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Review Article

Physicochemical analysis of Cinnamon Sticks (Cinnamomum Zeylanicum) and their Pathogen & Microbial Examination

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Abstract

Cinnamon is a medical herbs tree and all are a member of the genus *Cinnamomum* in the family Lauraceae. Cinnamon (Dalchini) is found in the Himalayan region, Ceylon and Malaya. It is used as an aromatic in a variety of medicine and foods. Due to which the food items are very tasty and full of quality. The cinnamon has different types of some physicochemical properties; foreign matter (28.56 % w/w) and total ash (58.2 % w/w) and water-soluble ash (28.3 % w/w) and water-soluble extractive (28.28 % w/w) and loss on drying (47.8 % w/w) and the help of TLC saw and identify that it has the same bioactive compound which has important medical properties and microbial & pathogen examination, which are useful in medicine and important food industries[1]. Cinnamon contains a variety of important derivatives with acids and it also has anti-inflammatory and antioxidant properties & it reduces the growth of cancer cells[2]. In physicochemical studies, we get information related to the structural characteristics of cinnamon as well as their physicochemical properties and can use them for our use likes the field of medicines, the field of human health, and food preservation.

Keywords: Cinnamon (Dalchini); Procyanidin A-Type and B-Type Linkages; Physicochemical properties; thin layer chromatography; Microbial limit & pathogen test.

INTRODUCTION

Cinnamomum Zeylanicum is relative to the Lauraceae family. Cinnamon is an evergreen herb tree found at an altitude of 30 feet and is brushy; leaves are dark green (Figure 1). This medicinal used in the treatment and prevention of many common ailments such as pimples, cough, and digestive troubles etc. There are different types of cinnamon species in the world but not only food but also significantly, to medicine and due to its medical properties, it is used in making various types of medicines [1].

Fig-1: Leaves of Cinnamon (Dalchini) plant and sticks

Cinnamon is a type of herb tree, whose components are cinnamaldehyde and tras-cinnamaldehyde and which is found in various types of the essential oil. This contributes to a variety of aromas and various biological activities. Both catechins and procyanidins are present in cinnamon barks which contribute significantly to the medical field. It has been observed by scientists that as procyanidins component there are procyanidins A-types and B-types linkage. varieties of different types of berries and Cinnamon extracted from procyanidins also have excellent properties of antioxidant activity [3] and the most widely used worldwide for species of medicinal and foodstuffs and with extreme properties [4]. Antioxidant properties as well as other active ingredients are found in various types of cinnamon it is found in the water-soluble portion of cinnamon, not in different types of cinnamon oils. More than 300 different types of essential cinnamon oils are still present as components [2, 5].

Cinnamon is used in ayurvedic medicine to treat various types of diseases like; rheumatism, aching joints, stiffness, urinary problems, and respiratory tract problems. Cinnamon powder mixed with tea when coughing drinking reduces or eliminates and also the effect of cough and colds to prevent it from taking hold fully (Figure 2). Cinnamon (Dalchini) is found in the Himalayan region [1]. It is also recorded that excess use of 'Dalchini' can cause liver damage due to the presence of a compound named 'coumarin' in it. The outermost layers consist of a well developed continuous band of characteristic stone cells. Parenchymatous cells contain phloem fibers, Phloem fibers, lignified, phloem parenchyma, and medullary rays, and lumen of fibers is greatly reduced[6]. Scattered in this region are also the secretary cells containing oil (oil cells), another characteristic feature of this bark, and large cells containing mucilage. Medullar rays, 1 to 3 cells wide and extend up to the stone cell layer where they may become wider.

Fig-2: Cinnamon sticks, powder of *Cinnamomum zeylanicum*

Physicochemical properties

"Physical science is the science that must clarify under arrangements of physical trials the purpose behind what's going on in complex bodies through substance activities". The useful properties of tests determine effectively by physicochemical properties such as loss on drying, water-soluble extractive, determination of a foreign matter, determination of total ash, etc [7, 8].

The physicochemical properties important to synthetic choices appraisal can be utilized to recognize physical dangers and to comprehend or anticipate a compound's natural destiny, human poisonous quality, and so on [9].

Determination of foreign matter

First of all, taken 150 gm of the sample (Cinnamon) and then tested the sample thoroughly and after that, we spread the sample in a thin layer. So that we can easily see foreign particles, which are present in the sample (Cinnamon) and after that analyzed the sample with the help of a lens. After the analyzed the sample is weighted again by disassembling the foreign particle and weighed it again by disassembling the foreign particle. Then calculated that by formula, what percentage of foreign particles present in the sample.

Determination of total ash

2.65 gm of the sample was incinerated in continuously weighted silica crucible at a temperature not exceeding 450 ^{0}C in a muffle furnace until it was free of carbon fumes until the release of the cooled carbon fume and the weight of the crucible+ash.

Determination of water-soluble ash

The ash obtained was boiled by inserting 25 ml distilled water for 2 min to 5 min. The insoluble matter was collected in an Ash less filter paper; (Whatman no.42), Washed with hot water, and ignited in the crucible, not exceeding the temperature 450 ^{0}C. After the weight of the remaining insoluble ash is subtracted from the weight of aggregate ash. So that we can easily calculate the water-soluble ash and it has been proved by various experiments that the percentage of ash found in water is done in the context of many dry drugs taken in the air.

Determination of water-soluble extractive

Taken 5 gm air-dried drug and mixed 100 ml specific strength of alcohol and kept it in a stable place for 3 hours in a closed conical flask and it always had to be shaking after 20 min to 25 min. filtering the solvent (alcohol) easily so that there is no loss, Evaporate 25 ml of the filtrate in a tarred, flat bottom shallow dish to dryness at 105^{0}C, weigh and the water-soluble extractive is then calculated by a formula.

Loss on drying

First of all taken the 5-gram drug, spread uniformly and thin layered in a shallow Petri dish. After then heated up to the temperature of 105^{0}C, cool in a desiccator, and after some time interval weight the drug. To get the excellent results, this process repeated

sometimes till two incessant weights were found constant. The fraction of weight loss was calculated based on the initial weight.

Determination of pH

pH of 1% solution

Dissolve an accurately weighed 1gm of drug inaccurately measured 100mL of water and use a standardized glass electrode to verify the sample pH.

pH of 10% solution

An experiment in the same manner as above taking 10gm of the drug instead of 1gm

Table-1: Physicochemical study of powder of *Cinnamomum Zeylanicum*

Parameter	Percentage (w/w)
Ash values	
Total ash	58.2
Water-soluble ash	28.3
Successive extraction values	
Water-soluble extractive	28.28
Alcohol soluble extractive	14.66
Loss on drying	47.8
pH values	
1% water soluble	6.23
10% water soluble	4.05
Foreign matter	28.56

Thin Layer Chromatography (TLC)

Thin-layer chromatography is a technique used to differentiate the different types of material that are mix in each other **(**Figure 3). It can also perform thin layer chromatography on a plastic seat and glass seat or aluminum foil, on which a thin layer of adsorbent material is made, usually aluminum oxide and silica gel or cellulose [10]. The thin layer of adsorbent material itself is known as the stationary stage [11].

Because different analysis climbs the thin layer plate chromatography on different various rates, so we can get partition through thin layer chromatography. TLC can be utilized to screen the progress of a response recognize compound present in a given blend and decide immaculateness of substance. Each spot has a retention factor (R_F) expressed as:

R_F = Distance traveled by sample/Distance traveled by the solvent

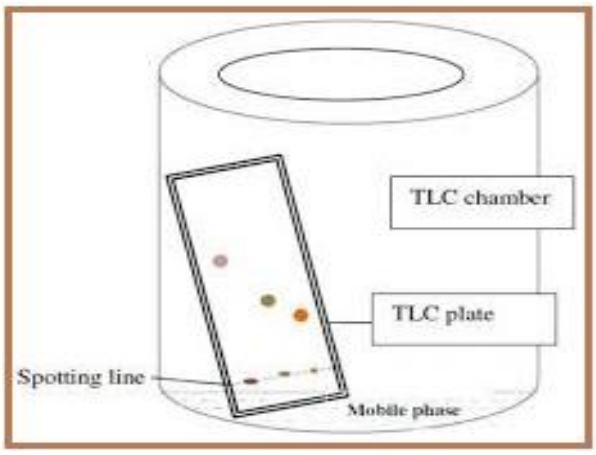

Fig-3: Thin layer chromatography

Extract 2 gm of the sample in a water bath for 25 min to 30 min with 10 mL of alcohol under reflux. After filtering a 5 ml concentration drug, spot on the chromatography and apply alcohol to the TLC plate, and extract. Use of different types of solvent like- ethyl acetate: methanol: H_2O, chloroform: ethyl acetate for developing the mobile phase to the plate distance of 8.2 cm. After the development of spots on the TLC plate, then go for dried in air and analysis in the under UV (366 nm). It shows major spots on TLC at R_F 0.28, 0.76, 0.24 (orange). Dip the plate in Vanillin-sulphuric acid reagent for 1 min followed by heating on 100 ^{0}C to 115 ^{0}C for about 2 minutes to 5 minutes and observe under visible light. The plate reveals large spots at R_F (0.98, 0.62, 0.54) (Figure 4) (A) (B).

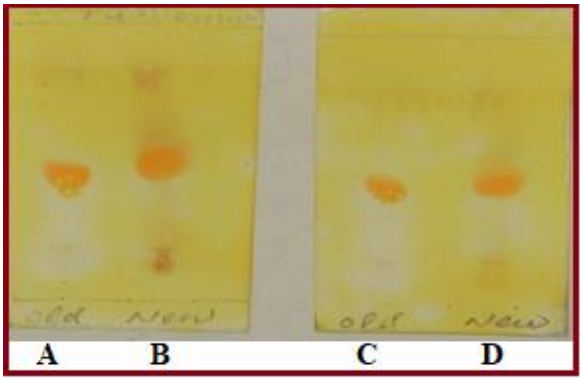

Fig-4: TLC (alcohol) of Cinnamon (*Cinnamon Zeylanicum*) (A) old, standard *Cinnamon Zeylanicum*, (B) sample of *Cinnamon Zeylanicum*. TLC (Chloroform) of Cinnamon (*Cinnamon Zeylanicum*) (C) old, standard *cinnamon Zeylanicum*, (D) sample of *Cinnamon Zeylani*

Microbial limit test

The microbial limit test is used for the qualitative and quantitative examination of specific types of micro-organisms (bacteria and fungi & *Escherichia coli*) present in the drug. This type of test should be conducted for drugs prepared by a smooth mixing portion from different types of ingredients. The experiments must be carried out rapidly because the

samples are diluted with a fluid medium. Mechanism of membrane filtration, method of pouring plate, method of spreading plate, and method of serial dilution (most likely method of numbering). Depending on the intent, an acceptable method should be taken from among those others. But I was analysis the microbial & pathogen limit test by "pour plate method" and "serial dilution method" in this research work.

Pour plate method

Use a Petri plate per diameter of 9 cm to 10 cm and take 1 ml test fluid and doing carefully diluted through distilled water and easily transfer 18 ml to 20 ml sterilized agar medium in all the Petri plates. Using soybean-casein digest agar medium for fungal detection, or one of Sabouraud dextrose agar, potato dextrose agar, for bacteria detection. After the medium is stabilized, keep in incubate it for 5 days at 30 ^{0}C to 35 ^{0}C for bacterial growth and incubate at 20 ^{0}C to 25 ^{0}C for fungal growth (Figure 5).

If there is a large number of colonies in the agar media and on receiving more than 300 bacterial colonies it was calculated based on viable counts and a plate should not contain more than 100 fungal colonies for fungal detection. If counts in a shorter incubation time than 5 days are considered reliable, these counts may be adopted.

Serial dilution method (most probable number method)

Using 12 test tubes: 9 containing 9 mL of digested soybean casein medium each and 3 containing 10 mL of control medium each using the nine tubes to prepare dilutions. Next, to each of the three test tubes, apply 1 mL of test fluid and combine to create a 10-fold dilution. Secondly, to each of the three test tubes, add 1 mL of each of the 10-time dilutions and mix to make a 100-time dilution. Each of the remaining three test tubes, add 1 mL of each of the 100-fold dilutions and combine to produce 1,000-fold dilutions. For at least five days at 30-35 ^{0}C, incubate all twelve test tubes. Not observe the microbial growth in the test tubes. If it is tough to decide the outcome or if the outcome is not accurate, take 0.1 mL of fluid from each of the nine test tubes and position it in an agar medium or fluid 24, incubate all the medium at 30 to 35 ^{0}C for 24 to 72 hours and check for the absence or existence of microbial growth.

Limits: Bacterial count limit and fungi (Yeast and Mold) count limit.

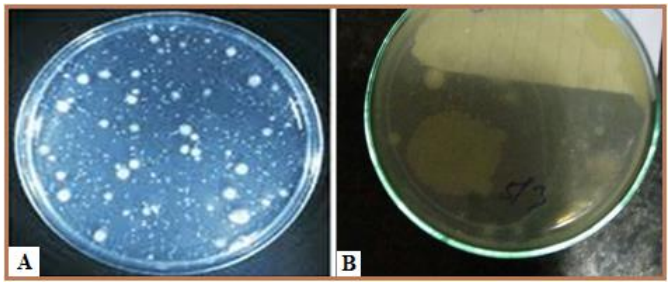

Fig-5: MLT for Bacterial growth (A) & MLT for Fungal growth (B)

Table-2: Microbial limits for raw materials

Microbial Parameter	Limit	Result
Total aerobic microbial count	Not more than 1000 cfu/gm	850 cfu/gm
Yeast & moulds	Not more than 500 cfu/gm	10 cfu/gm
Pathogen		
E.coli	Absent/mL	Absent/mL
Salmonella	Absent/mL	Absent/mL
Pseudomonas	Absent/mL	Absent/mL
Control Plate	Absent/mL	Absent/mL

Table-3: Pathogen result of different temperature

Sample	E.coli	Pseudomonas Aeuginosa	Salmonella	Staphylococcus Aureus
Spray preservative & stand at 60 ^{0}C for 2 hrs	Absent/ mL	Absent/ mL	Absent/ mL	Absent/ mL
C. Zeylanicum 60 ^{0}C for 3 hrs	Absent/ mL	Absent/ mL	Absent/ mL	Absent/ mL
C. Zeylanicum 60 ^{0}C for 5 hrs	Absent/ mL	Absent/ mL	Absent/ mL	Absent/ mL
C. Zeylanicum 80 ^{0}C for 2 hrs	Absent/ mL	Absent/ mL	Absent/ mL	Absent/ mL

RESULT

The purity of the isolated compound was analyzed by thin-layer chromatography. The spot on TLC chromatography developed with ethyl acetate gave a positive result in the Liebermann-Birched test and methanolic H_2SO_4 reagent indicating a steroidal substance. The physicochemical study shows that cinnamon stick, different types of important bioactive

components and need for natural components has led to cinnamon in various fields such as pesticides, medicines, pharmaceuticals, and particularly in the food processing industry, motivated to get special interest in all these. Different types of cinnamon oils have important types of biological activities such as; antimicrobial and antifungal properties etc. The total viable aerobic count is considered an important method for the determination of various types of mesophilic bacterial and fungal. It has been observed by research that it can also give negative results, even if it is in large numbers. There is a certain method for its tests which are considered important for counts of different types of micro-organisms.

DISCUSSION

A variety of laboratory findings and pharmacopoeia-based cinnamon trials are the traditional for the standardisation of *Cinnamon Zeylanicum*. These have also possessed antidiuretic, antiarthritis, and anticancer agents so the isolation and study of the group of the compound may help to demonstrate their specific properties. Physicochemical standardization is of paramount importance for the Unani medicine quality control. As the effectiveness of many medications depends primarily on its physical and chemical properties.

CONCLUSION

Different pharmacognostic evaluations serve as the standard for identification and distinguishing a plant from its adulterants. Preliminary phytochemical tests showed that methanolic extract of *Cinnamon Zeylanicum* has a greater amount of Phyto-constituents compared to the other three extracts i.e., petroleum ether, ethyl acetate, and chloroform extract. The powder drug exhibited different fluorescence characters due to the presence of different function growth groups. TLC could be helpful in the identification of the sample. In the present study, it has been claimed by many scientists that cinnamon has a variety of important therapeutic properties. Which are also useful for human society and health and other food industry and medical fields and also use for food preservation? It have also some medical properties such as likes; Antimicrobial & antifungal properties etc.

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Conflict of interest

Authors declare no conflict of interest.

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