



## Cytotoxic and antimicrobial potential of different varieties of ripe banana used traditionally to treat ulcers



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### ABSTRACT

Globally, plant as herbal medicine is used to treat different diseases. The goal of this present study was to assess the antimicrobial and cytotoxic potential of ripe banana extracts with various polarities which were grown in Oman. The powder banana samples were extracted with methanol by soaking mode. The solvent was removed by a rotary evaporator and the extract was fractionated by dissimilar polarities of solvents to give corresponding extracts. The antimicrobial and cytotoxic potential of the various polarities extracts of ripe banana were determined by modifying disc diffusion and brine shrimp lethality (BSL) bioassays. Each banana extract at different concentrations showed modest antimicrobial potential against two Gram (+) and two Gram (-) with the range of inhibition 0–13 mm. The results of cytotoxic potential revealed that the each banana extract at various applied concentrations killed the shrimp larvae. Among the extracts, the butanol and water extract gave the maximum cytotoxic potential with LC<sub>50</sub> value of 20.12 and 24.22 µg/ml, however, the lowest cytotoxic potential was obtained in the ethyl acetate extract with LC<sub>50</sub> value of 36.68 and 49.32 µg/ml. In our experimental results showed that the highest polarity banana extracts have significant antimicrobial and cytotoxic potential. In conclusion, the highest polarity extract could be a candidate for the use of antibiotics.

### 1. Introduction

Natural products are the main source of drugs and that is the only choice worldwide. Plants are reflected as one of the predominant natural sources and plants itself and its derived products are more dependable and safer source for its renewability. Therefore, the plant sources and its derived products are well accepted primary medicines and still, the plants and its formulated products are globally recognized as a main health care system. However, previously, researchers are interested to work with available plant sources to find an effective individual therapeutic agent [1]. The plant, fruits, and vegetables itself and its extracts are more effective compared to pharmaceutical ones which has no or major side effects [1]. Several scientific evidence showed that more intake of fruits and vegetables have a poorer risk of cancer [2]. Polyphenolics, alkaloids and glycoside compounds are the most important group of plant ingredients which is mandatory for the daily human diet. Those groups of compounds play a vital role to control the biological functions of the human body and it is directly related to the interference of cancer development [3]. Bananas are globally recognized crops available everywhere, including Oman. It is a cheapest and available crop and affordable for everyone. Several varieties with different size of bananas are available in Oman. Banana is

considered a major source of naturally nutritious food belongs to the Musaceae family. It originates from the western countries and currently, widely distributed and cultivated worldwide. First, it is spread to the Indian region during 600 BCE and later on, it spread globally. From the literature search reveals that banana is the oldest cultivated crop [4]. Nowadays, more than 100 countries are commercially cultivated regular basis due to the instant source of energy and nutrition [5,6]. According to the priority crops list, banana is the fourth position and it is the most significance traded crops in the global market [7,8]. The plant height is about 4 to 5 m and the leaves are spirally arranged. All parts of the selected plant showed significant medicinal values [5]. Most of the phytochemical studies showed that the banana extracts contain a high percentage of fatty acids, sterols and its ester, linoleic acid and its derivatives, and sugar derivatives [7–10]. Recently, some important bioactive chemical compounds were detected in the banana like vitamin B & C, oxalic acid, sulphate, starch, tannin, glycosides, phenolic derivatives, triterpenes, stigmast-7-methylenecycloartanol, stigmast-7-en-3-ol, lanosterol, and α-amyrin which are responsible to control biological function and activities [11–14]. Current literature searches reveal that the complex phytochemicals also present in the banana extracts which are more effective than the single ingredient to prevent cancer due to the additive effect [15,16]. The people are using

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traditionally the stem juice to treat epilepsy, hysteria, dysentery, and diarrhea [9]. The extract from flowers is a good medicine using traditionally to treat bronchitis, dysentery, and ulcers [17]. Since ancient times, the banana syrup has been used to treat diabetes, epilepsy, leprosy, fevers, hemorrhages, dysentery and diarrhea [17]. In India, the paste of mature and young leaves is used to treat burns, skin afflictions, dysentery, diarrhea and malignant ulcers [17]. The paste of the roots is also used to treat dysentery and other chronic diseases [17]. Omani are used traditionally to treat digestion, constipation, and diarrhea. However, there is no extensive scientific work was done on Omani banana species. Therefore, it is mandatory to know the toxicity and phyto-pharmacological activities of the locally grown banana that are collected from Sohar and Dhofar region. On this regards, the goal of this research work was to prepare various polarities ripe banana extracts from varieties ripe banana and to assess their antimicrobial and cytotoxic potential.

## 2. Materials and methods

### 2.1. Chemicals and reagents

Various polarities of solvent like hexane, ethyl acetate, butanol, methanol, chloroform and acetone were used collected from Sigma Aldrich Company, Germany and were > 98.3%, > 98.6%, > 96.9%, > 99.1%, and > 97.6% purity, respectively. The other necessary reagents and chemicals, especially NaCl and Na<sub>2</sub>SO<sub>4</sub> were used to purchase from BDH, UK. The levofloxacin and dimethyl sulphoxide (DMSO) were obtained from E. Merck, Germany.

### 2.2. Microorganism

Two Gram (+) bacteria: *Staphylococcus aureus* (*S. aureus*, Code no. 207) and *Streptococcus pneumoniae* (*S. pneumoniae*, Code no. 257) and two Gram (-) bacteria: *Escherichia coli* (*E. coli*, Code no. 337) and *Haemophilus influenza* (*H. influenza*, Code no. 236) were used in this present study. All the selected Gram (+ and -) bacterial strains were obtained from the local Nizwa Hospital, Nizwa, Oman.

### 2.3. Sample collection

The samples were collected from the garden at Dhofar and Sohar cities in the morning time on December 21, 2016. The ripe banana samples were taken at home for cleaning and slicing.

### 2.4. Banana extracts

The banana samples were sliced and it was dried at ambient temperature for 5 days under the electric fan. The dried hard samples were made coarse powder by a heavy-duty blender machine and the samples (350 gm) were packed with filter paper and kept in a beaker and added methanol (1 L) for 48 h. Then, the methanol was filtered by using a Buckner funnel and the solvent was removed by a rotary evaporator. Finally, the prepared banana extract (20 gm) was liquefied in water and successively fractionated with organic solvents with increasing polarity pattern to afford corresponding hexane, chloroform, ethyl acetate, butanol, and water extracts, respectively [18]. All mother solvents were evaporated by the same process and the dried fractions were used for the determination of pharmacological and toxicological activities.

### 2.5. Antimicrobial assay

The ripe banana extracts were used for the determination of antimicrobial potential by using slightly modified agar gel diffusion method



Fig. 1. Local Banana collected from Sohar.

against two Gram-positive bacteria: *S. aureus* (code no. 207) and *S. pneumoniae* (Code no. 257) and two Gram-negative bacteria: *E. coli* (Code no. 337) and *H. influenza* (Code no. 236) [18,19]. In this experiment, board-spectrum antibiotic levofloxacin and DMSO solvent were used as controls. The concentration of positive control was 1 mg/5 ml in DMSO. Filter paper 6 mm diameter was used as the paper discs. The determination of the antimicrobial potential of various polarity banana extracts at various concentrations (2, 1, 0.5 and 0.25 mg/ml) were used. Initially, all discs were sterilized and impregnated with each concentration of various banana extracts and place on the agar plates. Finally, all applied plates were incubated at 37 °C for 24 h. The antibacterial potential of each concentration extract has measured the zones of inhibition against the Gram (+ and -) bacterial strains. The experiment was replicated three times. The potential for each polarity ripe extract at each concentration was evaluated by using the following formula (Fig. 1).

Antimicrobial potential = Inhibition zone of ripe banana sample/Inhibition zone of standard

### 2.6. Cytotoxic assay

The cytotoxic potential was assessed of all polarities banana extracts of ripe banana samples by BSL method described by Weli et al. [20]. Initially, the shrimp eggs were hatched with artificial seawater for 24 h. After 24 h hatching, the nauplii was separated from the eggs and come out to the other compartment. Those active nauplii was used for cytotoxic potential in this experiment. Two types of banana samples at different concentrations (10, 100, 250, 500 µg/ml) were prepared by using water. One hundred (100) µl of each concentration ripe banana extract was placed in the test tube containing 4900 µl of artificial sea water and 10 nauplii of each test tube. After 24 h of incubation, the total number of alive nauplii was counted and recorded. The well-established formula was used to calculate the percentage of the lethality of brine shrimps.

## 3. Results

The ripe banana sample was collected from the farmer at Sohar and Dhofar, Oman. Then the samples were sliced and dried under an electric

**Table 1**  
Yield of crude extracts of Sohar and Dhofar ripe banana.

Extracts	Yield of ripe extracts (gm)	
	Sohar	Dofar
Hexane	0.28	0.35
Ethyl acetate	1.22	4.82
Chloroform	0.19	0.13
Butanol	0.97	0.11
Methanol	38.7	13.91
Water	0.82	3.85

The values are means  $\pm$  SD of three replicates.

fat at normal temperature for 5 days. The dried samples were made into coarse powdered.

### 3.1. Preparation of banana extracts

The banana extract was obtained from the powdered ripe banana with methanol by using a maceration method for five days. The extract was fractioned and separated individually by various solvents to give corresponding hexane, chloroform, ethyl acetate, butanol, methanol and water extract (Table 1).

### 3.2. Antimicrobial potential

The antimicrobial potential of six extracts of both ripe banana was determined by disc diffusion method with modification against clinically isolated (+ and -) bacterial strains [18,19]. The antimicrobial potential of six banana extracts was determined through the disc diffusion method against *S. aureus* (Code no. 207) and *S. pneumoniae* (Code no. 257) and negative *E. coli* (Code no. 337) and *H. influenzae* (Code no. 236) culture bacterial strains. Four different concentrations of each banana extract of both ripe bananas were used to determine the antimicrobial potential. Most of the extracts from both ripe bananas showed potential antimicrobial activity against the applied microbes and the results were presented in Table 2.

### 3.3. Cytotoxic potential

Four concentrations of each polarities extract from the Sohar and Dhofar cities showed significant cytotoxic potential against the brine shrimp larvae [20]. The percentage of mortality (%) was found in different polarities extracts of Sohar and Dhofar's ripe banana extracts shown in Table 3.

## 4. Discussion

Globally, people are using various traditional medicines as primary health care therapy to treat different ailments. The Omani people are also used all those various therapies to treat diseases. But, there is no scientific evidence of this used therapy to treat diseases. Recently, scientists/researchers are working on those traditionally available therapies to find out the biologically active ingredients including antimicrobial and anticancer agents through the plants including Omani medicinal plants. The banana is one of the major economical agricultural crops which is available worldwide. It has unlimited medicinal benefits. In Oman, people are using banana to treat digestion, constipation, and diarrhea. For this medical ground, we are intended to design our present study to evaluate the antimicrobial and cytotoxic potential of different types of locally grown ripe bananas. The

antimicrobial potential of the prepared crude extracts from both ripe bananas was determined against the locally available clinically isolated Gram (+ and -) bacterial strains which were collected from a local hospital. In our experimental results showed that both ripe banana extracts have moderate antimicrobial potential against all culture bacterial strains at all applied concentrations within the range of 0–13 mm. The chloroform ripe banana extract showed the highest potential against *E. coli* at the concentration of 2 mg/ml which was collected from Sohar. The other polarities banana extracts were collected from Sohar and Dhofar showed the medium potential against *E. coli* at all applied concentrations. However, water banana extract from Dhofar did not show any potential against *H. influenzae* and *S. aureus* of any applied concentrations. The chloroform extract from Sohar also did not show any potential at any prepared concentrations against *S. aureus* (Code 207). Similarly, the highest potential was obtained in the chloroform extract which was collected from Sohar against *E. coli* (Code 337) and the lowest in the hexane banana extract against *S. aureus*. Most of the ripe banana extracts collected from both places gave the moderate potential against *E. coli* at all prepared concentrations. However, almost of the prepared extract at various concentrations gave the minimum potential against the applied *H. influenzae*, *S. aureus* and *S. pneumoniae* strains (Table 2). All banana extracts collected from Sohar gave the lower potential against *S. aureus* compared to the Dhofar banana extracts. The butanol, methanol and water banana crude extract from Sohar did not give any potential against *H. influenzae* (Table 2). Due to lack of reference data, we are unable to compare our results. The antimicrobial potential of the banana extracts depends on various factors such as chemical compounds, doses of extracts and the sensitivity of the specific bacterial strains. The experimental results also showed that all banana extracts gave minimum potential that means all extracts from both bananas contains less concentration of bioactive compounds which is responsible for antimicrobial potential. Previous studies related to the antimicrobial potential of banana extract from other countries and different varieties showed that the extracts from the banana are active against some the Gram (+ and -) bacterial strains [6,21,22,15]. Our experimental results also showed that all bananas crude extracts are not significantly active against the applied bacterial strains. On the other hand, each banana extract was killed all shrimp larvae (percent mortality 100%) at the highest concentration of 500 ug/ml. The percent mortality (%) for all extracts of ripe banana at all concentrations was given in Table 3. The highest cytotoxic activities were shown in the butanol extract from Sohar and the water extract from Dhofar banana. However, the lowest potential was in the ethyl acetate of both ripe bananas. Moreover, there was an increase in the mean percentage of mortality with the increase in concentrations of the extract. In our antimicrobial and cytotoxic potential results showed low potential its also depend on the chemical compounds in the extract and the dose applied. These results are contrary to what has been reported for the cytotoxic potential of bananas extract collected from elsewhere [23,24,12]. This variation of cytotoxicity value could be due to the methodologies of sample collection, extraction and preparation of samples; however, the same study used the same BST assay by other investigations.

## 5. Conclusion

The antimicrobial and cytotoxic potential of Sohar and Dhofar ripe banana were evaluated by disc diffusion and BSL biassays. Both ripe banana samples collected from both cities showed significant antimicrobial potential against the selected isolated culture Gram (+ and -) bacteria. In addition, the prepared ripe banana extracts also showed good cytotoxic potential against brine shrimp method. The highest potential ripe banana extracts might be used as a natural safe medicine

**Table 2**  
Antimicrobial potential of different crude extracts from Sohar and Dhofar ripe banana samples.

Bacteria	Extract Conc. (mg/ml)	Hexane		Chloroform		Ethyl acetate		Butanol		Methanol		Water	
		Sohar	Dofar	Sohar	Dofar	Sohar	Dofar	Sohar	Dofar	Sohar	Dofar	Sohar	Dofar
<i>E. coli</i> (Code no. 337)	2	11 ± 0.12	10.5 ± 0.56	13 ± 0.14	9 ± 0.42	9 ± 0.12	7.5 ± 0.55	7 ± 0.15	9 ± 0.20	7 ± 0.12	8 ± 0.17	7 ± 0.13	7.5 ± 0.07
	1	11 ± 0.43	10 ± 0.23	8 ± 0.11	8.5 ± 0.33	8 ± 0.10	7 ± 0.18	7 ± 0.43	8.5 ± 0.07	8 ± 0.23	7.5 ± 0.10	6 ± 0.90	7 ± 0.21
	0.5	9.5 ± 0.89	7 ± 0.78	8 ± 0.09	6.5 ± 0.41	7.75 ± 0.11	6.5 ± 0.23	7 ± 0.55	8 ± 0.65	7.15 ± 0.15	6.5 ± 0.15	7 ± 0.10	6.5 ± 0.57
Control	0.25	9 ± 0.15	6.5 ± 0.23	7 ± 0.16	6 ± 0.10	9 ± 0.07	6 ± 0.32	7 ± 0.08	7 ± 0.14	7 ± 0.10	nd	6 ± 0.19	6 ± 0.16
	3	12 ± 0.32	8 ± 0.47	30 ± 0.10	25 ± 0.16	11 ± 0.23	29 ± 0.21	10 ± 0.25	9 ± 0.18	7 ± 0.10	nd	9 ± 0.12	22 ± 0.13
	2	7 ± 0.76	8 ± 0.28	8 ± 0.19	7.5 ± 0.19	8 ± 0.15	7.5 ± 0.10	8 ± 0.18	7.5 ± 0.15	8 ± 0.32	8.5 ± 0.09	9 ± 0.15	nd
<i>H. influenza</i> (Code no. 236)	1	6 ± 0.19	7.5 ± 0.26	6 ± 0.22	7 ± 0.31	8 ± 0.78	7 ± 0.19	8 ± 0.42	7.15 ± 0.11	7 ± 0.42	8.15 ± 0.54	9 ± 0.08	nd
	0.5	nd	7 ± 0.55	8 ± 0.41	6.5 ± 0.64	7.5 ± 0.52	6.5 ± 0.11	7 ± 0.08	7.15 ± 0.17	7 ± 0.12	8 ± 0.22	6 ± 0.10	nd
	0.25	nd	7 ± 0.16	8 ± 0.23	6.5 ± 0.55	7 ± 0.10	6.5 ± 0.45	6 ± 0.09	7 ± 0.19	nd	nd	6 ± 0.54	nd
Control	3	33 ± 0.18	26 ± 0.21	28 ± 0.32	27 ± 0.14	27 ± 0.10	27 ± 0.56	28 ± 0.23	27 ± 0.09	29 ± 0.15	33 ± 0.32	30 ± 0.87	25 ± 0.23
	2	6 ± 0.90	8 ± 0.12	nd	9 ± 0.17	6 ± 0.55	10 ± 0.23	8 ± 0.27	nd	8 ± 0.10	10 ± 0.10	8 ± 0.10	nd
	1	0 ± 0.39	7 ± 0.82	nd	8.5 ± 0.29	nd	9 ± 0.91	6 ± 0.14	nd	6 ± 0.35	8 ± 0.15	7 ± 0.10	nd
<i>S. aureus</i> (Code no. 207)	0.5	0 ± 0.55	7 ± 0.29	nd	6.5 ± 0.18	nd	9 ± 0.12	6 ± 0.17	nd	6 ± 0.34	7.5 ± 0.19	nd	nd
	0.25	0 ± 0.72	6 ± 0.65	nd	6 ± 0.15	nd	9 ± 0.16	6 ± 0.19	nd	6 ± 0.18	nd	nd	nd
	3	24 ± 0.15	31 ± 0.34	26 ± 0.17	29 ± 0.10	29 ± 0.23	29 ± 0.10	27 ± 0.10	25 ± 0.89	27 ± 0.13	nd	29 ± 0.16	29 ± 0.13
<i>S. pneumoniae</i> (Code no. 257)	2	8 ± 0.82	7.5 ± 0.41	7 ± 0.34	7.5 ± 0.06	7 ± 0.11	8 ± 0.22	nd	7 ± 0.10	8 ± 0.21	7 ± 0.11	nd	7 ± 0.09
	1	7 ± 0.24	7 ± 0.23	7 ± 0.14	7 ± 0.18	6 ± 0.18	6.5 ± 0.10	nd	6.75 ± 0.22	7 ± 0.19	7 ± 0.10	nd	6.5 ± 0.10
	0.5	6 ± 0.12	6.5 ± 0.24	6 ± 0.10	6 ± 0.52	6 ± 0.25	6 ± 0.09	nd	6.5 ± 0.37	7 ± 0.10	nd	nd	nd
Control	0.25	0 ± 0.12	6.5 ± 0.27	nd	6 ± 0.21	6 ± 0.72	0 ± 0.65	nd	6 ± 0.13	nd	nd	nd	nd
	3	31 ± 0.12	27 ± 0.56	27 ± 0.18	34 ± 0.32	33 ± 0.12	40 ± 0.15	31 ± 0.22	34 ± 0.17	26 ± 0.12	30 ± 0.17	30 ± 0.12	26 ± 0.19

nd = not detected; Each value is a mean of three biological replicates.

**Table 3**  
Percentage of mortality and lethal concentration (IC<sub>50</sub>) of different polarities Sohar and Dofar ripe banana samples.

Crude extract	Conc µg/ml	Mortality (%)		LC <sub>50</sub> (µg/ml)	
		Sohar	Dhofar	Sohar	Dhofar
Hexane	500	100	100		
	100	80	80	22.39 ± 011	27.88 ± 017
	50	60	50		
	10	30	10		
	Control	0	0		
Chloroform	500	100	100		
	100	70	80	29.74 ± 0.14	29.16 ± 0.10
	50	50	40		
	10	20	20		
	Control	0	0		
Ethyl acetate	500	100	100		
	100	60	50	36.68 ± 0.22	49.32 ± 0.16
	50	50	40		
	10	10	10		
	Control	0	0		
Butanol	500	100	100		
	100	90	80	20.12 ± 0.10	26.19 ± 0.57
	50	60	50		
	10	30	20		
	Control	0	0		
Methanol	500	100	100		
	100	80	70	29.15 ± 0.18	27.86 ± 0.23
	50	40	50		
	10	20	30		
	Control	0	0		
Water	500	100	100		
	100	80	50	27.94 ± 0.25	24.22 ± 0.18
	50	50	40		
	10	10	20		
	Control	0	0		

Each value is a mean of three biological replicates.

to treat different ailments. Our future studies will be designed for the isolation and structure elucidation for active compounds and their mechanism of action.

### Declaration of Competing Interest

None.

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