International Journal of Botany Studies
ISSN: 2455-541X; Impact Factor: RJIF 5.12
Received: 26-10-2020; Accepted: 11-11-2020: Published: 27-11-2020
www.botanyjournals.com
Volume 5; Issue 6; 2020; Page No. 274-276

Effect of plant derived catecholamines on growth of *Escherichia coli* and *Lactobacillus* sp.

Sandhya M[1], Jessy P[2]
[1] Department of Bioanalytical Sciences, Ramnarain Ruia Autonomous College, Matunga, Mumbai, Maharashtra, India
[2] Department of Botany, Ramnarain Ruia Autonomous College, Matunga, Mumbai, Maharashtra, India

Abstract
Biogenic amine comprises of molecules like dopamine, epinephrine and norepinephrine secreted by the adrenal medulla in response to stress and thereby directing the human body to respond to respond to the stress. Biogenic amines have been reported to occur in *Portulaca oleracea* and *Gomphrena globosa.* These catecholamines are known to induce growth in certain pathogenic species of *Escherichia coli*. The current study aims to study the effect of catecholamine present in *Portulaca oleracea* and *Gomphrena globosa* on the growth of *Escherichia coli* (wild type) and *Lactobacillus* sp.

Keywords: biogenic amine, catecholamines, *Gomphrena globosa, Portulaca oleracea, Escherichia coli, Lactobacillus* sp.

Introduction
Aromatic biogenic amines are basic nitrogenous compounds formed mainly by decarboxylation of amino acids or by amination and transamination of aldehydes and ketones. In human and animal cells, they play an important role as neurotransmitters, source of nitrogen and precursor for the synthesis of hormones, alkaloids, nucleic acids, proteins etc. Biogenic amines have been reported to occur in 44 plant families [5, 10, 7]. Though biogenic amines and their precursors as well as their derivatives have been reported to occur in plant families, no important metabolic role has been assigned to them [6]. Their role and action are partly understood. Many plant species in human diet contain biogenic amines and are reported to be active principle ingredients of many medicinal plants [7]. Catecholamine and precursors are reported in *Portulaca oleracea* and *Gomphrena globosa* [8].
Colonization of the intestine with micro-organisms plays an important role in stimulating the normal development of the immune system [4]. For the microorganisms to grow within the human body they need to adapt themselves to the biochemical environment and interact with the host in a number of complex ways. Quorum-sensing, signaling between bacterial cells for the purpose of communication, however is not restricted to bacterial cell-to-cell communication but also allows communication between microorganisms and their hosts [1]. Quorum sensing allows them to sense chemicals like hormones in the environment and thereby respond to these hormones [1].
Catecholamines are critical hormones secreted by the human body during stress like infection, adverse climatic conditions, physical stress etc. Host catecholamines released during stress can induce bacterial growth, enhance colonization to host tissue, and upregulation of virulence factors [2, 3].
The current study focuses on identifying whether bacteria can respond to plant catecholamines. The study works with catecholamines present in *Portulaca oleracea* and *Gomphrena globosa*. The crude extracts of these plants are checked for promoting growth of *Escherichia coli* and *Lactobacillus* sp.

Materials and methods
Materials
Extracts used: Inflorescence of *Gomphrena globosa*, flower of *Portulaca oleracea* were used for preparing extracts for the study. The plant material was collected from the garden of Ramnarain Ruia College, Matunga. Fresh Samples were used for preparing the extract. The two types of extracts were made: Acidic and methanolic.
Acidic extract was made by macerating 2gm of plant material in 1 ml of 0.1N HCl and the volume made upto 10ml with methanol. Methanolic extract was made by crushing 2gm of plant material in a clean mortar and pestle in 10 ml of methanol. The samples were kept for overnight extraction and filtered through a Whatmann filter paper no.41 the next day.
10 ml of both the extracts were evaporated to dryness and reconstituted in 10 ml of Dimethyl Sulphoxide (DMSO) stored at 4°C till further use.

Culture used: Culture was prepared in 0.85% sterile saline. The culture density was adjusted to 0.1 absorbance units (at 610 nm) using PharmaSpec UV 1700 (Shimadzu) and used for the present study.

Growth medium used: The medium for the study was Sterile Nutrient broth. 1.3 gm of dehydrated Nutrient Broth (HiMedia) was dissolved in 100 ml of Distilled water and autoclaved at 121º C and 15 psi for 20 minutes. 10% of Triphenyl Tetrazolium Chloride (TTC) was prepared in Distilled water and autoclaved at 121º C and 15 psi for 20 minutes. Sterile 10% of Triphenyl Tetrazolium Chloride (TTC) was added to sterile nutrient broth to get a final concentration of 0.2% Triphenyl Tetrazolium Chloride (TTC) in nutrient broth.

Graphs: Depicted values represent mean values of three readings. Error bars are standard deviation of the mean.

Statistical analysis: Statistical analysis was performed using SPSS 16.0 software. Mean values were compared

using ANOVA. A probability P-value of ≤0.05 ($P \leq 0.05$) was considered statistically significant.
In the table given in results, mean values followed by same alphabet in superscript (a, b, c, d....) within a column are not significantly different [11, 9].

Method for Quantitative measurement of bacterial growth by reduction of tetrazolium salts

The assay consisted of two sets run for *E. coli* or *Lactobacillus* sp. culture. Each set had a group of eight tubes run in duplicates.

a. Blank tube containing 3 ml of nutrient broth was inoculated with 0.5 ml sterile saline and 0.5 ml DMSO.
b. Positive control tube containing 3 ml of nutrient broth was inoculated with 0.5 ml culture of *E.coli* or *Lactobacillus* sp. respectively and 0.5 ml DMSO.
c. Standard tubes containing 3 ml of nutrient broth was inoculated with 0.5 ml of standard dopamine (100 ppb, 1 ppm) or standard epinephrine (100 ppb, 10 ppm) and 0.5 ml culture.
d. Sample set A-Sample tubes containing 3 ml of nutrient broth was inoculated with 0.5 ml of culture of *E. coli* and 0.5 ml of methanolic extract of *Gomphrena globosa* or acidic extract of *Portulaca oleracea*.
e. Sample set B- Sample tubes containing 3 ml of nutrient broth was inoculated with 0.5 ml of culture of *Lactobacillus* sp. and 0.5 ml of methanolic extract of *Gomphrena globosa* or acidic extract of *Portulaca oleracea*.
f. All the sets were incubated at 37º C for 24 hrs. A colour change from yellow to red (due reduction of TTC to formazan) indicated cell growth.
g. The readings were recorded at 480 nm in UV vis spectrophotometer. Standard deviations were determined and the results were statistically evaluated at 95% confidence interval.

Results

The culture of *Escherichia coli* and *Lactobacillus* sp. was subjected to treatments of standards dopamine, epinephrine as well as extracts of *Gomphrena globosa* and *Portulaca oleracea*. From the data obtained (Table.1, Fig.1) it is evident that standards were showing an increase in the growth of *Escherichia coli* and *Lactobacillus* sp. A marked increase in growth is registered for standard dopamine at 100 ppb concentration for *Escherichia coli*. However, for *Escherichia coli*, standard dopamine at 1 ppm concentration didn't show a significant difference in growth in comparison to positive control. This indicates that dopamine at low concentrations promotes growth in *Escherichia coli.* A marked increase in growth was recorded for standard dopamine at 1 ppm concentration for *Lactobacillus* sp. while 100 ppb standard dopamine didn't show a substantial effect. Similarly, epinephrine at a lower concentration of 100 ppb promotes growth of *Lactobacillus* sp. while no effect was seen for *Escherichia coli*. In presence of extracts of *Gomphrena globosa*, *Escherichia coli* and *Lactobacillus* sp. showed a growth of 0.454 absorbance units and 0.377 absorbance units respectively while in presence of extracts of *Portulaca oleracea*, *Escherichia coli* and *Lactobacillus* sp. showed a growth of 0.633 absorbance units and 0.317 absorbance units respectively. A marked increase in growth is reported for *Portulaca oleracea* and *Gomphrena globosa.* Although *Gomphrena globosa* extracts reported an increase in growth of the organisms in comparison to standards and positive control, it was not as effective as *Portulaca oleracea.* Table 1 represents the growth of organisms in the form absorbance readings at 480 nm. Fig 1 compares the growth of organisms in the presence of standards and extracts of *Gomphrena globosa* and *Portulaca oleracea.*

Table 1: Absorbance readings at 480 nm for *E.coli* and *Lactobacillus* sp. in presence of standard catecholamines and plant samples

Sample (with *E. coli)*	Absorbance at 480 nm ± S.D.	Sample (with *Lactobacillus* sp.)	Absorbance at 480 nm ± S.D.
Blank	0 ±0.000[a]	Blank	0 ± 0.000[a]
Positive Control	0.338 ± 0.054[b]	Positive Control	0.119 ± 0.000[b]
Std. Dopamine (100 ppb)	0.547 ±0.002[c]	Std. Dopamine (100 ppb)	0.190 ± 0.00[b]
Std. Dopamine (1 ppm)	0.348 ±0.026[b]	Std. Dopamine (1 ppm)	0.235 ± 0.008[c]
Std. Epinephrine (100 ppb)	0.388 ±0.017[b]	Std. Epinephrine (100 ppb)	0.234 ±0.009[d]
Std. Epinephrine (10 ppm)	0.364 ±0.013[b]	Std. Epinephrine (10 ppm)	0.208 ±0.071[b]
Extract of *Gomphrena globosa*	0.454 ± 0.011[d]	Extract of *Gomphrena globosa*	0.377 ±0.037[e]
Extract of *Portulaca oleracea*	0.633 ± 0.002[e]	Extract of *Portulaca oleracea*	0.317 ±0.036[f]

*S.D.: Standard deviation

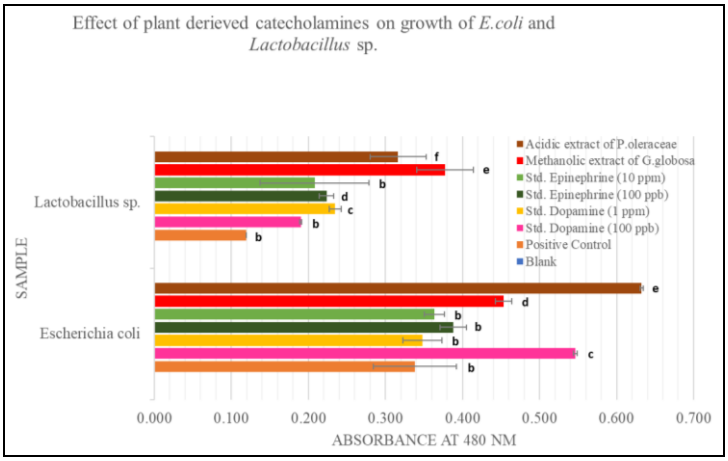

Fig 1: Effect of plant derived catecholamines on growth of *E.coli* and *Lactobacillus* sp.

Discussion

The present study aims to prove that the catecholamines produced in the plants are biologically active and can stimulate the cells to produce a response towards the hormones. David and Vanessa (2008) reported that bacteria can sense hormones, specifically epinephrine produced by the host. They hypothesized that the bacteria respond to these hormones which may help the cell in metabolism or virulence. The present study proves that *E. coli* and *Lactobacillus* sp. respond to dopamine while *Lactobacillus* sp. respond to epinephrine as well, when present at low concentrations. Menon and Pius (2016) have reported the presence of catecholamines in *Gomphrena globosa* and *Portulaca oleracea*. The plants showed a marked increase in the growth of the organisms which can be due to combined effect of the catecholamines present within them.

Conclusion

In conclusion, catecholamines in *Gomphrena globosa* and *Portulaca oleracea* promote growth in *Escherichia coli* and *Lactobacillus* sp. These plant extracts can therefore be used as stimulants in commercial manufacturing of probiotics.

Acknowledgement

The authors are thankful to the Principal, Ramnarain Ruia Autonomous College for the constant support and encouragement. We also thank the Head of the Department and the teaching and nonteaching staff of Department of Bioanalytical Sciences and Department of Botany for the help provided in successful completion of this work.

References

1. David H, Vanessa S. Inter-kingdom signalling: communication between bacteria and their hosts, Nature Reviews Microbiology. 2008; 6:111-120.
2. Freestone PP, Sandrini SM, Haigh RD, Lyte M. Microbial endocrinology: how stress influences susceptibility to infection, Trends Microbiol. 2008; 16:55-64.
3. Freestone PP, Lyte M. Microbial endocrinology: experimental design issues in the study of interkingdom signalling in infectious disease. Adv. Appl. Microbiol. 2008; 64:75-105.
4. Hai Ning Shi DVM, Allan Walker. Bacterial colonization and the development of intestinal defences, Can J Gastroenterol. 2004, 18(8).
5. Kimura M. Florescence histochemical study on Serotonin and Catecholamines on some plants. Japanese J Pharmacol. 1968; 18:162-168.
6. Kuklin AI, Conger BV. Catecholamines in plants. J Plant Growth Regul. 1995; 14:91-97.
7. Kulma A, Szopa J. Catecholamines are active compounds in plants. Plant Sci. 2007; 172:443-440.
8. Menon S, Pius J. Screening the members of Caryophyllales for Biogenic amine and its precursors, Bionano Frontiers. 2016; 9(2):194-196.
9. Panse VG, Sukhamte PV. Statistical methods for agricultural analysis, ICAR publications, New Delhi, 1967.
10. Smith TA. Amines in food. Food Chem. 1980; 6:169-200.
11. Zar JH. Biostatistical analysis, Dorling Kindersley, India, 1999.