



RESEARCH PAPER

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Antimicrobial activity of *Garcinia mangostana* using different solvents extracts

Ramesa Shafi Bhat*, Sood Al-Daihan

Biochemistry Department, College of Science, King Saud University, P. O. Box 22452, Riyadh 11495, Saudi Arabia

Key words: *Garcinia mangostana*, antibacterial activity, crude extract, disc diffusion method.

doi: <http://dx.doi.org/10.12692/ijb/3.10.267-272> Article published on October 21, 2013

Abstract

The present study investigates antibacterial activity of *Garcinia mangostana* in different solvents by disc diffusion method. Seven different extracts were prepared, using different solvents viz., methanol, ethanol, acetone, chloroform, ethyl ether, petroleum spirit and water. The result of antibacterial screening showed that all the extracts show moderate inhibition against pathogenic bacteria, both gram positive including *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Bacillus subtilis* and gram negative bacteria including *Escherichia coli* and *Pseudomonas aeruginosa*. Overall analysis of the antibacterial activity of tested samples revealed that the highest inhibitory activity was produced by ethanol extract (31 ± 0.37 mm) against *S. pyogenes* while minimal activity was found from ethyl ether extract (12 ± 0.00) against *B. subtilis*. Minimum inhibitory concentration (MIC) for *S. pyogenes* was 10 mg/ml for methanol extract.

*Corresponding Author: Ramesa Shafi Bhat ✉ ramesa.aftab@gmail.com

Introduction

Plants are used medicinally in different countries and are a source of many potent and powerful drugs. The traditional medicine is used in all parts of the world and has rapidly growing economic importance (Dash *et al.*, 2005; Agra *et al.*, 2007; Ushimaru *et al.*, 2007). The World Health Organisation (WHO) estimated that 80% of the population of developing countries still relies on traditional medicines, mostly plant drugs, for their primary health care needs (Gurib-Fakim, 2006). Globally researchers are using extracts of plants for their antibacterial, antifungal and antiviral activities (Bakht *et al.*, 2011 a, b, c and d).

G. mangostana a tropical fruit cultivated in the tropical rainforest of Southeast Asia has been used as a medicine for a great variety of medical conditions for hundreds of years. (Obolskiy *et al.*, 2009). The fruits of *G. mangostana* also named as 'queen of tropical fruits' is dark purple or reddish, with white, soft and juicy edible pulp with a slightly acid and sweet flavor and a pleasant aroma (Jung *et al.*, 2006). It has long been reported to contain multiple health promoting properties and is used to treat infections, reduce pain or control fever, and treat various and skin ailments, such as eczema and pruritus. (Akao *et al.*, 2008; Chin *et al.*, 2008; Gopalakrishnan *et al.*, 1997; Suksamrarn *et al.*, 2006 ; Chomnawang *et al.*, 2006; Ji X *et al.* ,2007; Yu L *et al.*, 2007). *G. mangostana* has long been reported to contain multiple health promoting properties. It contains high amounts of xanthenes, a class of polyphenolic compounds which were shown to have significant biological activities in vitro systems. (Ji X *et al.*, 2007; Mahabusarakam *et al.* 1987; Nilar *et al.*, 2002; Nilar *et al.*, 2005; Fu *et al.* ,2007) Xanthenes have antioxidant, antibacterial, antifungal, anti-inflammatory, antitumor, antiplatelet aggregation, antithrombotic, and vasorelaxant activities, prevent oxidative damage of low-density lipoprotein, histamine, and serotonin receptor blocker activity, and inhibit HIV (Ji X *et al.*, 2007). Although antibacterial activity of *Garcinia mangostana* have been reported (Dharmaratne *et al.* , 2013; Pedraza *et al.* , 2008 ;Suksamrarn *et al.* , 2006) but

antibacterial activity of its fruit extracted with different organic solvents in the increasing polarity order been found lacking in literature. Therefore the present study was undertaken taken to investigate the antimicrobial activities of different extracts from *G. mangostana* against some bacterial strains (both gram positive including *S. aureus*, *S. pyogenes*, and *B. subtilis* and gram negative bacteria including *E. coli* and *P.aeruginosa*).

Material and methods

Collection of plant material

Commercially available fresh fruit of *G. mangostana* free from disease were collected from Riyadh. The aril (i.e. the white part of the *G. mangostana* fruit) was removed. Collected material was washed thoroughly in running tap water, rinsed in distilled water and shade dried for one week in open air, crushed using mortar and pestle, reduced to powder using waring laboratory blender (MX-7011G) for 5 min at high speed and then stored in airtight closed bottles for two days before used for analysis.

Microorganisms

Bacteria cultures of *S. aureus*, *S. pyogenes*, *E. coli*, *B.subtillis*, and *P. aeruginosa*. (Clinical isolates) were obtained from Botany Department of King Saud University. The strains were maintained on agar slant at 4°C and activated at 37°C for 24 h on nutrient agar (Sigma-Aldrich, Germany) before any susceptibility test.

Extraction of plant material

Aqueous extraction

Ten gram of dry powder of samples was dissolved in 30 ml of 0.01 M HCl containing 0.15 M NaCl. (Ratio of sample/extract solution of 1:3 w/v). The residue was then removed by filtering through cheese cloth; the filtrate was then centrifuged at 8,100 x g, for 5 min. (Franco, 2006).

Other solvent extraction

Powdered sample of *G. mangostana* was successively extracted with different organic solvents in the increasing polarity order. Ten grams of powdered was

dissolved in 100 ml of solvent (Methanol, Ethanol, Acetone, Chloroform, Ethyl ether, Petroleum spirit) in a conical flask, plugged with cotton wool and then kept on a rotary shaker at 190–220 rpm for 24 h. The supernatant was collected slowly and evaporated in wide mouthed evaporating bowls at room temperature for 2–3 days till the final volume was reduced to one fourth of the original volume of the solvent used giving the concentration of 400 mg/ml. (Harborne,1973) and stored at 4 °C in airtight bottles.

Media preparation

Twenty three grams of nutrient agar (Sigma–Aldrich, Germany) was dissolved in 1000 ml of distilled water and bring to boil. Agar was then autoclaved for 15 min at 121° C and left to cool at room temperature. Once the LB medium was cooled (about 45 ° C), it was poured into Petri dishes. Each Petri dish was left on the flat surface for 30–40 min until completely set.

Table 1. Antibacterial activity of various extracts of test samples against bacterial species tested by disc diffusion assay.

Plant extract	<i>S. Aureus</i>	<i>S.Pyogenes</i>	<i>B.subtillis,</i>	<i>E. coli</i>	<i>P.Aeruginosa.</i>
Aqueous	13±0.60	12±0.66	13±0.00	18±0.15	16±0.00
Methanol	15±0.50	18±0.54	20±0.45	25±0.33	20±0.55
Ethanol	17±0.10	31±0.37	30±0.80	25±0.56	20±0.33
Acetone	30±0.25	15±0.23	17±0.65	20±0.00	20±0.11
Chloroform	14±0.60	17±0.00	15±0.44	20±0.70	18±0.10
Ethyl ether	13±0.80	14±0.70	12±0.00	15±0.31	14±0.00
Petroleum spirit	15±0.15	20±0.00	24±0.12	18±0.21	15±0.00
Kanamycin(30µg/disc)	26.5±0.33	28±0.57	21±0.15	20±0.33	25±0.10

Table 2. The MIC of ethanol extract of *Garcinia mangostana* against *S. pyogenes*.

Extract	<i>S. pyogenes.</i>				
Con. (mg/ml)	0	5	10	15	20
<i>G.mangostana</i>	-	-	+	+	+

Antibacterial activity

Antibacterial activity was assayed by disc diffusion method. For all bacteria strains, overnight culture grown in broth was adjusted to an inoculum's density of 100 µl: 0.1A600 culture containing $3.2 \cdot 10^8$ colony forming unit. Further, 20 µl was spread onto 20 ml of sterile agar plates by using a sterile cotton swab. The surface of the medium was allowed to dry for about 3 min. Sterile filter paper discs (5 mm in diameter) impregnated with different test extracts (100 µl disc) were then placed on the surface of inoculated agar plates. Kanamycin (30 µg/disc) was used as positive control. The plates were then incubated at 37°C for 24 h after which microbial growth was determined by measuring the diameter of the inhibition zone (mm) using a transparent scale. Each extract was analyzed in triplicate, the mean values are presented. Kanamycin disc (30 µg/disc) was used for comparing the bioassay.

Minimum inhibitory concentration

The Minimum Inhibitory Concentration (MIC) was determined by using serial dilution technique (Anu *et al.*, 2011) in this technique a large number of test tubes were used and each of the test tubes was filled with 1 ml of sterile nutrient broth media and graded doses of sample solution were added. Then these test tubes were inoculated with the selected organisms (inoculum contains 1×10^6 cells/ml) followed by incubation at 37°C for 24 hours to allow the growth of the bacteria. The test tubes which showed minimum concentration as well as clear content were selected. This lowest or minimum concentration was considered as Minimum Inhibitory Concentration (MIC).

Statistical analysis

The results were analyzed by using standard deviation (SD) statistical methods (Bhat, Al-daihan, 2013).

Results and discussion

Antibacterial activity

The antibacterial activity of seven extract of *G. mangostana* was assayed in vitro by agar disc diffusion method against 5 bacterial species. Table 3 summarizes the microbial growth inhibition of all extracts. The significant antibacterial activity of the active plant extracts was comparable to Kanamycin disc (30 µg/disc). In this study, the plant materials were sequentially extracted with different organic solvents in increasing polarity order. Among the tested extracts, acetone and ethanol extracts exhibited higher inhibition against all test bacteria compared to ethyl ether and petroleum spirit extracts of plant. Furthermore, the results produced by the aqueous extract were found to show minimal inhibition against all test bacteria.

Antimicrobial activities of plant extracts dependent on the solvent used for the extraction (Valgas, 2007.) Against all the tested bacterial strain, we observe organic extract showing much better antibacterial activities in contrast to aqueous extract, which may be because of high capacity of organic solvent to dissolve more organic and active antimicrobial compounds (Cowan,1999). The antimicrobial action of the aqueous extracts could be ascribed to the anionic components such as thiocyanate, nitrate, chlorides and sulfates besides other water soluble components which are naturally occurring in the plant material (Darout *et al.*, 2000).

Minimum inhibitory concentration

S. pyogenes was found most sensitive among all the strain therefore MIC was determined only against *S. pyogenes*. To determine MIC different concentration of methanol extracts were used against selected bacteria. Results are presented in table II.

Conclusion

G. mangostana possess significant antibacterial activity. Further research is needed for the isolation and identification of active principles present in the extracts which could possibly be exploited for pharmaceutical use.

Acknowledgement

This research project was supported by a grant from the “Research Center of the Center for Female Scientific and Medical Colleges”, Deanship of Scientific Research, King Saud University.

References

Agra MF, Freitas PF, Barbosa-Filh JM. 2007. Synopsis of the plants known as medicinal and poisonous in Northwest of Brazil. *Revista Brasileira de Farmacognosia* **17**, 114-140.

Akao Y, Nakagawa Y, Iinuma M, Nozawa Y. 2008. Anti-cancer effects of xanthenes from pericarps of mangosteen. *International Journal of Molecular Sciences* **9**, 355-370.

Bhat RS, Al-daihan S. 2013. Antimicrobial activity of Litchi chinensis and Nephelium lappaceum aqueous seed extracts against some pathogenic bacterial strains. *Journal of King Saud University – Science*.

<http://dx.doi.org/10.1016/j.jksus.2013.05.007>

Bakht J, Tayyab M, Ali H, Islam A, Shafi M . 2011a. Effect of different solvent extracted samples of *Allium sativum* on bacteria and fungi. *African Journal of Biotechnology* **10**, 5910-5915.

<http://dx.doi.org/10.5897/AJB11.232>

Bakht J, Islam A, Tayyub M, Ali H, Shafi M. 2011b. Antimicrobial potentials of *Eclipta alba* by disc diffusion method. *African Journal of Biotechnology* **10**, 7658-7667.

<http://dx.doi.org/10.5897/AJB11.454>

Bakht J, Ali H, Khan MA, Khan A, Saeed M, Shafi M, Islam A, Tayyab M. 2011 c. Antimicrobial activities of different solvents extracted samples of *Linum usitatissimum* by disc diffusion. *African Journal of Biotechnology* **10**, 19825-19835.

<http://dx.doi.org/10.5897/AJB11.229>

- Bakht J, Islam A, Shafi M.** 2011d. Antimicrobial potential of *Eclipta alba* by well diffusion method. *Pakistan Journal of Botany* **43**, 161-166.
- Chin YW, Jung HA, Chai H, Keller WJ, Kinghorn AD.** 2008. Xanthonones with quinone reductase-inducing activity from the fruits of *Garcinia mangostana* (mangosteen). *Phytochemistry* **69(3)**, 754-758.
<http://dx.doi.org/10.1016/j.phytochem.2007.09.023>
- Chomnawang MT, Surassmo S, Nukoolkarn VS, Gritsanapan W.** 2007. Effect of *Garcinia mangostana* on inflammation caused by *Propionibacterium acnes*. *Fitoterapia* **78(6)**, 401-408.
<http://dx.doi.org/10.1016/j.fitote.2007.02.019>
- Cowan MM.** 1999. Plant products as antimicrobial agents *Clinical Microbiology Reviews*, **12** .564–582.
- Darout IA, Christy AA, Skaug N, Egeberg PK.** 2000. Identification and quantification of some potentially antimicrobial anionic components in Miswak extract *Indian Journal of Pharmacology* **32** , 11–14
- Dash S, Nath LK, Bhise S, Bhuyan N,** 2005. Antioxidant and antimicrobial activities of *Heracleum nepalense* D Don root. *Tropical Journal of Pharmaceutical Research* **4**, 341-347.
- Dharmaratne HRW, Sakagami Y, Piyasena KGP, Thevanesam V.** 2013 Antibacterial activity of xanthonones from *Garcinia mangostana* (L.) and their structure–activity relationship studies. *Natural Product Research: Formerly Natural Product Letters* **27**, 10 , 938-941.
<http://dx.doi.org/10.1080/14786419.2012.678348>
- Franco OL, Murad AM, Jos LR, Mendes PAM, Prates MV, Jr CB,** 2006. Identification of a cowpea g-thionin with bacteria activity. *FEBS Journal* **273**, 3489- 3497.
<http://dx.doi.org/10.1111/j.1742-4658.2006.05349.x>
- Fu C, Loo AE, Chia FP, Huang D.** 2007. Oligomeric proanthocyanidins from mangosteen pericarps. *Journal of Agriculture and Food Chemistry* **55(19)**, 7689-7694
<http://dx.doi.org/10.1021/jfo71166n>
- Gopalakrishnan G, Banumathi B, Suresh G.** 1997. Evaluation of the antifungal activity of natural xanthonones from *Garcinia mangostana* and their synthetic derivatives. *Journal of Natural Products* **60(5)**, 519-524
- Gurib-Fakim A.** 2006, Medicinal plants: Traditions of yesterday and drugs of tomorrow. *Molecular Aspects of Medicine* **27**, 1-93.
- Harbone JB,** 1973. *Phytochemical Methods*. London: Chapman and Hill. 49-188.
- Ji X, Avula B, Khan IA.** 2007 Quantitative and qualitative determination of six xanthonones in *Garcinia mangostana* L. by LC-PDA and LC-ESI-MS. *J Pharm Biomed Anal* **43(4)**, 1270-1276.
- Jung HA, Su BN, Keller WJ, Mehta RG, Kinghorn D,** 2006. Antioxidant Xanthonones from pericarp of *Garcinia mangostana* (Mangosteen). *Journal of Agriculture and Food Chemistry* **54**, 2077–2082.
- Mahabusarakam W, Wiriyachitra P, Taylor WC.** 1987. Chemical constituents of *Garcinia mangostana*. *Journal of Natural Products* **50(3)**, 474-478.
- Nilar, Harrison LJ.** 2002 Xanthonones from the heartwood of *Garcinia mangostana*. *Phytochemistry* **60(5)**, 541-548.
- Nilar, Nguyen LH, Venkatraman G, Sim KY, Harrison LJ.** 2005. Xanthonones and benzophenones from *Garcinia griffithii* and *Garcinia mangostana*. *Phytochemistry* **66(14)**, 1718-1723.

- Obolskiy D, Pischel I, Siritwatanametanon N, Heinrich M.** 2009. *Garcinia mangostana* L.: a phytochemical and pharmacological review. *Phytotherapy Research* **23**, 1047–1065.
<http://dx.doi.org/10.1002/ptr.2730>
- Pedraza CJ, Cardenas RN, Orozco IM, Perez RJM.** 2008. Medicinal properties of mangosteen (*Garcinia mangostana*). *Food Chemistry and Toxicology* **46**, 3227-3239.
- Suksamrarn S, Suwannapoch N, Phakhodee W, et al.** 2003 Antimycobacterial activity of prenylated xanthenes from the fruits of *Garcinia mangostana*. *Chemical & Pharmaceutical Bulletin (Tokyo)* **51(7)**, 857-859.
- Suksamrarn S, Komutiban O, Ratananukul P, Chimnoi N, Lartpornmatulee N, Suksamrarn A,** 2006. Cytotoxic prenylated xanthenes from the young fruit of *Garcinia mangostana*. *Chemical & Pharmaceutical Bulletin* **54**, 301-305.
- Ushimaru PI, Mariama TN, Luiz C, Luciano B Di Ary FJ.** 2007. Antibacterial activity of medicinal plant extract. *Brazilian Journal of Microbiology* **38**, 717-719.
- Valgas C, Machado de Souza S, Smânia EFA, Artur Smânia Jr.** 2007. Screening methods to determine antibacterial activity of natural products. *Brazilian Journal of Microbiology* **38**, 369–380.
- Yu L, Zhao M, Yang B, Zhao Q, Jiang Y.** 2007. Phenolics from hull of *Garcinia mangostana* fruit and their antioxidant activities. *Food Chemistry* **104(1)**, 176-181.
<http://dx.doi.org/10.1016/j.foodchem.2006.11.018>