"Multifunctional Role of traditional medicinal plant for dengue fever in Bangladesh "-A comprehensive review

B. Pharm (Honors) Project Report

This Report in Partial Fulfillment of the Requirement for the Degree of Bachelor of Allied Health Science (Department of Pharmacy) .

Submitted To

Department of Pharmacy

Faculty of Allied Health Science

Daffodil International University

Submitted By

Student ID: 191-29-1504

Batch: 21st, Section: A

Department of Pharmacy

Faculty of Allied Health Sciences

APPROVAL

This is to certify that this project, titled "A Review Article on the Multifunctional Role of Traditional Medicinal Plants for Dengue Fever in Bangladesh," was submitted to the Department of Pharmacy, Faculty of Allied Health Science, Daffodil International University for partial fulfillment of the requirements for the degree of Bachelor of Pharmacy and approved in terms of its styles and contents.

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A C K N O W L E D G E M E N T

First and foremost, I would want to express my profound appreciation to **Allah** the Almighty for the knowledge he bestowed upon me, as well as the strength, peace of mind, and good health, and for allowing me to persevere through all the hardships to complete this project, Alhamdulillah.

I am grateful to my renowned teacher and supervisor, **Md. Mominur Rahman**, Lecturer, Department of Pharmacy, Daffodil International University, for his outstanding support, advice, and motivation in completing my project work. I will never forget his help and kindness.

I would like to specifically thank and express my gratitude to Prof. **Dr. Muniruddin Ahmed**, Professor and Head of the Department of Pharmacy at Daffodil International University, for his invaluable support and assistance in the ongoing development of the Department of Pharmacy at Daffodil International University. I am grateful for the opportunity to have you as both our instructor and the department head .

I will always be appreciative to my parents for providing me with spiritual guidance throughout my life . I couldn't do anything well without my parents' encouragement and direction.

I am incredibly grateful and fortunate to have all of my esteemed lecturers from the Department of Pharmacy at Daffodil International University providing me with ongoing support, encouragement, and direction.My gratitude and appreciation also extend to my dear friends and well-wishers who have always had my back and given me encouragement and support without condition.

D E C L A R A T I O N

I, MD.Mustafizur Rahman hereby declare that this project work entitled "Multifunctional Role of traditional medicinal plant for dengue fever in Bangladesh arrived at my observation under the supervision of Md. Mominur Rahman Lecturer, Department of Pharmacy, Daffodil International University, for the partial fulfillment of the requirements for the degree of Bachelor of Pharmacy. I further certify that, unless otherwise stated, this work has never before been submitted, in whole or in part, to any other university or institute for the purpose of conferring a different degree or professional qualification.

Supervised By

Md. Mominur Rahman

Lecturer Department of Pharmacy

Daffodil International University

Submitted By

MD.Mustafizur Rahman

ID: 191-29-1504

Batch: 21st, Section: A

Department of Pharmacy

Daffodil International University

Abstract

A serious infection spread by mosquitoes is the dengue virus (DENV). There are four different DENV serotypes (DENV1 through DENV4), however DENV5 is now also available. DENV is spread from person to person via the bite of female Aedes aegypti and Aedes albopictus mosquitoes that are infected. Recently, it has spread to more than a hundred countries, yet there are still no specific, effective treatments for it. There are no licensed vaccinations or anti-dengue medications that are useful in treating viral infections. In an effort to find organic chemicals that can be utilized as anti-dengue medications, researchers have been studying medicinal plants . Our focus is on herbal extracts since they have the potential to be safer, more effective, and less toxic than manufactured medications. In areas where dengue disease is endemic, many traditional Bangladeshi plants are used as remedies. Many traditional medicinal herbs in Bangladesh have been found to have potent antiviral properties, and some of them have even been utilized to cure patients who already had viral infections. In sufficient scientific and experimental detail, this review includes information on herbal plants that are typically found in tropical regions and that have been shown to have anti-dengue qualities. The search, which was conducted independently by the two writers and includes pertinent papers over the last 10 years, was conducted using PubMed, Google, and the Liberary Database. With future research on the plants covered in this study, we can hopefully find novel therapeutic targets for the treatment of dengue fever.

Keywords: Aedes aegypti , DENV5 , Aedes albopictus , PubMed

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Chapter One

1.1 Introduction

In the twenty-first century, dengue hemorrhagic fever is one of the most significant new tropical diseases. . In recent years, the prevalence of dengue, a virus spread by the Aedes mosquito bite, has skyrocketed. Currently, it is prevalent in over 125 countries, infects roughly 50–270 million people year, and kills a significant number of people. impact of the illness on the economy . In addition to the four DENV serotypes DENV-1, DENV-2, DENV-3, and DENV-4 that are now known, the DENV-5 serotype has also been found. Dengue viral infection is currently one of the most signification mosiquito-hbrone infection in significant humans . Dengue has been considered as the serious cause of morbidity and mortality in most tropical and subtropical areas, including Southeast and South Asia, Central and South America . Dengue fever is endemic in Bangladesh. The first serologically confirmed dengue fever (DF) patient was reported in the 1960. In Bangladesh, dengue fever is endemic. In 1960, a patient with dengue fever (DF) was reported for the first time. The disease known as "Dacca fever" (dengue) was first identified in Bangladesh .

Dengue is a fever virus that can affect people of all ages, including newborns, children, and adults, according to the World Health Organisation (WHO). 3 to 14 days after the infectious bite, the symptoms could start to show. It is not directly spread from one person to another, and symptoms can range from a slight fever to a high temperature that can be incapacitating, along with a strong headache, pain behind the eyes, muscle and joint pain, and a rash. At this time, the WHO does not suggest a dengue vaccine or any particular medicine to treat dengue. People with dengue fever should rest, drink a lot of water, and take paracetamol to lower their fever with the help of a doctor (WHO, 2016).

Due to its complicated bioactive ingredients and rich source of pharmacology, a medicinal plant has been used in traditional healing and as a source of treatment for a number of diseases. In the same way, there have been reports of many extracts and chemicals from medicinal plants that kill viruses. Also, natural goods are less dangerous and less expensive than synthetic drugs in general.

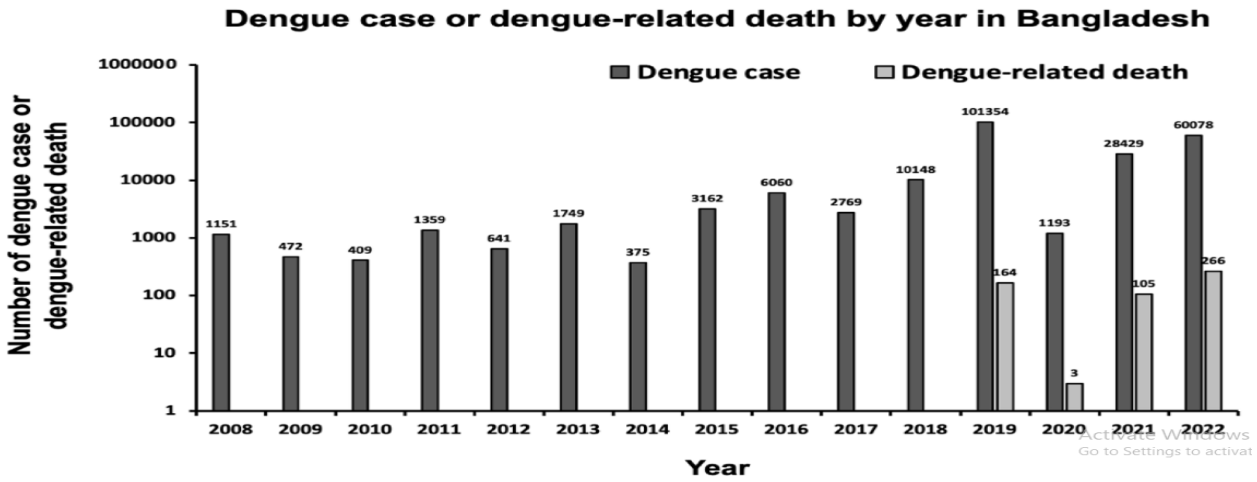


Figure-1 : 'Bangladesh's annual rate of dengue cases or deaths caused by the disease'

Methods:

We looked at the monthly records of cases sent to the Bangladesh Institute of Epidemiology, Disease Control, and Research from January 1, 2008, until December 15, 2022.

Results

According to what we found, there were 61089 proven cases of dengue in 2022, and 269 people died from it. This is the most people who died from this disease in one year since 2000. The fact that almost one-third (32.14%) of all dengue deaths in Bangladesh happened in 2022 (1 January–15 December) shows how dangerous this disease will be in the coming year. Also, we see that dengue is most likely to spread in Bangladesh during the months in the second half of the year. In 2022, Dhaka city and Chittagong will be hit the hardest (incidence: 63.07% vs. 14.42%; morality: 63.34% vs. 24.16%). This shows how important population density is in spreading this deadly disease.

Chapter Two

2.1 Purpose of the study

- The purpose of this endeavor is to fully comprehend the medical issue being investigated.
- To gain additional knowledge about the factors that could affect a potential medicinal plant cure for dengue fever.
- To gain a better understanding of the various diagnostic methods utilized to identify this illness.
- Minimize the body's toxicity.
- To get a comprehensive understanding of the illness, including its origin, symptoms, and indicators.
- To get a complete awareness of the illness, including its causes, symptoms, effects, and available nursing and medical treatments.Review the exhibition of a patient infected with dengue virus .
- The treatment of dengue disease with natural resources .

Chapter Three

3.1 Material and Method

Using PubMed, Google, and the library database, the two authors separately searched the literature. Among the search terms used were "dengue medication," "dengue herbal treatment," "dengue papaya leaf extract," and "dengue C. papaya." The PubMed database was searched for pertinent publications published within the last ten years, from 2002 to 2013. To discover any manuscripts that PubMed had not yet indexed, the same keywords were utilised in a Google search. A thorough search was also done by the authors in the library database for relevant articles that had been published in journals that weren't indexed in PubMed. For this analysis, a total of seven studies were chosen, including two randomised controlled trials, one case report, three case series, and one animal study. The study's findings will be published in 2021 or later.

Virus reservoirs

To boost viral stocks, monolayers of C6/36 cells were treated with blood serum that had been tested positive for DENV NS1. The cytopathic effect (CPE) of these cells was then observed after 6 to 8 days of incubation. At -80 °C for 20 minutes, the evident CPE flasks were gently tapped to thaw them. Three times, this process was completed. The cell debris was subsequently pelleted by spinning the cell lysate at 2000 g for 15 min. at 4°C. The DENV serotype was identified by standard RT-PCR following supernatant collection.

Crude plant extracts in water

Every single medicinal plant was gathered from the botanical garden. The entire plant was ground into a fine powder after being shade-dried. A 1:3 ratio of plant powder was soaked in dichloromethane for 18 hours at 4°C as the solvent. Whatman filter paper No. 1 was used to filter the crude extracts, which were then dried in a water bath at 80°C. The dried extracts are weighed and kept at a temperature of -20 °C until use. The leftovers from the dichloromethane extraction were then reextracted using 70% ethanol at 4°C for 18 hours with a plant-solvent ratio of 1:3. Whatman filter paper No. 1 was used to filter the crude extracts, which were then dried by evaporation in a water bath at 80°C. The dried extracts are weighed and kept at a temperature of -20 °C until use.

Predicting likely medication targets

Through careful examination of the metabolic pathways, it has been possible to identify the target genes that contribute to the host cell's induction of the anti-immune response. Therefore, the protein targets that perform the associated functions were discovered using UniProt-KB. Non-structural proteins like NSP2 and NSP3 and envelope proteins like E1 and E2 were chosen as potential CHIKV treatment targets. In addition, non-structural proteins like NS1 and NS5 as well as the envelope protein ENVP were chosen as possible DENV therapeutic targets.

The epidermis's Langerhans cells and keratinocytes become infected with the dengue virus when an infected mosquito bites a person. As a result, it spreads to numerous additional cell types, such as hepatocytes, dendritic cells, macrophages, endothelial cells, and monocytes. A larger number of pro-inflammatory cytokines and chemokines are produced by infected monocytes and dendritic cells. After an initial incubation phase of 3–7 days following DENV infection, symptoms appear as a sudden onset of fever accompanied by high viremia. Among these, some people go on to the crucial stage related to plasma leakage. This vascular leakage, which is a defining feature of DHF, results from an increase in vascular permeability brought on by endothelial dysfunction.

Study of Data

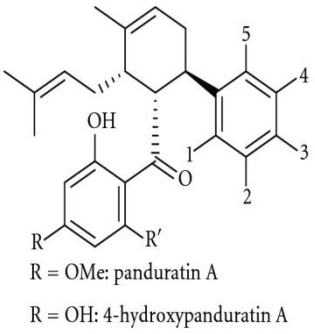
The mean percentages of cytotoxicity and infectiousness between the treatment group and the negative control were compared using a one-way ANOVA in SPSS version 23, with a p value less than 0.05 (p 0.05) being considered a statistically significant difference. The values of CC50 and IC50 were determined using the regression models obtained from concentration-percentage of viability and concentration-percentage.

Chapter Four

4.1 Overview of studies on plant species used as anti-dengue

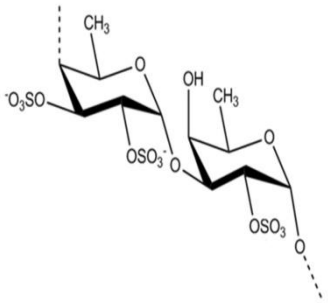
Serial No.	Scientific name	Bangladeshi name	Isolated compound	Parts of plant
1.	Andrographis paniculata Burm.f.	Green chiretta	Isolated compound - Andrographalide or methanolic extract	Stem and leaves
2.	Syzygium grenade	Bhattijam, dhakijam	Methanol, ethyl acetate, ethyl acetate-methanol	Leaves
3.	Boesenbergia rotunda	Finger Root	4-hydroxypanduratin A , panduratin A	Rhizome
4.	Carica papaya	Papaya	ethanol extract of C. papaya	leaves
5.	Alternanthera philoxeroides	Malancha	Fucoidan	Whole plants
6.	Leucaena leucocephala	Ipil-ipil	Galactomanan	Seeds
7.	Ocimum sanctum	Holy Basil, Tulsi	Methanolic extract	Leaves
8.	Azidarachta indica	Neem	4-hydroxypanduratin A , panduratin A	Leaves
9.	Cymbopogon citratus	Lemon Grass	Methanol	Whole plants
10.	Houttuynia cordata	Fish mint and heartleaf	Hyperoside	Whole plants
11.	Hylocerens undatus	Dragon fruit	Alkaloids,Phenols, Tannins, Saponin, Flavonoids,teroids,Amino acids,Glycosides	Fruits
12.	Momordica Charantia	Korola	n-hexane, ethyl acetate, and methanol	Heartwood, root-bark, Fruits
13.	Euphorbia hirta	Dudhiya, Boro Dhudi	Ethyl acetate extract	Rhizome and leaves
14.	Piper retrofractum	Chuijhal	Dichloromethane and ethanol extract	Whole plants
15.	Psidium guajava	Guava	Ethanol extract, Gallic acid, quercetin, catechin, naringin	Leaves

Table-1: Isolated compound collected in plant .

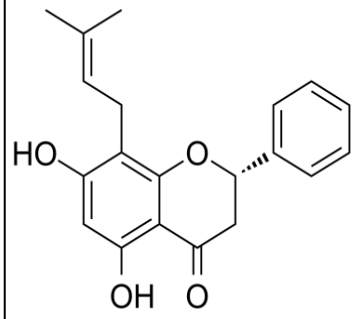


4 hydroxy panduratin A

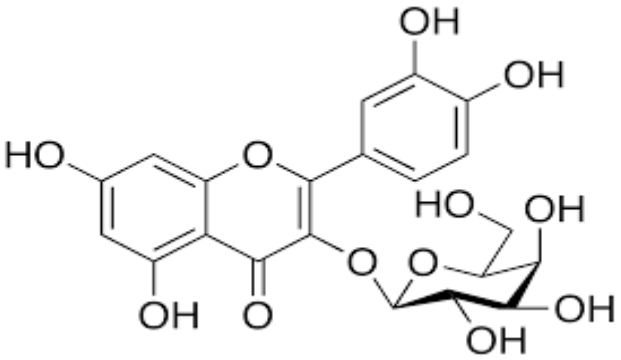
Andrographolide



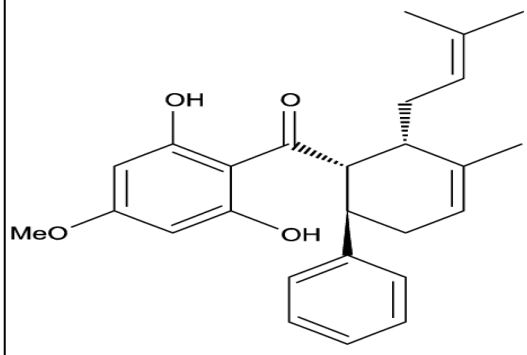
Fucoidan



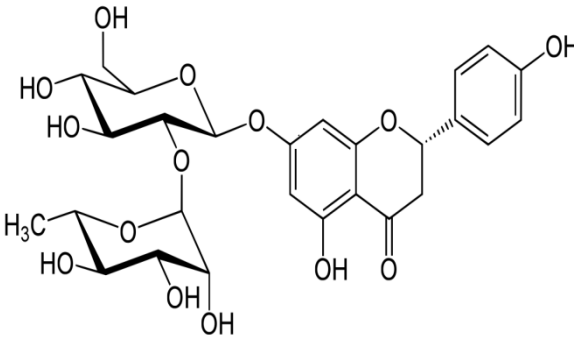
Galactomanan



Hyperoside



Panduratin A



Naringenin

Table-2: The structures of the anti-dengue chemicals that were taken from medicinal plants.

Due to their low or nonexistent side effects, the use of herbal medicine and medicinal plants to treat numerous diseases is expanding globally. The species of medicinal plants from various families that have been studied for anti-dengue action are described in the sections that follow. We also list species and their isolated chemicals that are utilised in conventional dengue therapies.

Alternanthera philoxeroides

Alternanthera philoxeroides is a member of the family Amaranthaceae. Aquatic plants are those that grow in or near water, and A. philoxeroides, also referred to as "Alligator Weed," is one of these. While originally from South America, it is now encroaching on Australia. Researchers looked at the dengue virus-inhibiting potential of A. philoxeroides extracts in vitro. Using an MTT assay, the cytotoxicity of A. philoxeroides towards C6/36 cell lines was evaluated. Coumarin extract from A. philoxeroides was the least harmful to cells, although petroleum ether extract from the same plant was the most effective against dengue virus.

Andrographis paniculata

Acanthaceae is a family of plants that includes the genus Andrographis paniculata. It is an annual plant that grows uprightly and is widely grown in southern and southeast Asia. It is indigenous to Sri Lanka and India. In Malaysia, it is referred to as "Hempedu Bumi" and has a bitter flavour. Investigations examining the methanolic extract of A. paniculata's maximum nontoxic dosage (MNTD) on Vero E6 cells were carried out in southern and southeast Asia. It is indigenous to Sri Lanka and India. In Malaysia, it is referred to as "Hempedu Bumi" and has a bitter flavour. On Vero E6 cells, tests were done to determine the methanolic extract of A. paniculata's MNTD (maximum nontoxic dosage). According to A. paniculata, the greatest dose that did not harm cells was between 0.05 and 1. The methanolic extract of A. paniculata showed the best antiviral inhibitory action on DENV-1 in an antiviral experiment based on cytotoxic effects.

Azidarachta indica

A Zingiberaceae plant, Boesenbergia rotunda belongs to this family. Chinese ginger is a herb that can be used in both cooking and medicine. It is widespread in China and Southeast Asia. The ability of several compounds extracted from B. rotunda to obstruct the dengue virus protease was tested on DENV-2. The cyclohexenyl chalcone derivatives of B. rotunda, 4-hydroxypanduratin A (1) and panduratin A (2), showed effective competitive inhibitory effects against DENV-2 NS3 protease with Ki values of 21 lM and 25 lM, respectively. The fact that 4-hydroxypanduratin A has a low Ki value suggests that it can in vitro inhibit DENV-2 NS3 protease.

Boesenbergia rotunda

The Boesenbergia rotunda is a member of the Zingiberaceae family. Chinese ginger is a herb that has both culinary and medicinal uses. It can be found all over Southeast Asia and China. On DENV-2, various chemicals isolated from B. rotunda were investigated for their ability to block the dengue virus protease. With Ki values of 21 lM and 25 lM, respectively, the cyclohexenyl chalcone derivatives of B. rotunda, 4-hydroxypanduratin A (1) and panduratin A (2), demonstrated good competitive inhibitory actions towards DENV-2 NS3 protease. The low value of Ki indicates that 4-hydroxypanduratin A has the ability to in vitro inhibit DENV-2 NS3 protease.

Carica papaya

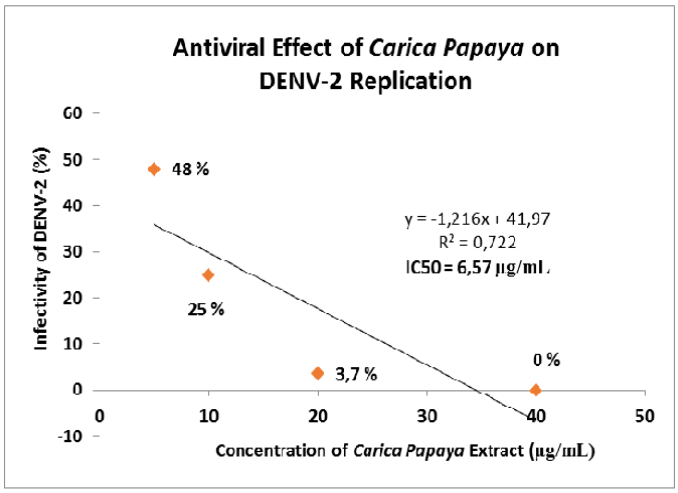


Figure-2: DENV inhibition following treatment with C. papaya: Linear regression curve .

The Carica papaya is a member of the Caricaceae family. It is an upright, quickly-growing, unbranched tree or shrub that is native to Central America and is grown for its edible fruits in Mexico and most tropical nations. Traditionally, C. papaya leaf has been used to treat DF. The leaf's ability to combat DF has been researched. In blood samples from a 45-year-old patient bitten by carrier mosquitoes, the aqueous extract of this plant's leaves showed potential effectiveness against DF by increasing the platelet (PLT) count, white blood cells (WBC), and neutrophils (NEUT). The PLT count increased from 55 9 10 3 /l to 168 9 10 3 /l, the WBC from 3.7 9 10 3 /l to 7.710 3 /l, and the NEUT from 46.0 to 78.3% after 5 days of oral administration of 25 mL of aqueous extract of C. papaya leaves to the patient twice daily. Increased platelets may result in less bleeding, preventing the development of the severe sickness known as DHF.

Cladogynos orientalis

Cladogynos orientalis is a member of the Euphorbiaceae family. It is a 2 m tall shrub with white stellate hairs that can be found throughout Southeast Asia, Malaysia, and Thailand. A Thai medicinal plant called Cladogynos orientalis was tested for its ability to inhibit dengue virus growth in vitro. The MTT technique was used to examine the dichloromethane ethanol extract of C. orientalis for anti-dengue activity against DENV-2 in Vero cells. According to the findings, DENV-2 was inhibited by 34.85% by the ethanol extract of C. orientalis at a concentration of 12.5 lg mL-1. Additionally, C. orientalis showed a 2.9% inactivated viral particle activity at a dose of 100 lg mL-1.

Cladosiphon okamuranus

Cladosiphon okamuranus is a member of the Chordariaceae family. It is a type of brown seaweed that is indigenous to Okinawa, Japan. Cladosiphon okamuranus fucoidan (3), a sulfated polysaccharide, was discovered to possibly prevent DENV-2 infection . A focus-forming assay was used to examine the viral infection in BHK-21 cells. Compared to untreated cells, fucoidan reduced infectivity by 20% at 10 lg mL-1. A carboxy-reduced fucoidan, on the other hand, diminished the inhibitory action on DENV2 infection by converting glucuronic acid to glucose.

Cryptonemia crenulata

The Halymeniaceae family includes the plant Cryptonemia crenulata. It is a marine species that can be found in the Pacific Islands, the Indian Ocean Islands, Southeast Asia, the Western Atlantic Islands, South America, Africa, and the Atlantic Islands. The reference polysaccharides heparin and DS8000 had IC 50 values of 1.9 and 0.9 lg mL-1, respectively, whereas the sulfated polysaccharides from Cryptonemia crenulata, galactan (4), had an IC 50 value of 1.0 lg mL-1 and was a selective inhibitor of DENV-2 multiplication in Vero cells. However, the substance is completely inert against DENV-1 and has no antiviral impact at all against DENV-3 and DENV-4. In comparison to treatment just during internalization (EC 50 = 5.5 0.7 lg mL-1), the inhibitory effect of C2S-3 against DENV-2 was slightly stronger when treatment was through adsorption (EC 50 = 2.5 0.1 lg mL-1) . Therefore, the inclusion of C2S-3 at both the adsorption and internalization stages resulted in a greater inhibitory impact.

Cymbopogon citratus

Cymbopogon citratus is a member of the Poaceae family. It is a tropical plant from Southeast Asia that is a kind of grass known as lemon grass. Based on the level of inhibition of DENV-1-infected Vero E6 cells and its cytopathic effects, Cymbopogon citratus was found to have antiviral activity. On DENV-1, the methanolic extract of C. citratus had a marginally inhibiting effect. With the use of an inhibition assay using the MTT method, this finding was further validated. C. citrate didn't exhibit any appreciable inhibition, though. The lowest MNTD content was also found in C. citratus, which was 0.001 mg mL-1. The plant C. citratus was discovered to be quite cytotoxic, with a maximal cytotoxicity of 0.075 mg mL-1.

Euphorbia hirta

The family Euphorbiaceae includes the Euphorbia hirta. In Java, Sumatra, Peninsular Malaysia, the Philippines, and Vietnam, it is a typical weed in garden beds, garden walkways, and wastelands. In the Philippines, DF is treated using a traditional medication known as gatas-gatas, a water decoction of leaves from Euphorbia hirta. After 24 hours, internal bleeding will stop and dengue fever will be treated. However, the exact mechanism of action is still unknown, and researchers are currently looking into the drug's antiviral capabilities and capacity to raise blood platelets. To treat DF, tea made from boiled E. hirta leaves is utilized.

Flagellaria indica

The family Flagellariaceae includes Flagellaria indica. It is a strong perennial climber that may be found in many of the Old World's tropical and subtropical areas, including Polynesia, Australia, Southeast Asia, and India. In Vero cells, Flagellaria indica was tested for its ability to prevent dengue. According to the antiviral assay results, 12.5 lg mL-1 of the plant's ethanol extract caused a 45.52% suppression of DENV-2 to be seen in vitro. The cytotoxicity of F. indica was assessed using MTT assays. The ethanol extract of F. indica had a CC50 of 312 lg mL-1. Therefore, our research suggests that F. indica may significantly affect DENV.

Gymnogongrus griffithsiae

Gymnogongrus griffithsiae belongs to the family Phyllophoraceae. It is a red seaweed found in Ireland, Europe, the Atlantic Islands, North America, South America, the Caribbean Islands, Africa, Southwest and Southeast Asia, Australia, and New Zealand. The inhibitory properties against DENV-2 of the sulfur-flavonoid polysaccharide from Gymnogongrus griffithsiae, kappa carrageenan, were evaluated in Vero cells. The compound effectively inhibits DENV-2 multiplication at the IC50 value of 0.9 lg mL-1, which is the same as the IC50 value for the commercial polysaccharide DS8000. However, the compound has a lower antiviral effect against DENV-3 and DENV-4 and is totally inactive against DENV-1.

Gymnogongrus torulosus

Gymnogongrus torulosus belongs to the family Phyllophoraceae. It is a red seaweed found in Australia and New Zealand. Gymnogongrus torulosus was investigated for its in vitro antiviral properties against DENV-2 in Vero cells . Galactan extracted from this plant was active against DENV-2, with IC50 values in the range of 0.19–1.7 lg mL-1.

Hippophae rhamnoides

The family Elaeagnaceae includes the species Hippophae rhamnoides. It is a deciduous shrub that can be found in Asia up to Japan and the Himalayas, as well as in Europe, including Britain, from Norway south and east to Spain. Human macrophages generated from contaminated blood were used to study the anti-dengue efficacy of extracts of Hippophae rhamnoides leaves. The results demonstrated that dengue-infected cells treated with H. rhamnoides leaf extracts were

able to sustain cell viability on par with ribavirin, a commercially available anti-viral treatment, as well as a drop and increase, respectively, in TNF-a and IFN-c. Further evidence of H. rhamnoides leaf extract's anti-dengue effectiveness came from a decline in plaque counts following treatment of infected cells.

Houttuynia cordata

Houttuynia cordata belongs to family Saururaceae. It is herbaceous perennial flowering plants growing between 20 and 80 cm, and is native to Japan, Korea, Southern China and Southeast Asia. Ethanol extract from Houttuynia cordata revealed an anti-dengue activity with 35.99 % inhibition against DENV-2 in Vero cells at a concentration of 1.56 lg mL-1. Aqueous extract of H. cordata showed effective inhibitory action against DENV-2 through direct inactiva- tion of viral particles before infection of the cells. A concentration of 100 lg mL-1 also effectively protects the cells from viral entry and inhibits virus activities after adsorption. HPLC analysis of H. cordata extract indicated that hyperoside was the predominant bioactive com- pound, and was likely to play a role in this inhibition.

Leucaena leucocephala

Leucaena leucocephala belongs to family Fabaceae. It is a species of Mimosoid tree indigenous throughout Southern Mexico and Northern Central America and the West Indies from the Bahamas and Cuba to Trinidad and Tobago. Galactomannans (7) extracted from seeds of Leucaena leucocephala have demonstrated activity against yellow fever virus (YFV) and DENV-1 in vitro and in vivo. Galactomannans are polysaccharides consisting of a man- nose backbone with galactose side groups, more specifically their structure consists of a main chain of (1 ? 4)-linked b- D-mannopyranosyl units substituted by a- D-galactopyr- anosyl units. L. leucocephala show protection against death in 96.5 % of YFV-infected mice. In vitro experiments with DENV-1 in C6/36 cell culture assays showed that the concentration producing a 100-fold decrease in virus titer of DENV-1 was 37 mg L-1 .

Lippia alba and Lippia citriodora

The Verbenaceae family includes Lippia alba and Lippia citriodora. South Texas, Mexico, the Caribbean, Central America, and South America are all home to these beautiful plants. On dengue virus serotype replication in Vero cells, essential oils from Lippia alba and Lippia citriodora had a significant suppressive effect. L. alba oil was shown to have a 50% reduction in virus plaque number values at concentrations between 0.4 and 32.6 lg mL-1, whereas L. citriodora oil had IC 50 values between 1.9 and 33.7 lg mL-1. The virucidal efficacy of L. citriodora essential oil against DENV-1, 2, and 3 was comparable to but weaker than against DENV-4, whereas L. alba essential oil was more efficient against DENV-2 than other serotypes. At 100 lg mL-1, it was shown that L. alba essential oil completely eliminated YFV yield.

Meristiella gelidium

The species Meristiella gelidium is a member of the Solieriaceae family. It is a marine species that can be found in South America, the Caribbean Islands, the North American mainland, and the Atlantic Islands. It was determined whether or not DENV-2 could be inhibited by the antiviral properties of kappa carrageenan in Meriella gelidium. The carrageenans that were isolated from M. gelidium had an IC 50 that fell somewhere in the range of 0.14–1.6 lg mL-1. According to the findings, both the M. gelidium extract and the fraction that was generated from it were significantly more effective at inhibiting DENV-2 than the reference polysaccharides, which were heparin and DS 8000.

Mimosa scabrella

Fabaceae is the family that includes the Mimosa scabrella plant. It is a native to the chilly, subtropical plateaus of Southeastern Brazil and has a rapid growth rate, reaching heights of 15–20 metres and having a diameter of up to 50 centimetres. Galactomannans derived from the seeds of Mimosa scabrella have been shown to have antiviral action in vitro and in vivo against the viruses YFV and DENV-1. In a study with mice infected with YFV, M. scabrella was shown to prevent death in 87.7% of the animals. Experiments conducted in vitro with DENV-1 in C6/36 cell culture assays revealed that a concentration of 347 mg L-1 resulted in a reduction in the virus titer of DENV-1 by a factor of one hundred.

Momordica charantia

The Momordica charantia plant is a member of the Cucurbitaceae family. It is a tropical and subtropical vine that can be found throughout Asia, Africa, and the Caribbean and is also known by the names bitter melon and peria (Malaysia). In vitro studies were conducted to determine the MNTD of the methanolic extract of Momordica charantia against Vero E6 cells. M. charantia found that a dose of 0.20 mg mL-1 was the highest level at which cells were not killed. In a test for antiviral activity based on cytotoxic effects, the methanolic extract of M. charantia demonstrated an inhibitory impact against DENV-1.

Ocimum sanctum

The Ocimum sanctum plant is a member of the Labiatae family. It is a shrub that is native to the tropical regions of both Asia and the Americas, and it is also an aromatic herb. Tea, which is typically made by boiling the leaves of the Ocimum sanctum plant, can be used as a preventative and medicinal treatment for DF. An analysis was done to determine the minimal non-toxic dose (MNTD) of a methanolic extract of O. sanctum for use against Vero E6 cells in vitro. However, there was not a discernible change in MNTD between O. sanctum and the control group. Based on its cytotoxic properties, the methanolic extract of O. sanctum showed a mild inhibitory impact on DENV-1.

Piper retrofractum

Piper retrofractum is a member of the Piperaceae family of plants. The fruit of this blooming vine, which is native to Southeast Asia, is the primary reason why it is cultivated in Indonesia and Thailand. Piper retrofractum was tested for its anti-dengue activity in Vero cells in an in vitro setting. The MTT assay was utilised to evaluate the dichloromethane ethanol extract's potential to prevent DENV-2 infection in cells. At a concentration of 100 lg mL-1, the P. retrofractum ethanol extract demonstrated an inactivated viral particle activity level of 84.93%. Previous research has demonstrated that an aqueous extract of long pepper, also known as P. retrofractum, possesses the highest level of efficacy when it comes to warding off mosquito larvae.

Psidium guajava

The Psidium guajava tree is a member of the Myrtaceae family. Native to Mexico, the Caribbean, Central America, and South America, this evergreen shrub or small tree can also be found in those regions. In most of the world's tropical and subtropical regions, it is one of the most common crops cultivated. It has been demonstrated through in vitro testing that the Psidium guajava leaf extract can prevent the growth of the dengue virus. Platelet counts were boosted to 100.000/mm3 in roughly 16 hours thanks to the usage of water that had guava leaves that had been boiled in it. This was done to prevent bleeding in DHF. In cases of deficiency of platelets, the ripe fruit or juice of P. guajava has been shown to have curative effects and can be used to treat the condition.

Quercus lusitanica

The Quercus lusitanica tree is a member of the Fagaceae family. Oaks of this species can be found naturally occurring in Morocco, Portugal, and Spain. It was discovered that the extract of Quercus lusitanica had a significant effect in inhibiting the reproduction of DENV-2 in C6/36 cells. The absence of cytotoxic effects indicates that the methanol extract of the seeds was able to totally inhibit the TCID 50 of the virus at its maximal non-toxic dosage of 0.25 mg mL-1. This inhibition ranged from 10 to 1,000 fold. With 10 TCID 50 of the virus, an extremely low dose of Q. lusitanica (0.032 mg mL-1) was able to completely inhibit the virus. After treatment with the extract, proteomics techniques were utilised to show that the action of Q. lusitanica was to downregulate NS1 protein expression in infected c6/36 cells. This was demonstrated by the fact that the extract inhibited the expression of the protein.

Rhizophora apiculata

The Rhizophoraceae family is comprised of the Rhizophora apiculata species. In Australia (Queensland and the Northern Territory), Guam, India, Indonesia, Malaysia, Micronesia, New Caledonia, Papua New Guinea, the Philippines, Singapore, the Solomon Islands, Sri Lanka, Taiwan, the Maldives, Thailand, and Vietnam, there is a kind of mangrove tree that may grow to

a height of up to 20 metres. There have been reports of the ethanolic extract of Rhizophora apiculata having anti-dengue effects when tested on DENV-2 in Vero cells. At doses of 12.5 and 100 lg mL-1, respectively, R. apiculata displayed inhibitory action and an inactivate particle activity of 56.14% and 41.5%d viral.

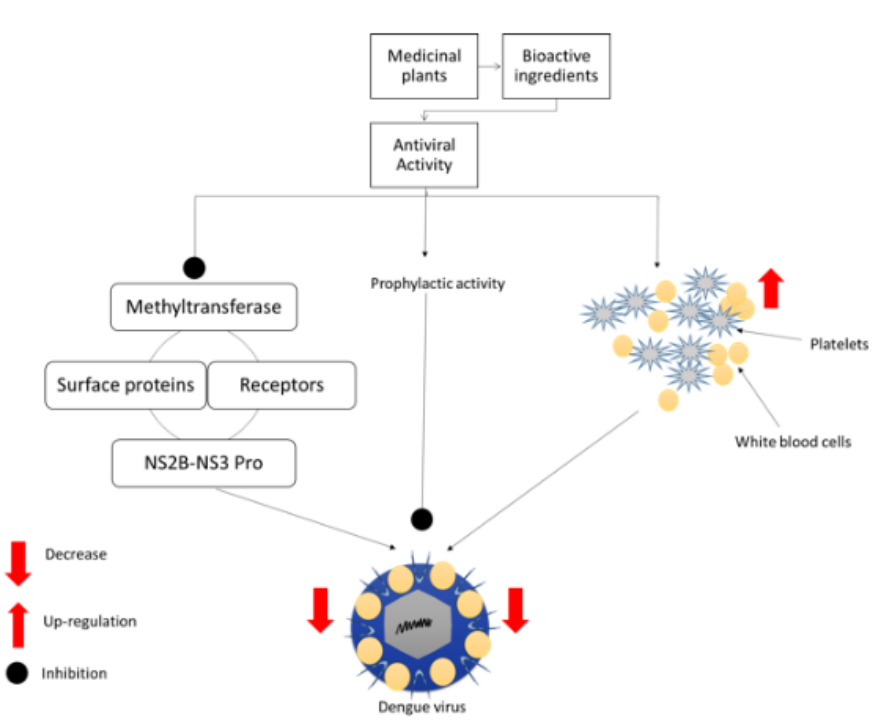


Figure-3: 'Anti-dengue effective of Medicinal plants.'

The figure above indicates that medicinal plants through their bio-functional ingredients can potentiate the inhibition viral enzymes such as methyltransferase and NS2B-NS3-Pro, other pharmacological benefits include prophylactic impact and rising platelets as well as WBCs count.

Chapter Five

5.1 Results & Discussion

Exploration to find the antiviral drugs to DENV has been developing rapidly and profoundly in many countries. Herbal medicines are a potential source for the development of new antiviral drugs, since they can be selected based on their ethno-medicinal use, for example, against infectious diseases . These plants produce a variety of phytochemical constituents with the potential to inhibit viral replication, and compounds from natural sources are of interested as possible sources to control viral infection.

From the various reports published in scientific literature, it appears that C. papaya L. leaf extract does have beneficial properties in dengue. It has been shown to bring about a rapid increase in platelet count. This could be possibly attributed to its membrane-stabilizing property. The flavonoids and other phenols present in the extract have been suggested to provide

the beneficial effects. One study found that the leaves of papaya plant are rich in several minerals. The researchers suggested that these minerals may balance the mineral deficiency caused by the virus and strengthen the immune cells against it . Earlier observations confirmed that flavonoids in plants and their derivatives possess antiviral activity . thus, in this study, the methanolic extracts were standardised based on total flavonoids content prior to antiviral assay. Another plant that recorded a low level of total flavonoids was the A. paniculata, which was examined for this study. Anti-atherosclerotic flavonoids such as 7-O methylwogonin, apigenin, onysilin, and 3,4-dicaffeoylquinic acid have been extracted from A. paniculata. These flavonoids have been tested and found to be effective. C. limon, on the other hand, had the third highest overall average concentration of flavonoids (33%) out of the six plants that were investigated. Flavonoids such as hesperidin, diosmin, and eriocitrin can be found in quite high concentrations in C. limon. The flavonoids citrunobin, cit-flavanone, and lonchocarpol-A were found in the root bark of Citrus species.

Current treatments for dengue

There is no effective drug or vaccine for the treatment of this potentially fatal disease. In 2020, the vaccine called Dengvaxia was developed and licensed for use in 11 countries. However, it is not effective for the protection from infection of all the four serotypes of dengue . There are multiple anti-DENV agents in various stages of development. Some are direct-acting antivirals namely RNA polymerase inhibitors, nucleoside analogues and protease inhibitors. Some anti-dengue agents are developing that target host-mediated translational modifications such as α glucosidase inhibitors. Appropriate dose of paracetamol is recommended for relieving the DF, and ibuprofen, aspirin, and naproxen (Aleve) should be avoided. Fluids replacement are very effective when patients suffering with dehydration. Fluid balance, electrolytes, nursing care, and blood clotting parameters should be taken care of a dengue patient. Patients should be hospitalized as per expand of symptoms.

Chapter Six

Summary and conclusion

The existing study focuses the information on numerous plant components and extract kinds that are used to treat dengue. However, some plants that have not yet been fully studied might have a wide range of therapeutic uses. Finding better, more potent, and less harmful anti-dengue medications requires the development of novel anti-dengue products from bioactive chemicals. In light of this, any in-depth investigation into the potential of plants with separated active components that have demonstrated anti-dengue action should involve additional in vitro and in vivo animal research, as well as toxicity and clinical investigations.

Uncertainty is surely caused by the lack of particular target treatments for dengue in the thoughts of those who are infected. Patients are not the only ones who become anxious in this situation; the professionals who treat the patients are also under strain. Additionally, it assisted Bangladeshi society in identifying alternative dengue prevention and treatment options. In different regions of Bangladesh, several plants and their preparations have been traditionally utilised to treat dengue. Numerous literary works have described how local communities and herbal healers use plants to combat dengue.When we analyzed scientific validation studies of evolving herbal alternatives, all the three methods and models (in vitro, in vivo and clinical trial) of efficacy testing against dengue viruses have been adapted by the Bangladesh researchers. However, number of such scientific validation for plants effective against dengue are very less in India, below double digits. About twenty- two plants from all over Bangladesh were found in literature search which were recommended for their use against dengue. However, only two to three plants/extracts have been tested scientifically and also have shown evidences of efficacy. There is a need to search more such herbal formulations which are being practiced at local level, document them properly and validate them scientifically to confirm efficacy, understand mechanistic action and safety so that they can be exploited for their anti-dengue potential. The potential herbal formulations being used by the communities and healers are the low hanging fruits which may provide alternative or adjuvant therapy if proper validation, value addition and product develop- ment steps are followed.

Moreover, such discoveries may lead to the development of highly efficient and safe anti-dengue treatments. However, to identify potential anti-dengue plants or compounds, knowledge of the mechanisms of virus infection need to be understood inorder to facilitate the search for and devel- opment of the most appropriate drugs. Further research is needed to determine how to target the most appropriate stages to prevent the spread of virus infection. Focusing on each phase in the life cycle of the virus, new compounds could prevent (1) infection of host cells, (2) the viral maturation process, (3) synthesis of viral RNA, or (4) the spread of viral particles.

Ethical Approval: No ethical approval needed.

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"Multifunctional Role of traditional medicinal plant for dengue fever in Bangladesh ''-A comprehensive review B. Pharm (Honors) Project Report This Report in Partial Fulfillment of the Requirement for the Degree of Bachelor of Allied Health Science (Department of Pharmacy) . Submitted To Department of Pharmacy Faculty of Allied Health Science Daffodil International University Submitted By Student ID: 191-29-1504 Batch: 21st, Section: A Department of Pharmacy Faculty of Allied Health Sciences APPROVAL This is to certify that this project, titled "A Review Article on the Multifunctional Role of Traditional Medicinal Plants for Dengue Fever in Bangladesh," was submitted to the Department of Pharmacy, Faculty of Allied

Health Science, Daffodil International University for partial fulfillment of the requirements for the degree of Bachelor of Pharmacy and approved in terms of its styles and contents. BOARD OF EXAMINERS Chairman _____ Professor Dr. Muniruddin Ahmed Professor and Head Department of Pharmacy Daffodil International University Examiners _ Internal Examiner-1 _____ Internal Examiner-2 _____ External i ACKNOWLEDGEMENT First and foremost, I would want to express my profound appreciation to Allah the Almighty for the knowledge he bestowed upon me, as well as the strength, peace of mind, and good health, and for allowing me to persevere through all the hardships to complete this project, Alhamdulillah. I am grateful to my renowned teacher and supervisor, Md. Mominur Rahman, Lecturer, Department of Pharmacy, Daffodil International University, for his outstanding support, advice, and motivation in completing my project work. I will never forget his help and kindness. 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My gratitude and appreciation also extend to my dear friends and well-wishers who have always had my back and given me encouragement and support without condition. ii DECLARATION I, MD.Mustafizur Rahman hereby declare that this project work entitled "Multifunctional Role of traditional medicinal plant for dengue fever in Bangladesh arrived at my observation under the supervision of Md. Mominur Rahman Lecturer, Department of Pharmacy, Daffodil International University, for the partial fulfillment of the requirements for the degree of Bachelor of Pharmacy. I further certify that, unless otherwise stated, this work has never before been submitted, in whole or in part, to any other university or institute for the purpose of conferring a different degree or professional qualification. Supervised By Md. Mominur Rahman Lecturer Department of Pharmacy Daffodil International University Submitted By MD.Mustafizur Rahman ID: 191-29-1504 Batch: 21st, Section: A Department of Pharmacy Daffodil International University iii Abstract A serious infection spread by mosquitoes is the dengue virus (DENV). There are four different DENV serotypes (DENV1 through DENV4), however DENV5 is now also available. DENV is spread from person to person via the bite of female Aedes aegypti and Aedes albopictus mosquitoes that are infected. Recently, it has spread to more than a hundred countries, yet there are still no specific, effective treatments for it. There are no licensed vaccinations or anti-dengue medications that are useful in treating viral infections. In an effort to find organic chemicals that can be utilized as anti-dengue medications, researchers have been studying medicinal plants . Our focus is on herbal extracts since they have the potential to be safer, more effective, and less toxic than manufactured medications. In areas where dengue disease is endemic, many traditional Bangladeshi plants are used as remedies. Many traditional medicinal herbs in Bangladesh have been found to have potent antiviral properties, and some of them have even been utilized to cure patients who already had viral infections. In sufficient scientific and experimental detail, this review includes information on herbal plants that are typically found in tropical regions and that have been shown to have anti-dengue qualities. The search, which was conducted independently by the two writers and includes pertinent papers over the last 10 years, was conducted using PubMed, Google, and the Liberary Database. With future research on the plants covered in this study, we can hopefully find novel therapeutic targets for the treatment of dengue fever. Keywords: Aedes aegypti , DENV5 , Aedes albopictus , PubMed iv Contents Chapter one Page No 1.1 Introduction 02 1.2 Dengue case 03 Chapter Two Purpose of the study 2.1 Purpose of the study 04-05 Chapter Three Material and Method 3.1 Material and Method 06-08 Chapter Four Overview of studies on plant species used as anti-dengue 4.1 Overview of studies on plant species used as anti-dengue 4.2 Isolated compound collected in plant 4.3 The structures of the anti-dengue chemicals that were taken from medicinal plants 4.4 Details of plants 09-10 10 11 12-19 Chapter Five Results & Discussion 5.1 Results & Discussion 20-21 5.2 Current treatments for dengue 21 Chapter Six Summary and conclusion v 6.1 Summary and conclusion 22-23 Chapter Seven Reference 24 7.1 Reference List of figures Figure-1 : 'Dengue case or dengue-related death by year in Bangladesh.' Figure-2: 'Linear Regression curve of the inhibition of DENV after treated with C. papaya.' Figure-3: 'Anti-dengue effective of Medicinal plants.' List of tables Table-1: Isolated compound collected in plant . Table-2: The structures of the anti-dengue chemicals that were taken from medicinal plants. vi Chapter One 1 1.1 Introduction In the twenty-first century, dengue hemorrhagic fever is one of the most significant new tropical diseases. . In recent years, the prevalence of dengue, a virus spread by the Aedes mosquito bite, has skyrocketed. Currently, it is prevalent in over 125 countries, infects roughly 50–270 million people year, and kills a significant number of people. impact of the illness on the economy . In addition to the four DENV serotypes DENV-1, DENV-2, DENV-3,

and DENV-4 that are now known, the DENV-5 serotype has also been found. Dengue viral infection is currently one of the most signification mosiquito-hbrone infection in significant humans . Dengue has been considered as the serious cause of morbidity and mortality in most tropical and subtropical areas, including Southeast and South Asia, Central and South America . Dengue fever is endemic in Bangladesh. The first serologically confirmed dengue fever (DF) patient was reported in the 1960. In Bangladesh, dengue fever is endemic. In 1960, a patient with dengue fever (DF) was reported for the first time. The disease known as "Dacca fever" (dengue) was first identified in Bangladesh . Dengue is a fever virus that can affect people of all ages, including newborns, children, and adults, according to the World Health Organisation (WHO). 3 to 14 days after the infectious bite, the symptoms could start to show. It is not directly spread from one person to another, and symptoms can range from a slight fever to a high temperature that can be incapacitating, along with a strong headache, pain behind the eyes, muscle and joint pain, and a rash. At this time, the WHO does not suggest a dengue vaccine or any particular medicine to treat dengue. People with dengue fever should rest, drink a lot of water, and take paracetamol to lower their fever with the help of a doctor (WHO, 2016). Due to its complicated bioactive ingredients and rich source of pharmacology, a medicinal plant has been used in traditional healing and as a source of treatment for a number of diseases. In the same way, there have been reports of many extracts and chemicals from medicinal plants that kill viruses. Also, natural goods are less dangerous and less expensive than synthetic drugs in general. Figure-1 : 'Bangladesh's annual rate of dengue cases or deaths caused by the disease' Methods: We looked at the monthly records of cases sent to the Bangladesh Institute of Epidemiology, Disease Control, and Research from January 1, 2008, until December 15, 2022. Results According to what we found, there were 61089 proven cases of dengue in 2022, and 269 people died from it. This is the most people who died from this disease in one year since 2000. The fact that almost one-third (32.14%) of all dengue deaths in Bangladesh happened in 2022 (1 January– 15 December) shows how dangerous this disease will be in the coming year. Also, we see that dengue is most likely to spread in Bangladesh during the months in the second half of the year. In 2022, Dhaka city and Chittagong will be hit the hardest (incidence: 63.07% vs. 14.42%; morality: 63.34% vs. 24.16%). This shows how important population density is in spreading this deadly disease. Chapter Two 2.1 Purpose of the study ‣ The purpose of this endeavor is to fully comprehend the medical issue being investigated. ‣ To gain additional knowledge about the factors that could affect a potential medicinal plant cure for dengue fever. ‣ To gain a better understanding of the various diagnostic methods utilized to identify this illness. ‣ Minimize the body's toxicity. ‣ To get a comprehensive understanding of the illness, including its origin, symptoms, and indicators. ‣ To get a complete awareness of the illness, including its causes, symptoms, effects, and available nursing and medical treatments.Review the exhibition of a patient infected with dengue virus . ‣ The treatment of dengue disease with natural resources . Chapter Three 3.1 Material and Method Using PubMed, Google, and the library database, the two authors separately searched the literature. Among the search terms used were "dengue medication," "dengue herbal treatment," "dengue papaya leaf extract," and "dengue C. papaya." The PubMed database was searched for pertinent publications published within the last ten years, from 2002 to 2013. To discover any manuscripts that PubMed had not yet indexed, the same keywords were utilised in a Google search. A thorough search was also done by the authors in the library database for relevant articles that had been published in journals that weren't indexed in PubMed. For this analysis, a total of seven studies were chosen, including two randomised controlled trials, one case report, three case series, and one animal study. The study's findings will be published in 2021 or later. Virus reservoirs To boost viral stocks, monolayers of C6/36 cells were treated with blood serum that had been tested positive for DENV NS1. The cytopathic effect (CPE) of these cells was then observed after 6 to 8 days of incubation. At -80 °C for 20 minutes, the evident CPE flasks were gently tapped to thaw them. Three times, this process was completed. The cell debris was subsequently pelleted by spinning the cell lysate at 2000 g for 15 min. at 4°C. The DENV serotype was identified by standard RT-PCR following supernatant collection. Crude plant extracts in water Every single medicinal plant was gathered from the botanical garden. The entire plant was ground into a fine powder after being shade-dried. A 1:3 ratio of plant powder was soaked in dichloromethane for 18 hours at 4°C as the solvent. Whatman filter paper No. 1 was used to filter the crude extracts, which were then dried in a water bath at 80°C. The dried extracts are weighed and kept at a temperature of -20 °C until use. The leftovers from the dichloromethane extraction were then reextracted using 70% ethanol at 4°C for 18 hours with a plant-solvent ratio of 1:3. Whatman filter paper No. 1 was used to filter the crude extracts, which were then dried by evaporation in a water bath at 80°C. The dried extracts are weighed and kept at a temperature of -20 °C until use. Predicting likely medication targets Through careful examination of the metabolic pathways, it has been possible to identify the target genes that contribute to the host cell's induction of the anti-immune response. Therefore, the protein targets that perform the associated functions were discovered using UniProt-KB. Non-structural proteins like NSP2 and NSP3 and envelope proteins like E1 and E2 were chosen as potential CHIKV treatment targets. In addition, non-structural proteins like NS1 and NS5 as well as the envelope protein ENVP were chosen as

possible DENV therapeutic targets. The epidermis's Langerhans cells and keratinocytes become infected with the dengue virus when an infected mosquito bites a person. As a result, it spreads to numerous additional cell types, such as hepatocytes, dendritic cells, macrophages, endothelial cells, and monocytes. A larger number of pro-inflammatory cytokines and chemokines are produced by infected monocytes and dendritic cells. After an initial incubation phase of 3–7 days following DENV infection, symptoms appear as a sudden onset of fever accompanied by high viremia. Among these, some people go on to the crucial stage related to plasma leakage. This vascular leakage, which is a defining feature of DHF, results from an increase in vascular permeability brought on by endothelial dysfunction. Study of Data The mean percentages of cytotoxicity and infectiousness between the treatment group and the negative control were compared using a one-way ANOVA in SPSS version 23, with a p value less than 0.05 (p 0.05) being considered a statistically significant difference. The values of CC50 and IC50 were determined using the regression models obtained from concentration-percentage of viability and concentration-percentage. Chapter Four 4.1 Overview of studies on plant species used as anti-dengue Serial No. Scientific name Bangladeshi name Isolated compound Parts of plant 1. Andrographis paniculata Burm.f. Green chiretta Isolated compound - Andrographalide or methanolic extract Stem and leaves 2. Syzygium grenade Bhattijam, dhakijam Methanol, ethyl acetate, ethyl acetate- methanol Leaves 3. Boesenbergia rotunda Finger Root 4-hydroxypanduratin A , panduratin A Rhizome 4. Carica papaya Papaya ethanol extract of C. papaya leaves 5. Alternanthera philoxeroides Malancha Fucoidan Whole plants 6. Leucaena leucocephala Ipil-ipil Galactomanan Seeds 7. Ocimum sanctum Holy Basil, Tulsi Methanolic extract Leaves 8. Azidarachta indica Neem 4-hydroxypanduratin A , panduratin A Leaves 9. Cymbopogon citratus Lemon Grass Methanol Whole plants 10. Houttuynia cordata Fish mint and heartleaf Hyperoside Whole plants 11. Hylocerens undatus Dragon fruit Alkaloids,Phenols, Tannins, Saponin, Flavonoids,teroids,Amino acids,Glycosides Fruits 12. Momordica Charantia Korola n-hexane, ethyl acetate, and methanol Heartwood, root-bark, Fruits 13. Euphorbia hirta Dudhiya, Boro Dhudi Ethyl acetate extract Rhizome and leaves 14. Piper retrofractum Chuijhal Dichloromethane and ethanol extract Whole plants 15. Psidium guajava Guava Ethanol extract, Gallic acid, quercetin, catechin, naringin Leaves Table-1: Isolated compound collected in plant . 4 hydroxy panduratin A Andrographolide Fucoidan Galactomanan Hyperoside Panduratin A Naringenin Table-2: The structures of the anti-dengue chemicals that were taken from medicinal plants. Due to their low or nonexistent side effects, the use of herbal medicine and medicinal plants to treat numerous diseases is expanding globally. The species of medicinal plants from various families that have been studied for anti-dengue action are described in the sections that follow. We also list species and their isolated chemicals that are utilised in conventional dengue therapies. Alternanthera philoxeroides Alternanthera philoxeroides is a member of the family Amaranthaceae. Aquatic plants are those that grow in or near water, and A. philoxeroides, also referred to as "Alligator Weed," is one of these. While originally from South America, it is now encroaching on Australia. Researchers looked at the dengue virus-inhibiting potential of A. philoxeroides extracts in vitro. Using an MTT assay, the cytotoxicity of A. philoxeroides towards C6/36 cell lines was evaluated. Coumarin extract from A. philoxeroides was the least harmful to cells, although petroleum ether extract from the same plant was the most effective against dengue virus. Andrographis paniculata Acanthaceae is a family of plants that includes the genus Andrographis paniculata. It is an annual plant that grows uprightly and is widely grown in southern and southeast Asia. It is indigenous to Sri Lanka and India. In Malaysia, it is referred to as "Hempedu Bumi" and has a bitter flavour. Investigations examining the methanolic extract of A. paniculata's maximum nontoxic dosage (MNTD) on Vero E6 cells were carried out in southern and southeast Asia. It is indigenous to Sri Lanka and India. In Malaysia, it is referred to as "Hempedu Bumi" and has a bitter flavour. On Vero E6 cells, tests were done to determine the methanolic extract of A. paniculata's MNTD (maximum nontoxic dosage). According to A. paniculata, the greatest dose that did not harm cells was between 0.05 and 1. The methanolic extract of A. paniculata showed the best antiviral inhibitory action on DENV-1 in an antiviral experiment based on cytotoxic effects. Azidarachta indica A Zingiberaceae plant, Boesenbergia rotunda belongs to this family. Chinese ginger is a herb that can be used in both cooking and medicine. It is widespread in China and Southeast Asia. The ability of several compounds extracted from B. rotunda to obstruct the dengue virus protease was tested on DENV-2. The cyclohexenyl chalcone derivatives of B. rotunda, 4- hydroxypanduratin A (1) and panduratin A (2), showed effective competitive inhibitory effects against DENV-2 NS3 protease with Ki values of 21 lM and 25 lM, respectively. The fact that 4- hydroxypanduratin A has a low Ki value suggests that it can in vitro inhibit DENV-2 NS3 protease. Boesenbergia rotunda The Boesenbergia rotunda is a member of the Zingiberaceae family. Chinese ginger is a herb that has both culinary and medicinal uses. It can be found all over Southeast Asia and China. On DENV-2, various chemicals isolated from B. rotunda were investigated for their ability to block the dengue virus protease. With Ki values of 21 lM and 25 lM, respectively, the cyclohexenyl chalcone derivatives of B. rotunda, 4-hydroxypanduratin A (1) and panduratin A (2), demonstrated good competitive inhibitory actions towards DENV-2 NS3 protease. The low value of Ki indicates that 4-hydroxypanduratin A has the ability to in vitro inhibit DENV-2 NS3

protease. Carica papaya Figure-2: DENV inhibition following treatment with C. papaya: Linear regression curve . The Carica papaya is a member of the Caricaceae family. It is an upright, quickly-growing, unbranched tree or shrub that is native to Central America and is grown for its edible fruits in Mexico and most tropical nations. Traditionally, C. papaya leaf has been used to treat DF. The leaf's ability to combat DF has been researched. In blood samples from a 45-year-old patient bitten by carrier mosquitoes, the aqueous extract of this plant's leaves showed potential effectiveness against DF by increasing the platelet (PLT) count, white blood cells (WBC), and neutrophils (NEUT). The PLT count increased from 55 9 10 3 /l to 168 9 10 3 /l, the WBC from 3.7 9 10 3 /l to 7.710 3 /l, and the NEUT from 46.0 to 78.3% after 5 days of oral administration of 25 mL of aqueous extract of C. papaya leaves to the patient twice daily. Increased platelets may result in less bleeding, preventing the development of the severe sickness known as DHF. Cladogynos orientalis Cladogynos orientalis is a member of the Euphorbiaceae family. It is a 2 m tall shrub with white stellate hairs that can be found throughout Southeast Asia, Malaysia, and Thailand. A Thai medicinal plant called Cladogynos orientalis was tested for its ability to inhibit dengue virus growth in vitro. The MTT technique was used to examine the dichloromethane ethanol extract of C. orientalis for anti-dengue activity against DENV-2 in Vero cells. According to the findings, DENV-2 was inhibited by 34.85% by the ethanol extract of C. orientalis at a concentration of 12.5 lg mL-1. Additionally, C. orientalis showed a 2.9% inactivated viral particle activity at a dose of 100 lg mL-1. Cladosiphon okamuranus Cladosiphon okamuranus is a member of the Chordariaceae family. It is a type of brown seaweed that is indigenous to Okinawa, Japan. Cladosiphon okamuranus fucoidan (3), a sulfated polysaccharide, was discovered to possibly prevent DENV-2 infection . A focus-forming assay was used to examine the viral infection in BHK-21 cells. Compared to untreated cells, fucoidan reduced infectivity by 20% at 10 lg mL-1. A carboxy-reduced fucoidan, on the other hand, diminished the inhibitory action on DENV2 infection by converting glucuronic acid to glucose. Cryptonemia crenulata The Halymeniaceae family includes the plant Cryptonemia crenulata. It is a marine species that can be found in the Pacific Islands, the Indian Ocean Islands, Southeast Asia, the Western Atlantic Islands, South America, Africa, and the Atlantic Islands. The reference polysaccharides heparin and DS8000 had IC 50 values of 1.9 and 0.9 lg mL-1, respectively, whereas the sulfated polysaccharides from Cryptonemia crenulata, galactan (4), had an IC 50 value of 1.0 lg mL-1 and was a selective inhibitor of DENV-2 multiplication in Vero cells. However, the substance is completely inert against DENV-1 and has no antiviral impact at all against DENV-3 and DENV-4. In comparison to treatment just during internalization (EC 50 = 5.5 0.7 lg mL-1), the inhibitory effect of C2S-3 against DENV-2 was slightly stronger when treatment was through adsorption (EC 50 = 2.5 0.1 lg mL-1) . Therefore, the inclusion of C2S-3 at both the adsorption and internalization stages resulted in a greater inhibitory impact. Cymbopogon citratus Cymbopogon citratus is a member of the Poaceae family. It is a tropical plant from Southeast Asia that is a kind of grass known as lemon grass. Based on the level of inhibition of DENV-1- infected Vero E6 cells and its cytopathic effects, Cymbopogon citratus was found to have antiviral activity. On DENV-1, the methanolic extract of C. citratus had a marginally inhibiting effect. With the use of an inhibition assay using the MTT method, this finding was further validated. C. citrate didn't exhibit any appreciable inhibition, though. The lowest MNTD content was also found in C. citratus, which was 0.001 mg mL-1. The plant C. citratus was discovered to be quite cytotoxic, with a maximal cytotoxicity of 0.075 mg mL-1. Euphorbia hirta The family Euphorbiaceae includes the Euphorbia hirta. In Java, Sumatra, Peninsular Malaysia, the Philippines, and Vietnam, it is a typical weed in garden beds, garden walkways, and wastelands. In the Philippines, DF is treated using a traditional medication known as gatas- gatas, a water decoction of leaves from Euphorbia hirta. After 24 hours, internal bleeding will stop and dengue fever will be treated. However, the exact mechanism of action is still unknown, and researchers are currently looking into the drug's antiviral capabilities and capacity to raise blood platelets. To treat DF, tea made from boiled E. hirta leaves is utilized. Flagellaria indica The family Flagellariaceae includes Flagellaria indica. It is a strong perennial climber that may be found in many of the Old World's tropical and subtropical areas, including Polynesia, Australia, Southeast Asia, and India. In Vero cells, Flagellaria indica was tested for its ability to prevent dengue. According to the antiviral assay results, 12.5 lg mL-1 of the plant's ethanol extract caused a 45.52% suppression of DENV-2 to be seen in vitro. The cytotoxicity of F. indica was assessed using MTT assays. The ethanol extract of F. indica had a CC50 of 312 lg mL-1. Therefore, our research suggests that F. indica may significantly affect DENV. Gymnogongrus griffithsiae Gymnogongrus griffithsiae belongs to the family Phyllophoraceae. It is a red seaweed found in Ireland, Europe, the Atlantic Islands, North America, South America, the Caribbean Islands, Africa, Southwest and Southeast Asia, Australia, and New Zealand. The inhibitory properties against DENV-2 of the sulfur-flavonoid polysaccharide from Gymnogongrus griffithsiae, kappa carrageenan, were evaluated in Vero cells. The compound effectively inhibits DENV-2 multiplication at the IC50 value of 0.9 lg mL-1, which is the same as the IC50 value for the commercial polysaccharide DS8000. However, the compound has a lower antiviral effect against DENV-3 and DENV-4 and is totally inactive against DENV-1. Gymnogongrus torulosus Gymnogongrus torulosus belongs to the family Phyllophoraceae. It is a red seaweed found in

Australia and New Zealand. Gymnogongrus torulosus was investigated for its in vitro antiviral properties against DENV-2 in Vero cells . Galactan extracted from this plant was active against DENV-2, with IC50 values in the range of 0.19–1.7 lg mL-1. Hippophae rhamnoides The family Elaeagnaceae includes the species Hippophae rhamnoides. It is a deciduous shrub that can be found in Asia up to Japan and the Himalayas, as well as in Europe, including Britain, from Norway south and east to Spain. Human macrophages generated from contaminated blood were used to study the anti-dengue efficacy of extracts of Hippophae rhamnoides leaves. The results demonstrated that dengue-infected cells treated with H. rhamnoides leaf extracts were able to sustain cell viability on par with ribavirin, a commercially available anti-viral treatment, as well as a drop and increase, respectively, in TNF-a and IFN-c. Further evidence of H. rhamnoides leaf extract's anti-dengue effectiveness came from a decline in plaque counts following treatment of infected cells. Houttuynia cordata Houttuynia cordata belongs to family Saururaceae. It is herbaceous perennial flowering plants growing between 20 and 80 cm, and is native to Japan, Korea, Southern China and Southeast Asia. Ethanol extract from Houttuynia cordata revealed an anti-dengue activity with 35.99 % inhibition against DENV-2 in Vero cells at a concentration of 1.56 lg mL-1. Aqueous extract of H. cordata showed effective inhibitory action against DENV-2 through direct inactiva- tion of viral particles before infection of the cells. A concentration of 100 lg mL-1 also effectively protects the cells from viral entry and inhibits virus activities after adsorption. HPLC analysis of H. cordata extract indicated that hyperoside was the predominant bioactive com- pound, and was likely to play a role in this inhibition. Leucaena leucocephala Leucaena leucocephala belongs to family Fabaceae. It is a species of Mimosoid tree indigenous throughout Southern Mexico and Northern Central America and the West Indies from the Bahamas and Cuba to Trinidad and Tobago. Galactomannans (7) extracted from seeds of Leucaena leucocephala have demonstrated activity against yellow fever virus (YFV) and DENV- 1 in vitro and in vivo. Galactomannans are polysaccharides consisting of a man- nose backbone with galactose side groups, more specifically their structure consists of a main chain of (1 ? 4)- linked b- D-mannopyranosyl units substituted by a- D-galactopyr- anosyl units. L. leucocephala show protection against death in 96.5 % of YFV-infected mice. In vitro experiments with DENV- 1 in C6/36 cell culture assays showed that the concentration producing a 100-fold decrease in virus titer of DENV-1 was 37 mg L-1 . Lippia alba and Lippia citriodora The Verbenaceae family includes Lippia alba and Lippia citriodora. South Texas, Mexico, the Caribbean, Central America, and South America are all home to these beautiful plants. On dengue virus serotype replication in Vero cells, essential oils from Lippia alba and Lippia citriodora had a significant suppressive effect. L. alba oil was shown to have a 50% reduction in virus plaque number values at concentrations between 0.4 and 32.6 lg mL-1, whereas L. citriodora oil had IC 50 values between 1.9 and 33.7 lg mL-1. The virucidal efficacy of L. citriodora essential oil against DENV-1, 2, and 3 was comparable to but weaker than against DENV-4, whereas L. alba essential oil was more efficient against DENV-2 than other serotypes. At 100 lg mL-1, it was shown that L. alba essential oil completely eliminated YFV yield. Meristiella gelidium The species Meristiella gelidium is a member of the Solieriaceae family. It is a marine species that can be found in South America, the Caribbean Islands, the North American mainland, and the Atlantic Islands. It was determined whether or not DENV-2 could be inhibited by the antiviral properties of kappa carrageenan in Meriella gelidium. The carrageenans that were isolated from M. gelidium had an IC 50 that fell somewhere in the range of 0.14–1.6 lg mL-1. According to the findings, both the M. gelidium extract and the fraction that was generated from it were significantly more effective at inhibiting DENV-2 than the reference polysaccharides, which were heparin and DS 8000. Mimosa scabrella Fabaceae is the family that includes the Mimosa scabrella plant. It is a native to the chilly, subtropical plateaus of Southeastern Brazil and has a rapid growth rate, reaching heights of 15– 20 metres and having a diameter of up to 50 centimetres. Galactomannans derived from the seeds of Mimosa scabrella have been shown to have antiviral action in vitro and in vivo against the viruses YFV and DENV-1. In a study with mice infected with YFV, M. scabrella was shown to prevent death in 87.7% of the animals. Experiments conducted in vitro with DENV-1 in C6/36 cell culture assays revealed that a concentration of 347 mg L-1 resulted in a reduction in the virus titer of DENV-1 by a factor of one hundred. Momordica charantia The Momordica charantia plant is a member of the Cucurbitaceae family. It is a tropical and subtropical vine that can be found throughout Asia, Africa, and the Caribbean and is also known by the names bitter melon and peria (Malaysia). In vitro studies were conducted to determine the MNTD of the methanolic extract of Momordica charantia against Vero E6 cells. M. charantia found that a dose of 0.20 mg mL-1 was the highest level at which cells were not killed. In a test for antiviral activity based on cytotoxic effects, the methanolic extract of M. charantia demonstrated an inhibitory impact against DENV-1. Ocimum sanctum The Ocimum sanctum plant is a member of the Labiatae family. It is a shrub that is native to the tropical regions of both Asia and the Americas, and it is also an aromatic herb. Tea, which is typically made by boiling the leaves of the Ocimum sanctum plant, can be used as a preventative and medicinal treatment for DF. An analysis was done to determine the minimal non-toxic dose (MNTD) of a methanolic extract of O. sanctum for use against Vero E6 cells in vitro. However, there was not a discernible change in MNTD between

O. sanctum and the control group. Based on its cytotoxic properties, the methanolic extract of O. sanctum showed a mild inhibitory impact on DENV-1. Piper retrofractum Piper retrofractum is a member of the Piperaceae family of plants. The fruit of this blooming vine, which is native to Southeast Asia, is the primary reason why it is cultivated in Indonesia and Thailand. Piper retrofractum was tested for its anti-dengue activity in Vero cells in an in vitro setting. The MTT assay was utilised to evaluate the dichloromethane ethanol extract's potential to prevent DENV-2 infection in cells. At a concentration of 100 lg mL-1, the P. retrofractum ethanol extract demonstrated an inactivated viral particle activity level of 84.93%. Previous research has demonstrated that an aqueous extract of long pepper, also known as P. retrofractum, possesses the highest level of efficacy when it comes to warding off mosquito larvae. Psidium guajava The Psidium guajava tree is a member of the Myrtaceae family. Native to Mexico, the Caribbean, Central America, and South America, this evergreen shrub or small tree can also be found in those regions. In most of the world's tropical and subtropical regions, it is one of the most common crops cultivated. It has been demonstrated through in vitro testing that the Psidium guajava leaf extract can prevent the growth of the dengue virus. Platelet counts were boosted to 100.000/mm3 in roughly 16 hours thanks to the usage of water that had guava leaves that had been boiled in it. This was done to prevent bleeding in DHF. In cases of deficiency of platelets, the ripe fruit or juice of P. guajava has been shown to have curative effects and can be used to treat the condition. Quercus lusitanica The Quercus lusitanica tree is a member of the Fagaceae family. Oaks of this species can be found naturally occurring in Morocco, Portugal, and Spain. It was discovered that the extract of Quercus lusitanica had a significant effect in inhibiting the reproduction of DENV-2 in C6/36 cells. The absence of cytotoxic effects indicates that the methanol extract of the seeds was able to totally inhibit the TCID 50 of the virus at its maximal non-toxic dosage of 0.25 mg mL-1. This inhibition ranged from 10 to 1,000 fold. With 10 TCID 50 of the virus, an extremely low dose of Q. lusitanica (0.032 mg mL-1) was able to completely inhibit the virus. After treatment with the extract, proteomics techniques were utilised to show that the action of Q. lusitanica was to downregulate NS1 protein expression in infected c6/36 cells. This was demonstrated by the fact that the extract inhibited the expression of the protein. Rhizophora apiculata The Rhizophoraceae family is comprised of the Rhizophora apiculata species. In Australia (Queensland and the Northern Territory), Guam, India, Indonesia, Malaysia, Micronesia, New Caledonia, Papua New Guinea, the Philippines, Singapore, the Solomon Islands, Sri Lanka, Taiwan, the Maldives, Thailand, and Vietnam, there is a kind of mangrove tree that may grow to a height of up to 20 metres. There have been reports of the ethanolic extract of Rhizophora apiculata having anti-dengue effects when tested on DENV-2 in Vero cells. At doses of 12.5 and 100 lg mL-1, respectively, R. apiculata displayed inhibitory action and an inactivate particle activity of 56.14% and 41.5%d viral. Figure-3: ‘Anti-dengue effective of Medicinal plants.’ The figure above indicates that medicinal plants through their bio-functional ingredients can potentiate the inhibition viral enzymes such as methyltransferase and NS2B-NS3-Pro, other pharmacological benefits include prophylactic impact and rising platelets as well as WBCs count. 19 Chapter Five 5.1 Results & Discussion Exploration to find the antiviral drugs to DENV has been developing rapidly and profoundly in many countries. Herbal medicines are a potential source for the development of new antiviral drugs, since they can be selected based on their ethno-medicinal use, for example, against infectious diseases . These plants produce a variety of phytochemical constituents with the potential to inhibit viral replication, and compounds from natural sources are of interested as possible sources to control viral infection. From the various reports published in scientific literature, it appears that C. papaya L. leaf extract does have beneficial properties in dengue. It has been shown to bring about a rapid increase in platelet count. This could be possibly attributed to its membrane-stabilizing property. The flavonoids and other phenols present in the extract have been suggested to provide the beneficial effects. One study found that the leaves of papaya plant are rich in several minerals. The researchers suggested that these minerals may balance the mineral deficiency caused by the virus and strengthen the immune cells against it . Earlier observations confirmed that flavonoids in plants and their derivatives possess antiviral activity . thus, in this study, the methanolic extracts were standardised based on total flavonoids content prior to antiviral assay. Another plant that recorded a low level of total flavonoids was the A. paniculata, which was examined for this study. Anti-atherosclerotic flavonoids such as 7-O methylwogonin, apigenin, onysilin, and 3,4-dicaffeoylquinic acid have been extracted from A. paniculata. These flavonoids have been tested and found to be effective. C. limon, on the other hand, had the third highest overall average concentration of flavonoids (33%) out of the six plants that were investigated. Flavonoids such as hesperidin, diosmin, and eriocitrin can be found in quite high concentrations in C. limon. The flavonoids citrunobin, cit-flavanone, and lonchocarpol-A were found in the root bark of Citrus species. Current treatments for dengue There is no effective drug or vaccine for the treatment of this potentially fatal disease. In 2020, the vaccine called Dengvaxia was developed and licensed for use in 11 countries. However, it is not effective for the protection from infection of all the four serotypes of dengue . There are multiple anti-DENV agents in various stages of development. Some are direct-acting antivirals namely RNA polymerase inhibitors, nucleoside analogues and protease

inhibitors. Some anti- dengue agents are developing that target host-mediated translational modifications such as α glucosidase inhibitors. Appropriate dose of paracetamol is recommended for relieving the DF, and ibuprofen, aspirin, and naproxen (Aleve) should be avoided. Fluids replacement are very effective when patients suffering with dehydration. Fluid balance, electrolytes, nursing care, and blood clotting parameters should be taken care of a dengue patient. Patients should be hospitalized as per expand of symptoms. Chapter Six Summary and conclusion The existing study focuses the information on numerous plant components and extract kinds that are used to treat dengue. However, some plants that have not yet been fully studied might have a wide range of therapeutic uses. Finding better, more potent, and less harmful anti-dengue medications requires the development of novel anti-dengue products from bioactive chemicals. In light of this, any in-depth investigation into the potential of plants with separated active components that have demonstrated anti-dengue action should involve additional in vitro and in vivo animal research, as well as toxicity and clinical investigations. Uncertainty is surely caused by the lack of particular target treatments for dengue in the thoughts of those who are infected. Patients are not the only ones who become anxious in this situation; the professionals who treat the patients are also under strain. Additionally, it assisted Bangladeshi society in identifying alternative dengue prevention and treatment options. In different regions of Bangladesh, several plants and their preparations have been traditionally utilised to treat dengue. Numerous literary works have described how local communities and herbal healers use plants to combat dengue.When we analyzed scientific validation studies of evolving herbal alternatives, all the three methods and models (in vitro, in vivo and clinical trial) of efficacy testing against dengue viruses have been adapted by the Bangladesh researchers. However, number of such scientific validation for plants effective against dengue are very less in India, below double digits. About twenty- two plants from all over Bangladesh were found in literature search which were recommended for their use against dengue. However, only two to three plants/extracts have been tested scientifically and also have shown evidences of efficacy. There is a need to search more such herbal formulations which are being practiced at local level, document them properly and validate them scientifically to confirm efficacy, understand mechanistic action and safety so that they can be exploited for their anti-dengue potential. The potential herbal formulations being used by the communities and healers are the low hanging fruits which may provide alternative or adjuvant therapy if proper validation, value addition and product develop- ment steps are followed. Moreover, such discoveries may lead to the development of highly efficient and safe anti-dengue treatments. However, to identify potential anti-dengue plants or compounds, knowledge of the mechanisms of virus infection need to be understood inorder to facilitate the search for and devel- opment of the most appropriate drugs. Further research is needed to determine how to target the most appropriate stages to prevent the spread of virus infection. Focusing on each phase in the life cycle of the virus, new compounds could prevent (1) infection of host cells, (2) the viral maturation process, (3) synthesis of viral RNA, or (4) the spread of viral particles. Ethical Approval: No ethical approval needed. Conflict of Interest: The author declare that they have no conflict of interest. Multilingual ORIGINALITY REPORT 34% 30% 12% 21% SIMILARITY INDEX INTERNET SOURCES PUBLICATIONS STUDENT PAPERS PRIMARY SOURCES 1 dspace.daffodilvarsity.edu.bd:8080 Internet Source 10 % 2 Submitted to Vet Nurse Plus Student Paper 10 % 3 mafiadoc.com Internet Source 3 % 4 doi.org Internet Source 2 % 5 www.florajournal.com Internet Source 2 % 6 www.phcogrev.com Internet Source 2 % 7 Submitted to Daffodil International University Student Paper 2 % 8 www.researchgate.net Internet Source 1 % 9 istiqur.blogspot.com Internet Source 1 % 10 Oby dulla, Sharifa Sultana, Md. Shohag Hosen. 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