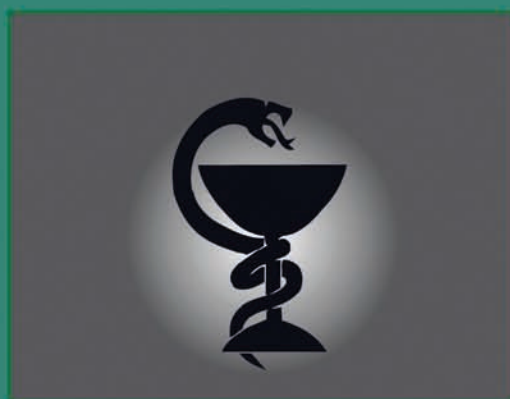


UDC 615

ISSN 0428-0296

PHARMACIA



3/2013

PUBLISHED BY BULGARIAN SCIENTIFIC PHARMACEUTICAL ASSOCIATION

Pharmacia

Vol. 60

No. 3

pp. 1-52

Sofia, November 2013

PHARMACIA

Volume 60

2013

Number 3

JOURNAL OF THE BULGARIAN PHARMACEUTICAL SCIENTIFIC SOCIETY

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Address of Editorial Board

Faculty of Pharmacy
2, Dunav str., Sofia 1000
Fax (02) 987 987 4

Editor in Chief: ☎(+359 2) 9236 505
E-mail: pharmacia_editor@pharmfac.net
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ANTIPYRETIC, ANTIVIRAL AND ANTI-THROMBOTIC PROPERTIES OF *EUPHORBIA HIRTA* AGAINST DENGUE FEVER

Rukhshan Khurshid¹, Mahjabeen Saleem², Sabiha Karim³ and Munnaza Mir¹

¹Department of Anatomy, Biochemistry and Pathology, Fatima Jinnah Medical College, Lahore, Pakistan

²Institute of Biochemistry and Biotechnology, University of the Punjab, Lahore-54590, Pakistan

³School of Pharmacy, University of the Punjab, Lahore, Pakistan

Abstract. Dengue is infecting 50 to 100 million people worldwide a year and study was tried to find out the effect of *Euphorbia hirta* called as TawaTawa on flu like symptoms and blood biochemical parameters especially thrombocytopenia. Blood samples were collected on the day of enrollment and subsequently using TawaTawa. In most of the male patients, platelet count was <25,000 while in females the count was >50,000. Hematocrit values (HCT) were >40% in males and less than 30-40% in females. Total leukocyte count (TLC) was observed in a range of 4000-11000/mm³ in both male and female subjects. IgMhaemagglutination antibody titer values greater than 1:160 were observed in 71% females and 50% males. AST level was found to be >40 IU/L in 38% female and 36% males while ALT level was >40 IU/L in 9% females and 12% males. Platelet count and TLC were increased non significantly after treatment with herbal water of *E.hirta* while HCT value was non significantly decreased after herbal use. Over 70% patients had slight recovery of platelet count and increased retrieval of leukopenia was detected after herbal therapy along recovery in fever and flu like symptoms.

Key words: Dengue, thrombocytopenia, *Euphorbia hirta*

Introduction

Dengue fever also known as breakbone fever caused by *Aedes aegypti* belonging to the RNA virus family of Flaviviridae. It is by far the most important arboviral pathogens in the tropics around the world, putting at risk of infection nearly a third of the global human population [1]. Dengue is endemic in more than 110 countries. The incidence of dengue increased 30 fold between 1960 and 2010. This increase is believed to be due to a combination of urbanization, population growth, increased international travel, and global warming [2]. The geographical distribution is around the equator with 70% of the total 2.5 billion people living in endemic areas from Asia and the Pacific [3]. In Pakistan, incidence of dengue is 48.7% and increasing since 1994 [4].

Dengue is transmitted by several species of mosquito. It prefers to lay its eggs in artificial water containers and feed on people rather than other vertebrates. Dengue can also be transmitted via infected blood products, person-to-person, through organ donation, from mother to child during pregnancy or at birth [5]. The binding of a virus to a host cell is a critical stage in the infectious process and the mechanism

involved in this process is remained largely unknown [6]. Once inside the skin, the virus genome is replicated in endoplasmic reticulum of Langerhans cells and produces, new viral proteins and new viruses are released [7]. Virus enters white blood cells which respond by producing interferons. These interferons are responsible for symptoms, like fever, the flu-like symptoms and the severe pains. Interferon also activates the adaptive immune system, which leads to the generation of antibodies against the virus as well as T cells that directly attack any cell infected with the virus. Various antibodies are generated; some bind closely to the viral proteins and target them for phagocytosis [8].

Dengue hemorrhagic fever presents in two phases: an initial phase, which is characterized by sudden onset of fever and a variety of signs and symptoms [9]. In severe infection, organs like liver and bone marrow can be affected. As a result, less blood circulating in the blood vessels leads to decrease blood pressure. Furthermore, dysfunction of the bone marrow leads to reduced numbers of platelets and increase the risk of bleeding and the other major complication of dengue fever [10].

Medical science has no remedy or vaccine to alleviate the situation. No specific drug to kill the dengue virus. Supportive measures to strengthen the body so that it can recover from the disease. The major means are to drink lots of water, monitor TLC, HCT and platelet count regularly. Eat soft, easily digestible foods and avoid dark colored foods, because to monitor the stool's color [11].

Native American tribes has been used a weed species, *Euphorbia hirta* Linn, commonly called tawa-tawa to treat dengue. It is reported to contain alkanes, triterpenes, phytosterols, tannins, polyphenols, and flavanoids[12]. Tawa-tawa apparently doesn't directly kill the dengue virus but it has immunomodulatory activity [13]. It acts by promoting the development of blood platelets; halts hemorrhage and prevent further bleeding. It may effect on the nausea and abdominal cramps. This herbal treatment makes the impact of dengue virus more bearable to the patient and hastens recovery [14]. As this is a fact that synthetic medicine and hospitalization is very expensive, present study was tried to find out the effect of water extract of Tawa-Tawa on the flu like symptoms and blood biochemical factors.

Materials and methods

Total 125 patients (55 females and 70 males) with confirmed dengue fever admitted in medical ward of Sir Ganga Ram Hospital, Lahore were involved in the study and the duration of study was 3-4 months. Patients were divided into 2 groups: group A (age >25 years) and group B (age <25 years). Blood samples were obtained on the day of admission and follow up samples took 24 hours after TawaTawa treatment. The input variables used were platelet count, hema-

tocrit, WBC count serum AST, ALT estimated by Auto analyzer. Serological assays of IgM/IgG were carried out by ELISA. All patients with a platelet count of less than 50,000 were included for analysis. IgMhaemagglutination antibody titres (>1:160) for dengue type 2 were interpreted as positive result. All other cases of acute febrile illness, not showing clinical features or hematological abnormalities of dengue fever, were excluded. Tea of TawaTawa was prepared by taking 5-6 whole plants, their roots were cut off and washed. The mixture was boiled for 1 minute in a slow rolling boil, allowed to cool and let to drink this water extract to dengue fever victims for 24 hour. Letter of consent was taken from each patient.

Statistical Analysis

Laboratory parameters were recorded on SPSS 11.0 program. Clinical laboratory data on the day of presentation and after using the herbal water extract were used to comprehend the effect of TawaTawa. Data was analyzed by Student's t-test for significance after dividing the patients into two groups.

Results

Platelet count, HCT and TLC before and after TawaTawa water extract use are presented in Table 1. Mean age of group A subjects was 53.89 years and of group B was 26.38 years. In group A, platelet count before using herbal water was 29,000 and after herbal water intake increased up to 41,145. HCT values were found to be 45.19 and 43.38% before and after the use of extracts of *E. hirta* and TLC values of 3500 and 4700 cells/mm³ were recorded before and after using herbal aqueous extract.

Table 1. Platelet count, HCT and TLC before and after usage of water extract of *E. hirta*.
Values are expressed as mean±SD

| Age group (yrs.) | Platelet before use | Platelet after use | HCT (%) before use | HCT (%) after use | TLC /mm ³ before use | TLC mm ³ after use |
|------------------------------------|---------------------|--------------------|--------------------|-------------------|---------------------------------|-------------------------------|
| Group A 53.89 ±10.94 (54) | 29,000 ±25,288 | 41,145* ±29,000 | 45.19 ±9.99 | 43.38 ±5.24 | 3500 ±250 | 4700 ±190 |
| Group B 26.38 ±6.55 (71) | 28,909 ±1566 | 31,153 ±23,518 | 47.09 ±6.68 | 45.51 ±3.01 | 4400 ±200 | 5010 ±210 |

*P< 0.05= Significant difference
No of cases in parenthesis

Table 2. Level of IgM, AST and ALT after use of water extract of *E. hirta*.

Values are expressed as mean±SD

| Groups | Age (yrs.) | IgM agglutination titer before use | IgM agglutination After use | ALT (IU/L) | AST (IU/L) |
|--------------|--------------|------------------------------------|-----------------------------|------------|------------|
| Group A (54) | 53.89 ±10.94 | > 1:160 | < 1:160 | 65.0±10.3 | 45.8±8.9 |
| Group B (71) | 26.38 ±6.55 | > 1:160 | < 1:160 | 69.0±9.8 | 47.7±7.8 |

*P< 0.05= Significant difference

No of cases in parenthesis

In patients of group B, platelet count before using herbal extract was 28,909 and following its dose increased up to 31,153. HCT value decreased from 47.09% to 45.51% after intake of aqueous extract of *E. hirta*. Subsequent ingestion of herbal tea resulted in decline of TLC from 4400 to 5010 cells/mm³.

The impact of TawaTawa herbal extract on IgM, AST and ALT levels in patients of both groups are shown in Table 2. It was observed that the IgM agglutination antibody titer was greater than 1:160 before the use of TawaTawa herbal water in both groups A and B. However, titer value was less than 1:160 on its subsequent use. AST and ALT levels were decreased non significantly in patients both groups after the usage of TawaTawa herbal water.

Discussion

In modern medicines, plants occupy a very important place as the raw material for some important drugs. Synthetic drugs are effective in controlling different diseases but these synthetic drugs are out of reach of millions of people. It is estimated that around 70,000 plant species have been used for medicinal purposes. The herbs provide the starting material for the synthesis of conventional drugs [15]. Dengue virus is an endemic in Pakistan, circulating throughout the year with a peak incidence in the post monsoon period [16].

Present study observed that younger patients with age <25 years were more vulnerable to disease than patients with age >25 years (age range 28-45 years). Similar results have been reported by Khan et al. [16] where the median age dengue patients were less susceptible while younger patients were prone to the fever. The study showed that IgM agglutination antibody titer was greater than 1:160 before the use of herbal water extract while less than 1:160 subsequently in the pa-

tients of both groups. Levels of AST and ALT were decreased non significantly after the use of TawaTawa water extract in both groups. Number of studies is in line. The study reported by Chacko & Subramanian [17] showed that deranged coagulation profile and serum glutamic pyruvic transaminase (SGPT) ≥40 IU are also the predictive of dengue shock [19]. Quiroz et al. [18] have reported the presence of IgM and IgG antibodies to dengue in 11% patients. High IgG levels are indicative of secondary infections [19]. The initial clinical and laboratory assessment can help in selecting appropriate investigations and empiric treatments for patients with imported fever [20].

According to Srikiatkachorn and Green [21], high viral load and intense activation of the immune system are associated with dengue hemorrhagic fever. Spontaneous bleeding was observed in 17 (8%) patients and found to be associated with increased serum alanine and aspartate aminotransferase levels and lower median platelet counts. A secondary immune response was significantly associated with both spontaneous bleeding and other severe clinical manifestations [22].

Our study is in agreement with a report which described that most abnormal WBC or platelet values found between 3-6 days after the onset of symptoms, whereas most abnormal AST, ALT values were observed later. On comparing the platelet count before and after using herbal water extract, it was found that count increased from 29,000 to 41,145 in Group A subjects, while a small increase in platelet count (28,909-31,153) was observed in patients belonging to Group B after herbal therapy.

TLC level increased from 3500/mm³ to 4700/mm³ in Group A subjects and 4400/mm³ to 5010/mm³ in Group B after herbal treatment. In contrast, in Group A, hematocrit value decreased from 45.19 to 43.38% after treatment with TawaTawa aqueous extract and

similar reduction pattern in hematocrit values was observed in Group B patients. Study also observed that flu like symptoms were markedly reduced. Our findings are confirmed by number of studies which reported the use of *E. hirta* the treatment of bronchial and respiratory diseases (asthma, bronchitis, hay fever, etc.) and in conjunctivitis. Its aqueous extract exhibits anxiolytic, analgesic, antipyretic, and anti-inflammatory activities [23,24].

General effect of *Euphorbia hirta* or TawaTawa has been described by a number of studies. The studies reported the antimicrobial activity specific to enteropathogens [13] and 45% immunomodulation activity through the regulation of nitric oxide production [25]. Flavonoids, as constituents of plant, are reported to be responsible for producing anti-inflammatory and humoral antibody responses [26]. The anti-inflammatory activity of *E. hirta* could be attributed to its ability to stabilize mast cell membrane, thereby inhibit the release of inflammatory mediators [27].

Our study is subjected to some limitations. It enrolled patients only during the initial 72 hours of illness, these algorithms may not adequately reflect clinical practice outside of a research setting where many patients present for medical attention after the first 72 hours of illness. We recognize that clinical algorithms cannot be replaced by good clinical management. Further validation using datasets from additional prospective cohort studies conducted in other dengue endemic regions is needed to establish the clinical utility of our algorithms in other populations.

Conclusions

Over 70% of patients had slight recovery of their platelet count. Leukopenia was more recovered after the use of aqueous extract of *E. hirta*. A Marked recovery in fever and flu like symptoms was observed. It is, therefore, concluded that early diagnosis and treatment of dengue illness not only has the potential to reduce morbidity and mortality, but could also reduce the economic impact of dengue illness by decreasing the duration of hospitalization and the number of patients who will develop shock. However, further research on the herbal treatment with *E. hirta* is needed to reach the better conclusion.

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**Corresponding author**

University of the Punjab, Lahore-54590, Pakistan
Institute of Biochemistry and Biotechnology
e-mail: mahjabeensaleem1@hotmail.com
