SUPPLEMENT

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Stimulation of Protein Expression Through the Harmonic Resonance of Frequency-Specific Music

Abstract

Purpose: The use of specific frequencies for specific individual amino acids may increase the potential energy of protein molecules in the medium [1]. The resonance would also increase the movement of particles in the cytosol, increasing the collisions necessary for the conduction of protein expression.

Methods: The clash of two waves that share frequencies will exhibit an increase in energy through an increase in amplitude [2]. The increase in energy would in turn increase the number of collisions forming the tRNA-amino acid, increasing the amino acid acquiry for ribosomes to improve intracellular efficiency in gene expression. To test the hypothesis, Red Fluorescent Protein (RFP) in transformated BL-21 strains of *E. coli* and p53 protein of MCF-7 were examined after exposure to sounds of specific frequencies.

Results: Through the exposure of the experimental systems to a sequence of sounds that match the frequencies of specific amino acids, the levels of RFP exhibition respective to the control groups in the bacterial medium increased two-fold in terms of RFU. The experiments that targeted the p53 protein with the 'music' showed a decrease in the cell prevalence in the MCF-7 type breast cancer cells by 28%, by decreasing the speed of tumour formation.

Conclusion: Exposure to 'music' that was designed through assigning a musical note for every single one of the twenty unique amino acids, produced both an analytical and a visible shift in protein synthesis, making it as potential tool for reducing procedural time uptake.

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Correspondence to: Ibrahim Yasir Orhan Address: 9210 Petersham Dr, Houston, TX, 77032 E-mail: w207725797@student.hccs.edu The cytoplasm, the region of the cell in which the main functional activity is conducted, has the complexity to conduct most of the intracellular functions [3]. Although there exists massive differences and specificities between tissues and cells in terms of organelles, function and niche activity, the common feature of all living material is the existence of proteins and their central role in cellular function, and the universality of amino acids [4]. All cells contain amino acids, which make up complex proteins that exhibit various functions. The cytoplasm, in which the intracellular functions of proteins take place, conducts a rotatory movement of the churn through its semi-fluid composition [5]. The amino acids that reside in the cytoplasm carry out movement through the rotation of the intracellular churn alongside the molecular interactions upon them; ATP usage acts as a biological instigator for the interactions of amino acids as well. The active and mass usage of the essential nature of amino acids necessitates the constant flow of high levels of energy, which in absence causes further irreparable harm to cellular function through disabling the conduction of protein activity [6].

Resonance is the tendency of a physical body (generally a linear body) to oscillate in greater amplitudes [7]. This principle can allow even bodies of relatively miniscule amplitudes individually to express high amplitude oscillations. An evaluation of amplitude as the energy level of the body, which holds true as energy translates into amplitude in a physical body, would also bring in energy as an important factor in the equation. The harmony of two waves to oscillate in higher amplitudes in specific correlating frequencies causes bodies to continue oscillating in that frequency after the resonance effect diminishes. This continuation can be in the form of an extended oscillation, with the relatively stable systems acting in a relatively non-oscillatory state due to the long periods in the wave like function of stable bodies.

Amino acids have specific resonance frequencies as well. The prupose of this study was to instigate an increase in amino acids through a resonance effect to increase their movement in the cytoplasmic churn. The amplitudes of amino acids would be increased through sending sound waves of respective frequencies, which would in turn increase the kinetic energy of the amino acids in the medium. The increase in kinetic energy would in turn increase the spontaneity of reactions stimulating further translation and protein expression in the cell medium. Aside from the increased spontaneity, the increased instability of amino acids would also increase their affinity to undergo reactions. This increase in cellular function would improve the progression of the cell cycle.

Methods

The theory crafted by the French physicist, composer and mathematician, Sternheimer, to convert sequences to specific music has not been put to test by the science community. Sternheimer inspected the specific frequencies for the tRNA-amino acid complex through inspecting with sound waves. Through his studies, he was able to identify the correlation between amino acids and musical notes, although after his studies the issue remained clandestine [8].

In the light of these studies, a 'protein music' for RFP and p53 were composed in accordance to the specific amino acid frequencies. The musical notes were aligned in order with the sequence of both respective proteins to be tested in the experimental process. The testing of RFP was to yield visible results in the BL-21 strains used in experimental procedure to be further analyzed with analytical measurement. The MCF-7 cell cultures that were exposed to the music created in lieu with p53 were to exhibit also exhibit a resonance stimulation to increase p53 production, which would reduce tumour expansion through the prevention of unregulated cellular division.

The system designed in theory was tested on the strains through transformating competent cells and measuring the lysates of these strains after 0, 6, 9, 12 and 15 hours. External factors in the medium that could temper with the were eliminated in the experimental process, such as other present proteins. The experimental procedure involving MCF-7 breast cancer cultures on the other hand were cultivated under the specifically composed music with the presence of a control group and the colony growth was measured under a microscope after 24 and 48 hours.

Results

The results show that the composed music had an effect on cellular function (Figure 1-4). This can be easily recognized through the visible red color of cultivated E. coli and the increase in the fluorescence rate in fluorometric measurements. The RFP concentration in transformated BL-21 lysates increased twicefold in terms of RFU (Figures 3 and 4). The three stars presented in the T-test for the numerical figures further proves the reliability of the data.

The experimental procedure regarding MCF-7 breast cancer cells were conducted through exposing the cells to a music specifically composed for the p53, which harbored results in which there were significant shifts in the cancerous cultures. The relatively lower number of cells in the cultures exposed to the music in respect to the control groups provides

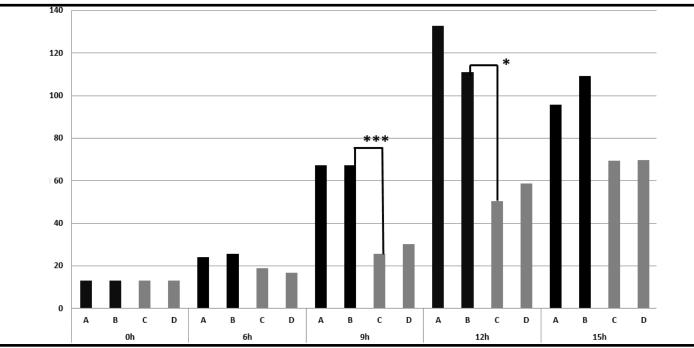


FIGURE 1. X axis, the time periods in which the measurements for the control group at 0, 6, 9, 12 and 15 hours; Y axis, the RFU values for RFP measurement. Groups A and B are samples applied the RFP protein music and Groups C and D are control groups. The samples were centrifuged and put under sonification to be lysed to acquire lysates for measurement. As the transformated RFP sequence, which was taken from Discosoma striata, was an mCherry type, the fluorometric excitation value was designated as 587 nm and the emission values as 610 nm through literature. The groups exposed to the music exhibit a twicefold increase in RFU value in respect to the control groups.2

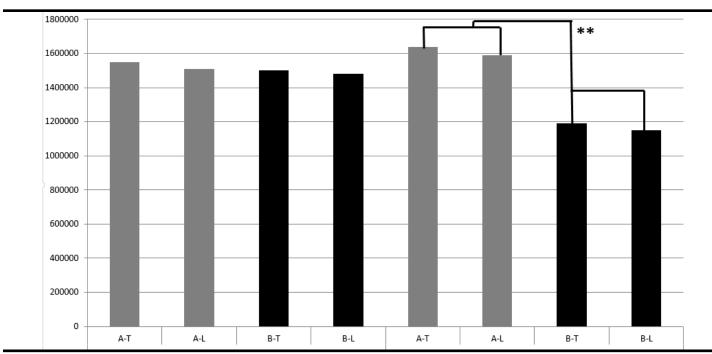


FIGURE 2. X axis, the time of measurement in hours as 24 and 48 hours; Y axis, the number of cells. Subject A is the control group whereas Subject B is MCF-7 wild-type breast cancer cells exposed to the p53 specific music. T stands for total number of cancer cells, L is for the total number of living cancer cells. The initial number of cells $(5x10^{5})$ cultivated in the medium show a 28% slower growth rate in Subject B



FIGURE 3. The microscope image of the MCF-7 wild-type breast cancer cell control group (Subject A) after 48 hours of incubation.

a very important result for the hypothesis in question. The p53 protein music that stimulated a 28% drop in the number of cancerous cells in the medium casted an effect visible to the eye under the microscope. Photometric and numerical data were plotted to further visualize the results (Figures 1 and 2).

Discussion

The hypothesis that there exists the applicability of harmonic oscillation to specific proteins through the usage of specially composed sound waves was to an extent proven and characterized. The outcomes show that theoretically the synthesis of all proteins in living domains can be elevated through music, as the amino acids in all living organisms are common, with the exception of formil methionine in bacteria [9], making this process almost universally applicable. Theoretically, another living domain can also show similarly elevated levels of activity as the functional purpose of the cytoplasm and the ribosome function show universality.

Protein expression is the most basic function in all organisms considered living; a basic function of life. Enzymatic activity is a function that allows essential functions like energy movement. Thus, the merits of expanding protein expression has limitless potential. When emerging fields, such as synthetic biology, are taken into account, the further improvements in functionality of molecular processes shows great promise. The shortening of the incubation times for certain cultivation processes is crucial for certain procedures and music provides a possible answer to ameliorate the associated problems.

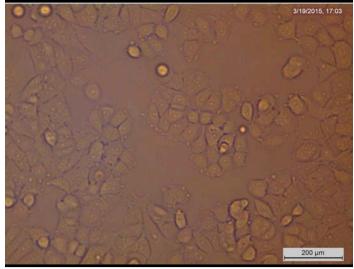


FIGURE 4. The microscope images of the MCF-7 wild-type breast cancer cells subjected to the p53 specific music (Subject B) after 48 hours of incubation.

The discussion of cancerous cells is not as straight-forward as the effects on the bacterial strains, as a multitude of factors are required to instigate cancer formation. Although targeting the p53 protein caused a marked decrease in the agility of cancer formation, the necessity for further investigation to elucidate the optimal composition of harmonical melodies to better suit the cancerous tissues is evident. MCF-7 was a specific cancerous cell type and the application of the harmonic sound therapy yielded results in a principally specific manner, meaning that this method could be used as a future alternative in cancer treatment to detrimental procedures bearing massive side effects.

Conclusion

In the future, this method could be tested with experimental setups using different organisms, targeting different proteins with the testing for optimal periods, amplitudes and future complications. This would enable the instigation of control over cellular functions in situ without any significant side effects that would otherwise cause a disruption of the 'naturality' of cellular function.

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