

Article

## Evaluation of Antibacterial Activity of the Mucilage of *Dioscorea esculenta* (Lour.) Burkill

A. Thajunnisha Begum<sup>1</sup>, S. Anbazhakan<sup>2,\*</sup>

<sup>1</sup> Department of Botany, A.V.C College (Autonomous), Mannampandal- 609 306, India

<sup>2</sup> PG and Research Department of Botany, Government Arts College, Tiruvannamalai-606 603, Tamil Nadu, India

\* Author to whom correspondence should be addressed; E-Mail: [sal1gac@yahoo.com](mailto:sal1gac@yahoo.com).

Article history: Received 21 August 2013, Received in revised form 26 September 2013, Accepted 30 September 2013, Published 8 October 2013.

**Abstract:** The present study was aimed to screen the antimicrobial activity of tuber mucilage extracted from *Dioscorea esculenta* by *in vitro*-well diffusion assay method against five bacterial strains, viz., *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pyogens*. The extracts showed antibacterial activity against *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* while no activity was showed by this extract against *Klebsiella pneumonia* and *Streptococcus pyogens*, where as maximum inhibition concentration (MIC) was observed in 50 µg concentration of the extract against *E.coli*. By this study, it was concluded that tuber mucilage of *D. esculenta* has antibacterial activity against human pathogens.

**Keywords:** medicinal plant; *Dioscorea esculenta*; tuber mucilage; antibacterial activity.

### 1. Introduction

Plants are integral part of human civilization. Medicinal plants are also been relied upon by over 80% of the world population for their basic health care needs. Despite of tremendous progress in

human medicines, infectious diseases caused by bacteria, fungi, viruses and parasites are still a major threat to public health. The impact is particularly large in developing countries due to relative unavailability of medicines and the emergence of widespread drug resistance (Zampini *et al.*, 2009). During the last two decades, the development of drug resistance as well as the appearance of undesirable side effects of certain antibiotics (Okemo *et al.*, 2003) has led to the search of new antimicrobial agents mainly among plant extracts with the goal to discover new chemical structures, which overcome the above disadvantages (Bouamama *et al.*, 2006). Current research on natural molecule and products primarily focuses on plants since they can be sourced more easily and be selected based on their ethno-medicinal uses (Arora and Kaur, 2007). Mucilage is widely used in pharmaceutical industries as thickeners, water retention agents, and emulsion stabilizers, suspending agents, binders and film formers. Apart from its use in finished medicines, newer uses have been found in the preparation of cosmetics, textiles and paint paper (Shirwaikar *et al.*, 2007). Hence the demand for these substances is increasing and new sources are getting tapped. India due to geographical and environmental positioning has traditionally been a good source for such products.

*Dioscorea esculenta* is species of yam belong to family of Dioscoreaceae known as Siruvallikilangu in tamil vernacular. It is an annual herb, which is cultivated widely in India. The high mucilage content of *Dioscorea esculenta* makes it an excellent demulcent that can be used for many applications. *Dioscorea esculenta* Nees in wall used as food and medicine by different ethnic groups of the world for the treatment of diseases have special significance from long time like swellings, inflammation etc. (Anonymous, 1981; Chopra *et al.*, 1956). Tuber mucilage can be used to heal and soothe inflammations and swellings. The present work is an attempt to investigate the antibacterial property for the mucilage against the selected bacterial strains.

## 2. Materials and Methods

### 2.1. Plant Material

The tuber of *Dioscorea esculenta* (yam) is the plant material for the present investigation and collected in the Vilamuthur village of Perambalur district, Tamil Nadu. Voucher specimen was deposited in the herbarium of the PG and Research Department of Botany, Government Arts College, Tiruvannamalai.

### 2.2. Extraction of Mucilage from Tuber

#### 2.2.1. Material preparation

The yam tuber was peeled, sliced (a thickness of about 5 mm), mixed with distilled water (3.5

times tuber volume), and then homogenized by using sterile pestle and mortar. The mixture was filtered through Whatman filter paper. The filtrate was mixed with distilled water (3.5 times cake volume), homogenized and filtered again as described above. The filtrates were combined and centrifuged at 10,000g at 4 °C for 20 min. The supernatant was used for isolation of the mucilage.

### 2.2.2. Mucilage extraction

The mucilage was prepared from the supernatant according to the method of Tsai and Tai (1984). The supernatant was gently agitated at 4 °C, to which 95% ethanol was simultaneously added at a rate of 4 mL/min for precipitating the mucilage. The mixture was then centrifuged at 10,000g at 4 °C for 20 min. Ethanol was added to the obtained supernatant for having precipitates. All the precipitates were collected and dissolved in distilled water, precipitated again by 95% ethanol and then centrifuged at 10,000g at 4 °C for 20 min. The dissolution of the precipitate and the centrifugation were repeated twice to possible impurities. The final precipitate was washed with acetone, and then dried at 100 mm Hg and 40 °C. It was also ground in sterile pestle and mortar to pass a 30-mesh (ASTM) sieve. The mucilage was stored in desiccators at 25 °C until use.

## 2.3. Antibacterial Studies

### 2.3.1. Media preparation

#### 2.3.1.1. Bacterial media (Muller Hinton Media)

The 36 g of Muller Hinton Media (Hi-Media) was mixed with distilled water and then sterilized in autoclave at 15 lb pressure for 15 min. The sterilized media were poured into petridishes. The solidified plates were pored with 5 mm diameter cork borer. The plates with wells were used for the antibacterial studies.

#### 2.3.1.2. Bacterial strains

The bacterial strains were obtained from the Microbial Type Culture Collection (MTCC), Institute of Microbial Technology, Chandigarh, India. Bacterial strains were *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pyogenes*.

### 2.3.2. Antibacterial activity of the plant extract

The mucilage obtained was used for present study. The mucilage was dissolved in sterile water to make the dilutions of 5, 10 and 25 µg. Each concentrations of the drug were tested against human bacterial pathogens by well diffusion method.

### 2.3.2.1. Well diffusion method

Antibacterial activity of the mucilage was tested using well diffusion method (Bauer *et al.*, 1996). The prepared culture plates were inoculated with different selected strains of bacteria using streak plate method. Wells were made on the agar surface with 5 mm cork borer. The extracts were poured into the well using sterile syringe. The plates were incubated at  $37\pm 2$  °C for 24 hours for bacterial activity. The plates were observed for the zone formation around the wells. Streptomycin was used as a positive control (Sokovic *et al.*, 2010). The zone of inhibition was calculated by measuring the diameter of the inhibition zone around the well (in mm) including the well diameter. The readings were taken in three different fixed directions in all 3 replicates and the average values were calculated.

## 3. Results and Discussion

The antibacterial activity of the mucilage extracted from the tuber of *D. esculenta*, on various bacterial strains was investigated and described with the minimum inhibitory concentrations (MIC). The results showed that tuber mucilage had an antibacterial effect against various bacterial strains (Table 1). Bacterial strains such as *E. coli*, *K. pneumonia*, *P. aeruginosa*, *S. aureus* and *S. pyogens* exist in humans are superficial contaminants that can cause a variety of serious infections. The maximum MIC is observed in 50 µg concentration of the tuber mucilage extract against *E. coli* while no antibacterial activity was observed with the bacterial strains *K. pneumonia* and *S. pyogens* by the all concentrations of mucilage extracts of *D. esculenta*. Commercial antibiotic streptomycin exhibited better antibacterial potential, with larger inhibition zones ranging from 11.00 to 26.00, 13.00-28.00 and 15.00-30.00 mm for 5 µg, 25 µg and 50 µg concentrations of the tuber extracts, respectively. Bacterial growth inhibition around the well is due to the release of diffusible inhibitory compounds from the tuber mucilage extracts because this plant has several medicinally important metabolites like diosgenin (De and De, 2005), dioscorin (Hou *et al.*, 1999), and antioxidants (Isamah *et al.*, 2000), steroids (Bruneton, 1995) and alkaloids. Some yams are used as medicines in oriental countries to prevent diabetes (Grindley *et al.*, 2002), thus yams are considered to be useful to human health and they also have nutritional superiority when compared with other tropical root crops. Several species of *Dioscorea* are amongst the principle sources of diosgenin, which can be converted to medicinally important steroids (Van Staden and Fowlds, 1992). These steroids are used as contraceptives and anti-inflammatory agents (Bruneton, 1995). Recently, Mulholland *et al.* (2002) reported the isolation of alkaloids of both the isoquinoline and isoquinuclidine types from *D. dregeana*. There was a report of two compounds with antimicrobial activity, 2,5 - dihydroxy - 4 - methoxy - 9,10 - dihydrophenanthrene and 7-hydroxy-2,4,6-trimethoxyphenanthrene, were isolated from the tuber of

*Dioscorea rotundata* (Ogundana *et al.*, 1984). These compounds may also cause the inhibition of bacterial growth in the present study in *Dioscorea esculenta*.

**Table 1.** Antibacterial activity of *Dioscorea esculenta* (Lour.) Burkill tuber mucilage against different human pathogens

S. No.	Bacteria	Zone of Inhibition (mm)					
		Mucilage (5 µg)	Streptomycin (5 µg)	Mucilage (25 µg)	Streptomycin (25 µg)	Mucilage (50 µg)	Streptomycin (25 µg)
1	<i>Escherichia coli</i>	17±0.06	26±2.10	21±2.00	28±0.95	25	30±1.00
2	<i>Klebsiella pneumonia</i>	-	11±0.90	-	13±0.94	-	15±0.90
3	<i>Pseudomonas aeruginosa</i>	7±0.30	15±1.20	9±0.65	19±0.18	11	20±0.82
4	<i>Staphylococcus aureus</i>	7±0.33	16±1.20	14±0.19	21±0.12	16	22±0.85
5	<i>Streptococcus pyogens</i>	-	13±1.25	-	14.5±0.70	-	16±0.75

Antibacterial activity was found in the methanolic extracts of the leaves and stems of *Cheilanthes viridis*, and *Dioscorea dregeana* tubers, both methanol and ethyl acetate extracts of *Dioscorea sylvatica* tuber bark, water and methanol extracts of leaves and stems of *Melianthus comosus*, and methanol and ethyl acetate extracts of leaves, stems and roots of *Vernonia colorata*. In general, these extracts were most active against Gram-positive bacteria, though tuber bark extract of *D. sylvatica* was active against *E. coli*, while extracts of *C. viridis*, *D. dregeana* and *V. colorata* were active against *P. aeruginosa*. Extracts of *Rothmannia capensis* and *V. colorata* had bacteriostatic activity against both *E. coli* and *K. pneumoniae*. These results were in line with those from previous screenings of medicinal plants for antimicrobial activity, where most of the active plants showed activity against Gram-positive strains only (Rabe and van Staden, 1997; Vlietinck *et al.*, 1995).

#### 4. Conclusions

The present study concluded that tuber mucilage extract of *D. esculenta* has antibacterial properties against three human bacterial strains such as *E. coli*, *P. aeruginosa* and *S. aureus*, while it has not inhibitory activity against *K. pneumonia* and *S. pyogens*. This study paves the way for further attention and research to identify the active compounds responsible for the plant biological activity in the *D. esculenta* tuber mucilage.

#### References

- Anonymous. (1981). *The Useful Plants of India*. Government of India, New Delhi.
- Arora, D. S., and Kaur, G. J. (2007). Antibacterial activity of some Indian medicinal plants. *J. Nat.*

*Med.*, **61**: 313-317.

- Bauer, A. W., Kirby, W. M. M., Sherris, S. C., and Turk, M. (1996). Antibiotic susceptibility of testing by a standard single disc method. *Amer. J. Clin. Pathol.*, **36**: 492-496.
- Bouamama, H., Noel, T., Villard, J., Benharref, A., and Jana, M. (2006). Antimicrobial activities of the leaf extract of two *Moroccan cistus* L species. *J. Ethnopharmacol.*, **104**: 104-107.
- Bruneton, J. (1995). *Pharmacognosy, Phytochemistry, Medicinal Plants*. Lavoisier Publishing, Paris, pp. 538-544.
- Chopra, R. N., Nayar, S. L., and Chopra, I. C. (1956). *Glossary of Indian Medicinal Plants*. CSIR, New Delhi.
- De, D., and De, B. (2005). Elicitation of diosgenin production of *Dioscorea floribunda* by theylene-generating agent. *Fitoterapia*, **76**: 153-156.
- Grindley, P. B. A., Omoruyi, F., Asemota, H. N., and Morrison, E. Y. A. (2002). Carbohydrate digestion and intestinal ATPases in streptozotocin induced diabetic rats fed extract of yam (*Dioscorea cayenensis*) or dasheen (*Colocasia esculenta*). *Nutr. Res.*, **22**: 333-341.
- Hou, W. C., Chen, H. J., and Lin, Y. H. (1999). Dioscorins, the major tuber storage proteins of yam (*Dioscorea batatus* Decne), with dehydroascorbate reductase and monodehydroascorbate reductase activities. *Plant Sci.*, **149**: 151-156.
- Isamah, G. K., Asagba, S. O., and Thomas A. E. (2000). Lipid peroxidation o-diphenolase, superoxide dismutase and catalase profile along the three physiological regions of *Dioscorea rotundata* Poir. *Cv. Omi. Food Chem.*, **69**: 1-4.
- Mulholland, D. A., Crouch, N., Decker, B., and Smith, M. T. (2002). The isolation of the Amaryllidaceae alkaloid crinamine from *Dioscorea dregeana* (Dioscoreaceae). *Biochem. System. Ecol.*, **30**: 183-185.
- Ogundana, S. K., Coxon, D. T., and Dennis, C. (1984). Natural antifungal compounds from the peel of yam tubers. In: Proceedings of the Sixth Symposium of the International Society for Tropical Root Crops. Lima, Peru; International Potato Centre, ARC Food Research Institute, Norwich, United Kingdom.
- Okemo, P. O., Bais, H. P., and Vivanco, J. M. (2003). In vitro activities of *Maesa lanceolata* extracts against fungal plant pathogens. *Fitoterapia*, **74**: 312-316.
- Rabe, T., and van Staden, J. (1997). Antibacterial activity of South African plants used for medicinal purposes. *J. Ethnopharmacol.*, **56**: 81-87.
- Shirwaikar, A. A., Prabu, L. S., Mahalaxmi, R., and Rajendran, K. (2007). Studies of disintegrant properties of seed mucilage of *Ocimum gratissimum*. *Ind. J. Pharma. Sci.*, **21**: 753-758.
- Sokovic, M., Glamoclija, J., Marin, P. D., Brkic, D., and Van-Griensven, L. J. (2010). Antibacterial

effects of the essential oils of commonly consumed medicinal herbs using an in vitro model. *Molecules*, **15**: 7532-7546.

Van Staden, J., and Fowlds, D. L. (1992). Micropropagation of medicinal *Dioscorea* species. In: *High-Tech and Micropropagation III. Biotechnology in Agriculture and Forestry*, Vol. 19, Bajaj, Y. P. S. (ed.), Springer-Verlag, Berlin, pp. 425-442.

Vlietinck, A. J., Van Hoof, L., Totte, J., Lasure, A., Vanden Berghe, D., Rwangabo, P. C., and Mvukiyumwami, J. (1995). Screening of a hundred Rwandese medicinal plants for anti-microbial and antiviral properties. *J. Ethnopharmacol.*, **46**: 31-47.

Zampini, I. C., Cuello, S., Alberto, M. R., Ordonez, R. M., Almeida, R. D., Solorzano, E., and Isla, M. I. (2009). Antimicrobial activity of selected plant species from the *Argentine puna* against sensitive and multiresistant bacteria. *J. Ethnopharmacol.*, **124**: 499-505.