International Research Journal of Microbiology (ISSN: 2141-5463)
Vol. 10(6) pp. 1-5, November, 2021
Available online @ https://www.interesjournals.org/microbiology.html

Research Article

Studies on Antimicrobial Activity of Anthocyanins Extracted from Red Sorghum (*Sorghum bicolor* .L) Bran

P.Suganyadevi[1]*, M Saravanakumar[2] and S. Mohandas[3]

[1]Department of Biochemistry, Karpagam Academy of Higher Education, Coimbatore, India.
[2]Department of Biochemistry, P.S.G College of Arts and Science, Coimbatore, India.
[3]Senior Scientists, Rasi Seeds, Salem, India.

E-mail: suganyadevibiotech@gmail.com

Abstract

Recently, natural products have been evaluated as sources of antimicrobial agents with efficacies against a variety of microorganisms. This study described the antibacterial activity of anthocyanins extracted from red *sorghum* bran on selected bacteria. The anthocyanins extracted by using acidified ethanol have shown highest antibacterial activity compared to methanol extracts. Among the selected bacterial cultures, the highest antibacterial activity was recorded against Staphylococcus aureus. Moderate antifungal activity was observed against *Aspergillus niger* and *Aspergillus fumigates.*

Keywords: Red *sorghum* bran, Antibacterial activity, Anthocyanins.

INTRODUCTION

Sorghum (*Sorghum bicolor* (L)) is the fifth leading cereal crop in the world and is used primarily in Asia and Africa as a food crop (Rooney, 2000). Speciality of *sorghum* have high levels of phytochemicals, including proanthocyanins (Awika; 2003, Hahn;1996), 3-deoxyanthocyanins(Gous; 1989, Awika; 2004a, Awika; 2004b), phenolic acids (Wansika; 1989), phytosterols (Singh; 2003) and policosanols (Huang; 2004) in their bran layers. In addition, *sorghum* bran is rich in dietary fiber (Rooney; 1992). This *sorghum* bran are potentially useful ingredients in various functional food applications and were shown to produce desirable attributes (e.g., attractive natural color) without adversely affecting other sensory properties of foods such as bread, cookies and expanded snacks (Acosta; 2003, Gordon; 2001, Mitre-Dieste; 2000).

Anthocyanins are belonging to water – soluble plant pigments and representatives of flavonoids. They are responsible for the blue, purple and red colour of many plant tissues. They play a definite role in the attraction of animals for pollination and seed dispersal, and hence they are of considerable value in the co-evolution of these plant-animal interactions (Han; 2006).

Antibiotic – resistant bacteria is still of world – wide concern. Since the use of antibiotics became wide spread over 50 years ago, bacteria have progressively developed resistance (Hsueh; 2005). Consequently, scientific efforts have been made to study and develop new compounds to be used beyond conventional antibiotic therapy.

The objective of the present study was to evaluate the antimicrobial activity of anthocyanins isolated from red *sorghum* bran.

MATERIALS AND METHODS

Samples

Sorghum bicolor was collected from the village area (Coimbatore district, India) and raised in the college campus under normal climatic conditions. The plant was identified and authenticated (No.BSI/SRC/5/23/2011-12/Tech.1486) by Botanical survey of India (BSI), Tamil Nadu Agricultural University (TNAU), Coimbatore, India. The bran was collected and was stored at -20^{o}C.

Anthocyanin extraction

The bran of red *Sorghum* were extracted by incubating with two solvent systems like methanol and with 1% Hydrochloric acid in methanol, overnight at room temperature, followed by a filtration through whatman filter paper no 4. Methanol was removed by a rotary evaporation under 35°C and the pigmented fraction extracts were stored for a further study.

Table 1: Antimicrobial influence of anthocyanin extracted from red *sorghum* bran using methanol solvent.

Extract	Microorganism	Concentration (mg/ml)	Zone of Inhibition (mm)	Streptomycin (1mg/ml)	Nystin (1mg/ml)
Anthocyanin extracted by using methanol solvent	*Staphylococcus aureus*	1.0	5 ± 0.01	1.70 ± 0.2	-
		2.5	6 ± 0.01		
		5.0	7 ± 0.02		
		7.5	8 ± 0.01		
		10.0	9 ± 0.01		
		12.5	9 ± 0.01		
	Klebsiella oxytoca	1.0	0	1.2 ± 0.1	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Escherichia coli	1.0	0	1.6 ± 0.2	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Klebsiella pneumonia	1.0	0	1.3 ± 0.2	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Pseudomonas aerginosa	1.0	0	1.7 ± 0.2	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Aspergillus niger	1.0	0		
		2.5	0		
		5.0	0		
		7.5	0	-	1.6 ± 0.02
		10.0	0.3 ± 0.01		
		12.5	0.4 ± 0.01		
	Aspergillus fumigates	1.0	0	-	1.86 ± 0.01
		2.5	0		
		5.0	0		
		7.5	0.5 ± 0.01		
		10.0	0.6 ± 0.01		
		12.5	0.8 ± 0.01		

Each value is the mean ± standard deviation of 3 replicate assays.

Microorganisms

Escherichia coli, Klebsiella oxytoca, Klebsiella pneumonia, Pseudomonas aeruginosa, *Staphylococcus aureus, Aspergillus niger and Aspergillus fumigatus* were the micro-organisms used and they were stored at freeze temperature until use.

Determination of antibacterial activity

An agar –well diffusion methods was employed for determination of antibacterial activity. The anthocyanin extracts were tested for microbial resistance using such technique with an inoculum volume equivalent to 0.5% Mc Farland's Standard in Mueller – Hinton agar and examined

Table 2: Antimicrobial influence of anthocyanin extracted from red *sorghum* bran using acidified methanol solvent.

Extract	Microorganism	Concentration (mg/ml)	Zone of Inhibition (mm)	Streptomycin (1mg/ml)	Nystin (1mg/ml)
Anthocyanin extracted by using acidified methanol solvent	*Staphylococcus aureus*	1.0	4 ± 0.01	1.70 ± 0.02	-
		2.5	5 ± 0.01		
		5.0	6 ± 0.01		
		7.5	8 ± 0.01		
		10.0	9 ± 0.02		
		12.5	10 ± 0.02		
	Klebsiella oxytoca	1.0	3 ± 0.01	1.2 ± 0.01	-
		2.5	3 ± 0.01		
		5.0	4 ± 0.01		
		7.5	5 ± 0.01		
		10.0	5 ± 0.01		
		12.5	6 ± 0.01		
	Escherichia coli	1.0	0	1.6 ± 0.02	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Klebsiella pneumonia	1.0	0	1.3 ± 0.02	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Pseudomonas aerginosa	1.0	0	1.7 ± 0.02	-
		2.5	0		
		5.0	0		
		7.5	0		
		10.0	0		
		12.5	0		
	Aspergillus niger	1.0	0	-	1.6 ± 0.02
		2.5	0		
		5.0	0		
		7.5	0.05 ± 0.01		
		10.0	0.06 ± 0.01		
		12.5	0.07 ± 0.01		
	Aspergillus fumigates	1.0	0	-	1.87 ± 0.02
		2.5	0		
		5.0	0		
		7.5	0.06 ± 0.01		
		10.0	0.07 ± 0.01		
		12.5	0.07 ± 0.01		

Each value is the mean ± standard deviation of 3 replicate assays.

after 24 hrs. The anthocyanin extracts containing 1 mg, 2.5 mg, 5 mg, 7.5 mg, 10 mg and 12.5 mg were dissolved in DMSO. Negative controls were prepared using DMSO solution. Streptomycin was used as positive reference standards to determine the sensitivity of each bacterial species tested and Nystin was used as reference standard for fungal cultures. The inoculated plates were incubated at 37°C for 24 hrs. Antibacterial activity was evaluated by measuring the inhibition zones formed on the medium were evaluated in mm of the tested bacterial cultures and for fungal culture it was incubated at room temperature for 48 hours. All the tests were performed in triplicates.

RESULTS AND DISCUSSION

Antibacterial activities of anthocyanin extracted by methanol solvent

One out of five bacteria used *Staphylococcus aureus* is Gram positive and four (*Escherichia coli*, *Klebsiella pneumoniae*, *Klebisella oxytoca*, *Pseudomonas aerginosa*) are Gram negative. There was significant variation in the antibacterial activities of anthocyanin extracts. In all four Gram negative bacterias the methanol extract of anthocyanin showed no inhibitory zone even at 12.5 mg concentrations. For *Staphylococcus aureus,* the zone of inhibition values of anthocyanin extracts were between 5-9 mm. The results of methanolic extracts on bacteria are presented in Table 1. The moderate antifungal activity was observed in *Aspergillus fumigates and Aspergillus niger.* (Table 1). The results observed in anthocyanin extracted from red *Sorghum* bran methanol extracts indicating that gram positive strain was more sensitive than gram negative. This observation can be attributed in the difference in the structure of bacterial cell wall. The less complex structure of the cell wall in the gram positive bacteria makes it more permeable to the antibacterial compounds (Chrissanthy *et al.* 2005).

Antibacterial activites of anthocaynin extracted by acidified methanol solvent

There was a significant variation in the antibacterial activities of anthocyanin extracted from acidified methanol. The acidified methanol extracts of anthocyanin samples showed inhibitory effect in gram positive and gram negative bacteria. The highest antibacterial activity was observed in *K. oxytoca* with zone of inhibition of 3- 6 mm with the concentration ranging from 1 to 12.5 mg. The moderate antifungal activity was observed in *Aspergillus fumigates and Aspergillus niger* (Table 2). The Suganya *et al.* 2011 reported the anthocyanin content was found to be higher in acidified methanol extract than methanol extract. Deividas *et al.* 2009 reported that phenolic compounds and anthocyanin content was higher in the extracts of skin and berry extracts of biliberry and blue berry showed highest inhibitory zone against gram negative bacteria than other extracts, which coincides with our results.

CONCLUSION

The present study confirms the potential antimicrobial activity of the extract of red *sorghum* bran. The presence of anthocyanin as major active constituents may be responsible for these activities. Further studies are necessary to isolate characterize the active constituents of the plant to evaluate their modes of action and render this species interesting for future.

ACKNOWLEDGMENTS

This work was supported by Department of science and technology, New Delhi, India (Grant No: 100/(IFD)/1398/2011-12) under Women Scientist Scheme.

REFERENCES

Rooney LW, Waniska RD (2000). *Sorghum* food and industrial utilization. In *Sorghum*: Orgin, History. Technology and Production, (1st ed); Smith, C.W., Frederiksen, R.A., Eds; John Wiley and sons: New York. 689-729.

Awika JM, Rooney LW, Wu X, Prior RL, Cisneroszevallos L (2003). Screening methods to measure antioxidant activity of *sorghum* (*Sorghum* bicolor) and *sorghum* products, J Agric Food chem. 51: 6657-6662.

Hahn DH, Rooney LW (1996). Effects of genotype on tannins and phenols of *sorghum*, cereal Chem. 63: 4-8.

Gous F (1989). Tannins and phenols in black. Ph.D. dissertation, Texas A&M University, College station, TX.

Awika JM, Rooney LW, Waniska RD (2004). Anthocyanins from black *sorghum* and their antioxidant properties, Food Chem. 90: 293-301.

Awika JM, Rooney LW, Waniska RD (2004). Properties of 3-deoxyanthocyanins from *sorghum*, J Agric Food Chem. 52: 4388-4394.

Wansika RD, Poe JH, Bandyopadhyay R (1989) Effects of growth conditions on grain molding and phenols in *sorghum* caryopsis, J Cereal Sci. 10: 217-225.

Singh V, Moreau RA, Hicks KB (2003). Yield and Phytosterol composition of oil extracted from grain *sorghum* and its wet-milled fractions, Cereal Chem. 80: 126-129.

Huang KT, Weller CL, Cuppett SL, Hanna MA (2004). Policosanol contents and composition of grain *sorghum* kernals and dried distillers grains, Cereal Chem. 81: 345-349.

Rooney TK, Ronney LW, Lupton JR (1992). Physiological characteristics of *sorghum* and millet brans in the rat model, Cereal Foods World. 37: 782-786.

Acosta D, Barron M, Riaz M, Waniska R, Rooney LW (2003). Factors affecting extrusion of white *sorghum* to produce whole gram snacks, AACC Annual Meeting, Portland. URL http;//WWW. aaccnet.org/meetings/2003/abstracts/a03ma305.htm.

Gordon LA (2001). Utilizationof *sorghum* brans and barely flour in bread, M.S. Thesis, Texas A&M University, College Station, TX.

Mitre-Dieste CM, Gordon LA, Awika J, Suhendro EL (2000). Cookies made with *sorghum* brans high in phenols and catechins, AACC 2000 Annual Meeting, Kansas City, MO. URL http;//WWW.aaccnet. org/meetings/2000//abstracts/a00ma203. htm.

Han KH, Sekikkawa KI, Shimada M, Hashimoto NH, Noda HT, Fukushima M (2006). Anthocyanin – rich purple potato flake extract has antioxiadant capacity and improves antioxidant potential in rats, British J Nutrition. 96: 1125-1133.

Hsueh P, Chen W, Luh K (2005). Relationships between antimicrobial use and antimicrobial resistance in Gram–Negative bacteria causing nosocomial infections from 1991–2003 at a university hospital in Taiwan, Int J Antimicrob Agents. 26: 463-472.

Deividas B, Antanas S, Ina J, Elicija S, Laurynas N, Valdimaras J (2009). Comparative study of anthocyanin composition, antimicrobial and antioxidant activity in bilberry and blue berry fruits, Actapoloniae Pharmaceutica Drug Res. 66: 399-408.

Chrissanthy P, Kalliopi S, Loannina A (2004). Potential antimicrobial activity of red and white wine phenolic extracts against strains of S.aureus, E.coli and Candida aldicans, Food Technol Biotechnol. 43: 41-46.