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Effect of Carica papaya leaves suspension in thrombocytopenia patients infected with dengue: A case control Study

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ABSTRACT

This study aimed to determine the prevalence of local dengue viral strains circulating in New Darband and isolate the thrombocytopenia-associated dengue strain. Platelet volume screening was conducted on dengue-infected patients before and after consumption of Carica papaya leaves suspension. Among 3733 positive patients, 23.5% females tested positive for IgG, IgM, 17.5% for IgG/NS1, and 40.6% for IgM/NS1. In males, 29% tested positive for IgG, IgM, 11.5% for IgG/NS1, and IgM/NS1 22.2% and 27.5% for NS1. Age groups analyzed included 18-30 years, 30-40 years, 40-50 years, and 50-60 years. The administration of Carica papaya leaves suspension resulted in a significant increase in platelet count in dengue-infected patients, with no adverse effects observed during the study. This indicates the potential of Carica papaya in protecting and regulating platelets against dengue virus complications. Additionally, the study suggested that NS1 is a reliable seromarker for accurately detecting dengue infection with thrombocytopenia. Carica papaya leaves suspension shows promise as a suitable remedy for increasing platelet count in dengue-infected patient in dengue-infected patients. However, further high-quality evidence from large clinical trials is necessary to determine its applicability.

Keywords: *Prevalence of Dengue virus; Carica papaya leaves suspension; IgG/IgM; NS1; FDA; GRAS; WHO; Experimental study*

INTRODUCTION

The dengue virus belongs to the family Flaviviridae within the genus Flavivirus. Flavivirus mostly caused arthropod-borne infections, including significant human microbes like dengue, Zika, yellow fever, and West Nile infections (Bok, Balakrishnan et al., 2020, Hasham, Ahmed et al., 2020). Sign and symptoms are high fever, sore muscles, a bad headache, feeling sick, throwing up, and a rash on the skin. In dengue cases, bone-break fever is a common symptom. In severe cases, bleeding comes from the nose, and mouth due to blood vessel bursts (Shrivastava, Alagarasu et al., 2022). Dengue virus infection reduces platelet count. In the case of dengue shock syndrome, frequent vomiting, heavy bleeding, and sudden blood pressure levels decrease. The symptoms of dengue hemorrhagic fever and dengue shock syndrome lead to death (Mohd Abd Razak, Norahmad et al., 2021).

Past studies suggested that the transmission of the different serotypes is cyclic, with unmistakable intermittently reappearing serotypes to overwhelm, and the presentation of new serotypes or genotypes prompting new scourges or episodes (Sathyapalan, Padmanabhan et al., 2020). The mosquito bites a dengue-infected person and sucks his or her blood; when the same mosquito bites a healthy person, the virus is injected into the person. In this way, mosquitoes transmit infection (Bukhari, Naveed et al., 2021, Rabaan, Bakhrebah et al., 2022). Dengue is the most common infection worldwide that mostly affects platelets. During infection, thrombocytopenia and platelet dysfunction are most commonly associated with dengue fever (Rabaan, Mutair et al., 2022). Dengue patients with symptomatic infections develop rapidly lifethreatening complications in which bleeding and plasma leakage are the most common. Because of this, the number of platelets dropped (Mwanyika, Mboera et al., 2021).

Medicinal plants are of importance in term of treatment and prevention (Afzal, Khan et al., 2023, Ahmad, Baig et al., 2022, Ahmed, Karobari et al., 2022, Ramzan, Karobari et al., 2022, Tariq, Ahmed et al., 2020). Carica papaya, also known as papaya, is widely cultivated as a vegetable or fresh fruit which grows in many

parts of tropical and subtropical soils, as well as soil in rainy zone areas (Wadekar, Nimbalwar et al., 2021). Carica papaya a member of the Kingdom Plantae, the Family Caricaceae, and the genus Caricae Species (Papaya). It has different species, and its name is given according to its present areas (Patil, Alagarasu et al., 2022). It has specialized tissues that help to support, and transport nutrients and water up and down the plants, structures are called xylem and phloem (Teh, Ahmad et al., 2022). Leaves are wide at 50-70 cm and 20-27 in diameter, mostly lobed with seven lobes (Sharma, Mishra et al., 2019).

Carica papaya is a tropical plant species that contain the natural medicinal benefits of natural remedies (Nor, Mohd et al., 2019). Papaya acts as a medicinal plant and has been reported to have antimicrobial and antioxidant activities for curing diseases, is due to the presence of secondary metabolites such as saponins, tannins, flavones, flavanols, and steroids (Subenthiran, Choon et al., 2013). Papaya suspension is used against several diseases such as inflammation, dengue fever, cancer, diabetes, and Alzheimer's disease (Dwivedi, Sonter et al., 2020).

The aim of the study was to find the frequency and prevalence of the local dengue viral stain circulating in the New Darband. Also, to isolate the thrombocytopenia associated-dengue strain. Study screened the platelets volume of the dengue infected patients before Carica papaya leaves suspension and after Carica papaya leaves suspension consumption. The current research investigated the efficacy of 1.1 g/ml Carica papaya leaves suspension in patient infected with dengue virus. Similarly purpose of the study was to prepare suitable suspension doses that are safe in consumption to human as well as effective dose against dengue viral strain among local population.

MATERIALS AND METHODS Study area and ethical conciderations

New Darband Township is located near the Indus River, one of the tropical areas of district Mansehra with 38-42°C temperatures. New Darband is in a rural area and is famous for ponds, rivers, and tube wells. This study was

conducted at Civil Hospital, New Darband Township.

The Ethical approval registration no MIC-UOH002 was received by the Advance Research Directorate of Human Ethics of a Civil Hospital, New Darband, as well as from the Department of Microbiology, The University of Haripur under registration no F20-2013. The concerned office permitted us to direct the clinical examination, testation and experimental study under the act and regulations of the Advance Research Directorate of Human Ethics. Furthermore, they supervised all the material before and after used in the study. They permit us to describe the cause and aim of the study. They issued an ethical letter to proceed with research work precisely.

Subject selection for Dengue antigen detection

We used 4500 patients for the research project who were suspected and positive for Dengue fever. Patients were classified into several groups based on age, sex, and area. Blood tests had been drawn from infected patients with dengue fever signs and symptoms as well as from suspected patients.

Subject selection for Carica papaya

We enrolled 100 dengue infected patients who were tested positive for dengue and 100 controls (healthy individuals). They were enrolled for the experimental study in the diagnostic center of civil hospital New Darband.

Specimen collection and processing

Fresh blood was obtained at about 3 mL in volume from patients by puncturing the vein. Blood was stored in EDTA tubes (K3) for complete blood profile such as Hemoglobin, Platelets, and white blood cells. 20 μ l of whole blood was added to the AccessBio® rapid diagnostic kit to each well for the detection of IgG/IgM and NS1 antigen.

Questionaire and informed consent consideration

We developed 200 questionnaires for experimental subjects. Each questionnaire carried a few questions according to the demographics, health, previous history, occupation, and dengue fever. Questionnaires were prepared in English and also translated into the local language. All the patients gave their answers to the questionnaire very accurately, as well as their signatures and contact number.

Serological markers detection

Blood samples of 4500 patients were tested in the diagnostic center, Civil Hospital New Darband, for the serological markers of dengue. All the tests were performed by the AccessBio® kit methodology.

AccessBio® rapid IgM/IgG and NS1 test

We used AccessBio®, a rapid vital diagnostic kit for the detection of IgG, IgM, and NS1 antigen tests for the dengue virus. The device detected different classes of antibodies (IgM, IgG), which are produced by the body against dengue virus. Perform a test 5-8 days after exposure for effective detection of IgM antibodies. If IgM detection occurs, it means an infection is an initial stage.

AccessBio® rapid NS1 Antigen

The AccessBio® NS1 kit can be bought as a diagnostic test to check for the presence of dengue viruses by testing for the non-structural proteins NS1. In this test, synthetically labeled antibodies were used for the detection of the NS1 protein portion of dengue.

Hematological analysis

We used hematology analyzer Sysmex kx-21 for the analysis of complete blood profiling. We used 2mL of whole blood to assess various blood parameters, including (hemoglobin, white blood cells, and different leukocytes, hematocrit, and platelets). All the samples were placed on the roller for 5 minutes at 37°C (room temperature) before the procedure. For the evaluation of the patient specimens for complete blood counts, tubes were loaded to a chamber of the Sysmex kx-21 and proceeded further.

Carica papaya suspension preparation

We followed the national pharmacopeia and international pharmacopeia for the dose calculation, weighting, and herbal suspension preparation. All procedures for Carica papaya leaves suspension, preparation and suspension dosage distribution were done according to the WHO, FDA, GRAS and EUFSA. The Carica

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papaya leaves were collected from Gulshanabad Street, New Darband.

Transportation of Suspensions

Carica papaya suspension was placed into a sterilized plain falcon tube with a capacity of 45 ml to 60 ml and sealed tightly with a screw cap before being transported to the experiment room under controlled conditions. (Supplementary figure S1)

Storage of Carica papaya Suspension

Carica papaya suspension was stored in the Biobase fridge for a day to maintain efficacy, proficiency, smell, and texture of Carica papaya leaves suspension. (Supplementary figure S1)

Carica papaya Suspension formulation

500 g leaves of Carica papaya leaves were blended to prepare the volume of the Carica papaya leaves suspension 450ml. the formulation and dose calculation was 1.1g/ml as per (12) (Table no.1). The total weight of the Carica papaya leaves suspension was divided by the total volume of the leaves as per pharmacopeia.

Inclusion criteria

We included patients in Carica papaya leaves suspension experiment who were confirmed positive for dengue. General signs and symptoms and warning signs and symptoms, including retro-orbital pain, purpura, and rashes. Also included the patients with >25,000/µl platelets.

Exclusion criteria

Those patients with hemorrhagic condition were excluded and patients with Less than $25,000/\mu$ l platelets.

Dosage distribution for Carica papaya leaves suspension

The dosing and distribution were carried out as per the national pharmacopeia and international pharmacopeia. The suspension was formulated with the help of a well-reputed pharmaceutical industry reference webpage (www.caripillmicro.com) and also analyzed by the pharmacist. A proper suspension was formulated. The dose schedule was chosen and categorized by the pharmacist as defined in the publication of an international or national company. The dose of suspension was decided based on the severity of the disease.

Dose distribution for control and patients

We included healthy individuals as controls in the experimental study. The study consultant recommended three spoons of Carica papaya leaves suspension three times a day. Also, these healthy individuals were monitored for adverse reactions, and toxicity. The Carica papaya leaves suspension dosage and distribution for control and patients has been described in Supplementary table S1.

Those patients who tested positive for dengue IgG/IgM. They recommended the same dose as prescribed for positive control due to less severe symptoms, average platelets destruction, and acceptable fever. They were recommended three spoons of Carica papaya leaves suspension three times a day.

Those patients who had warming signs and symptoms also had low platelets in their blood. Due to high-grade fever, body pain, and severe headache. Due to very low platelets, they were recommended four spoons three times a day.

These patients were tested for low platelet volume ($<50,000/\mu$ l) in the blood so they were recommended to continue their allopathic medicine alone with Carica papaya suspension. They were prescribed to administer the Carica papaya suspension in three spoons a day.

Statistical significance

The isolated data through questionnaires such as (Age, sex, areas, medical history, routine questions, and dengue-related questions were interpreted through Microsoft Excel such as Mean \pm S.D, chi-square, and T-pair test to find out the variance, correlation among different groups of patients' data based on age, gender, as well as dengue serological markers like IgG/IgM and NS1.

RESULTS

Prevalence portion of the study

The current study was conducted at a civil hospital in New Darband. The healthcare center has (20+) villages and towns with a large population rate. We gathered patients in the presence of concern physician and sent for further diagnostic examination to isolate the

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specific dengue NS1 antigen and IgG/IgM antibodies against dengue virus. The study tested 4500 patients in which 3733 were positive and 767 were negative. The study included only dengue positive patients who had signs and symptoms (fever was p = 0.007), (Rashes were p = 0.005), Headache was p = 0.007), Leukopenia was p = 0.005), (Retro-orbital pain was p = 0.001). (Table 1)

All serological markers (IgG, IgM, and NS1) were detected by the AccessBio® (United States of America) due to its highest specificity and sensitivity rates. We tested positive IgG, IgM, and NS1 in patient infected with dengue viral infection and found a higher seroprevalence of dengue viral infection in females were 14.1%, while males 75% across the population. The highest seroprevalence of dengue NS1 was in males while the lowest was in females due to a lot of males working in garages, boat operators, garage, and fishermen. While the smaller number of cases of dengue NS1 was found in females who worked outside the home. They also slept outdoors due to the warm ecosystem around their living area. (Supplementary figure S3)

Out of 3733 positive patients, females were tested positive for IgG, IgM (23.5%), IgG/NS1 (17.5%) and IgM/NS (40.6%), while males were tested positive for IgG, IgM (29%), IgG/NS1 (11.5%), and IgM/NS1 (22.2%) and NS1 were screened as (27.5%). We also reported the highly age-groups such as 18–30 years, 30–40 years, 40–50 years, and 50–60 years. The most infected or exposed group were under 30 years of age because these individuals were working outdoor and issues of poverty.

Experimental portion of the study Carica papaya suspension results

The study encompassed patients for Experimental study about n=100 that made an account for Control were 50, dengue positive patients were 50 with age group of 22-60 years as shown in (table 2).

Control (healthy individuals)

A group of healthy individuals (Control) with an average age of 22-60 years and an average weight of 41–81 kg participated in the study. Carica papaya was tested on a small population, such as healthy individuals (Control 50), to pass judge its safety and incidental effects and to track down

the right medication dose as shown in the (Table 3). The difference in M \pm SD of parameters of placebos such as platelets were recorded 33.1 (11.2), WBCs 1.1 (8), Hb% 16.4 (4), hematocrits 9.2 (7) whereas T=1.2819 and p=0.002. (Table 3)

Patients with IgG and IgM dengue positive

The difference in M \pm SD of parameters of IgG/IgM such as platelets were recorded 41.8 (15), WBCs 3.4 (11), Hb% 11.5 (6), hematocrits 10 (7) whereas T=1.1569 and p=0.03. (**Table 4 and Figure 1**)

Patients with NS1 dengue positive

The difference in M±SD of parameters of NS1 positive such as platelets were recorded 69.4 (34), WBCs 2.5 (12), Hb% 13.3 (4), hematocrits 4.1 (11) whereas T=1.1765 and p=0.02. (**Table 5**)

The difference in M±SD of parameters of IgG/IgM such as platelets were recorded 41.8(15), WBCs 3.4(11), Hb% 11.5 (6), hematocrits 10 (7) whereas T=1.1569 and p=0.03. The difference in M±SD of parameters of NS1 positive such as platelets were recorded 69.4 (34), WBCs 2.5 (12), Hb% 13.3 (4), hematocrits 4.1 (11) whereas T=1.1765 and p=0.02. The difference in M±SD of parameters of suspension + medication such as platelets were recorded 120 (70), WBCs 4.9 (10), Hb% 13.8 (4), hematocrits 2.0 (8) whereas T=1.1765 and p=0.02. (**Table 6 and Figure 2**)

DISCUSSION

We investigated the Seroprevalence in the circle of the civil hospital. The highest Seroprevalence of dengue-NS1 was reported in males due to working in tyre shops, garages, boat operators, and fishermen while lowest in females. They also sleep outdoors due to the warm atmosphere around their living area. All of them were used their water from open ponds and rivers instead of filtered or sealed water and worked outdoors. New Darband was highly affected due to its higher population rates, the Indus riverbank, and ponds are found in abundance numbers.

An experiment on papaya action on dengue was proposed by Sarker et al., (2021) (Sarker, Khan et al., 2021), in which the papaya suspension shows its effect on platelets directly by inhibiting

the activation and aggregation of platelets with neutralizing dengue viral infected plasma. In addition, Sharma et al., (2019) (Sharma, Mishra et al., 2019) observed a significant increase in the number of thrombocyte counts in Wistar rats after administering 150 mg/kg of aqueous suspension of papaya leaves orally for 72 hours.

Dengue virus Critical disease is characterized as the underlying or first infection with a firm serotype (Koul, Pudhuvai et al., 2022). The majority of critical episodes were asymptomatic or manifest as mild febrile illness, even though they can cause hemorrhagic fever in certain patients, particularly newborn children born to DENV-invulnerable parents. May prompt serious clinical suggestions, for example, dengue hemorrhagic fever (DHF) or dengue shock disorder (DSS).

In a cross-sectional studied by Khan et al., (2020) (Khan, Danish et al., 2020) about 274 healthy populations matured at 15 years and above were arbitrarily chosen to utilize a multistage inspection procedure. These people were in July–September 2012, utilizing a semi-structured survey, followed by drawing 3 ml of their venous blood for a dengue IgG test. A Nova-Tech ELISA unit with an affectability and particularity of (96.5% and 97.5%), respectively, was utilized for serology.

In a previous study, Raza et al., (2018) (Raza, Ashraf et al., 2018) Selected a total of 612 subjects with a presumed infection. Out of the 612 speculated cases (319 were discovered to be positive for dengue IgG, IgM, or both IgG and IgM). The overall light predominance of dengue viral antibodies IgM, as well as IgG, was (52.12%). Generally, of the (52.12%, 31.86%) were positive for dengue IgM, and (20.26% were positive for dengue IgG), while just 23 (3.75%)examples showed both (IgG and IgM antibodies). In the age group of 21–30 years, IgM antibodies (39.35%) were more common than IgG antibodies (22.42%). This is in contrast to the age groups of children (10 years) and the oldest people (51 years).

According to Shabbir et al., (2020) (Shabbir, Pilz et al., 2020) dengue Seroprevalence was altogether recognized in male patients n = 149, 87.13%, when contrasted with female patients n = 22, 12.87%, P = 0.0004. Patients in the age group (20–39 years, n = 114, 66.67%), were more inclined to dengue than those in the age group over (60 years). Sex, age classification, and region were critical elements in the dengue Seroprevalence P = 0.034. The general Seroprevalence of anti-dengue (IgG was 56.60%, n = 171 in dengue-suspected patients. Stale water repositories were found to be answerable for the spread of the infection, preferring mosquito rearing destinations.

From an aggregate of Khan et al., (2022) (Khan, Adil et al., 2022) 900 subjects, 46% were found to have positive IgG in their blood. Their outcomes revealed that a male offspring old enough to be over 10 years old was bound to be IgG positive. Other risk factors associated with seropositivity included family income and absence of illness, thrombocytopenia, and lack of hand washing.

In a study by Mukhtar et al., (2021) (Mukhtar, Mukhtar et al., 2021), authors Observed the seasonal changes in seropositivity in dengue patients from 2010 to 2013. They found that the most common time for dengue fever to start was in October, while the least common time was from January to April. Age-wise, younger subjects were more prone to the infection, with a mean age of (21 to 30 years). 64% of the total febrile seropositivity cases were male as compared to female patients noted at 36%.

Mubbashir et al., (2018) (Mubbashir, Munir et al., 2018) analyzed 378 cases; among these, 126 (33.3%) cases were positive for (anti-dengue IgG and IgM antibodies). Females were more affected, (266 70.4%) and the commonest age group affected in these adults was (37-54 years), (143 37.8%). A significant association at (p-value<0.5) was found in (gender, ethnicity, marital status, primary education, and unemployed subjects with seropositive tests.

Dengue is one of the most prevalent infections throughout the globe. About one-third of the world's population is at risk of developing the disease. Researchers have always been in search of an effective treatment against dengue infection. However, papaya leaf extract has been found to have an anti-thrombocytopenic, immuno-modulatory, and anti-viral effect that may provide relief from the symptoms (Chandrasekaran, Seetharaman et al., 2018).

According to Mohd Abd Razak et al., (2018) (Mohd Abd Razak, Mohmad Misnan et al., 2018) a study was conducted to check the efficacy of

papaya leaf extract against Dengue virus and its impact on platelet count increase. Platelet count was investigated in cyclophosphamide-induced thrombocytopenia in rats. A platelet specimen from the retro-orbital plexus was taken into consideration. The data showed a significant reduction in intracellular viral load as a result. Erythrocyte damage was also reduced. An increase in platelet count was also observed as a result of treatment with PLE.

Solanki and Trivedi et al., (2020) (Solanki and Trivedi, 2020) Designed study to evaluate the efficacy of Carica papaya leaf extract among pediatric subjects with dengue fever revealed that CPLE syrup; Caripill had a positive impact on thrombocytes upon consumption. A total of 294 subjects aged between 1 and 12 years were investigated for CPLE syrup efficacy for about 05 days. A subsequent increase in platelet count from days 03-05 with P=0.030 to P=0.023 was seen in the control group. Mohd Abd Razak et al., (2019) (Mohd Abd Razak, Norahmad et al., 2019) demonstrated a (p = 0.007) increased platelet count (482% \pm 284) among 26 of the CPLE-treated confirmed dengue adults with severe thrombocytopenia from day 03 compared to the placebo group.

CONCLUSIONS AND RECOMMENDATIONS

It has been determined that a suspension of the Carica papaya leaves suspension raises the platelet count in patients with dengue viral infection. No adverse effects found in the course of the study, show protective and regulate platelets against the complication of dengue virus. From current study it can be concluded that NS1 as good seromarker for the accurate detection dengue infection with thrombocytopenia. Oral administration of prescribed dose of Carica papaya leaves suspension to selected dengue patients does significantly increase the platelet count in patients with thrombocytopenia associated with dengue with no side effects and good tolerability. It is necessary to conduct test in vitro or in vivo studies prior to conducting any clinical trials with these phytochemicals in order to evaluate the potential therapeutic applications of these compounds.

Carica papaya leaves suspension can be regarded a suitable remedy for increasing platelet count in

dengue infected patients. However, high-quality evidence in the form of large clinical trials is required before a determination on whether to use such extract is applicable. The research that was addressed earlier offered evidence for the presence of bioactive phytochemicals in papaya leaves, which have the potential to play role in the prevention of dengue fever as well as in its treatment. As a result of these factors, Phytomolecules are anticipated to bring about a revolutionary change in the prevention and treatment of dengue fever within the next decade. Furthermore, they will offer a viable and efficient alternative to traditional medications. Besides all of the encouraging results that have been obtained from a number of biochemical, cell culture, animal, and a few human clinical trials, there is still a need for more in-depth research and pharmacological experiments to examine the possible role that papaya could play in the treatment of a number of distinct human chronic conditions.

REFERENCES

- Afzal M, Khan AS, Zeshan B, Riaz M, Ejaz U, Saleem A, Zaineb R, Sindhu HA, Yean CY. Ahmed N (2023). Characterization of Bioactive Compounds and Novel Proteins Derived from Promising Source Citrullus colocynthis along with In-Vitro and In-Vivo Activities. Molecules 28(4) 1743.
- Ahmad A, Baig AA, Hussain M, Saeed MU, Bilal M, Ahmed N, Chopra H, Hassan M, Rachamalla M. Putnala SK (2022). Narrative on Hydrogen Therapy and its Clinical Applications: Safety and Efficacy. Current Pharmaceutical Design 28(31) 2519-2537.
- Ahmed N, Karobari MI, Yousaf A, Mohamed RN, Arshad S, Basheer SN, Peeran SW, Noorani TY, Assiry AA. Alharbi AS (2022). The Antimicrobial Efficacy Against Selective Oral Microbes, Antioxidant Activity and Preliminary Phytochemical Screening of Zingiber officinale. Infection and Drug Resistance 15 2773.
- 4. Bok ZK, Balakrishnan M, Jong YX, Kong YR, Khaw KY. Ong YS (2020). The plausible mechanisms of action of Carica papaya on dengue infection: A comprehensive review. Progress in Drug Discovery & Biomedical Science 3(1).
- Bukhari B, Naveed M, Makhdoom SI, Jabeen K, Asif MF, Batool H, Ahmed N. Chan YY (2021). A comparison between organic and inorganic nanoparticles: Prime nanoparticles for tumor curation. Nano 16(13) 2130011.
- 6. Chandrasekaran R, Seetharaman P, Krishnan M, Gnanasekar S. Sivaperumal S (2018). Carica

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papaya (Papaya) latex: a new paradigm to combat against dengue and filariasis vectors Aedes aegypti and Culex quinquefasciatus (Diptera: Culicidae). 3 Biotech 8 1-10.

- Dwivedi MK, Sonter S, Mishra S, Patel DK. Singh PK (2020). Antioxidant, antibacterial activity, and phytochemical characterization of Carica papaya flowers. Beni-Suef University Journal of Basic and Applied Sciences 9 1-11.
- Hasham K, Ahmed N. Zeshan B (2020). Circulating microRNAs in oncogenic viral infections: Potential diagnostic biomarkers. SN Applied Sciences 2(3) 1-13.
- 9. Khan J, Adil M, Wang G, Tsheten T, Zhang D, Pan W, Khan MA, Rehman Iu, Zheng X. Wu Z (2022). A cross-sectional study to assess the epidemiological situation and associated risk factors of dengue fever; knowledge, attitudes, and practices about dengue prevention in Khyber Pakhtunkhwa Province, Pakistan. Frontiers in Public Health 10 923277.
- Khan NU, Danish L, Khan HU, Shah M, Ismail M, Ali I, Petruzziello A, Sabatino R, Guzzo A. Botti G (2020). Prevalence of dengue virus serotypes in the 2017 outbreak in Peshawar, KP, Pakistan. Journal of Clinical Laboratory Analysis 34(9) e23371.
- 11. Koul B, Pudhuvai B, Sharma C, Kumar A, Sharma V, Yadav D. Jin J-O (2022). Carica papaya L.: a tropical fruit with benefits beyond the tropics. Diversity 14(8) 683.
- 12. Mohd Abd Razak MR, Mohmad Misnan N, Md Jelas NH, Norahmad NA, Muhammad A, Ho TCD, Jusoh B, Sastu UR, Zainol M. Wasiman MI (2018). The effect of freeze-dried Carica papaya leaf juice treatment on NS1 and viremia levels in dengue fever mice model. BMC Complementary and Alternative Medicine 18 1-13.
- Mohd Abd Razak MR, Norahmad NA, Md Jelas NH, Afzan A, Mohmad Misnan N, Mat Ripen A, Thayan R, Zainol M. Syed Mohamed AF (2021). Immunomodulatory activities of Carica papaya L. leaf juice in a non-lethal, symptomatic dengue mouse model. Pathogens 10(5) 501.
- 14. Mohd Abd Razak MR, Norahmad NA, Md Jelas NH, Jusoh B, Muhammad A, Mohmad Misnan N, Zainol M, Thayan R. Syed Mohamed AF (2019). Preliminary study on the expression of endothelial cell biology related genes in the liver of dengue virus infected mice treated with Carica papaya leaf juice. BMC research notes 12 1-6.
- 15. Mubbashir H, Munir S, Kashif R, Nawaz HB, Abdul B. Baharullah K (2018). Characterization of dengue virus in Aedes aegypti and Aedes albopictus spp. of mosquitoes: A study in Khyber Pakhtunkhwa, Pakistan. Molecular Biology Research Communications 7(2) 77.
- 16. Mukhtar MU, Mukhtar M, Iqbal N, Nawaz Z, Bhatti A, Haq F, Arslan A. Rashid M (2021). The

emergence of Dengue Fever in Sheikhupura, Pakistan: its seroprevalence and risk factors assessment during 2014-2017. The Journal of Infection in Developing Countries 15(09) 1351-1355.

- Mwanyika GO, Mboera LE, Rugarabamu S, Ngingo B, Sindato C, Lutwama JJ, Paweska JT. Misinzo G (2021). Dengue virus infection and associated risk factors in Africa: a systematic review and meta-analysis. Viruses 13(4) 536.
- 18. Nor AN, Mohd R, Norazlan MM, Nur H, Umi RS, Amirrudin M, Ho C, Bazilah J, Nor AZ. Ravindran T (2019). Effect of freeze-dried Carica papaya leaf juice on inflammatory cytokines production during dengue virus infection in AG129 mice. BMC Complementary and Alternative Medicine 19(44).
- Patil P, Alagarasu K, Chowdhury D, Kakade M, Cherian S, Kaushik S, Yadav J. Parashar D (2022). In-vitro antiviral activity of Carica papaya formulations against dengue virus type 2 and chikungunya virus. Heliyon 8(12) e11879.
- 20. Rabaan AA, Bakhrebah MA, Nassar MS, Natto ZS, Al Mutair A, Alhumaid S, Aljeldah M, Garout M, Alfouzan WA. Alshahrani FS (2022). Suspected adenovirus causing an emerging hepatitis among children below 10 years: a review. Pathogens 11(7) 712.
- Rabaan AA, Mutair AA, Alhumaid S, Garout M, Alsubki RA, Alshahrani FS, Alfouzan WA, Alestad JH, Alsaleh AE. Al-Mozaini MA (2022). Updates on Measles Incidence and Eradication: Emphasis on the Immunological Aspects of Measles Infection. Medicina 58(5) 680.
- 22. Ramzan M, Karobari MI, Heboyan A, Mohamed RN, Mustafa M, Basheer SN, Desai V, Batool S, Ahmed N. Zeshan B (2022). Synthesis of silver nanoparticles from extracts of wild ginger (Zingiber zerumbet) with antibacterial activity against selective multidrug resistant oral bacteria. Molecules 27(6) 2007.
- Raza FA, Ashraf S, Hasnain S, Ahmad J. Iqbal M (2018). Dengue seroprevalence and its socioeconomic determinants in Faisalabad, Pakistan: A cross-sectional study. Revista da Sociedade Brasileira de Medicina Tropical 51 503-507.
- 24. Sarker MMR, Khan F. Mohamed IN (2021). Dengue fever: therapeutic potential of Carica papaya L. Leaves. Frontiers in pharmacology 12 610912.
- 25. Sathyapalan DT, Padmanabhan A, Moni M, P-Prabhu B, Prasanna P, Balachandran S, Trikkur SP, Jose S, Edathadathil F. Anilkumar JO (2020). Efficacy & safety of Carica papaya leaf extract (CPLE) in severe thrombocytopenia (≤ 30,000/µl) in adult dengue–Results of a pilot study. PLoS One 15(2) e0228699.

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- Shabbir W, Pilz J. Naeem A (2020). A spatialtemporal study for the spread of dengue depending on climate factors in Pakistan (2006– 2017). BMC Public Health 20(1) 1-10.
- 27. Sharma N, Mishra KP, Chanda S, Bhardwaj V, Tanwar H, Ganju L, Kumar B. Singh SB (2019). Evaluation of anti-dengue activity of Carica papaya aqueous leaf extract and its role in platelet augmentation. Archives of virology 164 1095-1110.
- Shrivastava N, Alagarasu K, Cherian S. Parashar D (2022). Antiviral & platelet-protective properties of Carica papaya in dengue. Indian Journal of Medical Research 156(3) 459-463.
- 29. Solanki SG. Trivedi P (2020). Evaluation of the efficacy of Carica papaya leaf extract on platelet counts in dengue patients. Journal of Advanced Scientific Research 11(Suppl 04) 62-65.
- 30. Subenthiran S, Choon TC, Cheong KC, Thayan R, Teck MB, Muniandy PK, Afzan A, Abdullah NR. Ismail Z (2013). Carica papaya leaves juice significantly accelerates the rate of increase in

platelet count among patients with dengue fever and dengue haemorrhagic fever. Evidence-Based Complementary and Alternative Medicine 2013.

- Tariq F, Ahmed N, Afzal M, Khan MAU. Zeshan B (2020). Synthesis, Characterization and antimicrobial activity of Bacillus subtilis-derived silver nanoparticles against multidrug-resistant bacteria. Jundishapur Journal of Microbiology 13(5).
- 32. Teh BP, Ahmad NB, Mohamad SB, Tan TYC, Mohd Abd Razak MRB, Afzan AB. Syed Mohamed AFB (2022). Carica papaya Leaf Juice for Dengue: A Scoping Review. Nutrients 14(8) 1584.
- 33. Wadekar AB, Nimbalwar MG, Panchale WA, Gudalwar BR, Manwar JV. Bakal RL (2021). Morphology, phytochemistry and pharmacological aspects of Carica papaya, an review. GSC Biological and Pharmaceutical Sciences 14(3) 234-248.

TABLE 1: Responses of the patients for generally asked questions at their open choice.

Questions	Yes	No	
Do you have stagnant water around your hou	78.2	21.7	
Do you sleep outdoor or indoors?	68.3	31.6	
Have you been diagnosed with dengue befor	12.0	88.4	
Where did you work for the last time?	Tyer shop	9.9	
	Fields	56.5	
	Not yet working	24.1	
	Students	9.3	
Do you have previous history?	34.0	65.9	

TABLE 2: Demographic variables included in the current study.

Subject Variables	Treatment Group (n=50)	Control Group (n=50)
Demographic Variables		
Male	25	25
Female	25	25
Age	22-60	22-60

TABLE 3: Effect of oral administration of Carica papaya suspension on Hematology parameters of Placebos (Control n=50).

Laboratory Parameters	Before Treatment	After Treatment	T-pair	p-value
	Placebos (Control group	Placebos (Control group	test	
	n=50)	n=50)	value	
Hematology Parameters			T-	0.002*
Platelets	211.3 (138)	212 (152)	1.2819	
White blood cells	5.1 (10)	5.3 (10)		
Hemoglobin	13.7 (4)	13.2 (4)]	
Hematocrits	41.2 (26)	38.2 (28)		

Mean (S.D), *significant at <0.005

TABLE 4: Effect of oral administration of Carica papaya leaves suspension on hematology parameters of patients with IgG and IgM dengue positive.

Laboratory Parameters	Before Treatment group (Patients n=30)	After Treatment group (Patients n=30)	T-pair test value	p-value
Hematology Parameters	((
Platelets	118 (63)	182.6 (109)**	T 1 15 CO	0.03*
White blood cells	4.1 (12)	5.3 (9)	T-1.1569	
Hemoglobin	14.6 (3)	16.2 (3)		
Hematocrits	56.1 (41)	43.6 (46)**		

Mean (S.D), *significant at <0.005

TABLE 5: Effect of oral administration of Carica papaya leaves suspension on hematology parameters of patients with NS1 positive.

Laboratory Parameters	Before Treatment group (Patients n=20)	After Treatment group (Patients n=20)	T-pair test value	p-value
Hematology Parameters	1.1765	0.02**		
Platelets	83.6 (51)	121 (98.5)**		
White blood cells	3.7 (12)	4.9 (8)		
Hemoglobin	12.4 (5)	12.8(5)		
Hematocrits	52.6 (41)	44(46)**		

Mean (S.D), *significant at <0.005

TABLE 6: Comparative Mean of difference and SD of difference of oral administration of Caricapapaya leaves suspensions on hematology parameters in Dengue-infected patients and placebos(n=100).

Laboratory Parameters	Positive Control group (n=50)	Treatment group IgG/IgM (n=30)	Treatment group NS1 (n=10)	Suspension Treatment Medication group (n=10)	+	T-pair test value	p- value
Hematology Parameters		(8		1.2293*	0.02*
Platelets	33.1 (11.2)	41.8	69.4 (34)	120 (70) *			
		(15)					
White blood cells	1.1 (8)	3.4 (11)	2.5 (12)	4.9 (10)			
Hemoglobin	±16.4 (4)	±11.5	±13.3 (4)	±13.8 (4)			
		(6)					
Hematocrits	±9.2 (7)	±10 (7)	±4.1 (11)	±2.0 (8) *			

Mean (S.D), *significant at <0.005

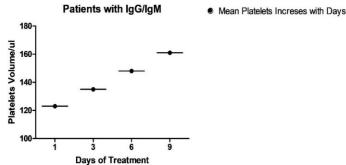
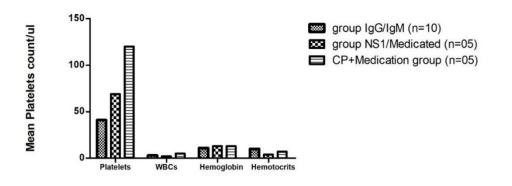


FIG. 1: Increase in the mean platelet counts during treatment.



Effect of Carica papaya on patients Blood parameters

FIG. 2: Comparative analysis of Carica papaya on Platelets of Dengue infected patient during treatment.