

Antimicrobial Activity of *Ananas comosus* Fruit Extract Against Some Pathogenic Bacteria

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Abstract

The present study investigates the use of agar well diffusion method and the inhibitory effect of *Ananas comosus* fruit extract on two gram- negative bacteria(*Klebsella* spp., *Salmonella typhi*) and one gram- positive bacteria (*Staphlococcus epidermidis*). The results showed that extract of *Ananas comosus* in concentrations 2.5%, 5%, 7.5%, 10% mg / ml, have significant inhibitory effect against bacterial species used in the study. It was indicated that there is a positive relationship between the concentrations of aqueous extract and bacterial sensitivity, whereas the extract is less active against gram positive bacteria as compared with gram negative bacteria.

Keywords: Ananas comosus; pathogenic bacteria; Antimicrobial Activity.

1. Introduction

There is an urgent and continuous need to discover new antimicrobial compounds with diversity in chemical composition and mechanisms of action, due to the alarming increase in the incidence of new infectious diseases and its reemergence. In addition to the evolution of antibiotic resistance in current clinical use [1]. Therefore, for the treatment of infectious diseases, many alternative and complementary compounds are discovered for safe therapeutic purposes.

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Nowadays, plant products considered as best solution for effective control of microbial infections, because of their side effects on body tissues are low [2]. Pineapple or *Ananas comosus* belongs to the family of Bromeliaceae, grown in several tropical and subtropical countries. It has been used as important food and in folk medicine to cure different disease[3]. Phytochemical analysis of pineapples juice showed the presence of antimicrobial substances such as phenolic compounds, alkaloids, Steroids , flavanoids, tannins, resins and enzymes such as bromelain. *In vitro* and *in vivo* studies demonstrate that bromelain exhibits various fibrinolytic, antiedematous, antithrombotic, and anti-inflammatory activities, however, its use as an antibacterial agent is yet to be studied[4]. Moreover, phenols, as the simple phenolic compound, one of the secondary metabolic compounds found in pineapple fruits, consisting one aromatic ring linking, one set of hydroxylated groups (OH) or more. This group carry anti-dioxin properties [5]. Toxicity of phenols on microscopic organisms depend on sites and the number of such hydroxylated groups, as more of these sites increase, the toxicity of phenolic compound increase [6].

2. Methods

2.1. Sample Collection and Preparation

Pineapple Fruits were taken from the domestic market. Cut to small pieces with weight of 20 grams, put it in panasonic processor with 200 ml distilled water. Left for 24 hours at room temperature. After that, by using several layers of medical gauze, the mixture was filtered. The solution was centrifuged ($3000 \text{ r} / \min$) for 10 minutes. Filtered with Whatman No. 1 filter paper in a sterile glass vessel,. The process was repeated for three times to ensure removing the disposal of the sediment. Stored the extract at 5°C [7]. The concentrations (2.5%, 5%, 7.5%, 10%) was prepared by taking appropriate amount of the extract with appropriate amount placed in a of sterilized distilled water.

2.2. Microbial Strains

The test organisms used in this study included, one gram positive bacteria (*Staphylococcus epidermidis*), and 2 gram negative bacteria(*Klebsilla spp., Salmonella.* typhi), isolated from 50 samples of urine from patients at Rahibat (The Nuns) and Al- Yarmouk Teaching Hospital in Baghdad . These strains were collected from laboratories of College of Science /Baghdad university.

2.3. Antimicrobial assay

Antimicrobial activity of concentrations (2.5%, 5%, 7.5%, 10%) of pineapple fruits extract were tested using an agar well diffusion method [6]. Mueller-Hinton agar (MH) was used. Microbial cultures that freshly grown at 37° C, were appropriately diluted in sterile distilled water to obtain cell suspension at ($1.5*10^{8}$ cfu/ml). 100µl of suspensions were spread on(MH), allow the inoculums to dry. Five wells of 5 mm diameter were made into the agar medium. Four wells were filled with 100 µl of different concentrations that prepared from pineapple fruits extract . The fifth well was filled with 100 µl of sterile distilled water. The inculam was allowed to diffuse into the medium for 1 h at room temperature. Plates of test organisms were incubated at 37° C for 24 h. Antimicrobial activity was estimated by measuring the inhibition zone diameter against the test organisms.

3. Results and Discussion

The wide spread of drug resistance in human pathogens against commonly used antibiotics became one of the major problems in the world that required a search for alternative antimicrobial substance from other sources including plants and microbes [8]. The traditional medicinal methods, especially the use of medicinal plants, still play an essential role to support the basic health need in the developing countries. In this manner, plants still to be a rich source of therapeutic agents[2]. Three identified clinical bacterial isolates(1 Gram-positive and 2 Gram-negative bacteria) were checked for their susceptibility pattern to pineapple fruits extract. The inhibitory effect of extract at concentrations of 2.5%, 5%, 7.5%, 10% for each of the different bacterial species were described in table-1. The results revealed that bacterial species under study are susceptible to pineapple fruit extract, and its sensitivity are increased with increasing of concentration of fruit extract. It was examined by the presence of inhibition zone diameter. This antibacterial activity is mainly due to the presence of high level of various active components such as phenolic compounds, alkaloids, flavanoids, tannins, resins and enzymes(bromelain) which play important role in plant defense, and showed higher antimicrobial activity that can used as strong antimicrobial agents for chemotherapy of multidrug resistant human pathogens[1, 5]. This result is matching with previous studies that showed the effectiveness of the pineapple extract is attributed to phenolic compounds that inhibit the growth of gram- negative and gram- positive bacteria by deposit proteins in the bodies of microbes by hydrogen bonds among phenolic hydroxyl groups, then by breaching the function of some important enzymes in the bodies of those bacteria [5,9,10]. It was indicated that the extract of pineapple fruits is less active against gram positive bacteria as compared with gram negative bacteria that may be attributed to the differences in cell wall structure [11].

Inhibition zone diameter (mm)				
Concentration				
Bacterial strains	2.5%	5%	7.5%	10%
Salmonella. typhi	15	20	22	25
721 1 11	10	1.5	10	20
Klebsilla spp.	10	15	18	20
Staphylococcus epidermidis	8	13	16	18

Table 1: The susceptibility pattern of pineapple fruit aqueous extract against some pathogenic bacteria

4. Conclusion

Pineapple fruits aqueous extract has antimicrobial activity against pathogenic bacteria under study. So, it can be used in the production of therapeutic materials (antimicrobial agents), thus reducing the incidence of resistance among pathogens that appear as a result of the misuse and overuse of antibiotics, and there is a positive relationship between the concentrations of aqueous extract and bacterial sensitivity.

5. Recommendations

Many more studies are needed to isolate and characterize the bioactive components present in pineapple fruits, and confirm ingredients effect *in vivo*.

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