reached the primary endpoint of platelet counts of above 150,000 by day 4. This platelets increase shows the self limiting nature of the disease which occurred in the majority of patients.

The question arises as to why colchicine is effective in increasing platelet counts in immune thrombocytopenic purpura and not in dengue fever? In dengue fever and dengue hemorrhagic fever, increased levels of many inflammatory mediators are found. Some of these are plasma soluble tumour necrosis factor, IL-8, IFN-α and IL-8 RANTES[14-21]. The effect of neutrophils as an inflammatory mediator in dengue is not well established. As colchicine primarily targets neutrophils, this may explain, the lack of response in the colchicine group. It can be postulated that direct marrow suppression by the virus is the dominant reason for decreased platelet counts in dengue fever, and in contrast to dengue hemorrhagic fever the immune mediated component plays a lesser role. This study excluded patients with suspected dengue hemorrhagic fever. Also, the effect of colchicine is largely mediated through microtubule depolymerization and disruption which are involved in the pathogenesis of diseases like gout, Familial Mediterranean Fever and Amyloidosis[22]. Hence the well-known use of colchicine in these diseases. Microtubules do not play a role in the pathogenesis of dengue viral infection, in contrast the disruption of vimentin results in inhibition of dengue virus infection, perhaps which is why colchicine is ineffective in dengue fever[23]. The results of this study are yet another proof that the mechanism of thrombocytopenia in dengue fever is complicated and poorly understood.

CONCLUSION

Colchicine does not increase platelet counts in patients with dengue fever. In the absence of effective anti-viral therapy and a commercially available vaccine, early detection and supportive therapy continues to remain the main stay of treatment.

REFERENCES

1. http://www.emro.who.int/pak/programmes/dengue-fever.html
2. http://www.who.int/csr/disease/dengue/impact/en/
3. http://www.who.int/mediacentre/factsheets/fs117/en/
4. Jessie K, Fong MY, Devi S, et al. Localization of dengue virus in naturally infected human tissues by immunohistochemistry and in situ hybridization. J Infect Dis. 2004; 189; 1411-1418.
5. Malasit P. Mongkolsapaya J, Kalayanarooj S, Nimmannitya S. Surface associated complement fragment (C3g) on platelets from patients with dengue infection. Southeast Asian J Trop Med Pub Health (abstract) 1990; 21: 705.
6. Wang S, Patarapotikul J, Innis BL, Anderson R. Antibody enhanced binding of dengue 2 virus to human platelets. Virology 1992; 223: 254-257.
7. Cao Xuan Thanh Phuong, Ngo Thi Nhan, Rachel Kneen, Pham Thi Thu Thuy, Chu Van Thien, Nguyen Thi Thuy Nga, Tran Thanh Thuy, Tom Solomon, Kasia Stepniewska, Bridget Wills and the Dong Nai study group. Clinical diagnosis and assessment of severity of confirmed dengue infections in Vietnamese children: Is the world health organization classification system helpful. Am J Trop Med Hyg February 2004 vol. 70 no. 2 172-179.
8. Strother SV, Zuckerman KS, LoBuglio AF. Colchicine therapy for refractory idiopathic thrombocytopenic purpura. Arch Intern Med. 1984 Nov; 144(11):2198-200.
9. Marwaha RK, Singh RP, Garewal G, Marwaha N, Prakash D, Sarode R. Colchicine therapy in immune thrombocytopenic purpura. Acta Paediatr Scand. 1990 Nov; 79(11):1118-20.

10. David C. Lye, Vernon J. Lee, Yan Sun, Yee Sin Leo. Lack of Efficacy of Prophylactic Platelet Transfusion for Severe Thrombocytopenia in Adults with Acute Uncomplicated Dengue Infection. Clin Infect Dis. (2009) 48 (9): 1262-1265.
11. Weisenberg RC, Borisy GG, Taylor EW. The colchicine binding protein of mammalian brain and its relation to microtubules. Biochemistry 1968; 7:4466–79.
12. Downing KH. Structural basis for the interaction of tubulin with proteins and drugs that affect microtubule dynamics. Annu Rev Cell Biol 2000; 16:89–111.
13. Cronstein BN, Molad Y, Reibman J, Balakhane E, Levin RI, Weissmann G. Colchicine alters the quantitative and qualitative display of selectins on endothelial cells and neutrophils. J Clin Invest 1995;96:994–1002
14. Mongkolsapaya J, Dejnirattisai W, Xu XN, et al. Original antigenic sin and apoptosis in the pathogenesis of dengue hemorrhagic fever. 9:921-927 Nat Med. 2003;9:921-927
15. Espina LM, Valero NJ, Hernandez JM. Increased apoptosis and expression of tumour necrosis factor-alpha caused by infection of cultured human monocytes with dengue virus. Am J TRop Med Hyg, 2003;69:
16. Gagnon SJ, Mori M, Kurane I, et al. Cytokine gene expression and protein production in peripheral blood momnuclear cells of children with acute dengue virus infection. J Med Virol, 2002;67:41-46
17. Green S, Vaughn DW, Kalayanarooj S, et al.Elevated plasma interleukin-10 levels in acute dengue correlate with disease severity.J Med Virol. 1999;59:329-334
18. Green S, Rothman A. Immunopathological mechanisms in dengue and dengue hemorrhagic fever.Current Opin Infec Dis. 2006; 19:429-436
19. Bethell DB, Flobbe K, Phuong CXT, et al. Pathophysiologic and prognostic role of cytokines in dengue hemorrhagic fever. J Infect Dis. 1998; 177:778
20. Avirutnan P, Malasit P, Seliger B, et al. Dengue virus infection of human endothelial cells leads to chemokine production, complement activation and apoptosis. J Immunol. 1998;161:6338-6346
21. Bokisch VA, Top FH Jr., Russell PK, et al. The potential pathogenic role of complement in dengue hemorrhagic shock syndrome. NEJM. 1973;289:996-1000
22. E. Ben-Chetrit, S. Bergmann, R. Sood. Mechanism of the anti-inflammatory effect of colchicine in rheumatic diseases: a possible new outlook through microarray analysis. Rheumatology (March 2006) 45 (3): 274-282. doi: 10.1093/rheumatology/kei140
23. Chen W, Gao N, Wang JL, Tian YP, Chen ZT, An J. Vimentin is required for dengue virus serotype 2 infection but microtubules are not necessary for this process. Arch Virol. 2008; 153(9):1777-81. Epub 2008 Aug 10.