



A DIDACTIC EXPERIMENT FOR REDUCTION OF SACCHAROMYCE CEREVISIAE IRRADIATED WITH MICROWAVE AT 2GHz USING PALM TREE CLASS VIVALDI ANTIPODAL ANTENNAS

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Abstract - Microwave radiation can be used in a variety of applications, including the reduction of microorganisms, can be applied in the healthcare sector, as well as in the food sector, can be used to improve food conservation, for example. For this purpose, the Antipodal Vivaldi Antenna Palm Tree class is ideal for radiating microwave signals, as it has low weight, simple construction, has high directivity, high gain in main lobe and low level of lateral radiation. Therefore, this article presents a low-cost didactic experiment to reduce *Saccharomyces Cerevisiae* through microwave radiation using the Palm Tree AVA. After 17 hours of exposure of the test culture medium to a 2GHz microwave signal with an average power of 1mW, a reduction in the growth of microorganisms (in dense yeast colonies) of the order of 88% was observed when compared to the control culture medium, free from radiation.

Keywords: Microwave Radiation Reduction; Yeast reduction; Palm Tree Antipodal Vivaldi Antenna; Colony Reduction.

INTRODUCTION

The use of microwave radiation on microorganisms has numerous functions, the main ones being: synthesis of metallic nanoparticles for nanotechnology applications; inactivation of microorganisms and enzymes; microbial reduction [1]-[17]. In particular, the latter can be applied to sterilize foods [3] such as green coconut water, which has two enzymes, polyphenol oxidase and peroxidase, responsible for oxidation and loss of quality after contact with the external environment [7]. In the same way, the microbial reduction system can be applied to the soil. This technique promotes microbiological reduction in the region existing in the applied area and allows researchers to study the DNA chains of the vegetation or carry out tests on the reaction of the local vegetation to the chemical products used in agronomy [4]. Another function in which this type of sterilization system can be used is in orthodontics, sterilizing maxillary prostheses to avoid contamination of the patient [9] and [10].

Thus, reduction by microwave radiation is based on the exposure of electromagnetic waves in the frequency range of 0.3 to 300.0 GHz [5]. The effect of microwaves on the reduction of microorganisms is partly linked to their heating, which occurs through the polar moment of the irradiated molecules, enabling the destruction of microorganisms at low temperatures [5].

Therefore, in this experiment it was necessary to use an electromagnetic wave transmitter that radiates microwaves directly into the microorganisms' cultivation medium. For this, two Antipodal Vivaldi (AVA) type antennas were used, the Exponential Slot Edge Antipodal Vivaldi Antenna (ESE-AVA), also known as Palm Tree AVA [18], and Fern-AVA [19]. By improving directivity and reducing the squirt of conventional AVA [18] and [19], these antennas are ideal for applying radiation to a bacterial culture. The Palm Tree AVA was used to radiate the culture

medium while the Fern AVA was used in conjunction with a wattmeter (power meter) to measure the signal radiated into the culture medium.

To better present the proposed concept, this work was divided as follows: The Development chapter presents how the experiment was set up to prove the concept of reducing microorganisms with a 2.0GHz signal; The Results and Discussions chapter presents and comments on the relevance of the data collected during the experiment; Finally, the Conclusion chapter summarizes all the information and results obtained in the work.

DEVELOPMENT

To carry out the experiment, Palm Tree AVA was used to reduce a culture made with dry and instant biological yeast Fermix from the Dona Benta brand, packaged in a 10g sachet, of the *Saccharomyces Cerevisiae* Meyen type with Sorbitan Monostearate emulsifier, produced in China and packaged in Brazil by batch number 796405 C3 04.

To create a culture with the yeast, 300ml of mineral water, which at 25°C has a pH of 6.65, electrical conductivity of 224µS/cm, with 133.33 mg/l of evaporation residues at 180° C, having in its chemical composition 56.46 mg/l of NaHCO₃, 0.3 mg/l of NO₃⁻, 10.6 mg/l of Ca²⁺, 1.66 mg/l of K⁺, 26.8 mg/l of Na⁺, 0.584 mg/l of Mg²⁺, 2.88 mg/l of Cl⁻, 30µg/l of Ba²⁺, 396 µg/l of Sr²⁺, 0.1mg/l of PO₄³⁻, and 1.76 mg/l of F⁻ from Frescca, 12 g of refined sugar from União and 200mg of biological yeast were incorporated for 1 minute using a 350W BMX350P mixer from Britânia, weighed on a precision digital scale from Aguia Urso, measuring 130x65x20 mm, capable of weighing items from 0.01 to 3000.0 g and with an estimated error of 0.01 g. Fig. 1 shows all the elements used in this first stage of the experiment.

Once the culture was created, Petri dishes containing 20ml of culture medium each were

prepared, one being separated as control medium and the other being subjected to microwave irradiation.



Figure 1 - The items and equipment used to create a culture of yeast strains are: mineral water, balance, biological yeast.

To carry out the reduction experiment, two Polylactic Acid (PLA) supports were assembled, made on a Fused Deposition Modeling (FDM) Fused Deposition Modeling (FDM) Computer Numerical Control (CNC) printer, one to position the Petri dish of the culture to be irradiated, and another to position the radiating antenna. To generate the signal that irradiated the yeast culture, a microwave signal generator (high frequencies) and a 20dB low noise amplifier (LNA) were used. Fig. 2 shows the setup used to irradiate the biological yeast culture with microwaves.

After preparation, the control Petri dish was placed out of direct view of the antenna, so that it would not receive direct irradiation, however no other additional measures were used to protect it from light and other variations in the environment of room 07 at Cubatão Campus of IFSP. In this same room, the radiation setup was set up, which exposed the irradiated plate for 17 hours. Both plates remained in the same room the entire time, whose average temperature was approximately 25° C.

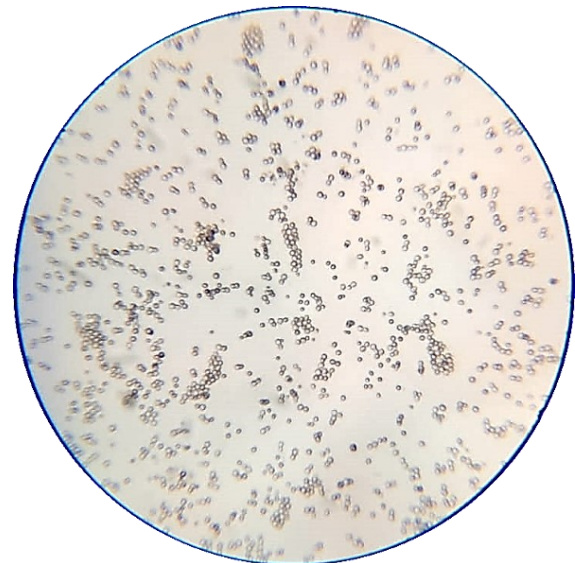
To measure the amplitude of the signal irradiated in the crop by the ESE-AVA, we used a Fern AVA, developed in [19], connected to a power meter.



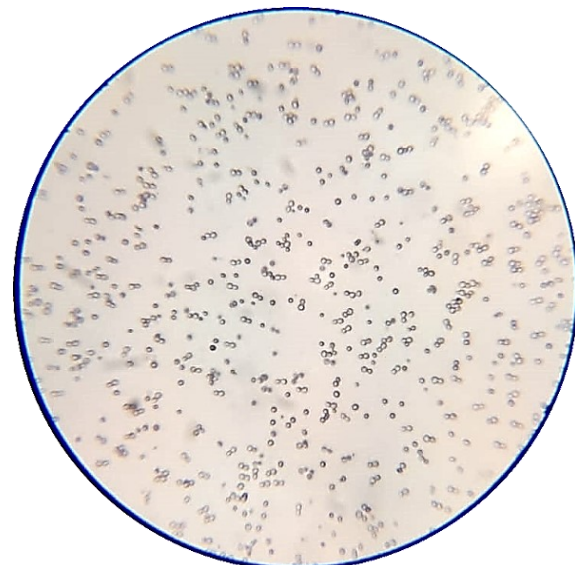
Figure 2 - Assembly made to irradiate microwave signals in the culture to be irradiated.

RESULTS AND DISCUSSIONS

This chapter presents the results obtained from the experiment structured in the Development chapter.



(a)



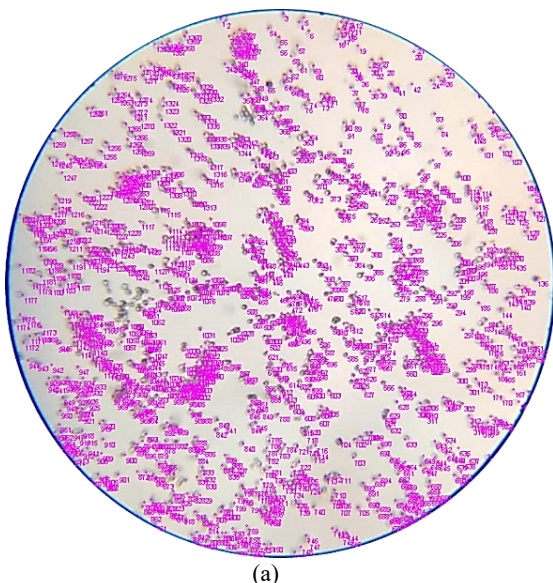
(b)

Figure 3 - Digital photographs, taken directly from the ocular lens of the optical microscope at thousand times magnification, of cultures after 17h of growth at 25° C. (a) Control culture medium. (b) Irradiated culture medium.

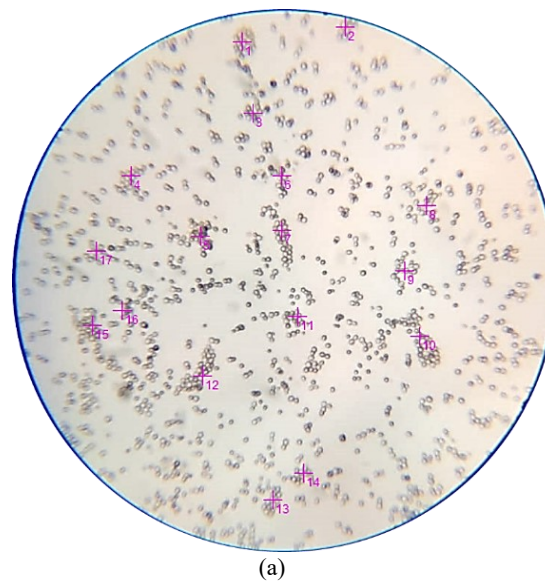
Fig. 3 presents two photos, one of the control medium (a) and the second of the irradiated medium (b), of cultures after 17h of growth at 25° C, with a thousand-fold magnification and it can already be seen that in both there are colony forming units (CFU), however in the first (control) there are CFUs with more than 11 individuals, in addition to visually having a greater density. To better study the two images, ImageJ was used, which is a powerful public domain medical image analysis program, developed by the National Institute of Health and based on Java [20] – [22].

TABLE 1 - Yeast count from digital photographs by microscope.

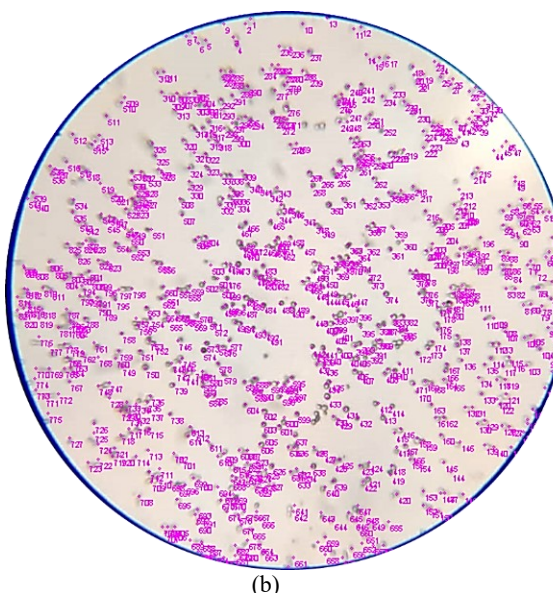
Petri dish	Total yeast	Count		
		CFU with 3 yeasts	CFU with 4 to 10 yeasts	CFU with more than 11 yeasts
Control	1360	31	45	17
Irradiated	828	10	39	2
Reduction	39%	10%	13%	88%



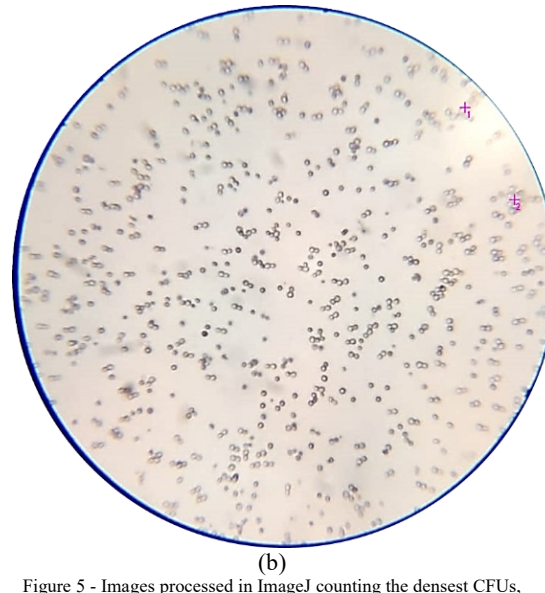
(a)



(a)



(b)



(b)

Figure 4 - Images processed in ImageJ containing the total yeast count in control (a) and irradiated (b) medium.

Figure 5 - Images processed in ImageJ counting the densest CFUs, from 11 or more yeasts, in the control medium (a) and in the irradiated medium (b).

Table 1 presents the results of yeast counts performed on images of the two cultures, control and irradiated respectively, using ImageJ. In the total yeast count, it was observed that the reduction was 39%. Less significantly was the reduction in CFUs of 3 yeasts and 4 to 10 yeasts, being 10% and 13% respectively.

In these two cases, it is observed that the radiation of a 2GHz microwave signal and 1mW of power after 17 hours of exposure, practically did not affect the emergence and maintenance of low-density CFUs. However, with regard to denser CFUs, with 11 or more yeasts, there was a significant reduction of 88%, which confirms that there are reduction effects, in denser CFUs, promoted by exposure to microwave radiation.

In Fig. 4, it is possible to observe the identification made by the ImageJ program of each yeast, both in the digital image of the control medium (a) and the image of the irradiated medium (b), both of which were obtained through a thousand-fold magnification using a microscope Aomekie optics.

Each yeast counted during the processing stage was identified with a small label containing the serial number in magenta color. When comparing the two images of the Fig. 5, it is observed that the count made by ImageJ digital processing, of the densest CFUs, that is, with eleven or more yeasts, is more evident in the control medium (a) than in the irradiated medium (b), the which contributes to the understanding that at this frequency and potency, the yeasts were affected in such a way that they were not capable of forming denser CFUs, which constitutes a significant reduction of 88%.

CONCLUSION

This work showed a low-cost didactic experiment to reduce *Saccharomyces Cerevisiae* through microwave radiation using the Palm Tree AVA. To prove this concept, an assembly was developed that consisted of using two AVAs, one to irradiate a microwave signal at 2.0 GHz in a culture of yeast strains and the other to measure the amplitude of the irradiated signal. After 17 hours of exposure of the test



culture medium to a microwave signal with an average power of 1mW, a reduction in the growth of microorganisms (in dense yeast colonies) of the order of 88% was observed when compared to the control culture medium, free from radiation.

Data collected at the end of the irradiation period, in the form of digital photographs taken directly from the ocular lens of the microscope set to 1000× magnification, were clear regarding the effects of exposure to microwave radiation on yeast, for a prolonged period, even with reduced power, which resulted in a significant reduction in the growth rate of dense colony-forming units. However, at the frequency, time and power chosen for radiation, it was observed that the impact of the reduction was smaller in less dense CFUs, in the order of 10% in the best case, which provides evidence that there is a correlation between the parameters chosen and the selectivity of the reduction capacity of denser CFUs. In this way, it is inferred that there is a window of opportunities regarding exploratory research in future developments of this research.

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