Determination of CNS Depressant and Analgesic activity of leaves of *Artocarpus chama.*

A Dissertation Submitted To the Department Of Pharmacy, East West University.
In The Partial Fulfillment of the Requirements for the Degree of Bachelor of Pharmacy.

Submitted by
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Declaration by the Research Candidate

I, Nasrin Jahan Billal, hereby declare that the dissertation entitled "CNS depressant and analgesic activity of leaves of *Artocarpus chama*" submitted by myself to the Department of Pharmacy, East West University, in the partial fulfilment of the requirement for the award of the degree Bachelor of Pharmacy is a complete record of original research work carried out by me during 2017, under the supervision and guidance of Meena Afroze Shanta, Senior Lecturer, Department of Pharmacy, East West University and the thesis has not formed the basis for the award of any otherdegree/diploma/fellowship or other similar title to any candidate of any university.

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Certificate by the Supervisor

This is to certify that the thesis entitled **"CNS Depressant and Analgesic activity of leaves of Artocarpus chama"** submitted to the Department of Pharmacy, East West University, in the partial fulfillment of the requirement for the degree of Bachelor of pharmacy was carried out by Nasrin Jahan Billal, ID# 2013-3-70-061 in 2017, under the supervision and guidance of Meena Afroze Shanta. The thesis has not formed the basis for the award of any other degree/diploma/fellowship or other similar title to any candidate of any university.

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Endorsement by the Chairperson

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Abstract

From different experiments, medicinal plant is using worldwide in order to dissolve drug as well as safer drug also. Methanol and pet ether extract of leaves of Artocarpus chama was assessed for CNS Depressant and Analgesic test on animal (mice) model. Administration of methanol extract and pet ether extract produced significant dose dependent CNS effect in Open field and Hole board test. Analgesic activity was evaluated by acetic acid induced pain method as well as formalin induced pain method. All the result of our experiment was statistically significant. In CNS depressant test the movement of the mice decreased in a dose depending manner comparing to the standard diazepam. Analgesic effect at writhing and formalin induced pain method comparing to control group was d\significant. The amounts of writhes decreased in acetic acid induced pain method as well as reduced licking and biting time in formalin induced licking and Biting method. Percent inhibition is 34.71% for 200 mg/kg body weight and 47.6% ($p < 0.05$) for 400 mg/k g body weight, whereas for Standard drug (Ibuprofen) the percent inhibition is 65.09% ($p < 0.001$) for 10 mg/kg body weight. In conclusion it can be said that Pet ether and Methanol extract of Artocarpus chama possesses good analgesic activity.

Dedication

DEDICATED TO MY PARENTS

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List of Abbreviation:

ACLM- *Artocarpus chama* leaf Methanol.

ACLP- *Artocarpus chama* leaf Pet Ether.

ml-milliliter

cm-Centimeter

kg-kilogram

g-gram

min-Minute

WHO-World Health Organization

Chapter 1: introduction

1.1 Overview

Medicinal plants, also called medicinal herbs, botanical drugs, or natural product drugs, have been discovered and used in traditional medicine practices since prehistoric time. From the different part of plant we get natural medicine. From the very beginning of the world people was willing to use natural medicine for different kind of disease.

In some develop countries, herbal medicines are very much popular like, USA, some countries of Africa. In Asia herbal medicines are more popular.

Natural Standard Research Collaboration was founded in 2000 to serve as a clearing house for information on evidence-based medicine covering numerous healthcare disciplines. This international effort involves authors, editors, and peer reviewers from multiple academic and research institutions.Medicinal plants such as *Aloe, Tulsi, Neem, Turmeric* and *Ginger* cure several common ailments. These are considered as home remedies in many parts of the country. It is known fact that lots of consumers are using Basil (Tulsi) for making medicines, black tea, in pooja and other activities in their day to day life.

In several parts of the world many herbs are used to honour their kings showing it as a symbol of luck. Now, after finding the role of herbs in medicine, lots of consumers started the plantation of tulsi and other medicinal plants in their home gardens.

Medicinal plants are considered as a rich resources of ingredients which can be used in drug development either pharmacopoeial, non- pharmacopoeial or synthetic drugs. A part from that, these plants play a critical role in the development of human cultures around the whole world. Moreover, some plants are considered as important source of nutrition and as a result of that they are recommended for their therapeutic values. Some of these plants include ginger, green tea, walnuts, aloe, pepper and turmeric etc. Some plants and their derivatives are considered as important source for active ingredients which are used in aspirin and toothpaste etc.(Khan.A.M, 2016)

1.2 Herbal medicine

Are Indigenous cultures (such as African and Native American) used herbs in their healing rituals, while others developed traditional medical systems (such as Ayurveda and Traditional Chinese Medicine) in which herbal therapies were used. Researchers found that people in different parts of the world tended to use the same or similar plants for the same purposes.In the early 19th century, when chemical analysis first became available, scientists began to extract and modify the active ingredients from plants. Later, chemists began making their own version of plant compounds and, over time, the use of herbal medicines declined in favor of drugs. Almost one fourth of pharmaceutical drugs derived from botanicals.Recently, the World Health Organization estimated that 80% of people worldwide rely on herbal medicines for some part of their primary health care. In Germany, about 600 to 700 plant based medicines are available and are prescribed by some 70% of German physicians. In the past 20 years in the United States, public dissatisfaction with the cost of prescription medications,

1.3 Medicinal plants

Currently, global health care is in crisis, the natural environment is undergoing widespread devastation, and indigenous people and their knowledge are disappearing at a rapid rate. There is an increasing amount of talk these days about sustainability. As this applies to medicinal plants, it means that users of the plants get good, viable remedies, the natural environment is enhanced and protected in the course of utilizing or trading in those plants, and people who work with the plants are able to flourish. Global Sustainability, as it applies to medicinal plants and products made from them, involves providing natural resources for human health needs in a manner that supports the health and diversity of the natural environment, and incorporates labor and wage practices that enable all people in the system to flourish. In a sustainable system, all life is supported and allowed to prosper.Global Sustainability. (Organization, 2010)

Approximately 25% of today's prescription drugs comes from plant extracts. Only about 15% of the known plant species have been screened for medicinal purposes. Most medicinal plants come from the Tropics. Most medicinal plants have been identified by the indigenous people by trial and error. 50% of the 250,000 plant species are from the Tropics. At least 10,000 species in the Tropics have not yet identified. 10,000 plant samples collected for screening against HIV and cancer. 2500 species and 200 families of the 3000 extracts analyzed, 170 contained agent. 62 were previously known as medicinal plants. 44 were entirely new. The most significant medicinal plants used for prescription drugs contain steroids or alkaloids. The rapid destruction of our tropical rainforests threatens the development of potentially useful drugs.

1.4 History of Medicinal plants

1.4.1 Pre-history

The use of plants as medicines predates written human history.Medicinal herbs were found in the personal effects of Ötzi the Iceman, whose body was frozen in the Ötztal Alps for more than 5,000 years. These herbs appear to have been used to treat the parasites found in his intestines.Archaeological evidence indicates that the use of medicinal plants dates back to the Paleolithic age, approximately 60,000 years ago.Carbon dating from ancient Babylon (Iraq) records that plants were cultivated plants as medicines 60,000 years ago.Written Materia medica of medicinal herbs go backapproximately 5,000 years in India, China and Egypt and atleast 2,500 years in Greece and Asia Minor.Twenty-five hundred years ago, Hippocrates (the father ofmedical literature), stated as part of his oath: "I will give no deadly medicine to anyone." Hippocrates used only food and herbs and is best known for the sayings:"Let your food be your medicine and let medicine who compiled lists of plants. (Michael,.2015)

1.4.2 Ancient history

Plants have been used for medicinal purposes long before recorded history. Ancient Chinese and Egyptian papyrus writings describe medicinal uses for plants as early as 3,000 BC.Plants have been used for medicinal purposes long before recorded history. (Michael,.2015)

1.4.3Middle age

Middle ages Medicine was extremely basic in an era when terrible illnesses such as the Black Death were killing nearly one third of the population. Medicine was limited. Physicians had no idea what caused the terrible illnesses and diseases. The Catholic Church believed that illnesses were a punishment from God for sinful behavior. Letting blood was conducted by cupping or leeches. There were no Antibiotics during the middle Ages and it was almost impossible to cure illness and diseases without them. Medicines in the middle Ages were made from herbs, spices and resins. The medicine was applied in drinks, pills, washes, baths, rubs, poultices, purges and ointments. (Michael,.15)

1.4.4 Early modern era

Recipe books and manuals for house-wives of the 16th and 17th centuries would often include home remedies and treatments alongside the more familiar domestic skills of cookery and needlework. Many of these "cures" were somewhat dubious, such as the use of chestnuts to treat people coughing up blood because the "tree is absolutely under the domain of Jupiter", as Nicholas Culpeper asserts in his 1652 work The English Physician. Home-made treatments were nevertheless very common. It was expected for most householders to have at least a basic knowledge of medicine and the ability to make remedies using only herbs and materials that could be gathered locally.The influx of modern science in this period began to define a new emphasis on personal diagnosis and cure instead of the more common preventative approach to illness. As professional doctors emerged, it is often presumed that household practice became more about culinary pursuits.While medicine in the 17th century was becoming an increasingly specialized. Formof knowledge, however, the market for medical recipes to make at

home was booming and the majority of care was still being performed by non-professionals, usually women. The Good House-wife Made Doctor, which was published in 1698 and features in the St John's exhibition. (Michael,.15)

1.4.5 Usage of medicinal plants in world

1.4.5.1 Ginger:In many Asian countries, especially in India, ginger is a part of daily diet. Because, the medicinal power of ginger can cure many diseases. The ginger root has many remedial actions including antibacterial, antiviral and antioxidant.

Health benefits of ginger:

- Ginger juice can balance digestive process.
- Improves the nutrients absorption power of body.
- Anti-inflammatory properties of ginger can cure joint pain.
- Ginger could keep away nausea in post-surgery.
- Help to fight against cancer, diabetics and asthma
- Clear and charge up the micro-circulatory channels of the body.

1.4.5.2 Lavender: The oil extracted from lavender flower has antibacterial and antiviral properties. It is also beneficial in many medical conditions like anxiety, stress and insomnia.

Health benefits of Lavender

- Lavender oil can keep away dandruff and can cool down your scalp.
- Help to get relief from dryness, itching and swelling of skin.
- One the best anti-depressant.
- Better solution for sun burn and headache.
- Provide prevention for airborne viruses.

1.4.5.3 Garlic: Garlic is a member of the onion family. It is uses in many countries in various dishes. Garlic is also a popular herb. It can heal a wide range of diseases. It is low in calories and rich in nutrients. Garlic contains Vitamin C, Vitamin B6, Manganese and Fiber. The Sulphur rich, strong pungent smell of garlic also can keep away insects and even snakes.

Health benefits of garlic

- Provide better protection against cancer infections.
- Help to Improves your immunity.
- Balances the digestive system and improves nutrient absorption power of the body.
- Reduces blood pressure and reduce chances of cardiovascular problems.
- Prevention against allergies and improves iron metabolism.
- Quick relief from toothache and detoxify your body.

1.4.5.4 Spinach: Spinach is the edible flowering plant native to Central Asia. This crispy, leafy vegetable also has powerful healing ability. It is a great source of vitamins and minerals. Spinach contains Vitamin K, Vitamin A, Manganese, Folate, Magnesium, Iron, Vitamin C, Vitamin B1, Zinc, Phosphorous, Vitamin B3 and Selenium.

Health benefits of spinach

- The presence of riboflavin, beta carotene and lutein in spinach improves cardiovascular system and nervous system.
- It protects your eyesight, strengthen your bones and fight against cancer.
- The antioxidant properties help to reduce the chances of DNA damages.
- Effective solution for diabetics and cancer.
- Reduce blood pressure and prevent the chances of insomnia, tumors, neuritis and obesity.
- Balance the cholesterol level in the body.
- The high concentration of Vitamin K in spinach protects your nerve system.

1.4.5.5 Catnip: Catnip or catmint native to Europe and central Asia. It is now largely cultivating in all parts of the world. Now it is widely popular in the world because of its health benefits.

Health benefits of catnip:

- It is a powerful detoxifier, help to sweat out toxic elements from your body.
- One of best natural medicine for headache and migraine.
- Relaxing property by calming down the nervous system.
- Quick relief from toothache.
- Balance digestive system.

1.4.5.6 Thyme: Thyme is widely used as an aromatic plant. Its flowers, leaves and oil also has many health benefits. In Ancient Egypt, the thyme was used for embalming and to heal many other medical conditions. Thyme plant has anti-fungal, anti-viral, anti-septic and anti-parasitic properties.

Health benefits of Thyme

- Better prevention against food-borne bacterial infections.
- Balance blood pressure level.
- Keep away the chances of colon cancer.
- Solution for skin problems such as dryness, redness and swelling.
- Quick relief from cold and cough.
- Act as relaxing herb by calming down the nervous system.

1.4.5.7 Tea tree: The tea tree oil is a popular herbal remedy. This herbal oil is derived from the leaves of tea tree native to coastal areas of Australia. The tea tree plants are highly priced in Australia. The tea tree oil has antibacterial, anti-fungal and antiviral properties. This herbal oil is nowadays used in soaps, lotions and shampoos.

Health benefits of Tea tree

- Tea tree oil is used for infection of nail, skin, mouth and nose.
- Make persistent body odor.
- Prevention against head lice.
- Quick relief from cuts and burns

- Faster dandruff cleaner and makes scalps remains cool.
- Cure headache, cold and toothache.

1.4.5.8 Lady fern: Lady ferns is a long, light greenly plant native to northern hemisphere. It is commonly used for decorations and to make a number of recipes. Lady Ferns can also power to heal a number of diseases. The roots and stems of lady ferns are used for medicinal purposes.

Health benefits of lady fern

- Recommended herb for the lack of appetite, fever and cough.
- Faster relief from cuts and sunburn.
- Prevention against harmful worms.
- Balance the digestive system.
- Prevention against Asthma, pneumonia and bronchitis.

1.4.5.9 Sage: Sages is a powerful herb with beautiful flowers and soft leafs. This plant grows in home gardens. The stem, flower and leaves of sage can cure a number of diseases in an effective way. The sage is very rich in nutrients and antioxidants.

Health benefits of Sage

- Improve your memory power.
- Can instantly reduce depression.
- Natural remedy for healing wounds.
- Balance stomach problems.
- Relief from toothache and keep away bad breath.

- Prevents the chances of infection to lungs, nose and throat.
- One of best antiseptic herbs.

1.4.5.10 Peppermint: Peppermint is a hybrid plant mainly cultivated in Europe. Peppermint oil is widely used as food flavor. This plant also has a calming effect and offers many other health benefits. Peppermint also has anti-bacterial and anti-fungal properties.

Health benefits of Peppermint

- Improves digestive system and nutrient absorption power of the body.
- The concentration of methanol in peppermint can prevent prostate cancer.
- Usage of peppermint oil could enhance memory power and alertness.
- Effectively prevent sinus infections.
- Quick relief from body pain.

1.4.5.11 Marigold: Marigold is a flowering plant that grows in wide range of soils. Marigolds are used in many summer dishes to add color. This plant can also cure many skin problems. It can also effectively reduce body scars.

Health benefits of Marigold

- Instant relief from fever.
- Can cure swollen body parts within short time.
- Solution for all wounds and burns.
- One of best herbs for headache and toothache.
- Anti-inflammatory properties of marigold help to cure allergies.
- Optimize the growth of new blood vessels and new skin issues.
- Reduce the chances of tumors.

1.4.5.12 Echinacea: Native to Central America, Echinacea is a flowering plant commonly known as cone flower. It is also a popular herb in the world. The leaves, flowers, stems and roots of echinacea can be used for medical purposes. This herb works as active chemicals in your body and fight against fungus, reduce flu and inflammation.

Health benefits of Echinacea

- Best anti-inflammatory herb for stiffness of joints.
- Improves immunity power of the body.
- Roots of Echinacea widely exported for medical purposes.
- Stimulate the growth of blood cells.
- Prevention against chances for cold, bronchitis and sore throats.

1.4.5.13 Dandelion: Dandelion is edible flowering plants widely grows in South America. The flowers, stem and leaves of dandelion plants also used in production of a number of medicines. Dandelion is a rich source of vitamins and nutrients. It is also used to make wine and coffee substitutes.

Health benefits of Dandelion

- Can cure liver problems.
- Act as a cleaning tonic for blood vessels.
- Balance blood sugar level and cholesterol level.
- Prevention against gallstones.
- Quick relief to ankle swelling.
- Improves the function of pancreas.

1.4.5.14 Chamomile: The chamomile flowering plant has a long tradition of using as an important herb. Its beautiful flowers contain a number of volatile oils including bisabolol, matricin, bisabolol A and Bisabolol B. Chamomile can heal many diseases with no side effects.

Health benefits of Chamomile

- Prevent diabetics and muscle spasms.
- Full care for skin.
- Reduce morning sickness during pregnancy.
- Fight against inflammation and teething problems.
- Prevention against bacterial affections.
- Balance digestive system and prevent the chances of stomach pain.
- Reduces chances of cancer.
- Help to grow thick black hair.
- Best herb choice for muscle relaxation.
- Maintain sugar balance in the blood and reduce heartburn.
- Eliminate dandruff and cool down the scalp.

1.4.5 Traditional use of medicine in Bangladesh

The rural population of Bangladesh has traditionally depended on folk medicinal healers for treatment of their ailments. These healers use medicinal plants as their primary source of medicinal formulations. Rural patients are more dependent on traditional or folk medicinal healers for treatment of urinary tract infections (UTIs) and sexually transmitted diseases (STDs) for a number of reasons including lack of access to modern medical facilities, clinging to traditional approaches, and finally hesitancy to relate this form of illnesses in front of unknown doctors. Since the traditional healer usually resides in the same village or in an adjoining area, the patient is more comfortable in seeking them for treatment. We conducted an ethnomedicinal survey among the traditional healers of various ethnic groups and in several regions of the country to obtain information on medicinal plants used to treat UTIs and STDs. Interviews were conducted in the local dialect or language about plant parts used, ailments treated, formulations, and dosages. Thirty-one species were reported by traditional healers as being used for UTIs, including leucorrhea, frequent or infrequent urination, cloudy urination and burning sensations during urination. Ten species were reported to be used against STDs like syphilis and gonorrhea (Shahadat *et al,* 2010).

1.5 Plant Review

1.5.1 Plant Name:*Artocarpuschama.*

1.5.2 Classification

Kingdom- Plantae

Phylum- Tracheophyta

Class - Magnoliopsida

Order - Rosales

Family – Moraceae

Genus- *Artocarpus*

Species –*Artocarpuschama.*

1.5.3 Genus of Artocarpus chama

The genus *Artocarpus* (Moraceae) comprises about 50 species of evergreen and deciduous trees. Economically, the genus is of appreciable importance as a source of edible fruit, yield fairly good timber and is widely used in folk medicines. The aim of the present review is to present comprehensive information of the chemical constituents, biological and pharmacological research on *Artocarpus* which will be presented and critically evaluated. The close connection between traditional and modern sources for ethnopharmacological uses of *Artocarpus* species, especially for treatment against inflammation, malarial fever, diarrhoea, diabetes and tapeworm infection. *Artocarpus* species are rich in phenolic compounds including flavonoids, stilbenoids, arylbenzofurons and Jacalin, a lectin. The extracts and metabolites of *Artocarpus* particularly those from leaves, bark, stem and fruit possess several useful bioactive compounds and recently additional data are available on exploitation of these compounds in the various biological activities including antibacterial, antitubercular, antiviral, antifungal, antiplatelet, antiarthritic, tyrosinase inhibitory and cytotoxicity. Several pharmacological studies of the natural products from *Artocarpus* have conclusively established their mode of action in treatment of various diseases and other health benefits.

Jacalin, a lectin present in seeds of this plant has a wide range of activities. Strong interdisciplinary programmes that incorporate conventional and new technologies will be critical for the future development of *Artocarpus* as a promising source of medicinal products. In the present review, attempts on the important findings have been made on identification; synthesis and bioactivity of metabolites present in *Artocarpus* which have been highlighted along with the current trends in research on *Artocarpus*

1.5.4Family of Artocarpus chama

Artocarpus chama lies under the moraceae family. The Moraceae are often called the mulberry family or fig family. They are a family of flowering plants comprising about 38 genera and over 1180 species. Most are widespread in tropical and subtropical regions, less so in temperate climates. The only synapomorphy within Moraceae is presence of laticifers and milky sap in all parenchymatous tissues, but generally useful field characters include two carpels sometimes with one reduced, compound inconspicuous flowers, and compound fruits. The family includes well-known plants such as the fig, banyan, breadfruit, mulberry, and Osage-orange. The 'flowers' of Moraceae are often pseudanthia (reduced inflorescences) (Wikipedia, 2017)

1.5.5 Plant Description: A genus of small to large evergreen trees. Laticiferous, all native of East Asia, Leaves leathery, flowers monoecious, composite, fleshy, globose or cylindrical edible fruit. Consisting of receipted and fleshy perianths and seeds. Some species also grown. Artocarpus chaplasha genus of small to large evergreen trees. Laticiferous, all native of East Asia, this plant attain a height of 30-40mt., and a girth of 3-5mt.Leaves leathery, flowers monoecious, composite, fleshy, globose or cylindrical edible fruit. Consisting of receipted and fleshy perianths and seeds. Some species also grown for timber.

Trees to 40 m tall, deciduous. Bark black, gray, or brown, coarse. Branchlets furrowed when dry, 4-8 mm thick, pubescence rust-colored to reddish yellow, hairs long and spreading to bent.Stipules amplexicaul. Leaves spirally arranged; petiole brown, 1.5-4.5 cm, densely pubescent; leaf blade elliptic, oblong, or ovate, 13-37 × 6-21 cm, abaxially densely rust-colored to grayish white pubescent but more densely so along veins,

adaxially glabrous or with sparse bent hairs, base broadly cuneate to rounded, margin entire or ± crenate, apex acute to shortly acuminate; secondary veins 9-18 on each side of midvein, apically curved, and joined together near margin, tertiary veins reticulate and with dark brown glandular points. Inflorescences axillary, solitary. Male inflorescences ellipsoid, ovoid, or clavate, 1.2-2.3 × 1-1.8 cm; bracts shield-shaped; pedicel ca. 2 mm, shortly pubescent. Female inflorescences globose to ellipsoid; bracts peltate. Style exserted. Male flowers: calyx lobes 2 or 3, ca. 5 mm, margin ciliate; filaments short; anthers ellipsoid. Fruiting syncarp yellow when young then rust-colored brown, ± globose, 5-6 cm in diam.; peduncle 1.5-4.5 cm, with short brown hairs; persistent calyx separating near top, with several persistent bracts. Drupes ellipsoid, ca. 10 × 6 mm.

1.5.6Stem: A large deciduous tree, up to 30 m. tall

1.5.7Leaf: Simple alternate, petiolate, lamina eliptic, ovate or obovate

1.5.8Fruit: A syncarp, globose, irregular lobed.(MIa, 2016)

1.6 Species of Artocarpus

1.6.1 Breadfruit (*Artocarpus altilis*)

It is a species of floweringtree in the mulberry and jackfruit family (*Moraceae*) originating in the South Pacific and eventually spreading to the rest of Oceania. British and French navigators introduced a few Polynesian seedless varieties to Caribbean islands during the late 18th century, and today it is grown in some 90 countries throughout South and Southeast Asia, the Pacific Ocean, the Caribbean, Central America and Africa. Its name is derived from the texture of the moderately ripe fruit when cooked, similar to freshly baked bread and having a potato-like flavor (J.R *et al,* 2013).

1.6.2Description

Breadfruit trees grow to a height of 26 m (85 ft). The trees are monoecious, with male and female flowers growing on the same tree. The male flowers emerge first, followed shortly afterward by the female flowers. Pollination occurs mainly by fruit bats, but cultivated varieties produce fruit without pollination. The compound, false fruit develops from the swollen perianth, and originates from 1,500-2,000 flowers visible on the skin of

the fruit as hexagon-like disks. Breadfruit is one of the highest-yielding food plants, with a single tree producing up to 200 or more grapefruit-sized fruits per season, requiring limited care. In the South Pacific, the trees yield 50 to 150 fruits per year, usually round, oval or oblong weight. Breadfruit is closely related to the breadnut, from which it might have been naturally selected. It is noticeably similar in appearance to its relative of the same genus, the jackfruit (*Artocarpus heterophyllus*) (J.R *et al*, 2013).

1.6*Artocarpus camansi*

The breadnut, is a medium-sized tree found in the mulberry family Moraceae. Native to Papua New Guinea, it is a relative of the breadfruit and is commonly used as a staple crop.Other common names for plant include *kluwih*in Indonesia, *chataigne* or *castaña* (French and Spanish for the unrelated but culinarily similar chestnut") in the Caribbean, *pana de pepita*, *kamansi* in the Philippines, *kapiak* in New Guinea, and *kos-del*(Ragone *et al*, 2012).

1.6.1Distribution and origins

Artocarpus camansi is endemic to New Guinea and possibly Indonesia and the Philippines.The ambiguity of the origins of this plant is a result of spread and domestication of multiple species of breadfruit, *Artocarpus camansi* included, as humans spread from island to island in the Pacific. There is speculation that breadfruit, *Artocarpus altilis*, the most widely used breadfruit, was selectively bred from *Artocarpus camansi* (Ragone *et al*, 2012).

1.6.2*Artocarpus integer*

Commonly known as **cempedak** (pronounced "chem-pe-dak"), is a species of tree in the family Moraceae, and in the same genus as breadfruit and jackfruit. It is native to southeast Asia, from Indonesia and the Malay Peninsula to the island of New Guinea. Furthermore, the tree has also been introduced to Queensland (Wikipedia.org, 2017).

1.6.4Description

Cempedak trees are large, evergreen trees. They can grow to a height of 20 m, although most only reach a dozen meters. The trees are monoecious, with male and female flowers growing on the same tree. There are many varieties, although few are named. The vigorously growing tree can bear heavy crops of fruit once or twice a year. (Wikipedia.org, 2017)

Chapter 2: literature Review

2.1 Chemical constituents isolated from *Artocarpus chama*

The preliminary phytochemical screening revealed that the methanol and pet ether fraction of fruits possess the presence of various bioactive components like flavonoids, alkaloids and carbohydrates(Ahmed *et al*, 2012).

2.2 Quantitative analysis of the phytochemical constituent of fruits of *Artocarpus chama*

2.2.1 Determination of total phenol content of fruits of *Artocarpus chama*

The pet ether extract of the fruits of *A. chama* Buch. was found to contain large amount of phenolics, 178.08± 2.05 mg/g Gallic acid equivalent(GAE) while methanolic extract contain moderate amount, 41.12 ± 1.83 mg/g GAE using Folin-Ciocalteau method (Ahmed *et al*, 2012).

2.2.2 Determination of total flavonoid content of fruits of *Artocarpus chama.*

The total flavonoid content of pet ether and methanol extracts were found to be 24.95 ± 0.36 and 25.71 ± 0.59 mg/ quercetin equivalent, respectively. These results suggested that the antioxidant activities of *A. chama* might be due to its flavonoid content since *A. chama* roots contains variety of prenylated flavonoids e.g. isoprenylated flavones, flavones (Yong-Hong *et al*, 2004).
(Ahmed *et al*, 2012).

2.3 Antioxidant property of the fruits of *Artocarpus chama*

2.3.1 DPPH^{-} radical scavenging activity

In DPPH radical scavenging assay both pet ether and methanol extract exhibited a concentration-dependent antiradical activity by inhibiting DPPHradical. Ascorbic acid, which is a well-known antioxidant, showed higher degree of free radical-scavenging activity than that of the plant extract at each concentration points. The IC_{50}value of

thecrude pet ether and methanol extract were 27.64 µg/ml and 39.08 µg/ml, respectively, while the IC_{50}value for the reference ascorbic acid was 12.70 µg/ml (Ahmed *et al*, 2012).

2.3.2 Cupric reducing antioxidant capacity (CUPRAC)

It was observed that at concentration level of 200 µg/ml, the reducing capacity of pet ether, methanol extract and ascorbic acid was 0.3115, 0.3545 and 0.744 µg/ml, respectively(Ahmed *et al*, 2012)

2.3.3 Reducing power antioxidant capacity

The reducing power capabilities of the plant extracts compared to ascorbic acid. Both extracts displayed good reducing power which was found to rise with increasing concentrations of the extracts. At 200 µg/ml concentration level, the absorbance of standard ascorbic acid, pet ether extract and methanol extract were 1.01, 0.52 and 0.60, respectively. Both the plant extracts showed almost similar reducing power capacity(Ahmed *et al*, 2012).

2.4 Quantitative analysis of the phytochemical constituent of seeds of *Artocarpus chama*

2.4.1Determination of total phenol content of seeds of *Artocarpus chama*

The methanolic extract of seeds of *Artocarpus chama*was found to contain high amount of phenolics, 61.04 mg/g gallic acid equivalent using Follin-ciocalteau method (Ahmed *et al*, 2013).

2.4.2Determination of total flavonoid content of seeds of *Artocarpus chama*

The total flavonoid content of methanolic extract was found to be 33.71 mg/g quercetin equivalent. Results told that the antioxidant activities of *A. chama*probably for its flavonoid content (Ahmed *et al*, 2013).

2.5Antioxidant property of the seeds of *Artocarpus chama*

2.5.1DPPH^{-} radical scavenging activity

The IC_{50}value of the crude methanol extract was 54.29 ± 1.98 µg/ml, while the IC50value for the reference ascorbic acid was 14.56. at concentration level of 200 µg/ml, the reducing capacity of methanol extract and ascorbic acid is 0.324 and 0.744, respectively (Ahmed *et al*, 2013).

2.5.2Cupric reducing antioxidant capacity (CUPRAC)

At concentration level 200 µg/ml, the reducing capacity of methanol extract and ascorbic acid was 0.324 and 0.744 respectively (Ahmed *et al*, 2013).

2.5.3 Reducing power antioxidant capacity

At concentration level 200 µg/ml, the absorbance of standard ascorbic acid and methanol extract was 1.01 and 0.56 respectively (Ahmed *et al*, 2013).

2.5.4Determination of total phenol content

The total phenolic content of plant extracts was determined using Folin–Ciocalteu reagent (Yu et al., 2002). Plant extracts (100 µl) were mixed with 500 µl of the Folin–Ciocalteu reagent and 1.5 mL of 20 % sodium carbonate. The mixture was shaken thoroughly and made up to 10 mL using distilled water. The mixture was allowed to stand for 2 hours. Then the absorbance at 765 nm was determined. (Ahmed.T *et al.*, 2013)

Chapter 3: Materials and Method

3.1 Preparation of the Plant Sample

3.1.1 Collection and Proper Identification of the Plant Sample

At first with the help of a comprehensive literature review*Artocarpus chama*selected for this investigation. The barks were collected from Botanical garden, Dhaka, Bangladesh during the month of December.

3.1.2 Preparation of plant sample

The experimented plant barks were separated from undesirable plant parts and dried in the sun shed for 5 days. After completion of drying, dried leaves were pulverized into coarse powder by suitable grinding machine. Powders were kept in clean airtight glass containers for further use. Here the grinder was properly cleaned so that contamination with previous ground material on other foreign matter can be avoided. The weight of leaf powder was 256 gm. Finally, it was placed in dry and cool area until experiment begins.

3.1.3 Extraction of powdered sample

Two glass jars were washed properly and then rinsed with methanol and pet ether as well as dried. After that the dried barks powder were put into the jar and methanol and pet ether were poured into it up to 1-inch height above the sample surface. The container was sealed with its content and kept for 4 days with occasional shaking and stirring. This shaking was done to get better extraction.

3.1.5 Procedure of Evaporation in Rotary Evaporator

- After the filtration process two parts were obtained namely ‘residual part’ and filtered part or filtrate‟.
- The filtered part, which contains the substance soluble in methanol was putted into a 1000ml round bottom flask and then the flask was place in a rotary evaporator.
- The evaporation was done at 50-degree Celsius for methanol and pet ether.
- The number of rotation per minute was selected as 60 rpm. The pressure of the vacuum pumper machine was 6bar.

- The water flow through the distillation chamber was also provided in a satisfactory flow rate.
- When the evaporation seemed to be satisfactory, then the methanol and pet ether extract were collected in a 50mL beaker.
- The extractions were collected from the evaporating flask and the solvents were collected from the receiving flask.
- The evaporator flask was rinsed by methanol and pet ether in case of the extract of methanol and pet ether extract.
- Then the beaker was covered with aluminium foil paper and kept for 60 minutes.
- Finally, the concentrated methanol and pet ether plant extract were found and stored in the laboratory refrigerator from which the extracts were used for many chemical investigations.

The extracts of methanol and pet ether were chosen for investigation and was labelled as-ACBM and ACBP (the methanol and pet ether extract of *Artocarpus chama* respectively)

3.2 Drugs

Diazepam, Ibuprofen and Indomethacin were used for current study

3.3 Experimental animal

For the pharmacological investigation 30 Swiss albino mice were collected from ICDDRB, Dhaka, Bangladesh. The average weights of the mice were 16 to 18 gm. Standard environmental situation was maintained to keep the mice. The condition was 55-65% relative humidity, 12 hours light/dark cycle and 24.0±0°C temperature. Also, sufficient amount of food and water was supplied all the time.

Figure 3.1: Swiss albino mice

3.4 Ethicalapproval:

Institutional animal ethical committee accepted the guidelines which were followed for animal test (Zimmermann, 1983).

3.5 Identification of animals during experiment

Each group consists of six mice and hence it is difficult to identify and observe at time six mice receiving same treatment. Thus, it was important to identify individual animal of a group during the treatment. The animals were individualized by marking: marked as M_1=mice 1, M_2=mice 2, M_3=mice 3 and so on with different colors.

3.6 Pharmacological investigation of plantextracts:

The following pharmacological investigations were done to determine the medicinal effect of the experimented extracts:

- CNS depressant activity
- Analgesicactivity

3.6.1 Study of CNS Depressant effect of ACLM and ACLP extracts:

CNS Depressant drugs are the agents which slow down the activity of brain. These types of drugs are prescribed by doctor for the treatment of panic attack, anxiety, insomnia etc. Mostly CNS Depressants activate GABA neurotransmitter. This helps in decreasing brainactivity.The CNS depressant action of *Artocarpus chama* plant extracts were

observed by comparing with the standard diazepam in the experimented rodents. CNS depressant activity was determined by using two techniques.

They are:

- Open fieldtechnique
- Hole boardtechnique

3.6.2 The Design of the CNS depressantExperiments:

In both methods 36 mice were chosen randomly and then divided into 6 groups. They were group G_1 to G_6 where 6 mice were in each group. A particular treatment was given to each group. Before this specific treatment, weight of every mouse was measured accurately as well as marked. Also, the dosage of the sample and standard were also settled according to bodyweight.

Group-G_1- ACLM 200 mg/kg

Group-G_2- ACLM 400 mg/kg

Group-G_3- ACLP 200 mg/kg

Group-G_4- ACLP 400 mg/kg

Group-G_5 Standard (Diazepam)

Group-G_6- Control (5 % CMC in distilled water)

3.6.2.1 Reagents, Chemicals andEquipment's

Table 3.1: Reagents, Chemicals and equipment's used for CNS depressant test

Reagents Chemicals and Equipment's	Source
Diazepam	Square Pharmaceuticals Ltd.
5 % CMC-low viscous (as suspending agent)	
Distilled water	Laboratory

Sterile disposable syringe (1ml, 100 divisions)	CHPL, India
Tuberculin syringe with ball shaped end	Merck, Germany
Electronic and digital balance	Denver Instruments M-220/USA

3.6.3 Preparation of drug and chemical solution:

At first standard which is considered to be diazepam at 1 mg/kg dose was prepared. Then required amount of diazepam was dissolved in water and according to body weight was given to each mouse orally.

Crude extract was also prepared at a dose of 200mg/kg and 400mg/kg by body weight of mice. Then doses were measured as well as 250 mg of CMC (low viscous) was added to every preparation. Then dose was administered according to their body weight.

Table 3.2: Test samples used in the estimation of CNS Depressant activity of *A.chama*plant.

Group	Treatment	Dose	Route of administration
Group 1 (Extract)	ACLM	200 mg/kg	Orally
Group 2 (Extract)	ACLM	400 mg/kg	Orally
Group 3 (Extract)	ACLP	200 mg/kg	Orally
Group 4 (Extract)	ACLP	400 mg/kg	Orally
Group 5 (Standard)	Diazepam	1 mg/kg	Orally
Group 6 (Control)	5 % CMC in Distilled Water	10 ml/kg	Orally

3.7 Open field test

Gupta's open field method (Gupta et al., 1971) was followed to carry out Open field test. The box was half square meter as well as divided into squares each. On the other hand, the box was black and white color like a chess board. The apparatus had a wall which was 40cm in height. For 3 minutes, each square was counted which was visited by mice. Also, during the study period, several results were taken on 0, 30, 60, 90 and 120minutes.

The procedure for evaluation of CNS depressant effect of *A. chama* plant by open field test is given below:

i. At first mice were weighed and after that categorized into 6 groups where 6 mice were in each group
ii. Thenby a long needlewhichwasattachedwithballshapedend, sample and standards were administered orally. This was done at 0 minutes.
iii. Number of squares which was visited by mice at 0 Minute was counted for 3 minutes
iv. Eventually after 30, 60, 90 and 120 minutes the number of times all the mice traveled from one compartment to another was counted for a duration of 3 minutes and afterwards the data was recorded for the two extracts of the plant.

3.8 Hole Board Test

The method described by (Takagi *et al*, 1971) was implemented for this study. Again, 36 mice were equally divided into 6 groups. The control group received 5% CMC in distilled water (10 ml/kg body weight), the standard group received Diazepam (1 mg/kg body weight) and the experimental groups received crude extract at 200 mg/kg and 400 mg/kg body weight. 16 holes, each of 4cm in diameter, were made at a plane plate of a woody table at a height of 1 foot from the ground. The number of poking and Deeping of mice through the hole was counted for a period of 3 minute after 0, 30, 60, 90, and 120 min of oral administration of theextract.

The procedure for evaluation of CNS depressant effect of *A. chama* plant by Hole board test is given below:

i. At first mice were weighed and after that categorized into 6 groups where 6 mice were in each group
ii. Thenby a long needlewhichwasattachedwithballshapedend, sample and standards were administered orally. This was done at 0 minutes.
iii. Number of holes which was visited by mice at 0 Minute was counted for 3 minutes
iv. Eventually after 30, 60, 90 and 120 minutes the number of times all the mice gave pocking and dipping was counted for a duration of 3 minutes and afterwards the data was recorded for the two extracts of the plant.

3.9 Analgesic activity of *Artocarpus chama* plantextracts:

Drug which is used to relieve pain is called analgesic drug. These drugs are also known as painkiller. The analgesic test was done by two methods. These two methods are: -

- Acetic acid induced writhingtechnique
- Formalin induced paintechnique.

3.9.1 Design of the analgesic experiment

36 mice were chosen anyway and divided into 6 groups where the groups were from G_1 to G_6 as well as 6 mice were in each group. Each group got a specific treatment. Before the treatment, each mouse was weighed properly as well as marked. Then the dosage of the test sample and control materials was also settled according to body weight.

Group-G_1- ACLM 200 mg/kg

Group-G_2- ACLM 400 mg/kg

Group-G_3- ACLP 200 mg/kg

Group-G_4- ACLP 400 mg/kg

Group-G_5 Standard (Diazepam)

Group-G_6- Control (5 % CMC in distilled water)

3.9.2 Acetic acid-induced writhingtechnique:

Acetic acid induced writhing test is a technique where analgesic behavior is observed. In this method (Ahmed *et al*, 2001) intra-peritoneally acetic acid was administered to the mice so that pain sensation generates. Here, indomethacin was considered as standard. At first the normal saline, extracts at a dose of 200 mg/kg and 400 mg/kg as well as standard drug were administered orally. After 30 minutes, the solution of 0.7% v/v acetic acid was administered intraperitoneally. After administration of solution of acetic acid, no writhing was counted for 5 minutes. After 5 minutes, writhing was counted for 15minutes. For that each mouse was placed on observation table and noticed the number of writhing of mice. The mice did not give full writhing all the time. They gave half writhing also. So, two incomplete writhing were counted as one completewrithing.

3.9.3 Reagents, Chemicals andEquipment's

Table 3.3: Reagents, Chemicals and equipment's used for acetic acid induced analgesic test

Reagents Chemicals and Equipment's	Source
Acetic acid	Merck, Germany
Indomethacin	Square Pharmaceuticals Ltd.
5 % CMC- low viscous (as suspending agent)	BDH Chemicals Ltd
Distilled water	Laboratory
Sterile disposable syringe (1ml, 100 divisions)	CHPL India
Tuberculin syringe with ball shaped end	Merck, Germany
Electronic and digital balance	Denver Instruments M-220/USA

3.9.4 Preparation of drug and chemical solution:

For the preparation of standard which is indomethacin at a dose of 1 mg/kg, specific quantity of this drug was suited.Then required amount of indomethacin was dissolved in water and then according to body weight was given to each mouse orally.

Crude extract was also prepared at a dose of 200mg/kg and 400mg/kg by body weight of mice. Then doses were measured as well as 250 mg of CMC (low viscous) was added to every preparation. Then dose was administered according to their body weight.

Table 3.4: Test samples used in the estimation of Analgesic activity of *A.chama*plant.

Group	Treatment	Dose	Route of administration
Group 1 (Extract)	ACLM	200 mg/kg	Orally
Group 2 (Extract)	ACLM	400 mg/kg	Orally
Group 3 (Extract)	ACLP	200 mg/kg	Orally
Group 4 (Extract)	ACLP	400 mg/kg	Orally
Group 5 (Standard)	Indomethacin	10 mg/kg	Orally
Group 6 (Control)	5 % CMC in Distilled Water	10 ml/kg	Orally

3.9.5 Procedure of analgesic activity of *Artocarpus chama* extract by acetic acid induced writhingtechnique

i. At first weight of the mice were measured as well as split into 6 groups where 6 mice were in each group
ii. Later at 0 hour orally standard and samples were administered with the help of ball shaped ended needle.
iii. Then acetic acid was induced to each mouse of group (G_1 to G_6) intraperitoneally after 30 minutes

iv. To confirm the acceptable absorption of administered specimen, 30 minutes interval was given

v. Afterwards 5 minutes of induced acetic acid, the writhing for each mouse were counted for 15 minutes

vi. Finally, the documented numbers of writhes were compared with standard group mice (Ahmed *et al*, 2001).

3.10Formalin induced pain method

Formalin test is another method (Sharma *et al*, 2010) by which analgesic activity is also observed. In this case, formalin injection is induced to mice's right hind paw. As a result, biphasic pain is produced. For standard, we choose ibuprofen which has pain sensation inhibition control. After that the standard was compared with test samples and control

3.10.1Reagents, Chemicals andequipment's:

Table 3.5: Reagents, Chemicals and equipment's used for formalin induced analgesic test

4

Reagents Chemicals and Equipment's	Source
Formalin	Merck, Germany
Ibuprofen	Square Pharmaceuticals Ltd.
5 % CMC- low viscous (as suspending agent)	BDH Chemicals Ltd
Distilled water	Laboratory
Sterile disposable syringe (1ml, 100 divisions)	CHPL India
Tuberculin syringe with ball shaped end	Merck, Germany
Electronic and digital balance	Denver Instruments M-220/USA

3.10.2 Preparation of drug and chemical solution:

For the preparation of standard which is ibuprofen at a dose of 10 mg/kg, specific quantity of this drug was suited.Then required amount of ibuprofen was dissolved in water and then according to body weight was given to each mouse orally.

Crude extract was also prepared at a dose of 200mg/kg and 400mg/kg by body weight of mice. Then doses were measured as well as 250 mg of CMC (low viscous) was added to every preparation. Then dose was administered according to their body weight.

Table 3.6: Test samples used in the estimation of Analgesic activity of *A.chama*plant.

Group	Treatment	Dose	Route of administration
Group 1 (Extract)	ACLM	200 mg/kg	Orally
Group 2 (Extract)	ACLM	400 mg/kg	Orally
Group 3 (Extract)	ACLP	200 mg/kg	Orally
Group 4 (Extract)	ACLP	400 mg/kg	Orally
Group 5 (Standard)	Ibuprofen	100 mg/kg	Orally
Group 6 (Control)	5 % CMC in Distilled Water	10 ml/kg	Orally

3.10.3 Procedure of analgesic activityof *Artocarpus chama* extract byformalin

i. The mice were weighed and then split into 6 groups where 6 mice were in each group

ii. Later at 0-hour standard and samples were administered to mice of group (G_1 to G_6) orally by a needle which contained a ball shaped end

iii. 20µl of 1% formalin was induced to each mouse in the right-hand paw after 30 minutes

iv. In first phase which is after the administration of formalin, numbers of licking and biting by mice are counted. The duration of first phase is 0 to 300 seconds.

v. In second phase, numbers of licking and biting by mice are also counted and the time was 15-30 minutes.

3.10.4 Counting of licking and biting of paws

Because of induced formalin, mice bite or lick the wounded paw and this was determined by help of stopwatch

3.11 Statistical Analysis

Total values which were obtained from the experiments are represented as mean ± standard error of the mean (SEM). Statistically obtained data was estimated by using ANOVA (Analysis of variance) and post-hoc Dunnett's test which was associated with SPSS program (SPSS 16.0, USA). Results were calculated by using Microsoft Excel 2016. Dissimilarity between the means of the various groups were measured significantly at $p < 0.05$, $p < 0.01$ and $p<0.001$.

Chapter 4: Result and Discussion

4.1.1 Result of CNS depressant activity in open field test

At 200 mg/kg and 400 mg/kg dose, experimental leave extracts were administered to mice. As a result, the movements of mice were reduced in a dose depending manner. Also, it was comparable with diazepam (standard). This movement lowering effect of extract on mice was observed at 30 min interval from zero minute up to 120 minutes. The extracts caused reduction in movement and this may be connected to CNS depression, as reduction or depression of movement is common to most antipsychotics.

Table 4.1: Data of CNS Depressant activity test of *A.chama* plant extracts by Open field method:

Group	Treatment	Dose	Number of movement					
			-30 min	**0 min**	**30 min**	**60 min**	**90 min**	**120 min**
Group 1 (Extract)	ACLM	200 mg/kg	221.83± 34.20*	86.83± 12.19*	79.00± 25.81*	71.00± 20.16***	38.67± 10.29***	37.33± 8.51***
Group 2 (Extract)	ACLM	400 mg/kg	66.17± 21.51	44.67± 10.41***	43.33± 10.01***	40.17± 7.17***	40.00± 4.12***	39.67± 8.61***
Group 3 (Extract)	ACLP	200 mg/kg	188.50± 28.16	99.0± 19.79	31.67± 6.65***	25.17± 6.87***	41.83± 9.44***	42.83± 13.43***
Group 4 (Extract)	ACLP	400 mg/kg	63.50± 16.96	44.67± 7.41***	19.33± 2.75***	14.00± 1.93***	23.00± 5.93***	53.50± 12.67***
Group 5 (Standard)	Diazepam	1 mg/kg	137.50± 18.23	137.50± 18.23***	36.17± 10.65	36.00± 8.11***	35.67± 10.87***	30.33± 15.95***
Group 6 (Control)	Distilled Water	10 ml/kg	130.83 ± 24.10	137.50 ± 18.23	126.00 ± 13.38	159.67 ± 6.32	162.50 ± 10.34	153.00 ± 9.41

Values are expressed as Mean ± SEM; n=6. One-Way Analysis of Variance (ANOVA) trailed by Dunnett's test. ***p<0.001; **p<0.01; *p<0.05 are considered to besignificant.

4.1.2 Result of CNS depressant activity in Hole board test

At 200 mg/kg and 400 mg/kg dose, experimental leave extracts were administered to mice. As a result, the number of poking and Deeping of mice at the hole board were reduced in a dose depending manner. Also, it was comparable with diazepam (standard). This lowering effect of extract on mice was observed at 30 min interval from zero minute up to 120 minutes. The extracts caused reduction in poking and Deeping and this may be connected to CNS depression, as a reduction or depression of poking and Deeping is common to most antipsychotics

Table 4.2: Data of CNS Depressant activity test of *A.chama* plant extracts by Hole- board method

Group	**Treatment**	**Dose**	**Number of movement**					
			-30 min	**0 min**	**30 min**	**60 min**	**90 min**	**120 min**
Group 1 (Extract)	ACLM	200 mg/kg	23.17 ± 2.21	8.17± 1.40	6.00± 1.21	6.67 ± 0.88	4.00± 0.86*	5.83± 1.19
Group 2 (Extract)	ACLM	400 mg/kg	14.33 ± 3.15	13.33 ± 2.14	10.50 ± 2.46	7.33 ± 0.95	6.67± 0.76	4.33± 1.12
Group 3 (Extract)	ACLP	200 mg/kg	22.67 ± 5.14	10.00 ± 1.83	8.67± 2.16	8.50 ± 2.40	8.50± 3.21	18.17 ± 4.25

Group 4 (Extract)	ACLP	400 mg/kg	14.00 ± 1.83	8.83± 0.91	6.00± 2.11	3.67 ± 0.99**	2.00± 0.77**	13.50 ± 1.67**
Group 5(Standard)	Diazepam	1 mg/kg	17.50 ± 3.70	5.50± 1.84*	5.17± 0.87*	4.00 ± 0.77*	3.67± 0.61*	3.33± 0.33
Group 6 (Control)	Distilled Water	10 ml/kg	20.83 ± 2.87	15.50 ± 1.12	11.50 ± 1.54	9.33 ± 1.33	9.50± 1.84	7.33± 1.48

Values are expressed as Mean ± SEM; n=6. One-Way Analysis of Variance (ANOVA) trailed by Dunnett's test. **p<0.01; *p<0.05 are considered to besignificant.

4.1.3 Result of Analgesic (Formalin induced) activity

The analgesic activity by formalin induced pain method was determined by counting paw licking and biting events. The leaf extract of the experimental plant at a dose of 200mg/kg and 400 mg/kg prevent the licking and biting activity of mice in a dose depending manner in the late phase compared to standard which was Ibuprofen. These events are given in the following table:

Table 4.3: Data of Formalin induced analgesic activity test of *A.chama* plant extracts

Group	**Treatment**	**Dose**	**Number of Paw leaking**	
			Early phase	**Late phase**
Group 1 (Extract)	ACLM	200 mg/kg	78.00±2.84	63.67±11.06*
Group 2 (Extract)	ACLM	400 mg/kg	56.33±9.63*	51.17±7.18***

Group 3 (Extract)	ACLP	200 mg/kg	59.17±3.25	53.83±4.87**
Group 4 (Extract)	ACLP	400 mg/kg	77.00±5.23	73.17±7.04
Group 5(Standard)	Ibuprofen	100 mg/kg	28.33±4.22	40.83±7.43
Group 6 (Control)	Distilled Water	10 ml/kg	82.50±10.97***	97.67±12.41***

Values are expressed as Mean ± SEM; n=6. One-Way Analysis of Variance (ANOVA) trailed by Dunnett's test. ***p<0.001; **p<0.01; *p<0.05 are considered to besignificant.

4.1.4 Result of Analgesic (Writhing) activity

In this test, the antinociceptive effects of plant *Artocarpus chama* were investigated by administering 200 mg/kg and 400 mg/kg dose to rodents. By applying this test, it was seen that there was significant effect of plant extract compare to standard drug (Indomethacin). Among the sample of crude extracts Group 2 and Group 8 showed better results compared to standard which was Indomethacin.

Table 4.4: Data of Analgesic (Writhing) activity test of *A.chama* plant extracts

Group	Treatment	Dose	Number of writhing
Group 1 (Extract)	ACLM	200 mg/kg	26.83±6.503418***
Group 2 (Extract)	ACLM	400 mg/kg	19.17±3.609401***
Group 3	ACLP	200 mg/kg	21.67±1.054093***

(Extract)			
Group 4 (Extract)	ACLP	400 mg/kg	19.17±2.056156***
Group 5(Standard)	Indomethacin	10 mg/kg	49.83±5.095205***
Group 6 (Control)	Distilled Water	10 ml/kg	6.5±1.875278

Values are expressed as Mean ± SEM; n=6. One-Way Analysis of Variance (ANOVA) trailed by Dunnett's test. ***p<0.001; **p<0.01; *p<0.05 are considered to besignificant.

4.2 Discussion

The present study was conducted to elucidate analgesic and CNS depressant activity of the methanol extract of *Artocarpus chama* leaves.It contains some of the phytochemicals such as flavonoids, alkaloids, carbohydrates and phenol. Most of the parts of this plant have medicinal benefits like the plant also can cure the pain and inflammation in the body.Some components like flavones , were isolated from the roots of *Artocarpus chama* which fight against breast adenocarcinoma and lung carcinoma.

For the different experiment diazepam, ibuprofen, indomethacin was used as standard. Diazepam was used for open field and hole board test as a standard. Ibuprofen was used for formalin test as standard. Indomethacin was used for writhing test as standard. Diazepam being a benzodiazepine class of drugs, it has sedative action. It increases neuronal membrane permeability to chloride ions by binding to stereospecific benzodiazepine receptors on the postsynaptic GABA neuron and enhancing the GABA inhibitory effects resulting in hyperpolarization and stabilization. The methanol and pet ether extract of plants gave action compared to diazepam, the movement of mice decreased in a dose dependent manner in open field. As phytochemical screening revealed that the plant contains flavonoid and flavonoid has CNS depressant activity so it might be flavonoid that is giving the depressant activity (Ahmed *et al*, 2012). In hole board test, it is found that the movements of the mice are decreasing, So, it also indicates that the plant has depressant activity. The results are also similar to diazepam. It can be concluded that the plant has depressant activity.

From the analgesic activity results, in case of formalin induce pain test ibuprofen was considered as standard. In Formalin induced pain test, formalin not only produce acute pain but also produces long lasting hyperalgesia (Zao *et at*, 2017).The plant extracts showed decreasing leaking and biting compared to standard, it may be due to the presence of phytochemical constituents which is reducing the number of writhing, so the methanol and pet ether extracts of leaves showed analgesic activity. Writhing test is a chemical method used to induce pain of peripheral origin by injecting of irritant (J. Pharmacol, 20120. Here we use acetic acid as irritant, which induced pain at the peritoneum.In case of writhing test, Indomethacin was considered as standard drug, it

reversibly inhibits the cox-1 and cox-2 enzymes, thus resulting in reduced synthesis of prostaglandin precursors, the methanol fraction of the leavesshowed very good writhing compared to standard, it might be flavonoid and alkaloid which is giving anti-inflammatory activity as the plant contain these constituents.

From our best knowledge, this is the first report of CNS depressant and analgesic activity of *Artocarpus chama* leaves.Now it can be concluded on the basis of results obtained from investigation that the plant may be useful as CNS depressant and analgesic agent. But our work was only preliminary effort. It will require additional detailed advanced investigation.

Chapter 5: Conclusion

5.1 Conclusion

We can find thousands of plants which are medicinally useful, *Artocarpus chama* is one of them. The plant showed analgesic and CNS activity. For determining the effects CNS (Open field and Hole board) And Analgesic (Formalin induced pain and Writhing test) test were done. The results of ACLM 200mg/kg, ACLM 400 mg/kg and ACLP 200 mg/kg, ACLP 400 mg/kg of CNS and Analgesic test are statistically significant. But it was only preliminary testing, we did not do phytochemical testing it is required further comprehensive exploration as well as depiction of active compounds and necessitates preformulating studies for expansion of a potential dosage form.

Based on the results of the present study, it can be proposed that the leaf part of *Artocarpus chama* in general methanol soluble fractions in particular, has less strong CNS depressant and analgesic properties. These results also may lend support to the relevant phytochemical and pharmacological works carried out so far on this plant. However, further studies are suggested to be undertaken to understand the underlying mechanism of the observed activities and to isolate, purify and characterise active phytochemical ingredient(s) responsible for these bioactivities in animal models.

The future goal of this study is to identify effective, cheap and available modalities to cope up with the upsurge of the dangers of diseases of different etiology in Bangladesh. Approaches may be developed to prevent and/or treat illness easily and effectively with readily available and cheaper resources. This research may be a platform for further investigation in this area. It is likely to show directions for the researchers to find ways out to save our lay people from the curse of diseases. Future endeavours in this area may open up exciting new therapeutic avenues.

Chapter 6: Reference

6.1 References

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