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Short Communication

Effects of Ultraviolet (UV) Radiations at Different Wave Lengths on the Microbial Load and Yeast Viability of Palm Wine

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Abstract
The effects of UV-radiation on the bacterial load and yeast viability of palm wine were investigated. In the studies 500ml of fresh palm wine sample each with initial yeast viability of 100% and bacterial load of 8.0 x 10^{15} Cfu/ml was exposed to UV-radiation at various wave lengths and time of 0 to 7hrs. The methlyene blue staining method was used to identify dead yeast cells based on the fact that dead cells absorbed the stain and were stained blue while the living cells reduced the stain and remained colourless. The viability of the yeast and bacterial load were determined on hourly basis for 7 hours. The result of the studies showed that bacterial load and yeast viability decreased with time and reached a minimal value of 3.0 x 10^{1} Cfu/ml bacterial count and 10% yeast viability. The optimum wave length at which the least bacterial count and yeast viability was obtained was 300nm. The organoleptic properties of the wine such as taste and flavor deteriorated slightly with time. The overall result had proved that UV-radiation, though bactericidal to some extent could not be used as an effective means of preserving palm wine. A plot of the number of surviving cell against the time of exposure to the radiation at various wavelengths showed a negative slope which implies that the death of a population of UV – irradiated cells demonstrates a log linear kinetics similar to thermal death.

Key words:

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Introduction

Palm wine is a traditional alcoholic beverage popularly drunk in tropical counties of the world including Nigeria. It is highly valued among the Igbos in south eastern part of Nigeria as number one alcoholic drink in traditional ceremonies. It is sourced from the saps of male inflorescence of *Elaeis Guinnensis*. The saps, which are rich in sugars, are fermented naturally by yeasts of the genera *Saccharomyces*. Lactic acid bacteria have been implicated to contribute to the characteristic flavor of fresh palm wine (Okafor, 2007). The sources of the yeast and bacterial micoflora include the air, knife, and palm wine keg, which the tapper uses in harvesting the saps. The alcoholic content increases with time due to the fermentative activity of the yeast. The major problem associated with the handling of the beverage is its short shelf life due to the uncontrolled metabolic activities of yeast and bacteria.

Ultraviolet radiation has been used successfully in disinfection of air and surfaces in food and beverage industries. The target organelles of the microorganisms are the nucleic acid bases (pyrimidine and thymine bases). The radiation causes dimerization of the DNA resulting in lysis and death of the organism.

A successful use of UV-radiation to control metabolic activities of these organisms will be a most welcome result.

Materials and Methods

The palm wine used for the study was sourced from Eke Obinagu Emene in Enugu East Local Government of Enugu state. The media and reagents used for the research work were supplied by the Resource concept laboratory Enugu. In the studies 500ml of fresh palm wine sample each was subjected to ultraviolet treatment in a uv-tank for seven hours. The viability of the yeast cells and bacterial count was determined every one hour for seven hours.

Methylene blue staining method was used to identify dead cells in the microscopic field. In this method one drop of the sample of the palm wine was placed on a clean haemocytometer slide and mixed with an equal volume of 1% methylene blue solution and allowed to react for one minute and then placed under X40 objective lens of a light microscope. Dead yeast cells were seen to be completely stained blue while live cells were colourless. This test was carried out on hourly basis for 7 hours. The number of live cells were counted in the microscopic field and calculated as percentage of the total cells (live and dead c ells) one hourly.

In determination of the bacterial count, Nutrient agar was prepared and cooled to 45^{o}C and mixed with 1ml of serially diluted sample (1:1000 dilutions). The inoculated plates were incubated for 48hrss at 37^{o}C. After incubation the discrete colonies were counted and used to calculate the bacterial load in colony forming unit per millilitre of the sample.

Result

The effects of Ultra-violet radiation on viability of yeast cells and bacterial load of palm wine were studied. The UV-radiation if effective for palm wine preservation should be able to destroy the microflorae of palm wine (yeast and bacteria) so that the quality of the beverage is retained, spoilage of palm wine arises from the uncontrolled metabolic activities of yeasts and bacteria. Metabolic activities of these organism result in production of such metabolites as ethanol, acetic acid, diacetyl and other off-flavour compounds.

In the studies methlyene blue staining method was used as recommended by 1.0B (1977) on the principle that living cells reduce methylene blue and remain colourless while the dead cells absorb the dye and are stained blue. The effect of the UV-radiation on bacteria was also determined by checking the hourly bacterial count of serially diluted samples in nutrient agar medium after incubation for 48hrs

In each investigation, 500ml of fresh palm wine with initial yeast viability of 100% was exposed to the radiation in a UV-tank at a wavelength of 100 to for 400mm for seven hours. The yeast viability and bacterial count were determined on hourly basis and recorded in table 1 to table 4 of the report. The result of the studies showed that bacterial count and yeast viability decreased with time and reached a minimal value of $3x10^{1}$ Cfu/ml and 10% respectively at the seventh hour of exposure at the optimal wave length of 300nm.

The taste and flavour profile of the palm wine decreased slightly with time. These results have shown that UV-radiation cannot be effectively used to preserve palm wine. It can also be deduced from the studies that the death of a population of UV-irradiated cells demonstrated a log-linear kinetics similar to thermal death as observed by Adams and Moss (1995). The target organelles by the UV-radiation on the microorganism are the nucleic acid bases (pyrimdine and thymine bases) resulting in dimerization of the deoxyribo nucleic acid (DNA) and death of cells.

The major problem with UV-radiation was its poor penetrability as reported by Adams and Moss (1995).

Table 1: Result of the effects of uv-radiations on bacterial load and yeast viability of palm wine at 100nm.

Time of exposure to UV-light (HRS)	Bacterial count in Cfu/ml	%yeast viability
0	8.0 x1015	100%
1	7.0x1012	70%
2	6.0 x1010	60%
3	5.0 x109	50%
4	4.0 x106	45%
5	3.0 x105	35%
6	3.0 x103	30%
7	3.0 $x10^3$	20%

Table 2: Result of the effects of Uv-radiations on bacterial load and yeast viability of palm wine at 200nm.

Time of exposure to UV-light (HRS)	Bacterial count in Cfu/ml	%yeast viability
0	8.0 x 10^{15}	100
1	6.0 x 10^{10}	65
2	5.0 x 10^8	55
3	5.0 x 10^7	45
4	4.0 x 10^5	40
5	3.0 x 10^4	35
6	3.0 x 10^3	20
7	3.0 x 10^2	15

Table 3: Result of the effects of Uv-radiations on bacterial load and yeast viability of palm wine at 300nm.

Time of exposure to UV-light (HRS)	Bacterial count in Cfu/ml	%yeast viability
0	8.0 x 10^{15}	100
1	6.0 x 10^6	60
2	5.0 x 10^5	50
3	5.0 x 10^4	40
4	4.0 x 10^3	35
5	3.0 x 10^2	20
6	3.0 x 10^1	10
7	3.0 x 10^1	10

Table 4: Result of the effects of Uv-radiations on bacterial load and yeast viability of palm wine at 400nm.

Time of exposure to UV-light (HRS)	Bacterial count in Cfu/ml	%yeast viability
0	8.0 x 10^{15}	100
1	6.0 x 10^8	65
2	5.0 x 10^6	60
3	5.0 x 10^5	50
4	4.0 x 10^3	45
5	3.0 x 10^2	40
6	3.0 x 10^2	25
7	3.0 x 10^2	20

Conclusion

Conclusively, the results of the studies have shown that UV-radiation though bactericidal cannot be used as effective technique for palm wine preservation. The major problem with the use of this technique is its poor penetrability as observed by other workers.

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