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Research Paper Biological Science

Effect of Audible Sound on Microbial Activity

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ABSTRACT

This study reports experimental results regarding the effects of audible sound on the growth of bacterial cultures (standard laboratory strains). Standardized culture suspensions of fixed density were used for inoculation throughout the experiment. First, the samples were incubated at 37°C on shaker for 2 hours. The samples were then transferred to a Styrofoam box to give sound treatment at room temperature of 25-30°C for five hours. Two sound pieces i.e the sound of Om (AUM) and a Mozart composition (The Magic Flute) were selected for this experiment. The growth of each bacterial culture was assessed by taking optical density readings at an interval of 30 mins to determine the growth curve. We found that both music clips were able to promote the growth of bacteria. In particular, the sound of Om demonstrated significant increase in cell density.

KEYWORDS : Sound, Acoustics, Microbial growth, *Om, Mozart*

Introduction:

The interaction between audible sound (20-20,000 Hz) and biological materials is usually neglected in the field of biological research. Apart from acoustic experiments, sound frequencies in the form of mantras hold significance in religious scriptures and the effect of these on brain activity, emotions and behaviour is a well studied phenomena. However, studies on effect of sound on single cell systems, at the molecular level, are rare.

The sound of *Om* (AUM) is regarded as the 'Sound of Universal Vibration'. Its significance is documented in Vedic scriptures and in many religions apart from Hinduism including Buddhism and Jainism. Numerous claims about health benefits of chanting OM have been made, but documented scientific experiments and replicable results are lacking.

Music composed by Mozart has been of research significance and claims have been made that it increases spatial-temporal reasoning. Apart from cognitive learning many diverse effects of Mozart compositions have been recorded. For instance a German sewage treatment plant plays Mozart music to break down the waste faster, reports the UK Guardian. Anton Stucki, chief operator of the Treuenbrietzen plant was quoted as saying, "We think the secret is in the vibrations of the music, which penetrate everything—including the water, the sewage and the cells."

Materials and methods:

Cultures

Following laboratory bacterial test cultures were used:

- Escherichia coli
- Staphylococcus aureus
- Saccharomyces cerevisiae

Cultures were obtained from previous stocks cultured and well maintained in the college laboratory and grown on Nutrient agar plates (medium) at 37°C for 24 hours in an incubator. The medium was prepared by suspending 10g of NA in 500ml distilled water and was autoclaved at 121°C for 20 mins. A culture suspension of 0.1 OD (at 540 nm on standard laboratory colorimeter) was made in sterile saline, out of the above sub-cultured stock.

Inoculation and Incubation:

18 (for part 1) + 27 (for part 2) side arm flasks (3 each of control, OM and Mozart exposure for each culture) containing 25ml each of NB were prepared in this experiment. The Nutrient broth medium was prepared by suspending a total of 10g of NB in 1250 ml distilled water, dispensing 25 ml into each sidearm flask and autoclaved at 121°C for 20 mins. Each flask was inoculated with the respective culture suspension such that final OD of culture in each side arm flask (irrespective of the culture) is 0.05 at 540nm. This was considered as the 0 hr reading.

Measurement:

After 0 hr, further readings were taken at an interval of 30 mins. Throughout the experiment, the flasks were kept in constant exposure to the respective sound pieces in the Styrofoam boxes at room temperature. Last reading was taken after a time interval of 5 hrs.

Exposure to Sound of OM

Initial experimentation was to study the effect of sound of OM on the 3 bacterial cultures chosen. In comparison to their respective controls, the cultures exposed to sound of OM showed consistent increase in the growth rates.

The results were a quantitative confirmation that the cultures exposed to OM showed a better growth rate than control cultures.

Exposure to Sound of Mozart

Once established that exposure to OM significantly raised the growth rate in cultures, another music clip i.e The Magic Flute of Mozart, was introduced in the scenario to compare its effect with effect of the sound of OM.

Clearly the growth rate of cultures exposed to OM was more compared to their counterparts exposed to Mozart.

Results:

Table 1: Readings for growth curve (Control)

Time (mins)	E.coli	S. cerevisiae	S. aureus
0	0.05	0.05	0.05
30	0.14	0.1	0.08
60	0.23	0.215	0.08
90	0.38	0.375	0.11
120	0.51	0.55	0.13
150	0.61	0.64	0.2
180	0.67	0.69	0.3
210	0.74	0.745	0.455
240	0.78	0.785	0.58
270	0.81	0.82	0.67
300	0.85	0.85	0.72

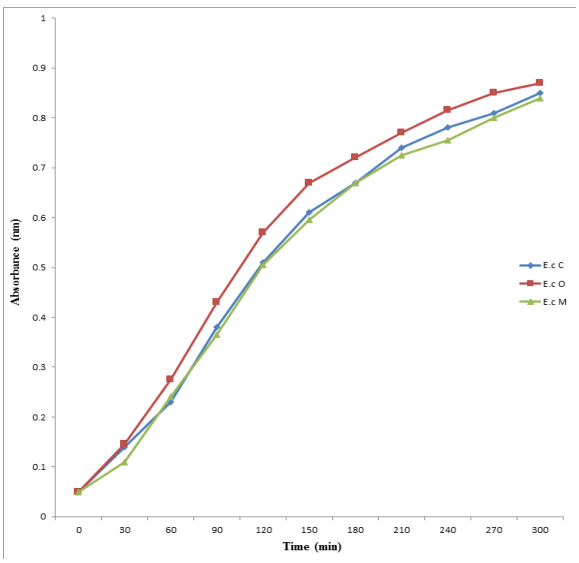
Table 2: Readings for growth curve (Exposure to Sound of OM)

Time (mins)	E.coli	S. cerevisiae	S. aureus
0	0.05	0.05	0.05
30	0.145	0.12	0.07
60	0.275	0.24	0.08
90	0.43	0.405	0.12
120	0.57	0.565	0.165
150	0.67	0.67	0.245
180	0.72	0.73	0.395
210	0.77	0.785	0.52
240	0.815	0.82	0.65
270	0.85	0.86	0.73
300	0.87	0.88	0.78

Table 3: Readings for growth curve (Exposure to Sound of Mozart)

Time (mins)	E.coli	S. cerevisiae	S. aureus
0	0.05	0.05	0.05
30	0.11	0.115	0.05
60	0.24	0.215	0.06
90	0.365	0.385	0.08
120	0.505	0.545	0.11
150	0.595	0.63	0.21
180	0.67	0.675	0.32
210	0.725	0.72	0.45
240	0.755	0.77	0.565
270	0.8	0.8	0.66
300	0.84	0.82	0.69

Graph showing Growth Curve of Control, OM exposed and Mozart exposed cultures of E.coli

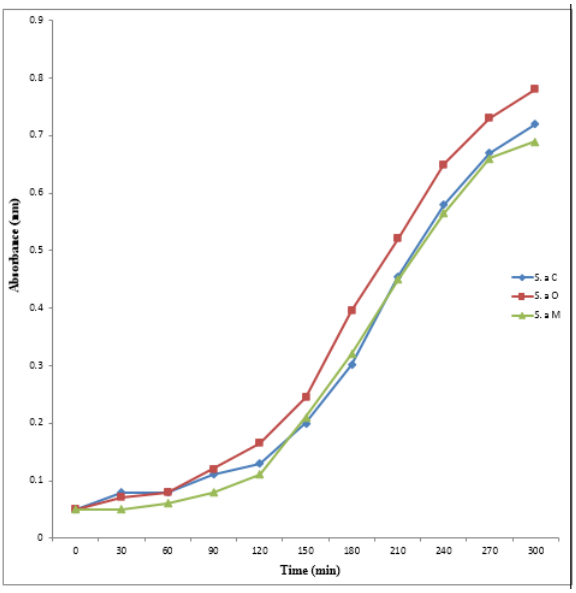


Key :
Ec C : Control

Ec O : OM exposed

Ec M : Mozart exposed

Graph showing Growth Curve of Control, OM exposed and Mozart exposed cultures of S.aureus



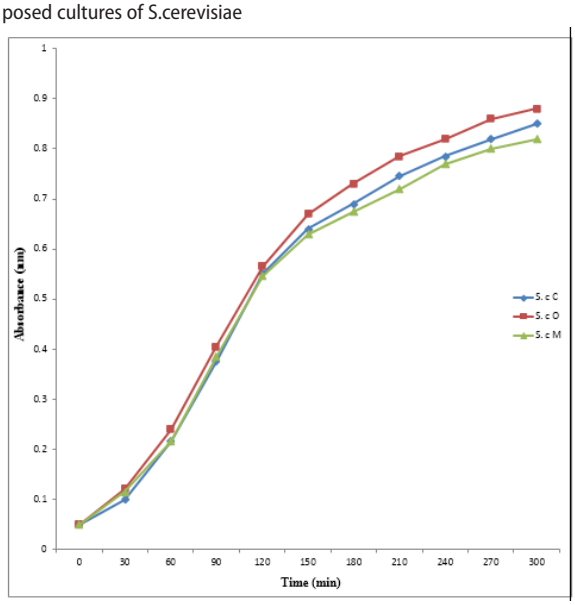
Key :

Sa C : Control

Sa O : OM exposed

Sa M : Mozart exposed

Graph showing Growth Curve of Control, OM exposed and Mozart exposed cultures of S.cerevisiae



Key :

Sc C : Control

Sc O : OM exposed

Sc M : Mozart exposed

Discussion:
As seen in the data and interpreted by the graphs, there is a clear rise in the growth curve of each culture that has been exposed to sound of OM in comparison to its Control counterpart. Likewise comparatively, OM exposed cultures grew faster than the ones exposed to the sound of 'The Magic Flute' of Mozart. Similar results have been obtained by some researches that are quoted below:

Shaobin G *et al.*[1] investigated the response of *E. coli* cells to the stimulation by audible sound. The results showed that the audible sound treatment significantly increases the colony formation of E. coli under the normal growth conditions.

Joanna Cho Lee Ying *et al.*[2] reported an experimental result regarding the effects of audible sound on the growth of Escherichia coli. Three different tonal frequencies were selected for sound treatment in this experiment which is 1 kHz, 5 kHz and 15 kHz. The growth of *E. coli* was assessed by their cell number through indirect viable cell counts and direct viable cell counts. In particular, the tonal sound of 5 kHz gave significant increase in cell number of E. coli for both growth media.

Hari Sharma *et al.*[3] compared the effects of 'primordial sounds' (Sama Veda from the Ayurveda system of natural health care), or hard rock music (AC/DC, "Back in Black"), and no sound on the growth of cells in culture. Primordial sound significantly decreased the average growth across cell lines ($p = 0.005$ ANOVA). In the presence of hard rock music, growth of cells was significantly increased ($p = 0.03$) but the effect was not consistent.

The possible cause of a growth spurt or increase in microbial activity is Acoustic Cavitation caused by subjecting a liquid to low frequency sounds. High frequency ultrasounds causes formation of microbubbles with kinetic energy enough to rupture cells.

William B. McNamara III *et al;*. 2015 studied that during acoustic cavitation experiments undertaken in the laboratory, one can recognise weak audible noise attributatble to cavitation bubbles. Just as a loudspeaker for music radiates sound by the vibration of its diaphragm, likewise a bubble radiates sound due to its vibration (pulsation) known as acoustic cavitation noise.

The evidence for the beneficial effects of music on reward, motivation, pleasure, stress, arousal, immunity, and social affiliation is mounting. We consider the evidence to be promising, yet preliminary, due to numerous confounds and limitations of many studies performed to date. The single cell systems can be used as models to infer these effects on brain cells, at a molecular level.

Conclusion:

Application of this research is novel and noteworthy. Standardizing frequencies of vibrations that give a good growth of the bacteria and which are culture specific would be a path breaking discovery. Many Industries could benefit out of it. If the correct kind of music is played to the cultures used in brewing or fermentation processes like cheese production, the process would become economical. The industry could save on a lot on important resources like electricity and media. Profits thus could be raised by increasing product quantity and cutting down other expenses by inserting the precise sound in their system.

The sewage treatment plant in Germany observed that their monthly expenditure of sludge removal is lowered substantially just by playing the tunes of "the Magic Flute" of the Mozart music. According to results of our experiment, the sound of OM gave a much higher cell density yield than Mozart. Thus clearly, the waste disposal systems could benefit from this little experiment.

Few research papers have stated a change in the antibiotic sensitivity of the cultures at specific frequencies, due to changes in their surface structures, for instance virulence factors like flagella or LPS layer. Standardizing frequencies of vibrations that increase antibiotic sensitivity of cultures could be an interesting approach to modern day prescriptions. Developing a musical medical theory, for once, doesn't seem a farcical thought!

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