An *In-vivo* Study on Anti-emetic and *In-vitro* Study on Anthelmintic Properties of *Cassia fistula*

By

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

Department of Pharmacy
Brac University
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Declaration

It is hereby declared that

1. The thesis submitted is my 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 "An *In-vivo* Study on Anti-emetic and *In-vitro* Study on Anthelmintic Properties of *Cassia fistula*" submitted by Tasnuva Shahreen Adrita (15146093) of Spring, 2015 has been accepted as satisfactory in partial fulfillment of the requirement for the degree of Bachelor of Pharmacy on August 22, 2019.

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Ethics Statement

The animals for antiemetic study were chick models which were collected from BLRI (Bangladesh Livestock Research Institute) with proper certification of permission from the institution. Then, earthworms were used in anthelmintic study which was collected from The University of Dhaka premises. The animal models used were used with the prior permission of the respective authorities. Most importantly, they were handled with care and taken to processes only required in the protocol, unnecessary hurting the animals was strictly avoided.

Abstract

Cassia fistula is a tropical and sub-tropical legume tree which is widely known for its antioxidant, anti-inflammaory, antitumor, hypolipidemic, antimicrobial, hepato-protective and many pharmacological activities. The present study focuses on the investigation of antiemetic and anthelmintic activity of crude methanolic extract of *Cassia fistula* using 2-4 days chicks (32-52gm) model and *Pheretima posthuma* model respectively. The chick samples were administered with copper sulphate by oral route of administration to initiate emesis among them. Afterwards they were administered with leaf extract (150 mg/kg) orally which exhibited significant antiemetic property (61.58%, 88.41% and 30.79%) compared with reference drug chlorpromazine (29.88%). *In-vitro* anthelmintic tests were done with earthworms (*Pheretima posthuma)* by using *Cassia fistula* leaf extract of different concentrations (60 mg/mL, 80 mg/mL and 100 mg/mL). Albendazole (20 mg/mL) was included as a standard reference and for control distilled water was used. The two concentrations of *Cassia fistula* extract which were 80 mg/mL and 100 mg/mL exhibited significant results which were superior than the standard used (albendazole).

Keywords: *Cassia fistula;* Emesis; Retching; Anthelmintic; Paralysis.

Dedication

This respectful work is dedicated to my parents for their benevolent love and inspiration.

Acknowledgement

First and foremost, I am very grateful to Almighty Allah for granting me the noble scope to attain my project and blessing me with great patience and robustness throughout the session.

I am thankful to my supervisor Faria Tahsin, Lecturer, Department of Pharmacy, Brac University for appointing me to perform my thesis project under her supervision. My deepest regards for giving me the chance and proper guidance throughout every steps and difficulties.

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List of Acronyms

mm	Millimeter
cm	Centimeter
SGOT	Serum Glutamic Oxaloacetic Transaminase
SGPT	Serum Glutamic Pyruvic Transaminase
mg	Milligram
ml	Millilitre
kg	Kilogram
DMSO	Dimethyl Sulfoxide
SEM	Standard Error of Mean
CNIV	Chemotherapy Induced Nausea and Vomiting
5-HT4	5-Hydroxytryptamine receptor 4

Chapter 1

Introduction

For centuries, parasites are of great distress in medicinal perspectives as it causes numerous complications for mankind and other living things. Though there have been advancement in treatments some adverse effects have always been as a major obstacle and gave rise to issues like drug resistances due to the non-selective adherence of the preventive measures or anthelmintics (Anbu, Murali, Sathiya, Saraswathy, & Azamthulla, 2015). On the other hand, emesis tends to be a process that keeps the body free from complications due to toxic compounds by flushing them out and preventing the reoccurrence of the situation. Induce of emesis can be the result of many chemotherapeutic agents. Emesis is usually comprised of nausea (decreased gastric motility and increased reverse peristalsis), retching (contraction of stomach antrum and relaxation of the fundus and cardia) and vomition (gastric or small intestinal contents are expelled out of the mouth). Although the exact mechanism is not yet developed and known it is suggested that the vomiting center and chemoreceptor trigger zone (different centers of the brain) found in the brain stem mainly controls emesis. These emesis control centers contain receptors for dopamine, serotonin, histamine (H1 and H2), opioids, acetylcholine and other endogenous neurotransmitters. It has been established in different studies that peripheral 5HT receptors gets activated and has very important role in copper sulphate induced emesis. Though significant progresses have been occurred in the treatment but effective management of nausea and vomiting is still not satisfactory (Ahmed, 2012). Medicinal plants can be considered as a great solution to the above mentioned problems. These plants are diversified and their demand is upsurging day by day due to less side effects and easy availability. Though world is enriched with lots of herbal drugs the quest for latest drugs with better potency with less aftereffects and affordability has always been a concern

for the scientific researchers. So, the researchers are working to find out effective but cheap drugs from traditional plants. In particular, the plant *Cassia fistula* has remarkable medicinal importance and it is widely allocated throughout Bangladesh and tropical areas around the world. *Cassia fistula* contains a range of biologically dynamic components such as flavonoids, flavon-3-ol derivatives, alkaloid, glycosides, saponin, anthraquinones, terpenoids, tannin, reducing sugar and steroids. These constituents showed more therapeutic activities such as antiemetic, anthelmintic, anti- inflammatory, antidiabetic, antioxidant hypolipidemic, hepato-protective, antimicrobial, antiulcer, antitumor etc. More significantly, there is no reported study on antiemetic and anthelmintic properties of *Cassia fistula* leaves. So therefore the study was formulated to analyze the antiemetic and anthelmintic properties on chick models and earthworms respectively (Ali, 2014).

1.1 Other species of *Cassia fistula*

Cassia grandis

Cassia grandis is a semi-deciduous tree which is medium in size having a rounded or spreading crown. Generally grows up to 20 metres tall with exceptional specimens to 30 metres. The bole is straight and cylindrical and can be up to 1 metre in diameter (Standley & Steyermark, 1946). The plant is collected from the wild, mainly used locally as a medicine. When the flower blooms, this seems to be one of the prettiest trees of Central America, especially along the pacific lowlands, similar as one of apple trees. People cultivate this tree as an ornamental and it is valued especially for its floral display. *Cassia grandis* can be found in open, brushy or forested hillsides and on simple forested plains, along roadsides as well. In pastures, it can be found at elevations below 900 metres (Lorenzi, 2002).

Cassia angustifolia

Cassia angustifolia has always been applied as laxative while it is familiar as medicinal plant from 1950. Fruits and the leaves are used for having pharmaceutical implementation to deal with the drastic or persistent constipation. It is equally implemented for epilepsy, haemorrhoids, derma abnormalities, tracheal complications, dermal infections, hemicrania and cardiac ailment. The observed chemical composition is- senna which carries anthraquinonoid compounds (sennoside A, B, C and D), saccaharide, flavonoid, naphthalene derivatives, phytosterols, waxes, tannins, essential oils, mineral salts, resins, and mucilage (Săvulescu, Georgescu, Popa, & Luchian, 2018). Side effects like serious rashes can be caused by this in infants if they mistakenly consume it. Senna glycosides are the active ingredients having the ability to deal with immune cells situated in the colon. It has fungicide property as well. The plant can be 60-80 cm high, the surface is glabrous to subglabrous and it is perennial. The leaves are alternate, even-pinnate, having length of 6-10.5 cm, with lanceolate leaflets having 5-9 pairs, having the entire edge, sharp end, length up to 4.5 cm and width around 3.5-10 mm. It has hairy surface on one side as well as the other, having a color of light green (Spiller et al., 2003).

Cassia javanica

Cassia javanica is tree which is grown up to 25-40 m tall, which makes it a small to medium sized tree. This tree is deciduous or semi-deciduous and the stock of tender trees seems to be polished or oriented with lots of divisions. Flower arrangement (inflorescence) is either unbranched or loosely branched with clusters of flowers. The shoot is oriented with leafy stems having short side branches which occur to be up to 16cm long. The flower arrangement consists of heavily flowering branches and they contain sepals up to 10 mm long which are green to dark red in color. The petals are almost 15-35 mm long, coloring whitish to red,

stamens are 10 in number, 2 of them are prolonged ones and have filaments 2 cm extended, 4 are smaller ones with filaments 1 cm extended. It has fruits which are pendent, cylindrical in shape, length of 20-60 cm and 2-2.5 cm in diameter. The fruits are indehiscent. Seeds are numerous in number, inserted in a horizontal saucer. *Cassia javanica* follows Troll's architectural model and the top meristem is terminated leaving the other meristems continuous. All the axes are plagiotropic. This tree shows polymorphism and several subspecies are identified by this. *Cassia javanica* flowers are found in October-December also the fruits grow in the parched time of the year. Flowering and fruiting can be noticed in a mast fruiting year in Malaysia (Orwa, Kindt, Jamnadass, & Simons, 2009).

1.2 *Cassia fistula* plant description

Cassia fistula is habitualy known as ‘Sonalu’ or ‘Bandarlati’ and different conventional system of medicines use this plant for various maladies since prehistoric times. Also, *Cassia fistula* is called the golden shower tree. *Cassia fistula* is a deciduous tree which is medium in size to be specific almost 10 m tall, has yellow flowers. The leaves are alternate, pinnate, 30-40 cm long with 4-8 pairs of leaflets which are ovate, having length of 7.5-15 cm and width of 2-5 cm. Fruits are saggy, cylinder like shaped, has brown color, septate, 25-50 cm in length, 1.5-3 cm in diameter, while have around 100 seeds. Seeds are lenticular, pale brownish in color and radiant (Ali, 2014).

Figure 1: Cassia fistula

Taxonomic Classification:

Kingdom - Plantae

Subkingdom – Tracheobinota

Super Division - Spermatophyta

Division - Mangoliophyta

Class – Magnoliopsida

Sub Class - Rosidae

Order - Fabales

Family - Fabacae

Genus - Cassia

Species - fistula

(Pawar, Patil, & Killedar, 2017).

Vernacular Names:

Bengali - Bundarlati, Sonalu, Soondali, Sondal

English - Golden Shower

Guajarati - Garmala

Hindi - Sonhali, Amultus

(Pawar et al., 2017).

1.3 Different parts of *Cassia fistula*

Leaves: The leaves of *Cassia fistula* are pinnate. They have smooth surface, they are 30 to 40 cm in length. Leaflets have ovate shape, 8 to 16 in number, having length of about 10 cm.

Flowers: Flowers contain nice fragrance and are dazzling yellow in color. Extended, lax racemes bear the flowers which are upto 50 centimeters long and on stalks 3 to 5 cm long. Smooth and deciduous calyx has a length of 6 to 8 milimeters. Petals have veins, are obovate in shape, 18 to 25 mm long also short-clawed at the center. The stamens are all presented with reproductive structures.

Fruits: Fruit is observed as an indehiscent pod, cylindrical in shape, up to 60 cm long and have thickness about 2.5 cm. They are dusky brown in color, saggy, has smooth surface and lustrous appearance.

Seeds: Seeds are countless in number which can vary from 20 to 25, inserted in black, sweet slush which is thoroughly partitioned by fine, transverse partitions. They are compact, elliptic, slightly pressed at the same time have a smooth, shiny appearance with yellowish brown color (Ali, 2014).

Cassia fistula flowers

Cassia fistula leaves

Cassia fistula fruits

Cassia fistula seeds

Figure 2: Different parts of Cassia fistula.

1.4 Chemical constituents of *Cassia fistula*

The plant *Cassia fistula* contains a good amount of phenolic antioxidants like flavonoids, anthraquinones, and flavan- 3-ol derivatives. This plant shows positive results for alkaloids, terpenoids, steroids, saponins, tannins, reducing sugars, carbonyl and phlobatanin. As this plant contains anthraquinone it has notable laxative actions. The seeds of this plant hold approximately 2% anthraquinones along with 50% carbohydrates, 24% crude protein, 6.5% crude fiber and 4.5% crude fat. The leaves have been recorded with 39.86% carbohydrates, 20% crude fiber, 15.9% crude protein and 6.65% crude fat. The stem bark is documented to

carry two flanol glycosides and one xanthone glycosides. Additionally, *Cassia fistula* also contains fistulic acid, fistuacacidin, chrysophanic acid, rhein, rheinglucoside, galactomannan, sennosides A and B, tannin, emodin, barbaloin, oxyanthraquinone compounds, phlobaphenes, lupeol, beta-sitosterol, and hexacosanol (Lee, Lee, & Kuo, 2001).

1.5 Background study

Anti-inflammatory effect of *Cassia fistula*

Fresh, fully fledged leaves of *Cassia fistula* are accumulated, identified, shed-dried and then pulverized with the help of a mechanical grinder. The grinded materials are then stored in a closed vessel until utilized. With the help of soxhlet extraction apparatus, the powdered leaf tissue was extracted with petroleum ether and then with benzene, chloroform and methanol. The terminal product was stored in acacia solution. As test animals, male albino rats were used and they were administered with different concentrations of plant extracts. Phenylbutazone was used as reference drug and normal saline for control groups. Different groups were treated intraperitoneally. Inhibition percentage of edema was then calculated from different groups of rats (Mandal et al., 2006).

Eventually, the methanolic extract of *Cassia fistula* seemed to exhibit remarkable anti-inflammatory property at different time interruptions against various moderators of inflammation (Mandal et al., 2006).

Antidiabetic effect of *Cassia fistula*

Fruits were assembled and the fruit pulp was unleashed manually. Fruit pulp was dried under shade and extracted with petroleum ether following simple maceration technique and the amount taken was 1kg. It was stored in a bottle made of amber glass with irregular shaking for 5 days straight. After 5 days, extract was run under rotary evaporator. Rats were used as test animals and a single intraperitonial injection of freshly prepared streptozotocin in specific concentration with normal saline was used to induce diabetes to them. In total 24 animals were used in the experiment in which 18 were diabetic surviving rats and the other 6 were normal rats. Total 30 animals were induced in five groups. The in vivo antidiabetic activity of the plant extracts was assessed using streptozotocin (which was used as a standard drug) model induced with diabetes. Various variables such as body weight, SGOT, SGPT, blood glucose levels, triglycerides and cholesterol were observed. Obtained data were evaluated in a statistical manner and the significance level was assessed in the respective charts. The potentiality of extracts in decreasing the glucose level was understood from observing the numerical values (Akhila & Na, 2015).

Hepato-protective activity of *Cassia fistula*

Cassia fistula leaves were assembled, dehydrated under shade, grinded and skimmed through a sieve of 40-mesh and then extracted with methanol. The extract was transit across an alumina column and removed with n-heptane and finally a semisolid mass was used for the experiment. Neutrosec was used in a specific dose as reference drug which is an accepted liver tonic. The semi-solid essence was eliminated in normal saline and given through oral route of administration to specific groups of animals. 40 male and female albino rats of were separated into four sections having 10 animals in every group and experimented. The yellow complexion indicated the existence of steroids, triterpenoids and anthraquinones. The

findings established the hepatotoxicity of paracetamol and the free-radical mechanism recommended for the poisonous consequences of this substance (Bhakta et al., 2001).

1.6 Aim and objectives of the study

Aim of the study

The aim of the antiemetic study was to estimate the percentage of inhibition by comparing the frequency of retching between control and test group of animals after oral administration.

The aim of the anthelmintic study was to observe and estimate the rate of time for initiating paralyzing and death of the earthworms.

Objectives of the study

The objectives of the study are to-

- Estimate the paralyzing time and then death time of earthworms over time period.
- Determine the retching time due to effect of copper sulphate.

Chapter 2

Methodology

2.1 Collection and Identification of plant material

Plant specimen of *Cassia fistula* has been accumulated from local area of Lalmatia, Dhaka, in January 2019. The plant was identified by the specialist of Bangladesh National Herbarium, Mirpur, Dhaka, Bangladesh and the identification number is- 3124.

2.2 Preparation of the plant material

The fresh leaves were segregated after collection from the unwanted parts such as stalks. Throughout the whole process, any kind of contamination was strictly avoided. The leaves were then washed with clean water again and again to make it free of dirt. They were then air dried and then shade dried for 7 days in an open space avoiding direct sunlight making them worthy for grinding. After grinding bristly textured powder was obtained and stored in an air-tight container in a dry place away from light and temperature until it was taken for the analysis (Bulbul et al., 2013).

2.3 Extraction process of the plant material

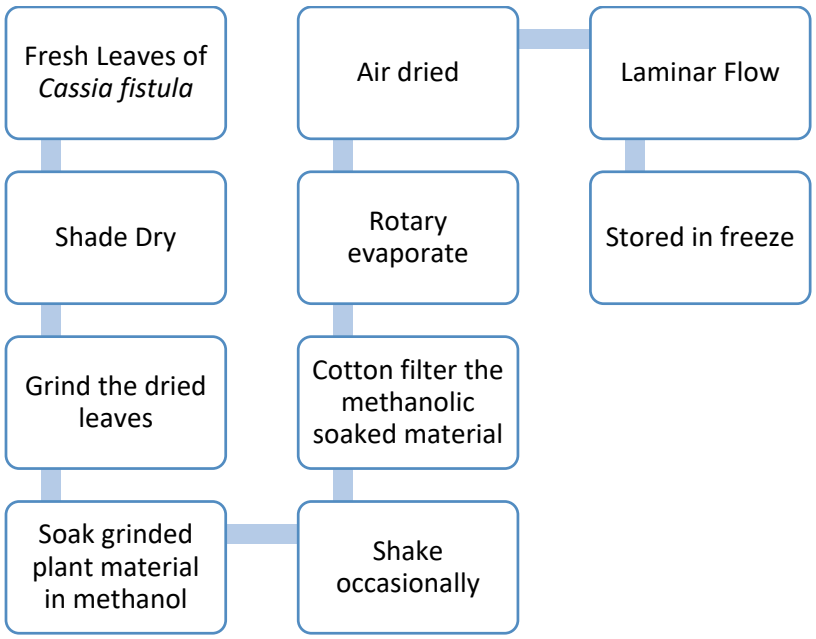


Figure 3: Extraction process of Cassia fistula

Figure 4: Cassia fistula leaves material in methanol

Methanolic extract of *Cassia fistula*

Leaves extract under laminar flow

Figure 5: Methanolic extracts of Cassia fistula leaves

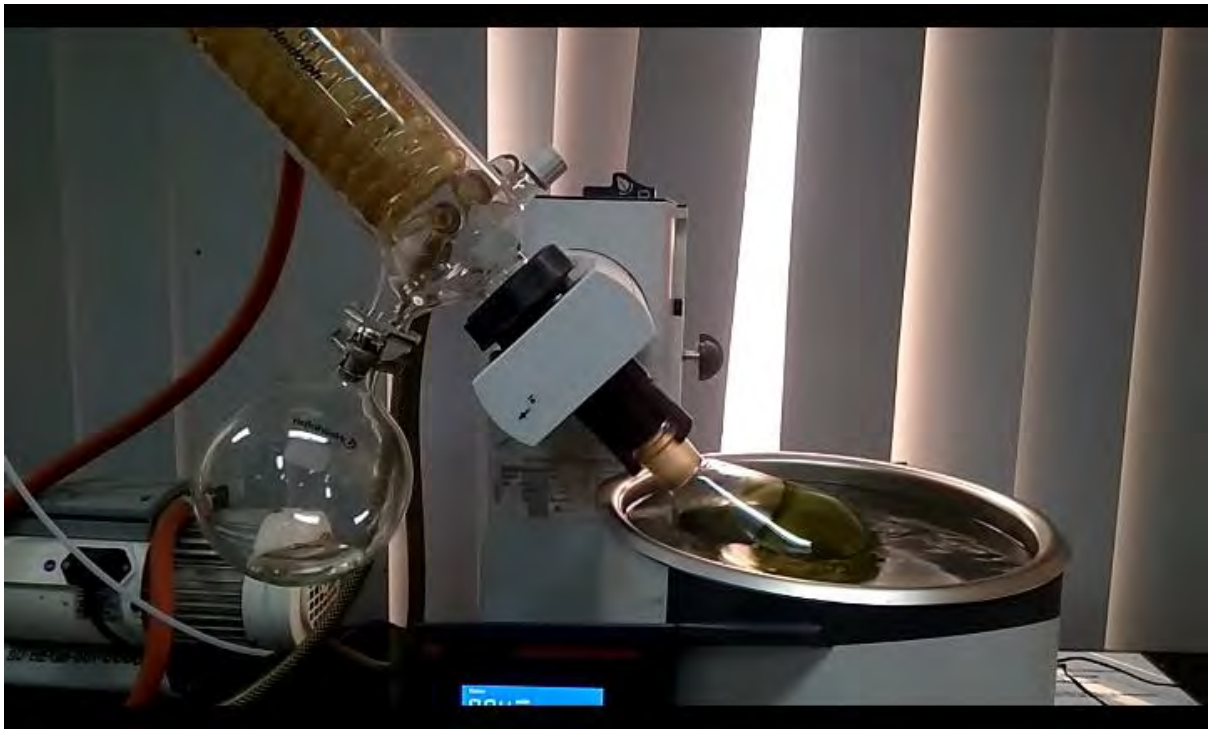


Figure 6: Extraction process of Cassia fistula by rotary evaporation

The dried and bristly powdered plant materials of *C. fistula* were then soaked in methanol and for soaking 200gm of powdered leaves, almost 1.3 liter of methanol was used. The methanol soaked plant materials were kept in air tight container made of amber glass. For straight 7 days they were preserved under cool, dark and dry conditions with occasional but vigorous shaking. The extract was then filtered through cotton filter and the filtrate achieved was then dried using rotary evaporator. After processing through rotary evaporator, a gel-like greenish mass was obtained which was kept for air drying first. Then it was kept in laminar flow which resulted the final product to be a heavily sticky tar textured dark greenish mass (Bulbul et al., 2013).

2.4 Worm collection and authentication

The collection and identification of earthworms named *Pheretima posthuma* (phylum: Annelida) was done from moist soil at University of Dhaka and they were first cleaned using

saline water so that soil and any dirt gets eliminated from them. The earthworms had an average length of 4-6 cm and width of about 0.2-0.3 cm as they were used under the protocol of the experiment.

2.5 Animals used in the experiment

Young chicks having an age of 2-4 days and weighing about 30-40 gm were collected from the own farm of Bangladesh Livestock Research Institute. Selected 20 chicks were isolated and specially nurtured from the day they were born for the experiment. The chicks were kept in a fasting condition for 24 hrs maintaining laboratory conditions which include ambient temperature including 12 hrs day-night cycles. All the experiments using the animals were accomplished according to the permission of Administrative Committee of Bangladesh Livestock Research Institute, Savar, Dhaka.

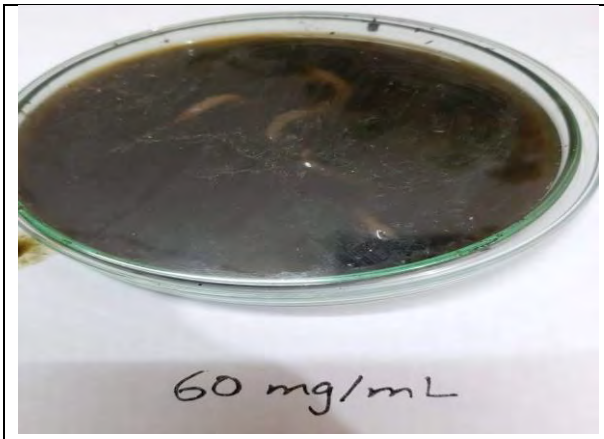
2.6 Chemicals

Two different types of solvents were used in the formulation. Pharmaceutical grade methanol was purchased from Merck, Germany and it was used as an organic solvent. Distilled water was prepared in the phytochemistry lab, Brac University and it was used as an aqueous solvent. Phosphate buffer solution, copper sulfate, dimethyl sulfoxide (DMSO) and polyoxyethylene sorbitan monooleate (Tween 80) was purchased from Merck, Germany and provided by technology lab and phytochemistry lab of Brac University. Albendazole was purchased from Eskayef Pharmaceuticals LTD. which was used as a reference drug for the anthelmintic test. Then again, chlorpromazine was purchased from Sanofi Bangladesh LTD. and was used as a reference drug for the antiemetic test.

2.7 *In-vitro* anthelmintic evaluation

In the experiment of anthelmintic assay, *Pheretima posthuma* were used as these earthworms have anatomical and physiological resemblance with human roundworm parasites of intestine

and fall under the same group which is Annelida. Before the experiment was ready to be done, the test solutions and solutions of standard drugs were freshly prepared which were then used in the experiment. Albendazole in a concentration of 20 mg/mL was used in the experiment as reference drug at the same time distilled water was used as control. All 20 earthworms were split into five groups having 4 members in each group. Each petri dish had 60 mL formulations accommodating a control group (distilled water), three different concentrations of *Cassia fistula* (60, 80 and 100 mg/mL in distilled water) and standard drug formulation (albendazole-20 mg/mL) and they were prepared freshly maintaining proper rules for the experiment. The paralysis time of each group of earthworms was recorded when no movement of any type was observed besides such conditions like vigorous shaking or contact with high temperature. The death times were noted at the same time when the worms completely stopped moving even when they were shaken vigorously or submerged in warm water having a temperature of approximately 50°C (Bulbul et al., 2013).



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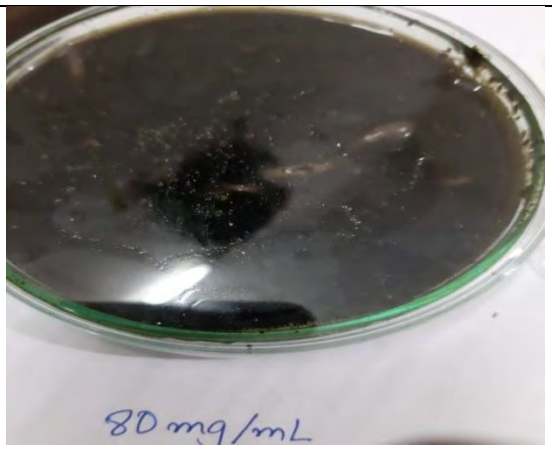

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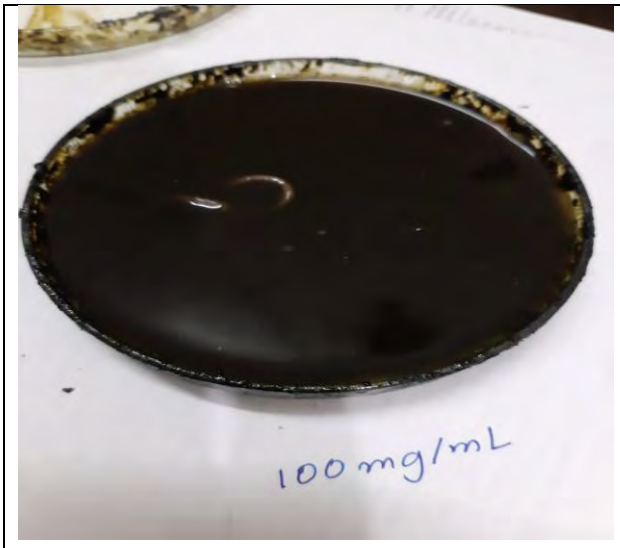

C

D

E

Figure 7: Pheretima posthuma treated with distilled water, albendazole and different concentrations of Cassia fistula leaves extracts (A=Earthworms in 60 mg/mL Cassia fistula leaves extract; B=Earthworms in 80 mg/mL Cassia fistula leaves extract; C=100 mg/mL Cassia fistula leaves extract; D=Earthworms in albendazole; E=Earthworms in distilled water

2.8 *In-vivo* antiemetic evaluation

Evaluation of the antiemetic activity was done by the calculation of the average decrease of retching number of the chicks. The selected 20 chicks were age of 1 week and they were split into 5 batches having 4 members of chicks in every batch and they were kept aside maintaining room temperature which is 25°C. The *Cassia fistula* leaf extracts were dissolved into a solution of 0.9% saline solution having 5% DMSO and 1% Tween 80 in it and it was provided in three different concentrations which were 50 mg/kg, 100 mg/kg and 150 mg/kg and they were given the dose orally. Then, the control group of chicks was provided with

only the 0.9% saline solution containing 5% DMSO and 1% Tween 80. Chlorpromazine was used as a standard drug and was provided to the reference group of chicks in a concentration of 150 mg/kg. All of the groups were then administered with 50 mg/kg concentrated copper sulphate and it was given orally as well. The initiation of antiemetic effect was recorded by observing the minimization in the retching count in the treated group comparing to the control one. The rule for calculating the inhibition percentage:

If, A = Frequency of retching in control groups

B = Frequency of retching in test groups

Then, Inhibition (%) = (A-B/A) x 100

(Ahmed & Onocha, n.d.).

Figure 8: Chick models treating with saline solution, chlorpromazine and different concentrations of Cassia fistula leaves extract

Chapter 3

Results

3.1 Inhibition in retching count from antiemetic study

Table 1 expresses the antiemetic activity of different concentrations of *Cassia fistula* leaves extract on chick models. The perceived retches count was control (82), chlorpromazine used as standard (91.5), 150 mg/kg concentrated *Cassia fistula* leaves extract (31.5), 100 mg/kg concentrated *Cassia fistula* leaves extract (9.5) and 50 mg/kg concentrated *Cassia fistula* leaves extract (56.75) (Figure 6). The 150 mg/kg, 100 mg/kg and 50 mg/kg concentrated methanolic extract of Cassia fistula showed 61.58%, 88.41% and 30.79% inhibition of retches respectively which is shown in (Table 1).

Table 1: The antiemetic activity of Cassia fistula leaves extract

Treatments (mg/kg)	**Number of retches (Mean±SEM)**	**% Inhibition of retches**
Standard (chlorpromazine) (150mg/kg)	57.5 ± 4.03	29.88%
***Cassia fistula* extract (50mg/kg)**	56.75 ±3.17	30.79%
***Cassia fistula* extract (100mg/kg)**	9.5 ±5.72	88.41%
***Cassia fistula* extract (150mg/kg)**	31.5 ±6.38	61.58%
Control (normal saline solution)	82 ±7.67	-

*SEM= Standard error of mean

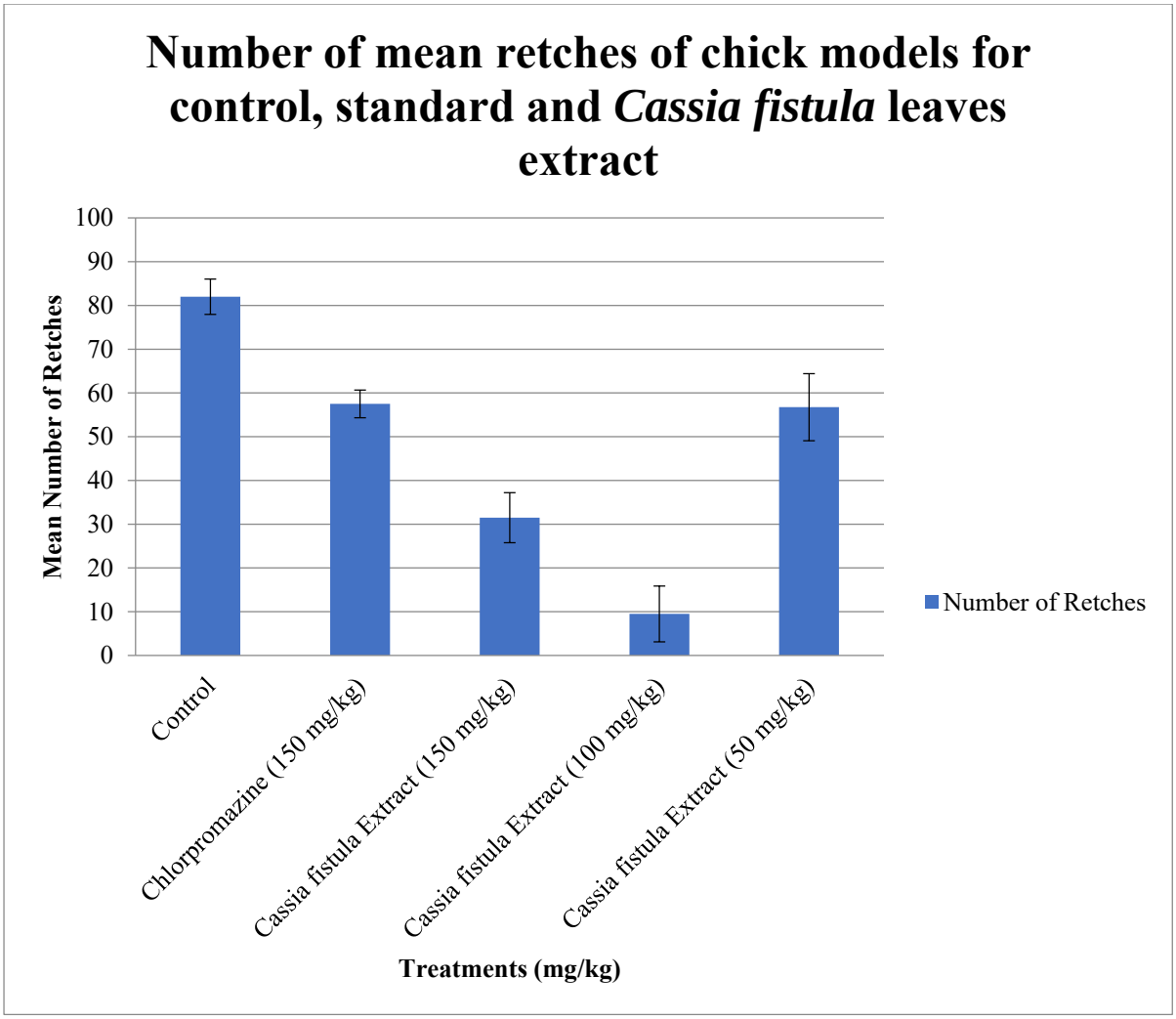

Figure 9: Number of mean retches of chick models for control, standard and Cassia fistula leaves extract.

3.2 Death of earthworms due to anthelmintic activity

Table 2 demonstrates the anthelmintic activity found in different concentrations of *Cassia fistula* leaves extract when applied on earthworms (*Pheretima posthuma*). The identified paralysis time for different concentration such as 60 mg/mL, 80 mg/mL and 100 mg/mL were 17.5, 14.5 and 9.75 minutes respectively. Also the observed death time for these was 20.25, 40.25 and 64.25 minutes respectively. The standard group which was treated with albendazole (20 mg/mL), exhibited paralysis time and death time at 23 and 63.25 minutes respectively (Figure 7).

Table 2: The Anthelmintic activity of Cassia fistula leaves extract

Treatments (mg/mL)	Time taken for paralysis in min. (Mean±SEM)	Time taken for death in min. (Mean±SEM)
Standard (albendazole) (20 mg/mL)	23 ± 1	63.25 ± 1.97
***Cassia fistula* extract (60 mg/mL)**	17.5 ±0.5	64.25 ± 5.10
***Cassia fistula* extract (80 mg/mL)**	14.5 ± 0.29	40.25 ± 5.10
***Cassia fistula* extract (100 mg/mL)**	9.75 ± 1.03	20.25 ± 3.09
Control (distilled water)	-	-

*SEM = Standard error of mean

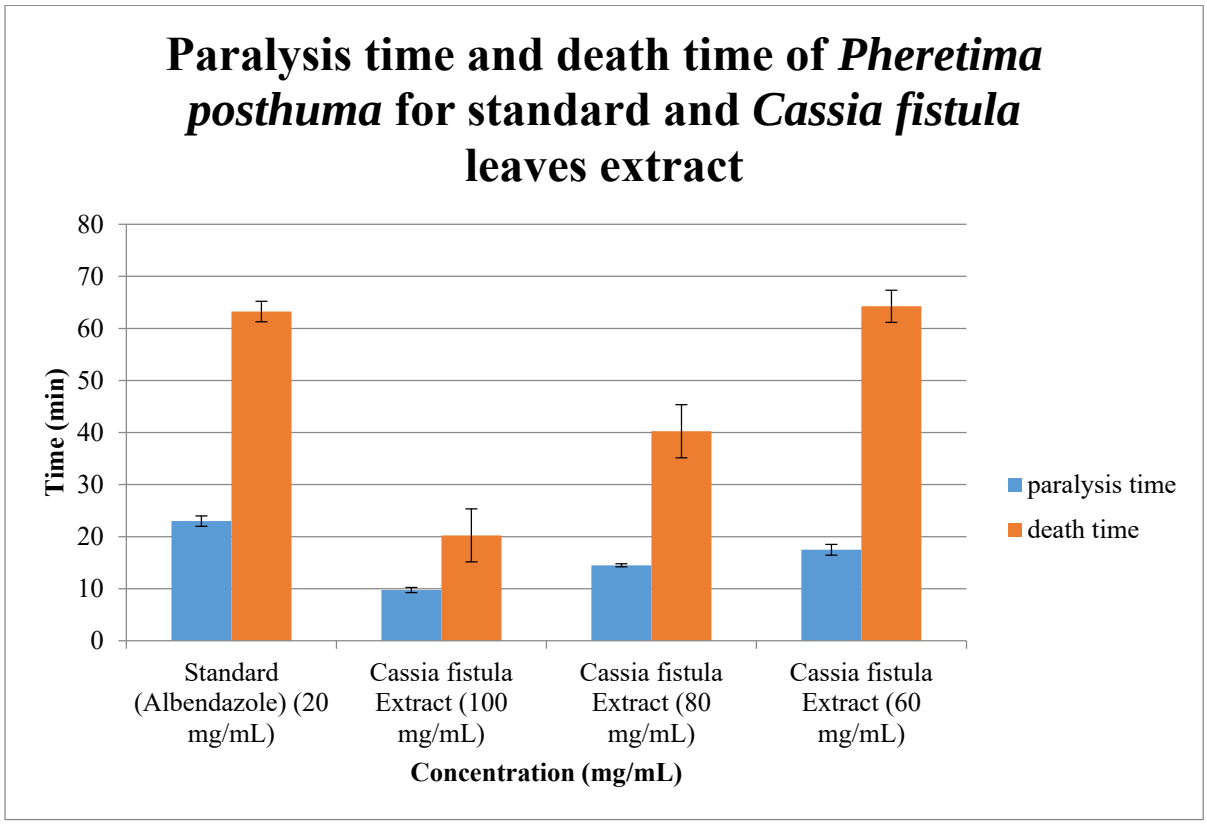

Figure 10: Paralysis time and death time of Pheretima posthuma for standard and Cassia fistula leaves extract.

Chapter 4

Discussion

4.1 Comparative discussion on the antiemetic study

Based on the results we observed and comparing the results of *Cassia fistula* leaves extracts with the standard which was chlormromazine, we can concur to the decision that *Cassia fistula* leaves extracts have shown remarkable antiemetic property (61.58%, 88.41% and 30.79%) compared to chlorpromazine (29.88%).

Copper sulphate played the significant role in inducing emesis among the chick models while the attained antiemetic property from *Cassia fistula* leaves extract is still in a vague state. Even so, the chick models were administered with copper sulphate orally and the *Cassia fistula* leaves extract could actually defend the effect of emesis in them by causing inhibition in retching count (Bulbul et al., 2013).

A lot of factors can actually be considered for inducing retches such as the aftereffects of the cancer treatments which may include Chemotherapy. Vomiting and nausea can be prompted due to CNIV (Chemotherapy induced Nausea and Vomiting). One confirmed fact states 5-HT4 receptors located on the periphery helps to initiate the emesis by copper sulphate. Also the drugs used in cancer treatments cooperates the vomiting zone by triggering them which undergoes further complex situations and ultimately causes retching (Bulbul et al., 2013).

However, *Cassia fistula* leaves extracts seemed to give better antiemetic results compared to chlorpromazine, which is already known for its remarkable effects as antiemetic agent. Most importantly, no established researches have been done with the antiemetic activity of *Cassia fistula* leaves extracts. As the results stated here the significant possibility of *Cassia fistula* leaves extract as an antiemetic that too better than the reference drug, further studies and

researches need to be conducted to understand the accurate mode of action working behind this.

4.2 Comparative discussion on the anthelmintic study

If we observe the results achieved from the anthelmintic study of *Cassia fistula* leaves extract and compare it with the reference drug (albendazole) we can come to the conclusion that *Cassia fistula* leaves extract has exhibited incredible anthelmintic results compared to albendazole.

The human race and animals have been severely damaged by parasitic helminthes and they have to undergo several complications related to growth, sometimes serious infections and other forms of maladies due to these helminthes. There are lots of different kinds of helminthes throughout the world which also have been carried by human and animals themselves by traveling and other processes. Bunch of researches and studied have been done to limit the aftereffects of the infections caused by these helminthes but there is hardly any effective medicine that will completely terminate the problem. The drawbacks are mainly either the established drugs are responsible of serious aftereffects or they convert the infections to their worst possible form which affects the body in a horrible way. These obstacles lead humans into thinking about finding an alternative way of remedy than the conventional ones which finally ended to herbal medicines. The activity analysis of the medicinally active plants paved the way to limitless possibilities of having a solution about getting an effective anthelmintic (Partap, Kumar, Kumar, Sharma, & Jha, 2012).

The present study of *Cassia fistula* leaves extract on earthworms exhibits an encouraging result compared to the reference drug (albendazole) that this can be a very prominent solution in the future in case of nematode control.

Chapter 5

Conclusion

The results of this study did specify that *Cassia fistula* leaves extract has the therapeutic potency to show against copper induced emesis on chicks and also as an anthelmintic against *Pheretima posthuma* (earthworms). Among the different concentrations, 150 mg/kg concentration of *Cassia fistula* leaves extract exhibited significant antiemetic effects on copper induced emesis on chick models compared to chlorpromazine. The lower concentrated extracts did not show significant result. *Cassia fistula* leaves extracts revealed outstanding results as an anthelmintic against earthworms compared to albendazole. Surprisingly, in this test, all three concentrations (60 mg/mL, 80 mg/mL and 100 mg/mL) showed significant results and specially the higher two concentrated extracts which are 80 mg/mL and 100 mg/mL exhibited very good results. Further in vivo studies and additional researches on this purpose are required to demonstrate *Cassia fistula* as a worthwhile and pharmacologically potent drug.

Chapter 6

Future Work

- The specific mode of action which is followed by the *Cassia fistula* leaves extracts to exhibit the antiemetic and anthelmintic property are still not very clear. For that, further studies are needed on this particular aspect so that the progress of these studies can be ensured for the development of the leaves as a drug. Furthermore, phytochemical screening needed to be done under strict supervision and examination so that the exact chemical constituent responsible for the anthelmintic and antiemetic properties can be evaluated and determined.
- Chemotherapeutic agents have severe side effects like nausea and vomiting which increases the sufferings of the cancer treatments which is already unbearable. Hence, further studies can be done to develop this *Cassia fistula* leaves extract as a drug so that an easily available herbal drug can be there to reduce the side effects of the antineoplastic agents.
- *Cassia fistula* pod extracts show a very minimum toxicity levels which is LD_{50} of 6600mg/kg (Akanmu, Iwalewa, Elujoba, & Adelusola, 2010). From this, future studies can be done on the test of toxicity levels of *Cassia fistula* leaves extracts.
- More *In-vivo* studies should be conducted on anthelmintic and other properties which have not been proved yet.

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