

EFFECT OF MUSIC ON GROWTH AND PIGMENT PRODUCTION OF *BREVIBACTERIUM* SP

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ABSTRACT

Human beings experience and feel the music for joy and happiness. Even animals also feel the sound. Recently, the concept of musical vibrations of various ragas of Indian Hindustani and Carnatic music have been evaluated for its impact on the growth of the microorganisms and its metabolite production. In this context, the present study was framed out to screen the effect of tollywood music (Top lesi poddi from the film Iddarammayilatho, with a sound frequency of 600-1000 Hz, and Priyathama Neevachata Kushalama from the film Guna with a sound frequency ranging from 100- 600 Hz) ranging from 60-90 decibels on marine *Brevibacterium* sp. that produce yellow pigment. The present study showed that the sound frequency ranging from 600-1000Hz has significant effect on growth and pigment production of *Brevibacterium* sp. The biomass was found to be increased three times and pigment produced greater when compared to untreated sample.

Keywords: Sonic stimulation, Music and *Brevibacterium* sp.

INTRODUCTION

Sound is repeated pressure wave which travels through matter. In developed world, sound is spread all over the environment in different forms that may be in the form of music and noise produced by living things or artificial things. There have been few reports (Hassanien *et al.*, 2014) describing the effect of noise and music on plant and animals. Previous studies (Matsushashi *et al.*, 1998) indicated that microorganisms have the ability to produce, sense and respond to sound. Recent studies showed that the alteration of microbial growth and production of some important metabolites are influenced by sound. Much more is remained to be investigated on how microbes interact and behave to external sound field. In this regard, the present work was undertaken to investigate the effect of sound on growth and pigment production of marine *Brevibacterium* sp.

METHODS AND METHODOLOGY

Test organism

The organism selected for the study was marine *Brevibacterium* sp. which was a gram-positive

and yellow pigmented bacterium. Selected bacterium was grown in Zobell marine broth 2216(Himedia) and incubated at temperature of $28 \pm 2^\circ\text{C}$.

Music Treatment

Test bacterium was subjected to music treatment as described by Sarvaiya and Kothari, 2015. The speaker and flask containing culture was separated by a distance of 15 cm in a sound proof chamber with a speaker connected to power supply producing the sound ranging from 60-90 decibels which was measured using a digital sound pressure tester level meter (Generic 1082476). Control was maintained as same as treated sample but without power supply. In the present study, the famous Tollywood songs (Top lesi poddi of Iddarammayilatho film and Priyathama Neevachata Kushalama of Guna film) were selected, with the frequency between 100-1000 Hz in major and minor part of music containing frequency of 20-20000 Hz which is audible sound range measured by wave pad sound editor master edition. Zobell marine broth

2216(HIMEDIA) was prepared and distributed into 4 flasks (100ml each), 1st flask was maintained as Control (un-inoculated broth), 2nd flask was maintained as untreated (inoculated with bacterium but it was not given music treatment). The 3rd flask was maintained as treated sample (TS 1) which was treated with "Top lesi poddi" song with a frequency of 600-1000 HZ and the 4th flask was treated as treated sample (TS 2) which was subjected to "Priyathama neevachata kushalama" song with frequency 100- 600 HZ, where experiment done using triplicates.

Measurement of bacterial growth

Turbidometry is the method used to measure the growth of bacteria. The amount of scattering is directly proportional to the biomass of cells present and indirectly related to cell number. Turbidity was measured using spectrophotometer at 620 nm for 25 hours with the time interval of one hour.

Estimation of biomass by dry weight determination

The bacterial cells from the culture broth were separated by means of centrifugation at 10000 rpm at 4°C for 10 minutes. The recovered pellet was transferred to a pre-weighed dry filter paper using a clean spatula and then placed in an oven at 55°C get the dry weight. Growth in terms of biomass accumulation was expressed as mg/ml of the culture medium.

Extraction of pigment:

The recovered pellet was resuspended in distilled water. The pigment was then extracted with methanol by repeated centrifugation at 10000 rpm at 4°C for 10 minutes. The extraction was repeated until all the pigment had been extracted totally and the pellet become colorless. The methanol extracts were pooled and concentrated by evaporation.

RESULTS AND DISCUSSION

In the present study the effect of sound was tested on growth and pigment production of marine yellow pigmented *Brevibacterium* sp. Growth and pigment production was found to be Increased growth rate was observed in treated cultures. Growth was measured by turbidometry, using spectrophotometer at 620 nm. Optical density was recorded for every one hour for 24 hours. It was observed that the Stationary phase of the treated sample (TS 1) was achieved at 12th hour, whereas the

Stationary phase in treated sample (TS 2) was observed at 15th hour. The stationary phase of untreated sample was observed at 19th hour as shown in **Graph-1**. The results of present study showed that the Stationary phase of the treated samples was observed early stationary phases due to increase in substrate utilization in the broth which indirectly shows the increase of bacterial growth. Pornpongmetta and Thanuttamavong (2010) reported microbial substrate utilization of aerobic bacteria from municipal wastewater treatment plant to be affected by music; similarly, same results were observed in the present investigation.

Biomass determination

The biomass of treated samples is found to be more than untreated sample by 2.8 - 3.5 times and this growth was considered to be more as given in **Table-1**. As audible sound treatment increased the colony forming ability of *Escherichia coli* (Shaobin G et al., (2010)) in the same context in the present investigation we got to see good growth of cells in treated samples.

Pigment Production

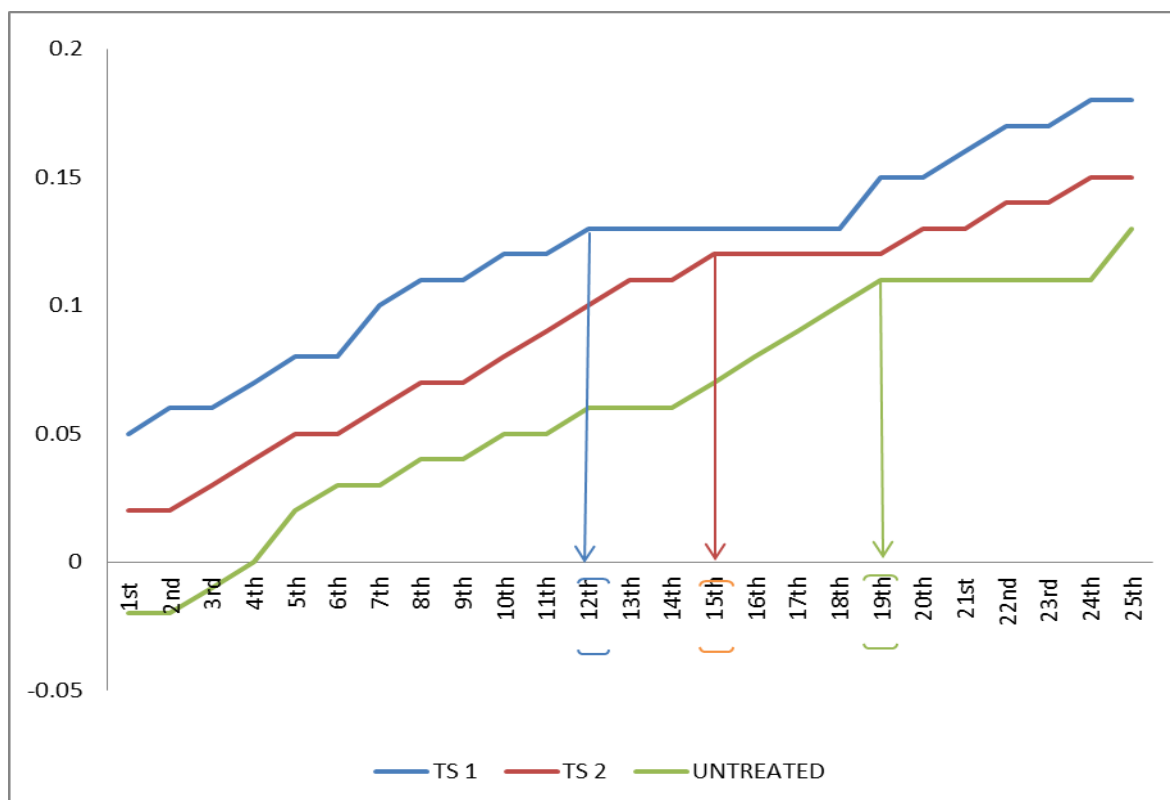
Pigment production was increased due to music treatment. When compared to the all the 3 samples TS 1 observed to be greater as in **Table 2**. *Chromobacterium violaceum* and *Saccharomyces cerevisiae* shown increased production of violacein and alcohol respectively due to music treatment (Niral Sarvaiya and Vijay Kothari, 2014). *Pseudomonas aeruginosa* treated with different sonic frequencies altered its pigment (pyoverdine) production (Vijay Kothari et al., (2017)) as we observed in our studies.

CONCLUSION

The results of the present study revealed that bacteria can be affected by sonic stimulation. External sound stimulation can be considered as an important factor for bacterial growth and pigment production. If we can study, how a particular microbial species inside human host responds to a given sonic stimulation, long run sonic waves can have therapeutic implications.

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Graph 1: Growth curve of *Brevibacterium sp*

Table 1: Biomass production of *Brevibacterium sp.* showing treated samples and untreated samples

Biomass	Untreated (Mean ± SD)	TS 1 (Mean ± SD)	TS 2 (Mean ± SD)
Dry weight (mg)	0.843 ± 0.009	2.7 ± 0.06164	2.1 ± 0.01

Table 2: Pigment production of *Brevibacterium sp.* showing treated samples and untreated samples

Pigment	Untreated (Mean ± SD)	TS 1 (Mean ± SD)	TS 2 (Mean ± SD)
Dry weight (µg)	35.6 ± 0.005	50 ± 0.003	45.3 ± 0.0064

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