Screening of Anthelmintic and Insecticidal Activity of *Flacourtia jangomas* Fruit pulp

By

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A thesis submitted to the School of Pharmacy in partial fulfillment of the requirements for the degree of Bachelor of Pharmacy (Hons.)

School of Pharmacy
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Declaration

It is hereby declared that

1. The thesis submitted is my/our own original work while completing degree at BRAC University.
2. The thesis does not contain material previously published or written by a third party, except where this is appropriately cited through full and accurate referencing.
3. The thesis does not contain material which has been accepted, or submitted, for any other degree or diploma at a university or other institution.
4. I have acknowledged all main sources of help.

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Approval

The thesis titled "Screening of Anthelmintic and Insecticidal Activity of *Flacourtia jangomas* Fruit pulp" submitted by Shah Md. Nasrul Islam 19146035 of Spring 2019 has been accepted as satisfactory in partial fulfillment of the requirement for the degree of Bachelor of Pharmacy on March 09, 2023.

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Ethics Statement

There were no human participants, specimens, or tissues used in this study. There were also no cephalopods, vertebrates, or vertebrate embryos or tissues used in this research. This study involves earthworms and rice insects.

Abstract:

The study aimed to evaluate the anthelmintic and insecticidal activity of *Flacourtia jangomas* fruit pulp (MFJ). The plant's anthelmintic property was tested on *Pheretima posthuma* (Earthworm), while its insecticidal property was tested on *Tribolium castaneum* (Rice insects). For the anthelmintic study, In comparison to albendazole, all crude extracts showed significa nt anthelmintic activity at different concentrations (25–75 mg/mL), which resulted in the paralysis and death of earthworms. The fruit pulp's methanolic extract was shown to be more effective at the highest dose. The extract had the highest death rate (44.44%) in the insecticidal test. The results raise the possibility of using *F. jangomas* as a natural anthelmintic and insecticide. There has to be further research done to corroborate these findings and prove that the plant extract is safe and useful to use.

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Keywords: *Flacourtia jangomas, Tribolium castaneum, Pheretima posthuma,* Anthelmintic, Insecticidal, Helminths, methanolic extract.

Dedication

I want to dedicate this project to my parents for their continuous support.

Acknowledgment

All praise to Almighty Allah, and I would like to commence by expressing my gratitude to Him for the continuous blessings.

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List of Acronyms

WHO	World Health Organization
BC.	Before Christ
N. sativa	*Nigella sativa*
STHs	Soil-transmitted helminths
MA	Mechanism of Action
ML	Macrocyclic lactones
nAChR	Nicotinic acetylcholine receptor
GluCls	glutamate-gated chloride channels
AChE	Acetylcholinesterase
F. jangomas	*Flacourtia jangomas*
T. castaneum	*Tribolium castaneum*
P. posthuma	*Pheretima posthuma*
MFJ	Methanolic extract of *F. jangomas*

Chapter 01

Introduction

Plants have been widely used as a source of medicine and have been crucial to health care and disease prevention for thousands of years. Initially, they comprised the majority of folk or ethnomedicine, practiced in countries like China, the Middle East, Africa, and South America in addition to India (Sasi et al., 2018).

Using plants as a source to cure diseases is not new to mankind. Almost every civilization in the world has knowledge of using plants for healing. Phytotherapy, or previously known as herbalism, refers to the practice of treating patients with plant extracts(Houghton, 1995). Researchers have found the first records of the use of the herbal plant in medical treatments from Chinese scriptures from 2800 BC. Not only that but the usage of plants as a source of medicine by the Indians, Chinese, Egyptians, Greeks, Romans, and Syrians is documented in ancient texts that date back roughly 5000 years (Pandey et al., 2011).

After Friedrich Wilhelm Sertürner synthesized morphine from *Papaver somniferum* in 1804, natural items underwent intensive testing for their potential as medicines. In recent years Pharmaceutical industries have been using plants as a source to develop new biologically active molecules or as a lead compound for a new drug.

A total of 100 anticancer drugs have been created between 1981 and 2006, 25 of which are derivatives of plant-based natural products, 18 of which are products that mimic natural compounds, 11 candidates are generated from natural product pharmacophores, and 9 of which are pure plant-derived natural products. A lot more study has been done recently on the chemistry of natural compounds obtained from marine species (Vicente, 2012).

From recent statistics, Around a quarter of all pharmaceuticals in the United States, include plant-based chemicals. Roughly 121 pharmaceuticals were developed in the last century using traditional knowledge from various sources. Herbal medicine, sometimes called phytomedicines, is gaining popularity because of its ability to restore damaged biological systems and combat harmful microorganisms without causing harmful side effects (Pandey et al., 2011).

1.1 Medicinal Plants in Bangladesh

There is no reason to emphasize the significance of medicinal plants in Bangladesh. Moreover, in underdeveloped rural regions and other locations with limited access to conventional medical care, the use of medicinal plants as a primary form of treatment remains crucial.

Folk medicine in Bangladesh is based on naturally occurring ingredients, cultural traditions, and religious rites. Other traditional medical techniques include Ayurvedic and Unani, which make use of scientific usage of pharmacological procedures and technology. However, only roughly 20-25% of the population has access to modern healthcare facilities, while the remaining 75-80% of the rural population still relies on traditional medical practitioners from within the community for their medical treatment (Haque et al., 2018). As part of the official recognition of the Unani and the ayurvedic medicine system and to assure the availability, commercial manufacture, and marketing of quality Unani and Ayurvedic Medicine and Drugs, the government of Bangladesh included them in the National Drug Policy that year, 1982 (Rahman & Fakir, 2013).

Medicinal plants may be found growing wild in Bangladesh in a variety of environments, including woods, shrubs, and marginal ground near canals, as well as other locations. It is generally acknowledged as a very essential component of basic health care because many indigenous peoples have a long legacy of herbal medicine systems that are based on the variety of abundant local plants (Rahman et al., 2013).

The majority of medical practitioners in Bangladesh are known as "Kaviraj," and they are the ones who practice traditional medicine. Over 87,000 villages can be found in Bangladesh, and the majority of these villages are home to at least one Kaviraj. Their generation-old Knowledge about medicinal plants might be helpful for further scientific research in the hunt for better pharmaceutical drugs derived from medicinal plants that are utilized and with little to no side effects.

About 5000 plant species have been identified in Bangladesh, because of the country's fertile soils, tropical climate, and varied growing seasons. On the whole, about 500 of these species are thought to provide some sort of healing effect (Chowdhury et al., 2009). Some of them are given below:

Green chiretta: Locally known as “Kalomegh”. Scientific name is“*Andrographis paniculata* ”. The whole body of green chiretta is used in the treatment of Cholera, diarrhea, Diabetes and liver-related problems.

Billygoat weed: Locally known as “Fulkuri " and scientific name is “ *Ageratum conyzoides L. "* The stem and leaf are mainly used. When the leaves and stems of the plant are cooked, they can be used as a strong laxative, antipyretic, antiasthmatic, analgesic, anti diarrheic, anti-inflammatory, and headache. The leaves are applied to the skin to treat disease. Cancerous growths can be treated using flower buds.

Neem Tree: Neem tree's scientific name is "*Azadirachta indica*". Inflammation of the gums, fever, gingivitis, smallpox and problems related to the spleen are among the diseases that can be treated using various parts of the plant.

Malabar Nut: Locally known as "Basak". The scientific name is "*Justicia adhatoda*". This plant's bark, roots, and leaves are all utilized in medicinal preparations. Used to treat a variety of health conditions, including breathing difficulties, coughing, colds, nasal congestion, sore throats, asthma, bronchitis, various infections of the upper respiratory tract, bleeding problems, and so on.

Thankuni: Scientific name of thankuni is "*Centella asiatica (L)*". Leprosy, skin diseases, and mental disorders are treated by administering the whole plant. Leaf juices can be beneficial in the treatment of indigestion, cleansing of the body, reduction of inflammation, ulcer wound healing, and other conditions.

Arjun Tree: Scientific name is "*Terminalia arjuna*". In the treatment of medical conditions such as hypertension and heart disease, the bark of the Arjun tree is used.

Belleric Myrobalan: Locally known as "Bahera" and scientifically known as "T*erminalia bellirica*". In the treatment of cuts, wounds, and skin ailments, bark juices are taken orally.

colds, coughs, and other respiratory issues can be alleviated with the usage of fruit powders.

Myrobalan: Local name is "Horitoki". The scientific name of myrobalan is "*Terminalia chebula*". Fruit of myrobalan is administered for reducing fevers, relieving cough, colds,and respiratory symptoms, while the bark is utilized for urinary issues. Patients with diabetes, heart disease, and gastrointestinal issues are also given the fruit

Madagascar Periwinkle: Locally known as "Noyontara". Scientifically known as " *Catharanthus roseus*". The Leaf of Madagascar Periwinkle is made into a paste and used for the treatment of diabetes (Rahman et al., 2013b).

1.2 Helminths

Helminths are the most prevalent type of parasite that may infect humans. The word "helminth" originates from an ancient Greek word that simply means "worm." Worms that are either free-living or parasitic might be classified as helminths. The Nematoda, sometimes known as roundworms, and the Platyhelminthes, also known as flatworms, are the two phyla that include the majority of parasitic worms. The most common types of parasitic flatworms are called Trematoda, also known as flukes, and Cestoda, also known as tapeworms. Helminth infections can be passed on to people by a variety of vectors, including food, water, soil, arthropods, and mollusks. Helminths are capable of infecting each organ as well as the organ system.

They are most common in the intestines, but may also be discovered in the liver, lungs, blood, and other organs, as well as infrequently the brain (Md et al., 2019).

More than 1.5 billion individuals, or 24 percent of the global population, have an infection caused by soil-transmitted helminths (STHs), according to the World Health Organization. Coinfection with schistosomes and soil-transmitted helminths is prevalent in Africa, China, East Asia, and the Americas, although infections may be found all throughout the tropics and sub-tropics (Romero-Benavides et al., 2017).

Helminths are separated into their respective categories based on the external and internal morphology of their eggs, larvae, and adult stages. The categories are:

Trematode: Also called flukes. The adult stages of flukes take the form of flatworms that resemble leaves. Oral and ventral suckers are prominent and aid in holding the body in place. All flukes, with the exception of the blood fluke, are hermaphroditic (having both male and female reproductive organs).

Cestode: Also known as tapeworms. Tapeworms are hermaphroditic flatworms that live in the intestinal lumen as adults. They are long and segmented. Larvae, which can be cystic or solid, live outside of the intestines in various tissues.

Nematodes: Also referred to as roundworms. Both adult and juvenile roundworms share the same spherical shape and are bisexual. They can be found both in and outside of the intestines (Castro, 1996).

1.3 Anthelmintic Activity of Plants

Anthelmintic properties means the ability to cure infection or diseases caused by parasitic worms or helminths. Traditional medicines have significant potential as a means through which the public might gain access to effective anthelmintic medications that are not only readily available but also cost-effective. Numerous plant-based or plant-derived remedies have been used historically to treat helminthic diseases. An ideal anthelmintic drug would have a wide range of efficacy, little host toxicity, and low cost. There is currently no synthetic medicine available that can fulfill this need. Side effects such as nausea, gastrointestinal difficulties, and giddiness have been observed with even the most commonly used medications like Piperazine salts (Mali & Mehta, 2008).

Some of the plants with anthelmintic activity are given below:

***Carica papaya* (Papaya):** Which belongs to the Caricaceae family. Locally known as Pepe. The anthelmintic action of papaya aqueous extract was demonstrated against the roundworms *Ascaris lumbricoids* and *Ascardia galli*. The anthelmintic benzyl isothiocyanate was extracted from its extract. Benzyl isothiocyanate was able to kill the parasites by reducing their energy consumption and disrupting their motor activities.

***Nigella sativa* (Black seed):** Locally known as Kala Jira**.** It belongs to the Ranunculaceae family. Thymoquinone, dithymoquinone-cymene, and alpha-pinene are the three primary bioactive compounds found in *N. sativa*. The essential oil of this seed was tested for its anthelmintic ability against four different worm species (earthworms, tapeworms, hookworms, and nodular worms), and it showed promising results only against tapeworms and earthworms.

***Punica granatum* (Pomegranate):** Locally known as Anar/Dalim and belongs to the *Punicaceae* family. In traditional medicine, the plant's root and stem bark are utilized for their astringent and anthelmintic properties. The alleged anthelmintic properties of an alcoholic extract of its stem bark were tested. Dose-dependent action was observed, preventing *Haemonchus contortus* eggs from hatching into filariform larvae. Clinical trials of the herb have shown its effectiveness in preventing nematodiasis in calves. Alkaloid Pelletierine has been found in the stem bark (Mali & Mehta, 2008).

***Ocimum sanctum* (Holy Basil)**: Holy basil, locally known as Tulsi, belongs to the Lamiaceae family. contains volatile oil, the primary components of which are eugenol and beta-caryophyllene, in addition to a variety of monoterpenes and sesquiterpenes. Eugenol and essential oil both exhibited significant anthelmintic action in vitro against *Caenorhabditis elegans* (Mali & Mehta, 2008).

***Azadirachta indica* (Neem):** The leaves, seeds, and oil from the neem tree have been used in Ayurvedic and traditional medicine systems to treat a variety of parasitic infections, which also include worms. Studies have shown that compounds in neem, such as azadirachtin, have anthelmintic activity against various species of worms (Jamra et al., 2014).

1.4 Anthelmintic Compound from Plant Extract

There are several plant-derived compounds that have been studied for their anthelmintic properties. Some examples include

- **Santonin:** This compound is extracted from the flowers of the plant *Artemisia cina* (santonica) and has been traditionally used to treat tapeworm infestations. Studies have shown that santonin has anthelmintic activity against pork tapeworm (*Taenia solium)* and other species of tapeworms. However, because to concerns over its toxicity, it has been phased out of production. Excessive santonin consumption is associated with a visual distortion called xanthopsia, and in extreme situations, it can lead to blindness (Sakipova et al., 2017).
- **Berberine:** This alkaloid compound is extracted from a variety of plants, including *Berberis vulgaris* (Barberry), *Argemone Mexicana* (Mexican prickly poppy), and *Coptis chinensis* (Chinese goldthread). Berberine has been traditionally used to treat parasitic infections and studies have shown that it has anthelmintic activity against various species of worms, including tapeworms, roundworms, and flukes. Studies have shown strongyloidicidal activity of Berberine against the *Strongyloides venezuelensis* in an in-vitro model at low concentrations (Elizondo-Luévano et al., 2021).

- **Emodin:** This anthraquinone compound is extracted from a variety of plants, including *Rheum palmatum* and P*olygonum cuspidatum*. Emodin has been traditionally used to treat parasitic infections and studies have shown that it has anthelmintic activity against various species of worms, including tapeworms and roundworms.
- **Curcumin:** The rhizome of the plant *Curcuma longa* is the source of this polyphenolic compound. Curcumin is also known as turmeric. It has been traditionally used to treat various kinds of ailments, which also include parasitic infections. Studies have shown that curcumin has anthelmintic activity against various species of worms, including roundworms and flukes.
- **Gingerols:** Active ingredients of *Zingiber officinale* (ginger). Studies have shown that movement of *Ascaridia galli* was decreased by the administration of ginger methanolic extract, and the wormicidal impact of this extract is concentration-time dependent. The in vitro survival of *Ascaridia galli* was inhibited by all ginger preparations tested (Bazh & El-Bahy, 2013).

1.5 Anthelmintic Drug

Medications called "anthelmintics" are used to treat worm infections (helminths). Anthelmintics are medications that eliminate worms from the body by killing or expelling them, and they do not harm the host in any meaningful way. Drug companies have made sluggish progress in discovering and developing anthelmintic drugs despite the widespread presence of parasitic worms. The fact that most people with helminth infections reside in poorer countries makes it harder to find a profitable medicinal market to tackle this issue. Anthelmintics are classified into the following:

Table 01: Anthelmintic drugs and their mechanism of action

Class Name	Mode of action	Examples
Benzimidazoles	It has been demonstrated that benzimidazoles specifically bind to parasite beta-tubulin with a high affinity and impede the process of microtubule polymerization. This ultimately leads to the breakdown of cellular structure and finally death of the parasite (Abongwa et al., 2017).	Albendazole, Mebendazole, and Thiabendazole
Imidazothiazoles	act as agonists of the nicotinic acetylcholine receptor (nAChR). Imidazothiazoles bind to nAChRs on the muscles of the body wall, which results in spastic paralysis and the worm's expulsion from the host through the feces (Abongwa et al., 2017).	Levamisole
Tetrahydropyrimidines	has an action mechanism that is comparable to that of imidazothiazoles and are typically categorized as nicotinic agonists (Abongwa et al., 2017).	pyrantel, oxantel, and morantel
Cyclooctadepsipeptides	have an inhibitory impact on pharyngeal pumping via binding to the latrophilin (LAT-1) receptor. It also targets the calcium-activated potassium channel (SLO-1) (Abongwa et al., 2017).	PF1022A and emodepside

Spiroindoles	functions as nAChR antagonist, paralyzing the host and triggering the parasite's excretion (Abongwa et al., 2017).	Derquantel
Macrocyclic lactones (ML)	glutamate-gated chloride(GluCls) channel selective agonists. When GluCls are activated by ML, mobility and pharyngeal pumping are stifled (Abongwa et al., 2017).	Milbemycins, Abamectin, ivermectin, doramectin
Amino-acetonitrile derivatives	nematode-specific nAChRs agonists (Abongwa et al., 2017).	Monepantel
Tribendimidine	bephenium-sensitive B-nAChRs are the only ones that this drug targets preferentially (Abongwa et al., 2017).	amidantel,tribendimidine

1.6 Insecticidal Properties of Plants:

The danger posed by pests and insects including mosquitoes, rats, parasitic worms, and cockroaches has long been recognized and countered by humanity. In order to safeguard crops against weeds, illnesses, and insect pests, ancient man used a wide range of control measures, including cultivation techniques, mechanical activities, and the use of organic and inorganic chemicals. Multiple insecticidal compounds were utilized to manage pest populations between 500 B.C. and the 19th century AD (Koma, 2012).

Insecticidal property refers to the ability to kill insects. Insecticides are a class of pesticides that are specifically designed to kill or control the population of insects. They are widely used in agriculture, forestry, and residential, commercial, and industrial settings to control pests. Insecticides can be synthetic or natural and can come in many forms such as liquid sprays, dusts, baits, and granules(Tembo et al., 2018).

Many plants produce natural compounds that have insecticidal properties, these compounds are called secondary metabolites, and they are used by the plants to protect themselves against herbivorous insects and other potential threats. Some examples of plants that are known to have insecticidal properties include

- **Neem (*Azadirachta indica*):** This plant is indigenous to areas of Asia and India. The compound azadirachtin is present in neem tree leaves, seeds, and bark which shows insecticidal effects. These compounds prevent insects from growing and reproducing normally, which reduces their capacity for survival and procreation.
- **Pyrethrum (*Chrysanthemum cinerariaefolium*):** This plant is indigenous to East Africa and is commercially produced for its insecticidal effects. Pyrethrins, which have insecticidal properties, are found in the flowers of the pyrethrum plant. These compounds cause paralysis and death in insects by disrupting their nervous systems.
- **Nicotine (*Nicotiana tabacum*):** This plant is native to the Americas and is known for its toxic properties. The leaves of the nicotine plant contain compounds called alkaloids, which have insecticidal properties. These compounds disrupt the nervous system of insects, causing paralysis and death.
- **Pawpaw tree (*Asimina tribola*):** A plant used medicinally in many different indigenous groups around the world, especially in Africa and the Americas, has been scientifically

examined and proven to have anticancer and insecticidal qualities. Seeds and bark have been studied for their insecticidal properties, with most attention given to the primary bioactive component, asimicin (Koma, 2012)

1.7 Plant-Derived Compound with Insecticidal properties

Many plants produce natural compounds that have insecticidal properties, these compounds are called secondary metabolites, and they are used by the plants to protect themselves against herbivorous insects and other potential threats. Some examples of plant-derived compounds with insecticidal properties include

- **Azadirachtin:** This compound is found in the leaves, seeds, and bark of the neem (*Azadirachta indica*) and it has insecticidal properties that disrupt both growth and reproduction of insects, making them less able to survive and reproduce.

  Mechanism of Action (MA): Azadirachtin acts by inhibiting the Prothoracicotropic hormone. It also disrupts plant phagostimulant (a compound that attracts insects) activity by reducing cholinergic transmission (Souto et al., 2021).

- **Pyrethrins:** This group of compounds is found in the pyrethrum plant flowers (*Chrysanthemum cinerariaefolium*) and it has insecticidal properties that eliminate the insect by disrupting the nervous system which causes paralysis.

  MA: Serves as a voltage-gated sodium channel modulator in the nervous system. It is capable of causing a delay in sodium channel closure, leading to excessive neuroexcitation, motor dysfunction, and eventually death (Du et al., 2013).

- **Nicotine:** This compound is found in the leaves of the tobacco plant (*Nicotiana tabacum*) and it has insecticidal properties that disrupt the nervous system of insects, causing paralysis and death.

  MA: Simulate the effects of acetylcholine by functioning as an agonist at the receptor for that neurotransmitter. This causes an increase in sodium ions and the production of an action potential. However, because AChE is not capable to metabolize nicotine. Which cause the prolonged activation produced by nicotine results in overactivity of the cholinergic transmission, which ultimately results in seizures, paralysis, and ultimately death (Green et al., 2013).

- **Rotenone**: This compound is found in the roots of plants from the genus *Lonchocarpus*, it has insecticidal properties that act as a contact and stomach poison to insects.

  MA: Rotenone exerts its insecticidal activity by blocking Complex I associated with the mitochondrial respiratory chain (Souto et al., 2021)

- **Coumaran:** Also known as 2,3-dihydrobenzofuran, found in *Lantana camara* from the *Verbenaceae* family.

  MA: blocks the action of the acetylcholinesterase (AChE) enzyme. The breakdown and recycling of acetylcholine is the responsibility of. Increased acetylcholine levels in the synapse cleft as a result of inhibition lead to restlessness, excitability, tremors, and death as a result of the prolonged attachment of the neurotransmitter to its postsynaptic receptor (Rajashekar et al., 2014).

- **Ryanodine:** An bioactive compound of *Ryania speciosa* (Family:Salicaceae). Mainly available in the roots and woody stems.

  MA: induces skeletal muscle cells' sarcoplasmic reticulum calcium channels to open. When triggered, calcium channels flood actin and myosin protein filaments with calcium ions, causing the skeletal muscles to contract and eventually become paralyzed (Nauen, 2006).

1.8 *Flacourtia jangomas* Description

1.8.1 Taxonomical Classification of *Flacourtia jangomas*

Kingdom : Plantae

Subkingdom : Viridiplantae

Infrakingdom : Streptophyta

Superdivision : Embryophyta

Division : Tracheophyta

Subdivision : Spermatophytina

Class : Magnoliopsida

Superorder : Rosanae

Order : Malpighiales

Family : Salicaceae

Genus : *Flacourtia*

Species : *Flacourtia jangomas*

(*Flacourtia Jangomas (Lour.) Raeusch.* | *Species*, n.d.)

1.8.2 Synonyms

- *Flacourtia cataphracta* Roxb. ex Willd.,
- *Roumea jangomas* Spreng,
- *Stigmarota jangomas* Lour,
- *Xylosma borneense* Ridley (Lim, 2013).

1.8.3 Local and Common Name

Table 02: Local and common name of F. jangomas (Lim,2013)

Language/Country	Vernacular Names
Arabic	Zarnab,Talis fir.
Assamese	Phinel, Polian,Paniyal
Bengali	Tali,Bara Baichi
Burmese	Mak Kyen, Naywe.
Chinese	Yun Nan Ci
English	Indian Plum
Gujarati	Talispatra
Kannada	Chankali, Goraji, Charichali
Nepalese	Talispatri

Thai	Makwen Khwai, Ta Khop Khwai Ta Khop Thai
Vietnamese	Bô Quân, Hông Quân, Muôn Quân

1.8.4 Distribution and Origin

F. jangomas is native to Southeast Asia, specifically in countries such as India, Bangladesh, Sri Lanka, Myanmar, Thailand, Malaysia, Indonesia, and the Philippines. The plant is also found in other parts of the tropics such as Africa and South America, likely due to its historical use as a food crop and ornamental plant. In some places, it is considered an invasive species. The plant prefers tropical and subtropical regions with warm temperatures and high humidity and can be found in a variety of habitats including forests, along riverbanks, and in disturbed areas (*Flacourtia Jangomas (Lour.) Raeusch.* | *Species*, n.d.-b).

1.8.5 Botanical Description

Trunk: While older trunks and branches may be thorn-free, their younger counterparts are often heavily equipped with simple or branching woody thorns. The bark peels off in tiny scales and can be any color from a pale brown or gray to a copper red or pinkish buff. Some younger branches have white, elevated spots.

Leaf: The leaves are generally wedge-shaped at the base and range in form from narrow-ovate to ovate-oblong, very rarely ovate-lance-like or long obtuse acuminate. They are 7-10 x 3-4 centimeters and have a little toothing pattern. The upper side is shiny and smooth, while the underside is largely dull. The leaf stalk ranges in length from 6-8 mm.

Flowers: There are two types of flowers, male and female, and they grow on different plants. These blooms provide a pleasant aroma. Male flowers may be anywhere from 15 to 30 millimeters in diameter and about 5 to 15 millimeters in height. Each male flower has four or five small greenish petals that are rather wide with rounded ends. These male flowers lack petals but have an abundance of bright yellow stamens. Similarly to the male flowers, the female flowers (10–15 mm in length) have a small number of blooms with comparable sepals. In addition to lacking petals, the ovary of these female flowers is usually fused together into a single structure and their styles number between four and six.

Fruit: Fruits have a spherical form and range in size from 15 to 25 millimeters. It is a fruit that may be eaten and has a content that is both juicy and meaty (Sasi et al., 2018).

Figure 01: Flower of F. jangomas (Flacourtia Jangomas (Lour.) Raeusch. | Species, n.d.-c)

Figure 02: F. jangomas Fruit (Flacourtia Jangomas (Lour.) Raeusch. | Species, n.d.-c)

1.8.6 Chemical Constituent of *F. jangomas*

Numerous chemical compounds, including flavonoids, tannins, saponins, alkaloids, and terpenoids have been discovered to be present in *F. jangomas*. It is believed that these compounds help the plant's therapeutic benefits.

According to some research, the leaves and fruit of *F. jangomas* contain flavonoids such as quercetin, kaempferol, and myricetin. Therapeutic activity of some common chemical compounds are given below:

Tannins: are present in the plant and are well-known for their astringent properties..

Saponins: discovered in the fruit and leaves. This compound has anti-inflammatory and immunostimulatory properties

Alkaloids: exhibit a variety of medicinal properties, including pain relief, anti-tumor, and anti-inflammatory actions.

Terpenoids: Known for their anti-inflammatory, anti-tumor, and anti-bacterial properties, have also been discovered in the plant.

To properly understand the variety of chemical compounds in *F. jangomas* and their potential therapeutic capabilities, additional study is necessary.(Talukder et al., 2012; Dutta & Borah, 2017b; Lim, 2013).

1.8.7 Therapeutic uses of the plant

F. jangomas have been traditionally used in Southeast Asian medicine for various therapeutic properties. Some of the reported properties include:

- **Anti-inflammatory:** According to studies, chemical compounds found in *F. jangomas* have anti-inflammatory properties that may be helpful in the treatment of conditions including rheumatoid arthritis and osteoarthritis. (Sasi et al., 2018).
- **Antioxidant:** Antioxidants present in high amounts in the plant may help protect cells from damage brought on by free radicals. This could possibly be used in the treatment of certain cancers (Talukder et al., 2012).
- **Antimicrobial and Antibacterial:** According to certain research, F. jangomas possesses antibacterial properties and can prevent the proliferation of several types of bacteria and fungus (Sarker et al., 2011).
- **Wound healing:** The plant has been traditionally used to promote wound healing, and some studies have found that compounds present in *F. jangomas* can promote tissue regeneration and wound healing (Sasi et al., 2018).
- **Antidiabetic:** Some studies have reported that compounds present in *F. jangomas* can reduce blood sugar levels and may have potential therapeutic uses for the treatment of diabetes (Singh & Singh, 2010).

However, it is important to note that many of these properties have not been studied extensively and more research is needed to understand the full range of therapeutic properties and potential applications of *F. jangomas*.

1.9 Parts of Plant Used in This Study

For this study fruit pulp of *F. jangomas* fruits was used.

1.10 Literature Review

The reports from earlier studies on plant roots, stems, bark, and leaves are provided below:

Sarker et al., 2011: The result showed that *F. jangomus* showed good bactericidal action against gram-positive as well as gram-negative bacteria.

Singh & Singh, 2010: The findings demonstrate that methanolic extract from the plant's leaves and the stem have antidiabetic action and also plays a role in restoring biological parameters that have been changed.

Hasan et al., 2012: The experiment's results suggest that the plant extract of *F. jangomus* possesses strong antioxidant capability, which fits with traditional usage in a variety of illnesses and conditions and with a number of well-established scientific studies.

Dutta & Borah, 2017: The study uncovered the fruits of F. jangomas's nutraceutical potential due to the abundance of bioactive chemicals important to human health. Furthermore, the results of the study show that the crude extract of the fruits contains bioactive chemicals with essential medical applications.

A. K. Singh et al., 2010: As a result of this finding, it is safe to say that the ethanolic extract of *F. jangomas* is being used to control diabetes in ethnomedical settings. The results of this experimental investigation on lab animals suggest that the plant may have antidiabetic effects.

1.11 Rationale of the project

It has become crucial for scientists and researchers to find new potent medicinal compounds from natural sources like plants in order to reduce the adverse effects of synthetic drugs and drug resistance. One of the most often utilized medicinal plants in conventional medicine is *F. jangomas*. Practitioners of alternative medicine have been utilizing this plant for a range of medical issues.

The goal of this study is to identify the anthelmintic and insecticidal properties of this plant as well as to scientifically verify the claim made by traditional healers. In order to develop safer and more effective anthelmintic and insecticidal compounds from a natural source, further investigation of this plant is recommended. This study assesses the anthelmintic effect of fruit pulp crude extract in *Pheretima posthuma* and the insecticidal effect in *Tribolium castaneum*.

1.12 Aim of the project

In vitro anthelmintic and insecticidal activity of *F. jangomas* was the focus of this investigation.

1.13 Objective of the Project

Because natural materials, such as medicinal plants, are readily available for investigation as potential new drugs, the area of pharmaceutical research has paid a significant amount of attention to these kinds of substances in recent decades. These days, numerous pharmacological lead compounds are extracted from plants rather than being synthesized in a lab. This is due to the fact that plant-based pharmaceuticals are less expensive to treat, less poisonous, and have fewer adverse effects than their synthetic counterparts. Aside from this, herbal medications are inexpensive, risk-free, and socially acceptable in many cultures. Because of this, herbal medicines captured the interest of researchers all over the world during the early stages of the herbal medications industry's growth.

After carrying out this experiment, it was determined beyond a reasonable doubt that our experimental plant had bioactive chemicals. We look at the plant components, specifically the fruit flesh, and find that it already possesses a great deal of potential therapeutic value in the realm of herbal medicine.

1.12 Study Protocol

The purpose of this research was to investigate the potential therapeutic benefits of *F.jangomas* extracts. It is necessary to have a research protocol that describes how to carry out the trial. The procedures for the study are presented in the following:

1. Collection of *F. jangomas* plant fruits.
2. Cut the fruit into small pieces and make fruit pulp with an electric blender.
3. Preparation of *F.jangomas* fruit pulp extract by soaking in methanol.
4. Screening of anthelmintic property of crude extracts *Pheretima posthuma* (Earthworms).
5. Screening of insecticidal activity on *Tribolium castaneum* (Rice Insects).

Chapter 02

Method & Materials

2.1 Preparation of *F. jangomas* Extracts

2.1.1 Collection and identification

In order to conduct the research, fresh fruits of the *F .jangomas* plant were collected from the Jahangirnagar University campus, Savar, Dhaka, Bangladesh. A taxonomist from the Bangladesh Herbarium in Mirpur, Dhaka, was responsible for the identification and verification of *F .jangomas* (DACB Accession No:87043).

2.1.2 Preparation of Plant samples

First, the fruits of the experimental plant were rinsed in running tap water to eliminate any dirt, and then they were washed again in distilled water to sterilize. The fruits were then cut into small pieces and then it was made into a pulp by using a laboratory electric blender.

2.1.3 Sample Extraction

A clean glass jar was taken. A second methanol wash was performed, and then it was dried. After that, 250g of pulp sample was weighed and then added to the container. Then, 1 L of methanol was added to each jar before being firmly sealed to prevent air leakage. The container sat at room temperature (about 22–25 degrees Celsius) for a whole week. A little shaking and stirring improved the extraction process on occasion. However, after a few days, a double layer developed, with methanol solution at the top and plant extract (methanol-soaked double-layer sample solution) at the bottom.

2.1.4 Methodology

Here, the entire *F.jangomas* extraction procedure essentially consisted of three phases, and they were:

I. Collection of *F. jangomas* fruit

II. Blending the fruit into pulp

III. Extraction in methanol

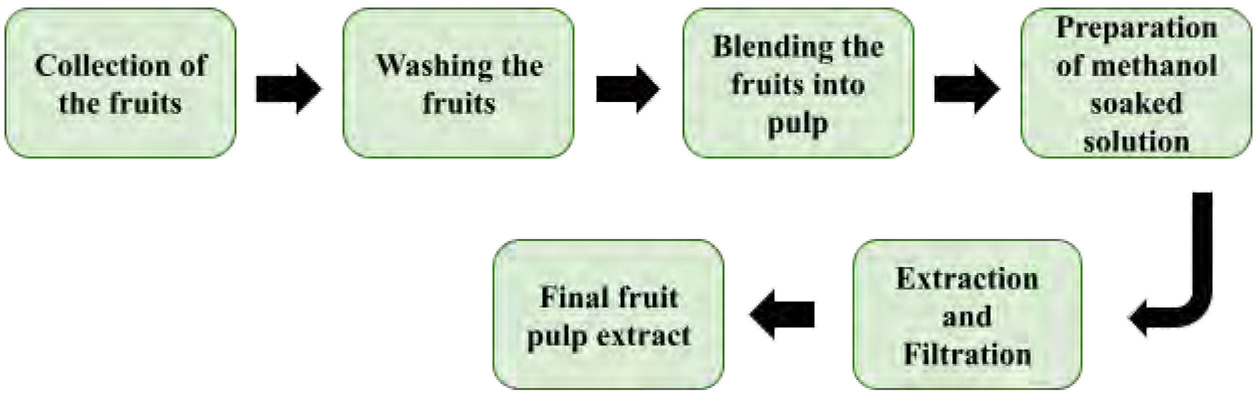

Figure 03: Steps of extraction of F. jangomas fruit pulp

2.2 Experiment Procedure for Anthelmintic Activity Screening

2.2.1 Standard Drug

This research involved the use of Albendazole, a drug prescribed for the treatment of neurocysticercosis.

2.2.2 Reagents

I. Methanol

II. Saline

III. Tween 20

2.2.3 Experimental Helminths

It was chosen to conduct anthelmintic research on mature *Pheretima posthuma (P. posthuma)* earthworms. Because of the similarity in its anatomy and physiology with the roundworm in the human intestine

2.2.4 Earthworms Collection

The earthworms were obtained from a nursery. The worms were cleaned with normal saline to remove any debris before they were used in the anthelmintic activity testing. Information on the worms used in the study is provided below:

Length: maximum 3-5 cm

Width: maximum 0.1-0.2 cm

Weight: 0.8-3.04 g

2.2.5 Preparation of Standard Drug and Extract

In order to conduct an in vitro investigation, three different methanolic extracts of *F. jangomas* were produced, with concentrations of 25 mg/mL, 50 mg/mL, and 75 mg/mL respectively. Following that, solutions of Tween 20 at a concentration of one percent were added to the methanol extract samples, and finally, the mixture was diluted with normal saline. In this particular research project, the control group consisted of tween-20 and normal saline, whereas the standard treatment was albendazole.

2.2.6 Anthelmintic Activity Screening

All *P. posthuma* that were collected were given a normal saline bath. This was performed to get them used to be in a laboratory setting before any tests were conducted. After that, the correct procedures were followed to prepare the standard and test solutions. To conduct this experiment, we have employed freshly made standards and controls. Tween 20 (1%) solution was diluted with normal saline to provide test samples with concentrations ranging from (25 mg/ml) to (75 mg/ml). Once that was done, earthworms of similar size were collected and split into five groups. These five groups were each let loose in a 30 ml portion of the test compound. This time around, six adult *P. posthuma* made up each group.:

1 st group - Control (Tween-20 and normal saline)

2nd group - Standard drug (Albendazole)

3rd to 5th group - *F. jangomas* solvent extracts at 25 mg/ml, 50 mg/ml, and 75 mg/ml concentrations.

As a result, it is evident that the first group contains tween-20 in addition to the normal saline used as the experiment's control. Group two received the usual care with the 20mg/ml concentrated reference medication albendazole. Additionally, the third through fifth groups received treatments with solvent extracts of *F. jangomas* at various dosages (25 mg/ml, 50 mg/ml, and 75 mg/ml). Anthelmintic action on P. Posthuma was observed, and the times required for the experimental worms to become paralyzed and die were recorded. Here, the period of paralysis was measured as the duration during which the worms were unable to detect any movement other than when they were violently agitated. The worms' time of death was measured when they ceased moving despite being shaken ferociously in the petri dish and submerged in hot water at a temperature of 50°C, at which point their body colors started to fade.

2.3 Experiment Procedure for Insecticidal Activity Screening

2.3.1 Preparation of Extracts

The test samples were made into six concentrations for the experiment: 2.5 mg/ml, 5 mg/ml, 10 mg/ml, 20 mg/ml, 40 mg/ml, and 50 mg/ml, respectively) by combining the extracts with the appropriate solvents.

2.3.2 Experimental Insects

Tribolium castaneum was chosen as the experimental insect to assay the insecticidal activity

2.3.3 Collection of Rice Insects

From the stock culture of Crop Protection and Toxicology Laboratory, Sher-e-Bangla Agricultural University in Bangladesh insects were collected.

2.3.4 Insecticidal Activity Screening

The test was conducted on 60mm Petri dishes. The extract was initially dissolved in each solvent to create the sample solutions (2.5 mg/ml, 5 mg/ml, 10 mg/ml, 20 mg/ml, 40 mg/ml, and 50 mg/ml).

They were then poured into the bottom of the petri dish and let dry. In each of the prepared Petri plates, insects were then unleashed.

The same settings and timing were used for a control experiment in which the solvent alone was added to the petri dish.

Petri dishes were set up in a safe location at room temperature once everything was set up. The entire experiment was periodically watched, and data was collected. Mortality was initially seen after 30 minutes from the start and subsequently after 1, 2, 4, 8, and 12 hours of exposure.

2.4 Statistical Analysis

Each value (experimental result) is reported as a mean ± standard error of the mean (SEM). Here, we examined all of the collected data with ANOVA (analysis of variance) and then Dunnett's test. It's possible, nevertheless, that the values are significant when $P<0.01$.

Chapter 3

Result

3.1 Results of Anthelmintic Activity

Depending on the concentration of the medicinal substance *F. jangomas* in our trial, the parasites displayed loss of motility, responsiveness, paralysis, and even death. Albendazole is used as the reference standard for determining the anthelmintic activity of different concentrations of methanolic extracts of *F. jangomas*

Table 03: Anthelmintic Activity of F. jangomas Fruit Pulp at Different Concentration

Treatment	Conc. (mg/ml)	Time taken for paralysis (min) X±SD	Time taken for death (min) X±SD
Control (normal saline)			
Standard drug (Albendazole)	20	40.67±1.53	49.00±2.64
Methanolic extract of *F. jangomas*	25	60.67±6.51*	80.33±7.51*
	50	56.33±2.51*	68.33±5.03*
	75	41.66±4.04*	53.00±1.73*

Values are expressed as Mean ± S.E.M. (n=3). Data were analyzed by ANOVA followed by Dunnett's test, *P< 0.001 compared with the standard drug

The table shows that at 20 mg/mL, the standard dose, The paralysis time was 40.67±1.53 min and the death time was 49.00±2.64 min.

Worms in the control group (those given only normal saline) were monitored for 24 hours and showed no signs of paralysis or death.

Anthelmintic activity of MFJ at 25, 50, 75 mg/mL concentrations showed time taken for paralysis are 60.67±6.51, 56.33±2.51, and 41.66±4.04 min and the time taken for death are 80.33±7.51, 68.33±5.03, and 53.00±1.73 min, respectively

3.2 Result of Insecticidal activity:

The percentages of the extracts' mortality rate were discovered using Abbott's technique. For the purpose of calculating the rice insect death rate, the following equation was utilized

Pr = (Po - Pc \ 100 - Pc) x 100

Where, Pr = Corrected mortality%;

Po = Observed mortality%;

Pc = Control mortality %,

Three different extracts were used in this instance to track the death rate of rice insects. They were closely watched for almost 48 hours.

The graph below shows the percentage of inhibitions that were achieved. The concentrations of the three extracts are utilized as the baseline for the graph.

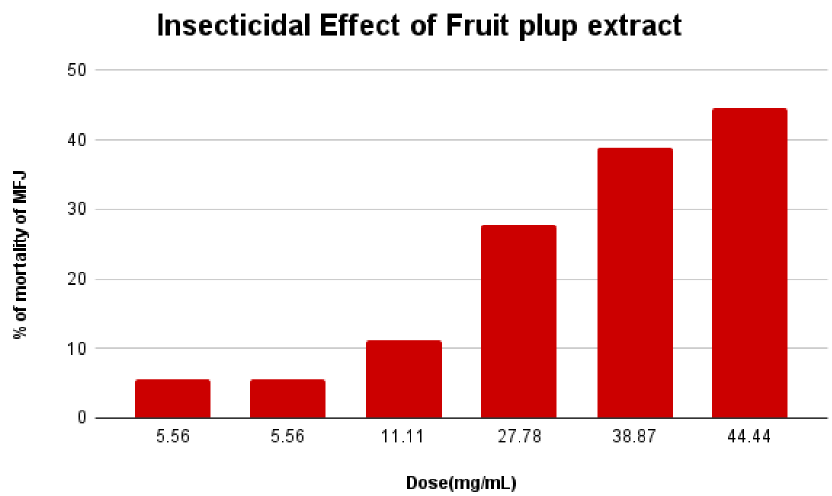

Figure 04: Graph showing the insecticidal effect of fruit pulp extract

The horizontal axis represents the Dose(mg/ml)and the vertical axis represents the %mortality of MFJ. The graph above makes it very obvious that the proportion of rice insect death rises with higher dosages. The fruit pulp of the *F. jangomas* has the highest mortality rate of around 44.44% at dosages of 50 mg/ml of methanol extract. At 2.5 mg/ml and 5mg/ml the mortality rate is around 5.56%. Also, at 10mg/ml concentration, the mortality rate is 11.11%. Moreover, at 20mg/ml and 40mg/ml the mortality rate is 27.78% and 38.87%% respectively.

Chapter 4

Discussion

For a long time, people and animals have turned to plant extracts to treat their ailments. People have always had to rely on natural resources to provide for their most basic needs, and medicines to cure a wide range of illnesses are no exception. Many traditional medical systems have their roots in plants, and this is especially true of the more advanced ones. Many ground-breaking pharmaceuticals, such as papaverine (an anti-diabetic medicine), quinine (an anti-malarial drug), reserpine (an anti-hypertensive drug), etc., have their origins in plants(Cragg & Newman, 2013). This inspired us to check the pharmacological activity of *F. jangomas* fruit pulp. In this research, the anthelmintic and insecticidal properties of *F. jangomas* fruit were assayed in vitro.

Among human pathogens, helminth infections inflict the greatest global illness burden, surpassing even those caused by malaria and tuberculosis, which are more common in underdeveloped countries (Hotez et al., 2008). From the result of the anthelmintic experiment, the standard drug Albendazole was shown to have a rapid worm-influencing effect, causing flaccid paralysis that ultimately led to worm death via peristalsis. For the most part, albendazole binds with free tubulin and provides selective prohibition of the polymerization that impedes glucose absorption of microtubules by the parasites (Md et al., 2019b). However, the anthelmintic action of the *F. jangomas* plant is due to its secondary metabolites, which include phenols, alkaloids, tannins, flavonoids, saponins, steroids, terpenes, etc. Our study found that 75 mg/ml of methanolic meat extract (MFJ) was as effective as the standard albendazole and was much more effective than lower concentrations of MFJ extract. The results showed that the extract was dose-dependent, as 75 mg/ml produced rapid paralysis and death in the worms.

Based on these findings, it appears that *F. jangomas* MFJ extract has therapeutic potential for treating intestinal worm infections.

Furthermore, Humans have been fighting off insect pests that affect crops, animals, and people since the dawn of civilization. People have employed natural insecticides for thousands of years to combat insects that pose harm to human health or that compete with humans for food and fiber. In the 20th century, synthetic pesticides have replaced natural ones as the primary tool for controlling insects. Synthetic pesticides altered insect management, but they also brought with them new risks. The dangers and side effects of using synthetic pesticides on humans are greater. They are not biodegradable, thus they are poisoning our water supply and threatening our biodiversity in addition to making their way into the food chain. Because of this, researchers are actively pursuing plant-based pesticides, and official policy encourages the increased use of natural insecticides (Coats, 1994). From the result of the insecticidal experiment, it is evident that the Fruit flesh of *F. jangomas* has insecticidal properties. *F. jangomas* flesh methanolic extracts insecticidal activity on *T. castaneum* (Rice insects) is proportional to dose. From the experiment results we observed that the higher the concentration of extract the higher the mortality of *T. castaneum* .Sugars, saponins, phytosterol, phenol, flavonoids, and tannins must be present in medicinal plants for them to have insecticidal action. As a result, we may conclude that the tested plant leaves of crude extracts exhibit a toxic effect on the evaluated bugs.

In conclusion, the results of our experiment showed that *F. jangomas* Fruit possesses both anthelmintic and insecticidal activity. Future research might investigate the phytochemical profile of F. *jangomas* in order to isolate the anthelmintic-active ingredient.

Chapter 5

Conclusion

To maintain human health and well-being, medicinal plants are essential. Since ancient times, these plants have been utilized in traditional medicine to treat a wide range of diseases and disorders. The significance of medicinal plants has increased as awareness of the harmful effects of synthetic pharmaceuticals has grown.

The fact that medicinal plants provide a natural alternative to synthetic drugs is one of their most important advantages. Contrary to synthetic drugs, which are frequently created in a lab from chemicals, medicinal plants contain natural materials that are more readily absorbed by the body. Because of this, they are a safer and better choice for many people, especially those who have chronic diseases. Additionally, the fact that medicinal plants may have fewer adverse effects than synthetic drugs is another significant advantage. They can also be used in conjunction with other herbal treatments to maximize benefits and reduce risks. Additionally, herbal supplements are frequently less expensive than manufactured ones. The cost of synthetic medications varies. On the other hand, medicinal plants are readily available and can often be grown and harvested sustainably, reducing the cost for both the consumer and the environment.

In this experiment, we choose the fruit of *Flacourtia jangomas* to investigate its anthelmintic and insecticidal activity. Crude extract of this plant fruit shows great anthelmintic activity against *P posthuma* (earthworm). Also, it shows promising insecticidal activity against *T. castaneum* (rice insect). *F. jangomas* is a valuable resource for traditional medicine. In traditional medicine, the plant is used to treat a variety of conditions. Further research is needed to fully understand and harness the potential of this plant

To sum up, the significance of medicinal plants cannot be emphasized. These plants have a long history of use in traditional medicine, provide a natural alternative to synthetic drugs, are usually more affordable, have fewer side effects, and are also widely available. The promotion of health and well-being for all can benefit greatly from the use of medicinal plants, whether alone or in conjunction with other natural medicines.

Future Plan:

To conduct our research, we used only extracts of *F. jangomas* fruit pulp that had been preserved in methanol. Future extraction methods and sophisticated screening may yield more bioactive molecules, more narrowly targeted components, and more effective medication alternatives for treating parasite illnesses. Taking these measures will aid in identifying the various bioactive chemicals by their structural makeup and evaluating their effectiveness. There are two broad processes thought to account for the anthelmintic and insecticidal actions of herbal medications. Plants' bioactive substances interact with the parasite in one way, while the host immune system is involved in the other. But in our research, we are mainly interested in the first mechanism of action, which involves plant bioactive chemicals interacting with parasite worms and insects. Therefore, a thorough screening of *F. jangomas*'s many components can lead to the identification of other, potentially anthelmintic bioactive chemicals. In addition, studies and comparisons of anthelmintic activity against other microbes are possible. This will shed light on the extracts' anthelmintic activity against different bacteria and their usefulness in treatment.

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