

Cardamom (*Elettaria cardamomum*) Seeds Extract as Antimicrobial and Wound Healing Agent

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Abstract: recently, the interest in plants is increased in different approaches of science, as it can be used to reinforce multiple materials of industry, as well as exhibit therapeutic properties to many pathologies and pathogens, including anti-diabetic, anti-cancer, antibacterial, antifungal, antiviral, antioxidants, and modulating various disorders. The importance of plants is gained from the presence of rich materials with multiple functions (most importantly therapeutic functions) called phytochemicals. Cardamom (*Elettaria cardamomum*) is an herb used as spice and exhibit many biological benefits. Our goal was to prepare an acetone extract of the cardamom seeds and use it as antibacterial, antifungal, and wound healing agent. The antimicrobial activity was investigated against two Gram negative bacterial strains *escherichia coli* as well as *klebsiella* sp, also two Gram positive bacterial strains *Staphylococcus epidermidis* and also *Staphylococcus aureus*, as well as one fungi (*Candida albicans*). Our results have shown a good activity of the acetone extract of the cardamom seeds against Gram positive as well as Gram negative bacteria as well as fungi. However, the most powerful effect was obtained against *Candida albicans*, which make the antifungal properties of cardamom extract over the antibacterial properties in acetone solvent. Furthermore, the effect of acetone extract of cardamom seeds on wound healing was significant, in which the healing was faster in rats whom treated with the extract. As it can exhibit these therapeutic potentials, cardamom can be used in the natural pharmacology field.

Keywords: Cardamom, antibacterial, antifungal, wound healing.

1. Introduction

Herbs are a collection of food additives that have been used for centuries as flavors and seasoning agents, as well as folk remedies and food preservatives, to improve the sensory quality of dishes. These spice elements provide dishes a distinct flavor, fragrance, or piquancy, as well as color, in order to pique the appetite. Many spices have historically been recognised to have therapeutic qualities, including tonic, carminative, stomachic antispasmodic, and antihelminthic [1]. Despite the fact that these findings are mostly

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empirical, they have been given pharmaceutical uses in the indigenous medical system. Spices and aromatic herbs are gaining popularity in both the industry and scientific study due to their antibacterial properties, particularly against food-borne bacterial infections [2]. Many bacterial infections have been shown to be efficiently controlled by extracts from plants such as cinnamon, clove, garlic, mustard, onion, and oregano [3, 4]. The essential oil portion of spices and herbs contains the majority of the antibacterial chemicals. According to Shan et al. (2005) [5,] more than half of the 46 spices tested had antibacterial activity against the pathogens *Staphylococcus aureus* and *E. coli*. *Shigella* species as well as *E. coli* enteropathogens have been discovered to be sensitive to extracts of numerous culinary spices [6]. In the presence of spice extracts, the growth of multiple drug-resistant pathogenic bacterial strains such as *Salmonella typhi* (D1 Vi-positive), *Salmonella typhi* (G7Vi-negative), *Salmonella paratyphi A*, *Escherichia coli* (SS1), *Staphylococcus aureus*, *Pseudomonas fluorescens*, as well as *Bacillus licheniformis* is inhibited [7]. Antimicrobial compounds in spices were more responsive to Gram-positive bacteria than antimicrobial compounds in spices were to Gram-negative bacteria [8].

Natural compounds have lately been investigated for their antibacterial and anti-inflammatory characteristics as potential oral infection therapy agents. The perennial aromatic plant cardamom (*Elettaria cardamomum*), which is traditionally used as a culinary component, is widely cultivated in southern India, Sri Lanka, Tanzania, and Guatemala. Flavonoids, alkaloids, terpenoids, anthocyanins, and phenolic compounds are all abundant in this plant [9, 10]. Cardamom has been used for centuries to treat a wide range of diseases, including asthma, digestive issues, as well as chronic jaundice [11]. Cardamom has been demonstrated to have a variety of pharmacological effects, including antioxidant, anti-inflammatory, anti-cancer, and antibacterial qualities [12-16]. Therefore, our goal was to investigate the effects of cardamom seeds extract on *escherichia coli*, *klebsiella* sp, *Staphylococcus epidermidis*, *Staphylococcus aureus*, and *Candida albicans*. Also, to explore the potentials of using cardamom seeds extract as a wound healing agent.

2. Materials and Methods

2.1. Preparation of cardamom seeds extract

Cardamom seeds were purchased from a local herbs shop. The seeds were grinded by the mortar and 200g of the dry powder were mixed with 500mL acetone (Merck, Germany). The beaker was sealed by a watch glass and placed in the oven at 37 °C for 3 days. The solution then was filtered and evaporated to obtain a concentrated extract of the cardamom seeds.

2.2. Antibacterial test

The acetone extract of the cardamom seeds was examined against two Gram negative bacterial strains *escherichia coli* and also *klebsiella* sp, as well as two Gram positive bacterial strains *Staphylococcus epidermidis* also *Staphylococcus aureus*, as well as one fungi (*Candida albicans*). In Petri dishes, well diffusion method was used. Two wells in the agar medium were made in a radius of 5mm. In these wells 50µL and 100µL of the cardamom seeds extract were added to the corresponded wells. The plates were incubated at 37 °C for 24 hours, as well as the inhibition zones were determined in mm.

2.3. Wound healing in rats

A paraffin cream was prepared from the concentrate extract of cardamom seeds. A volume of 50 or 100mL of the cardamom seeds extract were mixed with paraffin at 70 °C under continuous stirring for 2h.

Three groups of healthy laboratory rats were placed in a cage and pre-conditioned for the experiment for 3 days. Each group was contained 10 male rats, and divided according to the treatment as control group, and two groups of cardamom seeds extract (50mL and 100mL). The dorsal area of the rats was circled, as well as the area was localized with a 10% lidocaine sprayer to produce a wound in a radius of 1.0 cm using a surgical blade, leaving the incision open until redness indicated acute inflammation. The rats were treated on a daily basis, and observations made during the treatment procedure were recorded. The diameter of wound was measured each day for 14 days, and the results were statistically processed by using analysis of variances (ANOVA) test, where the values were considered significant at $P \leq 0.05$.

3. Results and Discussion

3.1. Antimicrobial effects

Table 1 contains the inhibition zones created by the presence of 50 and 100µL acetone extract of cardamom seeds. The acetone extract solution of cardamom has shown to have an antimicrobial effect against all of the strains that have been used in this study. Upon the use of 50µL of the extract, cardamom seeds were caused an inhibition zones 7mm for *E.coli*, 6.5mm for *klebsiella* sp., 7.1mm for *S. epidermidis*, 6.8mm for *S. aureus*, and 8mm for *Candida albicans*. While the 100µL of the cardamom seeds extract have shown inhibition zones as 7.8mm for *E.coli*, 9mm for *klebsiella* sp., 8.1mm for *S. epidermidis*, 7.6mm for *S. aureus*, and 8.5mm for *Candida albicans*. Cardamom seeds extracts were effective to inhibit the growth of Gram positive and Gram negative bacterial strains, while the most affected microbe was the fungi *Candida albicans*. Hence, we can presume that acetone extract of cardamom seeds has antifungal activity more than its antibacterial activity.

Table 1: Inhibition zones of caused by cardamom seeds extract.

Type of microbe	50µL Cardamom extract	100µL Cardamom extract
<i>E. coli</i>	7	7.8
<i>klebsiella</i> sp.	6.5	9
<i>S. epidermidis</i>	7.1	8.1
<i>S. aureus</i>	6.8	7.6
<i>Candida albicans</i>	8	8.5

The antimicrobial activity of cardamom seeds were reported in several previous studies [2, 17-20]. The cardamom has been reported to contain high quantity of therapeutic materials such as flavonoids and vitamin C [21]. Flavonoids are shown to exhibit strong antibacterial activity through different mechanisms such as damaging the bacterial membrane, reducing the synthesis of nucleic acids, and reducing the energy metabolism [22].

3.2. Wound healing

Table 2 contains the information that obtained from the observations of rats post inductive injury. The significant effect of both 50mL cardamom seeds cream (1.64 ± 0.05 cm) and 100mL cardamom seeds cream (1.67 ± 0.09 cm) were started after 7 days from the treatment compared to control (1.76 ± 0.05 cm). Furthermore, the effect of 100mL acetone cardamom seeds extract cream (1.13 ± 0.14 cm) and 50mL acetone cardamom seeds extract cream (1.12 ± 0.1 cm) was observed to be highly significant ($P < 0.01$) after 14th day from injury which have shown complete healing with no sign of inflammation. The differences between

using different volumes of cardamom was not significant, which may be attributed to the mild applicable potentials of the material.

Table 2: The effect of cardamom seeds extract on wound healing in rats.

Healing Period (day)		Control rabbits	50mL Extract	100mL Extract	p-value
1 st	Diameter (cm)	2.0±0.0	2.0±0.0	2.0±0.0	-
	Observations	Redness	Redness	Redness	
3 rd	Diameter (cm)	1.94±0.05	1.91±0.07	1.93±0.07	0.582
	Observations	Redness	Light Redness	Light redness	
5 th	Diameter (cm)	1.86±0.05	1.83±0.07	1.84±0.07	0.568
	Observations	Inflamed area	Clot	Clot	
7 th	Diameter (cm)	1.76±0.05	1.64±0.05	1.67±0.09	0.002*
	Observations	inflammation	Clogged	Clogged	
10 th	Diameter (cm)	1.68±0.06	1.55±0.08	1.56±0.12	0.006*
	Observations	Clogged	Small clot	Small clot	
14 th	Diameter (cm)	1.45±0.08	1.12±0.1	1.13±0.14	<0.001*
	Observations	Clogged	Small clot	Small clot	

Mean ± Standard deviation; * Significant at P equal or less than 0.05

The seeds of cardamom plant are full of various phytochemicals that exhibit many therapeutic potentials such as flavonoids, alkaloids, and polyphenolic compounds [23, 24]. An optimum wound healing dressing or agent protects the wound tissue from bacterial infection, reduces inflammation and induces cell proliferation to aid in the reconstruction of damaged tissue [25]. Our results have shown that cardamom seeds extract have an antimicrobial activity against different microbes. Cardamom has also an action as an antioxidant [26] as free radicals are considered the major cause of inflammation during wound healing process [27].

4. Conclusion

Cardamom seeds are full of phytochemical compounds with various medical potentials. Our results have shown a good activity of the acetone extract of the cardamom seeds against Gram positive and also Gram negative bacteria as well as fungi. However, the most powerful effect was obtained against *Candida albicans*, which make the antifungal properties of cardamom extract over the antibacterial properties in acetone solvent. Furthermore, the effect of acetone extract of cardamom seeds on wound healing was significant, in which the healing was faster in rats whom treated with the extract. As it can exhibit these therapeutic potentials, cardamom can be used in the natural pharmacology field.

5. Highlights

The study examines the therapeutic potential of cardamom, highlighting its antimicrobial and wound healing properties. The acetone extract of cardamom seeds displayed a significant effect against bacteria and fungi, particularly *Candida albicans*, and enhanced wound healing in rats. The study suggests the possible application of cardamom in natural pharmacology.

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