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RESEARCH ARTICLE

EFFECT OF LEMON JUICE ON THE KINECTICS OF FERMENTATION OF PALM WINE

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ABSTRACT

The fermentation of palm wine at varying temperature (0,30 and 45°c) have been studied by monitoring spectrophotometrically the absorbance of glucose with Dinitrosalicyclic acid (DNS) at 560nm wavelength for five days. The effect of lemon juice (citrus limonum) as additive on the fermentation was also studied. The introduction of lemon juice shows that the additive inhibits the fermentation of palm wine. The kinetics of uninhibited and inhibited fermentation of palm wine is found to be a pseudo first order with rate constants, k_{un} =0.45, k_{in}=0.36 respectively at room temperature (30°c). These values of k vary with temperature. Average value of activation energy (Ea) of the inhibited fermentation reaction was found to be 6.892J/mol and this was higher than the average value of activation energy obtained for the uninhibited fermentation reaction which was 3.504J/mol. Delayed fermentation of palm wine as needed by local Nigerians consumers of fresh palm wine can be achieved by addition of lemon juice.

Key words: Fermentation, Lemon juice, Activation Energy (Ea), and Specific Rate Constant

INTRODUCTION

Palmwine is the fermented sap of the oil palm (Elaeis gunensis) and the Raphia palm (raffia hookeri). It is an alcoholic beverage obtained from the fermentation of the sugary sap of various palm species [1]. Raphia hookeri is hapanzanthic, ie after a period of vegetative growth; it produces flowers and fruits only once and dies off after the fruit is matured. Esechie, (1978) has reported that raphia palm wine tapped at the base of the inflorescence is composed of sugar, protein, titrable organic acids, alcohol and macrominerals [2] . Further analysis of the sap (sugar) by paper chromatographic methods revealed that the sap contains sucrose, fructose, glucose and raffinose. Quantitatively sucrose contributed over 95% of the sugar [1]. Bottled palmwine preserved by pasteurisation at 70°C for 40 minutes without the addition of chemicals has been shown to posses the three characteristics of good palmwine (fresh sugary taste ,whitish colouration and vigorous effervescence) even after nine months of storage [2].

Kinectics of fermentation of sugar from other sources apart from palmwine have been studied by earlier investigations [3,4]. There is a consensus of opinion by these investigators that the rate of fermentation of simple sugars differs from source to source but the kinectic order of the fermentation process seems to be similar being specifically pseudo first order. However, the effect of citrus lime juice on the kinectic of fermentation of palm wine has not been reported elsewhere. The thrust of this work therefore is to study the kinectic of fermentation of fresh Nigerian palmwine and to investigate the effect of lime juice as preservative adjunct on the kinectic of the fermentation of raffia palmwine. People in the south eastern Nigeria relish fresh raphia palm wine for its wild euphoric intoxicating effect and for its assumed medicinal purpose for treatment of eye problems (Ndon,2007) and for fast breast milk production in young mothers especially primiparous women[2]. Palmwine drinkers in Nigeria have been trying to preserve the freshness of Raphia palmwine by several methods including addition of citrus lime juice. However, the effect of this additive has not been studied empirically. There is no report in literature on the kinetics of the fermentation of raffia palmwine and the effect of citrus lime juice on the kinetics.

MATERIALS AND METHOD

Sample collection

Fresh samples of raphia palmwine were randomly collected in clean labelled plastic bottles from local tappers in Utu Abak, Abak Local Government area Akwa ibom state, Nigeria and transported to University of Uyo central laboratory in an ice cubed box for kinectic study. The palm was tapped traditionally as described by Tulley, 1964 .

Laboratory method

Reducing sugar levels during the fermentation process was monitored by introducing 10ml of 5% dinitrosalicyclic (DNS) reagent into 10ml of the sample (samples with lime and those without) collected from stock [5]. Using D-glucose as standard, the calibration curve was made when 0.1ml of D-

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glucose was dissolved in 50ml of distilled water to obtain the stock. Using the equation $C_1V_1=C_2V_2$ where C and V represent concentration and volume, while subscript 1and 2 represent stock and standard solution respectively. These standards were prepared from different concentrations: 0.2, 0.4, 0.6, 0.8 and 1.0. 1ml of DNS reagent was added to each glucose standard and heated for 5 minutes. After cooling, red colour was developed and absorbance was taken at 560nm wavelength using spectronic 20 spectrophotometer, after a blank DNS solution had been used to zero the instrument.

Experimental procedure

100ml of the sample was measured into six different conical flasks of 250ml capacity. Then 50ml of the lime juice was added to three of the six conical flasks. Samples without lime were labelled "N" while samples + lime were labelled "NL". The conical flasks (NL) were corked with the foil and shaken for proper mixture. The samples (N, and NL) were allowed to ferment at different temperatures (0^o, 30^oC and 50^oC). 1ml of each in thermostated water bath of the fermenting samples (N and NL) was removed at 24 hours interval using pipette into six labelled different test tubes for colour development. The variation of reducing sugar content of the fermenting sample was monitored spectrophotometrically at λ =560nm. The absorbance was taken for each sample daily and their corresponding values of concentration were determined from the standard calibration curve drawn with D-glucose. The data obtained were fitted by trial and error into some integrated kinectic equation $2.303\log A_o/A_t = kt$ to deduce the rate constant k of fermentation and order[6]. Arrhenius plot of logk against 1/T was made and the activation energy (Ea) of the fermentation was evaluated from the slope. The data fitted best into a first order kinectic equation of the type $2.303 \log A_o/A_t = kt$.

Where A_o=concentration of glucose at zero time
A_t =concentration of glucose after t days
K= experimental rate constant
t= time duration of fermentation in days.

RESULTS AND DISCUSSION

Fermentation of palmwine uninhibited and inhibited by lemon juice at various temperatures have been studied. It was been observed that both fermentation processes resemble a consecutive reaction in which initial product leads to the formation of a new product B. Observations also showed that fermentation at 30^oc for both the inhibited and uninhibited fermentation of palmwine was faster than those at other two temperatures (0 and 45^oc). A possible explanation for this is that endogemic enzymes which are responsible for fermentation perform maximally at moderate temperature, that is to say, adverse temperature inhibits the activities of these micro-organisms[7]. At 0^oC which is the freezing point of water the enzymes are rendered inactive, possibly lowering the fermentation rate, while at 45^oC, it is possible that some of the enzymes responsible for fermentation must have died or denatured due to high temperature. Figure 1 shows the effect of lemon juice on fermentation at room temperature which inhibits the fermentation rate as compared to the fermentation of palmwine without additive. In order to determine the order of the uninhibited and inhibited fermentation of palmwine, attempts were made to fit data obtained from concentration of reducing sugar with respect to zero, first and second order. By far the inhibited and uninhibited fermentation reactions were found to obey pseudo first order kinetics at all temperatures (0^oC,30^oC,and 45^oC). By first order equations (In A/At =kt), a plot of $\ln A/A_t$ against t gives a straight line graph with slope= rate constant, k. In order to calculate the activation energy of the fermentation of palmwine, the Arrhenius equation was used[8].(k=exp(-Ea/RT) where k is the rate constant obtained from the slope of kinectic plots, Ea is the activation energy of palmwine , R is the gas constant and T is the temperature in kelvin units. From the equation k=exp(-Ea/RT) or log k=-Ea/2.303RT, a plot of log k against 1/T produces a straight line with slope equal to –Ea/2.303R. an Arrhenius plot for the fermentation of both palmwine without additive and palmwine with lemon juice shows that from the slope of lines on the plots ,the calculated values of Ea for the inhibited fermentation was found to be 6.892kJ/mol and this was higher than the value of Ea for the uninhibited fermentation which was 3.504kJ/mol. The results obtained have corroborate those of similar studies by Ekop and Etuk(1988); Ekop(1997)[9,10].

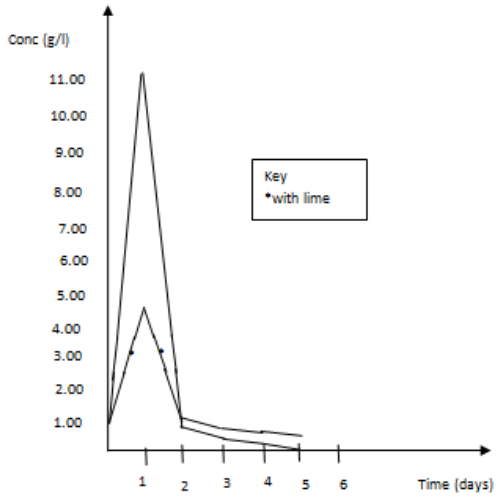

Fig 1: a graph of conc. against time for both sample without additive and sample with lime at 30^oc

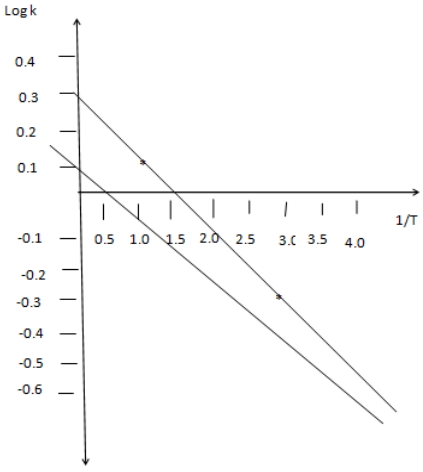

Fig 2: a graph of log k against 1/T for both sample without additive and sample with lime

Conclusion

The effect of lemon juice on the fermentation of palm wine has been observed. The study showed that lemon juice acts as inhibitor in the fermentation of palmwine. The effect of temperature on the fermentation of palmwine has been monitored spectrophotometrically by absorbance of glucose with dinitrosalicyclic acid (DNS) at 560nm wavelength. The data of palmwine follows a pseudo first order. The fermentation is optimal at room temperature (30^oC). There is an adverse effect on the fermentation of palmwine at high temperature (45^oC) and low temperature (0^oC). The slow on fermentation of the initial stage of less than three days have been assumed to be an induction period where the starting material (complex sugar) is converted to simple sugar (glucose). Average values of activation energy (Ea) of the inhibited fermentation of palmwine (6.892KJ/mol) was higher than average values of activation energy obtained for the uninhibited fermentation of palmwine(3.504kJ/mol).

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