Cinnamon Zeylanicum Extracts: A Study of Phytochemicals, Physicochemical Properties, and Antibacterial Effects

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ABSTRACT

Cinnamon (*Cinnamomum zeylanicum*), the evergreen tree of tropical area, a member of family Lauraceae, has been used in day to day routine as a spice and condiment in India. Cinnamon is widely used as a spice due to its distinct odour of essential oils. Literature review on cinnamon revealed that it chiefly contains essential oils and all other categories like cinnamic acid, cinnamaldehyde and cinnamate. It has good anti-inflammatory, anti-oxidant, anti-ulcer, anti-microbial, hypoglycemic and hypolipidemic potential. Cinnamon is found very safe and is being used as spice for ages. In the present study the bark of Cinnamon were analysed for the phytochemical, pharmacognostical evaluation and antibacterial activity using standard methods. Phytochemical screening included qualitative chemical examinations and tests for determination of primary and secondary metabolites. Pharmacognostic evaluation included examinations of morphological characters, determination of leaf constant, ash value, powder analysis, and extractive values were carried out. The plant has effective pharmacological action. The medicinal properties of this plant were attributed to its variety of active phytochemical constituents. Although this plant had received interest for the phytochemical investigations since many years, more work has to be done on its isolation and characterization for exploring the immense medicinal potential of this plant.

KEYWORDS: Cinnamon, phytochemical, pharmacognostical, antibacterial.

I. INTRODUCTION

Indian flora serves as a land of spices and spices are being used in food preparations since ancient times. It has been well established that spices not only impart good flavor and pungent stimuli but also contribute to medicinal, antimicrobial and antioxidant properties (Agarwal *et al*., 2014). Due to these multiple potential properties, spices serve as an ideal food preservative and will aid in prolonging the shelf life of foods by preventing rancidity through their antioxidant activity (Patel *et al.,* 2011). Another interesting fact is that the natural potential of spices contributes to a safe intake for food products as well as improves the shelf life of food products (Sharma *et al*., 2012). Cinnamon bark is one among the commonly used Indian spices in food preparations. It has been reported that when Cinnamon bark is used as natural preservative, it prevents decomposition of the products.

Cinnamomum zeylanicum Blume, a member of the family Lauraceae, is a tropical evergreen tree, native to Sri Lanka and the Malabar Coast of India, called differently in different languages such as dalchini in Hindi, cannelle in French, kaneel in German, canela in Spanish, yook gway in Chinese and kurunda in Sinhalese. The botanical name Cinnamomum is derived from the Hebraic and Arabic term amomon, meaning fragrant spice plant. In India, Southeast Asia, United States and in the European countries, cinnamon is used for flavouring foods, beverages, boiled beef, pickles, chutneys and ketchup. Medicinally, cinnamon is used in the treatment of diarrhoea, flatulent dyspesia, poor appetite, low vitality, kidney weakness and rheumatism, influenza, cough, bronchitis, fever, arthritic angina, palpitations, hypertension and nervous disorders, stimulating the circulatory system and capillary circulation, spasms, vomiting and controlling infections, reducing blood sugar levels in diabetics and as a skin antiseptic. It is sometimes used alternatively with damiana (*Turnera diffusa*) to promote conception. It is proven to be particularly effective against some species of toxicogenic fungi as well as respiratory tract pathogens, including species belonging to the genera *Aspergillus*, *Candida*, *Cryptococcus* and *Histoplasma* (Jakhetia *et al*., 2010).

Research investigation by Vangalapati *et al*., 2012 reported that Cinnamon bark is rich in cinnamaldehyde which has been proven to be active against many pathogenic gram positive and gram negative bacteria. Jakhetia *et al*., 2010 suggested that spices can serve as the excellent alternatives to chemical food additives due to their antimicrobial properties. The antioxidant and antimicrobial properties are very important to preserve the quality of food material which provides safety to the consumers (Sangal, 2011; Jain, 1987).

Fig 1: Various useful parts of Cinnamomum zeyanicum

Since this spice naturally occurs in varying habitats, it is naïve to expect a great magnitude of variation in the concentration and composition of chemical ingredients in different parts of the bark. However, the extent to which the chemical composition varies in populations adapted to varying habitats is not known. Thus, detailed studies are required to examine this aspect.

In view of its multiple uses, this spice needs to be widely cultivated in most of the areas where climatic conditions favour its optimum growth. In this way, a maximum yield of its different usable parts could be achieved to derive the maximal amount of commodities of a multifarious nature for the welfare of mankind (Leela *et al*., 2008; Senanayake *et al*., 2003). Hence looking all these facts, it is important to determine its therapeutic, pharmacological and various other properties which are useful to mankind. The aim of the current investigation was to determine various Phytochemical constituents, Physiochemical and Antibacterial activities on different extracts of *Cinnamon zeylanicum* (Dalchini)

Objectives of Research:
The Objectives of the study were as follows:

1. Procurement of plant material and preparation of various extracts.
2. Phytochemical Profiling of various plant extracts
3. Physiochemical Analysis
4. Determination of Antibacterial Activity of various extracts against pathological strains.

II. MATERIALS AND METHODS

1. Chemicals

All chemicals and reagents were of analytical grade and used without further purification.

2. Procurement and authentication of plant material

Cinnamomum zeylanicum bark were collected from local market and authenticated by Botanist of Life Science Department.

3. Preparation of bark extracts by successive extraction method

Bark were air dried at room temperature for 3 weeks to get consistent weight. The dried barks were later ground to crude powder. Fifty grams of crude powder of bark was taken in soxhlet apparatus. Successive extraction with different solvents (Chloroform, Butanol, Methanol, Ethanol, and Aqueous) was carried out. Extracts were filtered using funnel and Whatman No. 1 filter paper. Each filtrate was concentrated to dryness under reduced pressure at 40°C through evaporator and stored at 4°C for further studies.

4. Qualitative estimation

For the quantitative estimation 100 g of powdered barks was successively extracted in a soxhlet apparatus with various solvents like (Chloroform, Butanol, Methanol, Ethanol and Water). The extracts were dried on water bath, weighed and colour of the extracts was also observed. The different extracts were subjected to qualitative estimation for the presence of various phytoconstituents (Sharma and Pracheta., 2013).

5.Phytochemical Profiling

Preliminary screening for the presence of phytoconstituents (Primary and Secondary metabolites) of all the extracts was carried out using standard conventional procedures (Pande and Pathak *et al*., 2010).

Pharmacognostical and fluorescence analysis

Pharmacognostical parameters such as foreign organic matter, loss on drying, total ash, acid insoluble ash, water soluble ash, moisture content and crude fibres contents were performed as per Indian Pharmacopoeia (Yogesh *et al*., 2010). Fluorescence analysis of the bark powder (Usman *et al*., 2010) was carried out with different chemical reagents in day (254 nm) and UV light (365 nm).

6. **Screening of antibacterial activity**:

Microbial strains-

The bacterial species used for the test were

- *Escherichia coli*
- *Klebsiella*

Culture media and inoculums preparation

Nutrient agar\ broth (Microbiology Laboratory) were used as the media for bacterial culturing. To 1 ml of mother culture of respective bacterial strains were inoculated in nutrient broth in aseptic condition and then incubated at 37˚C for 24 h. Mac Concy Agar was further used for preparation of plates.

Test Sample preparation

The concentration (100 mg\ml) of all five extracts (Chloroform, Butanol, Methanol, Ethanol, and Aqueous)of bark were prepared. The concentration was prepared in their respective solvents. The **antibacterial** activity of *Cinnamomum zeylanicum* was screened by disc diffusion method (Jakhetia *et al*., 2010).

Disc diffusion method

1. Firstly prepared the extract disc by using WhatmanNo-1 filter paper with the help of punching machine and then autoclaved.
2. Soaked the discs in already prepared different concentrations of extracts and left overnight.
3. Prepared the petriplates with suitable agar media (already prepared and autoclaved).
4. Spreaded the bacterial strains on their respective agar media.
5. Test extract loaded disc were placed on respective bacterial and fungal lawn and then incubated at suitable temperature i.e. 37˚C for bacteria
6. After incubation period, the zone of inhibition was measured and recorded.
7. Similarly standard antibiotic disc of Amoxycillin was used instead of test extract for comparative study of test extracts.

Statistical analysis
The inhibition zones were calculated as mean±SD (n＝3).

III.RESULTS

1. Yield of extracts from successive extraction of *Cinnamomum zeylanicum* Bark

The yield of successive extracts (g) is shown in Table 3.1. The amount of the chloroform extract obtained from the extraction was 21 % w/w yield, butanol extract was 12.8 % w/w yield, Hydromethanol extract 15 % w/w yield, Hydroethanol extract 20 % w/w yield and aqueous extract was 25 % w/w yield. Extracts are shown in Fig 3.1

Table 3.1: Phytoprofile and Yield of Extracts from various solvent extraction of Cinnamomum zeylanicum Bark (100g)

S. No	Name of Extract	Polarity Index	% Yield (w/w)	Consistency	Nature
1	Chloroform	4.1	21%	Oily	Semi Solid
2	Butanol	4.0	12.8%	Dry	Semi Solid
3	Hydromethanol	5.1	15%	Sticky	Crisp solid
4	Hydroethanol	5.2	20%	Sticky	Crisp solid
5	Aqueous	9.0	25%	Dry	Solid

Fig 3.1: Whole bark, Powdered Bark and various extracts of Cinnamomum zeylanicum

2. Phytochemical screening of successive extracts of *Cinnamomum zeylanicum* bark

The presence or absence of different phytoconstituents viz. carbohydrate, glycoside, protein, tannins, saponins, flavonoids and terpenoids were detected by the phytochemical screening methods with different chemical reagents. Phytochemical components are responsible for both pharmacological and toxic activities in plants. These metabolites are said to be useful to a plant itself but can be toxic to animals, including man (Paliwal *et al*., 2011; Sharma *et al*., 2011). The presence of these chemical constituents in this plant is an indication that the plant, if properly screened, could yield drugs of pharmaceutical significance. This is better supported by the fact that members of the family of this plant have been known to be involved in ethnomedicine in the management of various ailments (Sharma and Paliwal, 2013a, b).

The result showed the presence of alkaloids, phenolics, flavonoids, saponin, tannin, lignin, protein, carbohydrates, suberin, glucoside, flavin, and traces amount of oil & sugars in the successive extract of ***Cinnamomum zeylanicum* bark** and the result of phytochemical test is presented in Table 3.2.

All these phytochemicals possess good antioxidant activities and has been reported to exhibit multiple biological effects including anti-inflammatory and antitumor activities.

Table 3.2: Qualitative Phytochemical screening of successive extracts of Cinnamomum zeylanicum Bark

Plant Constituents	**Tests Performed**	**Name Of Extracts** **Chl Met Et But Aq**
Alkaloids	Mayer's Test	++ ++ ++ ++ ++
Saponins	Frothing Test	+ + + + +
Phytosterols	Libermann-Burchard's Test	+ + + + +
Phenolic Compounds	Ferric Chloride Test	+ + + + +
Tannins	Ferric Chloride Test	++ ++ ++ ++ ++
Flavanoids	Ammonia Test	++ ++ ++ ++ ++
Terpenoids	Salowski Test	+ + + + +
Phlobatannins	Hydrochloride Test	++ ++ ++ ++ ++
Ascorbic Acid		+ + + + +
Steroids	Lieberman test	++ ++ ++ ++ ++
Cardiac Glycoside	Keller Killani Test	+ + + + +
Proteins and Amino Acids	Millon's Test Biuret Test Ninhydrin Test Xanthoproteinic Test	+ + + + + + + + + + + + + + + + + + + +
Carbohydrates	Molish's Test Benedict's Test	+ + + + + + + + + +
Oils and Fats	Stain Test Soap test	+ + + + + + + + + +

3. Organoleptic Characters of *Cinnamomum zeylanicum* bark

The macroscopic study is the morphological description of the plant parts which are seen by naked eye or magnifying lens. Organoleptic evaluation can be done by means of sense organs, which provide the simplest as well as quickest means to establish the identity and purity to ensure quality of a particular drug (Sharma and Pracheta, 2013). Organoleptic characters such as shape, size, colour, odour, taste and fracture of stem bark, are evaluated (Table 3.3).

Table 3.3: Organoleptic Character of Cinnamomum zeylanicum Bark

S. No	**Variable**	**Result**
1	Bark Colour	Dull Yellowish Brown
2	Bark Shape	Wavy Longitudinal
3	Powdered Bark	Dark brown
4	Odour	Pleasant
5	Taste	Sweet

4.Physiochemical Evaluation

Identification and evaluation of plant material using various analysis techniques is one of the simplest and cheapest methods to establish the correct identity of the source materials. The parameters which are studied are moisture content, loss on drying, total ash, acid-insoluble ash, alcohol and water-soluble extractive values etc. (Paliwal, 2015). The proximate analysis revealed that water soluble extractive values of leaves was 29.75, alcohol soluble extractive values of leaves was 35.21, total ash value was 9.75, acid insoluble ash was found to

be 2.50 and sulphated ash was 37.35. Loss on drying was 25.52, foreign organic matter was found to be 7.56 and pH was slight acidic, 5.1 were observed in fresh bark (Table 3.4).

Table 3.4: Physiochemical Analysis of Cinnamomum zeylanicum Bark

S. No.	Parameters	Values obtained w/w on dry weight basis
1	Total Ash Value	9.75
2	Water Soluble Ash	29.75
3	Acid Insoluble Ash	2.50
4	Alcohol Soluble Ash	35.21
5	Sulphated Ash	37.25
6	Loss on Drying	25.52
7	Foreign Organic Matter	7.56
8	pH	5.1

5. Fluorescence Analysis of various extracts

The result of fluorescence studies the extracts is compiled in Table 3.5.

Fluorescence is an important phenomenon exhibited by various chemical constituents present in plant material. Many phytochemicals fluorescence are seen when suitably illuminated. The fluorescence colour is specific for each compound. A nonfluorescent compound may fluorescent if mixed with impurities that are fluorescent. Some constituents show fluorescence in the visible range in day light. The ultra violet light produces fluorescence in many natural products (e.g. alkaloids like berberine), which is not visible in day light. If the substances themselves are not fluorescent, they may often be converted into fluorescent derivatives after reacting with different reagents hence some crude drugs are often assessed qualitatively in this way and it is an important parameter of pharmacognostical evaluation (Chanda, 2014; Tatiya *et al*., 2012).

Table 3.5: Florescence Characteristics of Different extracts of Cinnamomum zeylanicum Bark

S.No	Name of Extract	Under Ordinary Light	Under UV Light(366nm)
1	Chloroform	Light Brown	Brown
2	Butanol	Dark Brown	Brown
3	Methanol	Dark Brown	Yellow
4	Ethanol	Dark Brown	Dark Brown
5	Aqueous	Brown	Black

6.Determination of Antibacterial Activity

The results of the disk diffusion test revealed that the various extracts of cinnamon showed different degrees of growth inhibition, depending upon the bacterial strains (Table 3.6). The chloroform extract of cinnamon showed notable antibacterial activity against Gram–positive bacteria. It is well known that most spices are more active against Gram–positive bacteria than Gram–negative bacteria. This study showed that chloroform and hydoethanol extracts of cinnamon were more effective against Gram–positive bacteria in vitro. The sequence of antibacterial activity against *E. Coli* is as follows with their zone of inhibitions:

Chloroform extract (2.5)>Hydroethanol (1.7)> Butanol (1.5)>Hydromethanol (1.3)> Aqueous (1.2). The sequence of antibacterial activity against *Klebseilla* is as follows with their zone of inhibitions:**Chloroform extract (2.3)>Hydroethanol (2.0)>Hydromethanol (1.8)> Butanol (1.7)> Aqueous (1.0).** Gram–positive organisms were more sensitive to chloroform extract, therefore, it can be useful to control food contamination by microbes.

However, there are some limitations in using spices like cinnamon, such as

a. the antibacterial activity is decreased when spices are added to food materials containing protein, carbohydrate, and fat, and
b. the strong flavor.

The flavor of the food products may not be acceptable by some consumer groups if large amounts of spices are added to the products to inhibit the food borne pathogens (Vangalapati *et al*., 2012, Domadia *et al*., 2007; Prasad *et al*., 2004).

Table 3.6:Antibacterial activity of Cinnamomum zeylanicum bark extracts determined by disc diffusion method on specific media for each test microorganism.

Cinnamon bark extracts	**Diameter of growth inhibition zones (cm±*SD*)**				
	Chloroform	**Hydromethanol**	**Hydroethanol**	**Butanol**	**Aqueous**
Escherichia coli	2.5±0.57	1.3±0.42	1.7±0.42	1.5±0.42	1.2±0
Amoxycillin	2.2±0.57	2.5±0.57	1.8±0.00	2.5±0.57	1.0±0.42
Klebsiella	2.3±0.13	1.8±0.57	2.0±0	1.7±0.57	1.0±0.57
Amoxycillin	1.2±0.42	0.5±0.57	1.1±0.57	1.7±0.42	1.0±0.57

IV. DISCUSSION

From ancient times spices have played a major role in the lifestyle of people from certain parts of the world. They have served numerous roles through history, including as coloring agents, flavoring agents, preservatives, food additives and medicine. The active phytochemicals derived from these spices have provided the molecular basis for these actions. Globalization has made these spices easily available and increasing their popularity. One widely used cooking spice which has potentially significant medicinal effects is cinnamon. There are reports of *C.zeylanicum* being imported to Egypt from china as early as 2000 BC. It has been mentioned in the bible (Exodus and proverbs) and in Chinese text written 4000 years ago. In native ayurvedic medicine, cinnamon is considered a remedy for respiratory, digestive and gynaecological ailments.

In the present study the bark of cinnamon were analysed for the phytochemical and physiological and antibacterial evaluation using standard methods against pathogenic bacterias. Phytochemical screening included qualitative chemical examinations and tests for determination of primary and secondary metabolites. The medicinal properties of this plant were attributed to its variety of active phytochemical constituents. The emergence of resistant bacterial and fungal strains due to overuse of antibiotics is a cause of worldwide concern. The use of plant extracts and phytochemicals with known antimicrobial properties may have great significance in therapeutic treatments (Pandey and Gupta, 2014; Paliwal *et al*., 2011; Vaya *et al*., 1997; Middleton and Kandaswami, 1992)

Although this plant had received interest for the phytochemical and antimicrobial investigations since many years, more work has to be done on its isolation and characterisation for exploring the immense medicinal potential of this plant.

V. CONCLUSION

Medicinal plants are the richest bioresource of drugs for traditional systems of medicine, modern medicines, Nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs (Paliwal, 2015). Aromatic plants are a source of fragrances, flavours, cosmeceuticals, health beverages and chemical terpenes. Medicinal plants are important for pharmacological research and drug development. Over three-quarters of the world population relies mainly on plants and plant extracts for health care. One fifth of all the plants found in India are used for medicinal purpose. Out of these the bark of Cinnamon is widely used as a spice due to its distinct odour of different compounds. The research and experimentation was done on its phytochemistry and various pharmacognistic and antimicrobial properties of the spice. On comparison of the antimicrobial activities of all the five extracts tested against the bacterial strains, it was finally concluded that chloroform cinnamon extract emerged as the potent agent exhibiting even much higher antibacterial activity than the standard antimicrobial drug Amoxicillin. The need of the hour is to perform more and more screening of the natural products or plant parts that may open the possibilities of finding new clinically effective antibacterial compounds against the resistant bacterial pathogens. Hence extensive research is required to find out the mechanism of action of other compounds in cinnamon and exploit their therapeutic potential to combat various diseases. Therefore, Cinnamon plays an important role in modern system of medicine as a multipurpose medicinal spice.

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