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ABSTRACT

In the aim to investigate the optimization of the process of production of brandy from sweetened juice with pulp of ginger (Zingiber officinale Roscoe) using Saccharomyces cerevisiae yeast at Brazzaville, the employment of the content of cane-sugar and cell density is studied. The pH of the juice of pulp of fresh ginger tested is valuated at 6.5 and the soluble extract appreciated at 3.75 Brix about. The good development of the fermentation process of the wort of the sweetened juice with pulp of ginger admits the addition of sugar at an optimal value of 21.5 °Brix producing a distillate titrating 33 °GL of ethanol. Superior or inferior dose
1. Introduction

The ginger has been used since several centuries for its medicinal role at human1, 18, 26; it is a medicinal plant used against the high blood-pressure and stimulates our digestive system. At the fish of breeding in aquaculture, it procures effects of resistance against infectious Bacteria (Aeromonas hydrophilia, Helycobacter pylori) 18, 28, 33. The aromatic character of rhizome of ginger (Zingiber officinale Roscoe) 24, 30, 31, 32 is very interesting on the food plan 5, 29, particularly in India, in Chine (respectively first and second world producing), in Thailand, at the Japan and Nigeria. The ginger is used in the pastry - work.

A ginger liqueur is produced in Charente (France) and an arranged rhum at the ginger at Madagascar.

This spice plant grows in the tropical regions, particularly in Congo Brazzaville where its transformation into sweetened beverage, locally called ‘’tangawissi’’, is artisanaally exploited.
Produced in a rural environment, the rhizome of ginger is ramified, thick, fleshy and very aromatic. It is commonly commercialized in Congo at fresh state. It is rich in starch (60%) 2, 6 and its natural extract juice31 is non-fermentable by yeast25, 34. The process of leaded fermentation of wort from the extract of the juice of ginger added of cane-sugar by exogenous yeast action8, 9, 17 conduct at an alcoholic wort which the distillation process furnishes a nicely brandy appreciated at Brazzaville.

In this study, evolution of parameters relative at the kinetics of disappearance of soluble extract in the course of development of the yeast fermentation process of sweetened wort of ginger (Zingiber officinale Roscoe) and alcoholmetric title of ethylic distillate obtained are evaluated.

1. Material and methods
1.1. Material
i) System of fermentation

The systems used for effecting the process of tests of fermentation are bottled in glass of 1 liter of capacity, higher than large (height = 31 cm and diameter = 8 cm) and flat bottom. The charge of wort tested corresponds to 80 % of the volume of the bottle.

ii) Vegetal material and food additives

The vegetal material used is constituted of an ensemble of 12 lots of rhizome of ginger (Zingiber officinale Roscoe) weighing each around 407, 5 g ±40 g.

These 12 lots of rhizome of ginger, originating of the valley of Niari, at around 200 kilometers at west of Brazzaville, are sampled at random in the heart of Total markets and PK-MFilou respectively in the districts II of Bacongo and VII of MFilou at Brazzaville. Brown cane-sugar of Saris-Congo (a measure of 5 kg) obtained on the market at Brazzaville is used in variable quantity relative at each test in triple.

A pure strain of yeast Saccharomyces cerevisiae (Saf-instant mark, France) is used for seeding the wort; it is obtained at Brazzaville market equally.

1.2. Methods of elaboration alcohol of ginger
The rhizome of ginger used is cleaned, weighed and grind. The ground of ginger obtained is mixed in a hot sugar solution. After thermic cooking, the mixing is cooled at ambient temperature and then filtered. The sweetened wort of an extract of the pulp of ginger is seeded of yeast and leaned fermenting.

At the end of the ethylic fermentation, the fermented wort of ginger is distilled (Figure1).

For the preparation of the ginger wort, two methods are employed.

i) Methods of preparation of ginger wort

First method of preparation of wort for the optimization of the sugar dose

Three types of wort differently added of cane-sugar making respectively 31.75 °Brix, 21.5 °Brix and 14 °Brix, are prepared in triple.

The ground ginger obtained is cooked in a pot containing 15 liter of a cane-sugar hot aqueous solution during an hour. After cooling down process at ambient temperature, each mixing is sifted and the waste pressed.

A volume of 1.6 liter of sugar wort obtained is introduced in a bottle of two liters, and then seeded with 13.4 x10⁶ cells/ml before-hand active, during 15-30 minutes at ambient temperature in 100 ml of aqueous solution at 2 % of cane-sugar.

A second method of preparation of the wort for optimization of the rate of the seeding

Four types of wort differently seeded according to cell density of yeast used7, 13 used, are tested in triple.

After grinding of rhizome of ginger cleaned, the ground is added of water. The juice is extracted by sifting and pressing of the waste, then cooked at fire during 30-45 minutes of ebullition. It is cooled down during a night at ambient temperature.

The next day, it is warmed up, then standardized at 20.5 °Brix by addition of cane-sugar on juice of pulp of ginger and finally pasteurized at 85 °C during 20 minutes for destroying the endogenous flore4, 10 and 11.
ii) Launching and realization of the process of fermentation.

A volume of 0.8 liter of sweetened extract obtained is cooled down and poured in a bottle of one liter. It is then seeded with a stump pure of commercial yeast (“Saf-Instant”, France) dosed respectively at 26.8 x 10^6 cells/ml for the trial E1, 13.4 x 10^6 cells/ml for E2, 6.7 x 10^6 cells/ml for E3 and 1.6 x 10^6 cells/ml for E4. The inoculum is beforehand stirred up during 15-30 minutes in 100 ml of a sweetened solution of 2 % of cane-sugar.

After seeding of sweetened wort with the preparation of stump yeast stirred up, the process of alcoholic fermentation is then initiated.

iii) Distillation of ethylic fermented wort

At the end of the fermentation process, the fermented wort of ginger is distillate.

For every essay of distillation process in the laboratory, a volume of 300 ml of alcohol fermented wort is put in a balloon of 500 ml.

In the course of distillation process, the distillate is collected by the fraction of volume of 100 ml into a gauged phial in glass.

1.3. Methods of physical-chemical and microbiological analysis

a) Determination of pH of soluble extract of juice of pulp.

For the determination of pH of the soluble extract of juice with pulp of every lot of ginger, the rhizome is cleaned in the water and then desiccated by means of blotting-paper. It is finely grated and the mash obtained is placed on a cloth of muslin, and then pressed manually by twisted fringe. The trouble juice obtained is filtered by means of a filter-paper 10.

The pH of the samples is measured by use of a pH-meter (Martini Instruments) beforehand calibrated at pH7 with distilled water. As samples one note the extracted juice of the rhizome, the mixing of ground and sugar before and after boiling, the ginger wort in the course of the fermentation process and the distillate.
b) Evaluation of extract juice with pulp of ginger, soluble extract

The measure of soluble extract of the samples is determined by use of a refract meter Brix at 20°C according to Navarre27.

c) Evaluation of cell density of yeast

To evaluate the cell density of the yeast destined at the feeding of the sweetened wort of ginger, the technique of counting to optic microscope is used11.

d) Determination of alcoholmetric title of the first fraction of volume of distillate by alcoholmeter.

For every first fraction (100 ml) of distillate collected, the alcoholmetric title is determined at 20°C by means of a Gay-Lussac alcoholmeter which the scale goes from 0 at 100°GL14.

1.4. Statistical Analysis of Results

Within the qualitative appreciation of juice of rhizome and of the kinetic study of the idyllic fermentation process of wort of ginger in the course of fermentation, then for evaluating of the alcoholmetric title of every essay, the following statistical values are considered: mean, standard-deviation, coefficient of variation and interval of confidence (mean± standard-deviation). The method based on the law of Gauss-Laplace in bell is used as presented it Larrieu20 with modification, in view to appreciate the relativity of the analysis and operations.

2. Results and discussion

2.1. Characterization of the pulp of rhizome of ginger of the Niari valley

The mass of the rhizome, the volume, the pH and the soluble extract of natural juice from the pulp of rhizome of ginger of the Niari valley in the fresh state are valuated (Tableau 1).

The relation of the volume of juice extracted from the pulp on a la Mass of rhizome of ginger tested is estimated at 0.55 ± 0.04/1 milliliter/gram.

The pH of the juice of de the pulp of the rhizome of fresh ginger is estimated at 6.5 ± 0.2 and its soluble extract evaluated at 3.75 ± 0.25 °Brix.
2.2. Evolution of soluble extract of sweetened wort in the course of the traditional process of ethylic fermentation.

During the fermentative incubation of the wort of ginger the profile of consumption of soluble extracts march of manner descendant according to three essays tested (Figure 2).

Residual extract of wort is especially more high in the case of the essay initially more dense (17.8 °Brix of residual extract obtained from the essay N°1 with 31.75 °Brix for the initial extract) in comparison with the essay more light (9°Brix of residual extract for the essay N°2 starting off with 21.5 °Brix and 4°Brix of residual soluble extract soluble essay N°3 with initially 14 °Brix).

2.3. Appreciation of the value of the alcoholometric title of the distillate generated by distillation of fermented wort.

The alcoholometric title of the distillate obtained after distillation process of the ethylic fermented wort vary according to the type of essay tested (Figure 3).

The essay N°2, originating from sweetened wort at 21.5 °Brix, produce more ethanol (33 °GL) in comparison with other essays tested (30 °GL obtained with the sweetened wort N°1 evaluated at 31.75 °Brix and 18 °GL with the sweetened wort N°3 initially dosed at 14 °Brix).

2.4. Influence of the rate of yeasting on the development of the fermentation process of the wort of ginger

Le profile of consumption of soluble extract presents downward speed for all rates of feeding of yeast carried out on wort of ginger (Figure 4).

With the rate of inoculation elevated of 26.8 x106 cells of yeast/ml, the content of soluble extract of the wort decreases quickly, into 3 days of fermentation process, from 19.5 °Brix at 6.5 °Brix. But it decreases more slowly, into 6 days of fermentation process, from 19.5 °Brix at 6 °Brix with the rates of inoculation more feeble of 6.75 x106 cells/ml for the essay N°3 and of 1.6 x106 cells /ml for the essay N°4.
On the other hand, the diminution of the pH during the fermentation process is lower with the creasing of the rate of feeding of yeast tested on the wort of ginger (Figure 5).

This diminution of the pH is more rapid when the rate of seeding of wort is higher (from pH 6.8 this value decreases rapidly in one day of fermentation process at pH 4.6 with the rate of inoculation of 26 x106 cells/ml) in comparison to the case of inoculation relatively more feeble (from pH 6.8 at pH 5 in one day with a setting of 1.6 x106 cells /ml).

2.5. Effect of the rate of yeasting of the wort on the conversion process of the cane-sugar into ethanol

In the course of the fermentation process of the sweetened wort of ginger at 19.5°Brix, the effect of the rate of yeasting is appreciated according to the conversion process of the sweetened extract into ethanol. The values of the alcoholometric title and that of the pH of the distillate obtained by distillation of the wort are in relationship with the rate of inoculation employed for realizing the ethylic fermentation of the sweetened wort of ginger (Tableau 2).

From the soluble extract submitted in the fermentation process of the sweetened wort, the alcoholometric title of ethylic distillate obtained vary very few (CV = 3.47 %) according to the different rates of seeding employed (31.75 °GL with an inoculum of 26.8 x 106 cells of yeast/ml in comparison at 34,6 °GL obtained from inoculation with 1.6 x 106 cells of yeast/ml). But the value of the pH of alcholic distillate is enough constant (CV = 0.12 %) for the four essays tested.

The pulp of the rhizome of ginger is a medium at acid character (pH= 6.6 ± 0.1), but poor in soluble sugar (3.75 ± 0.25 °Brix). The juice of ginger doesn’t permit to the yeast to assure particularly the ethylic fermentation process.

The initial content of sugar added to the wort of ginger corresponding at a soluble extract of 21.5 °Brix is an optimal value in the wort and conduct to maximal production of ethanol (33 °GL). For an initial value of content of sugar equal or superior at 31.75 °Brix of soluble extract, the quantity of ethanol formed (30 °GL) exerts an antibiotic effect on the yeast cells and an effect of flocculation, leading to slowing and at the stopping of the process at 17.78 °Brix21.
For the dissolution of the cane sugar during the ebullition process, the operation of sugaring must effect on the hot extract of ginger 20 minutes before the end of the cooking of the wort in order to assure the inactivation of endogenous flora4, 10, 11, and 22.

The valorization of this local know-how is necessary to make in view to promote the knowledge on the useful of the biochemical transformation of tropical agro-resources3, 12, 15, 16, 19, 22 and 23.

An inoculation rate more high favor the speed of the fermentation of wort with optimal density of content sugar given (20.5 °Brix).

The residual extract more low noted at the end of the fermentation process of sweetened wort and the alcoholometric title of the distillate efferent the highest possible, reveal a good activity of fermentation. They are obtained in the course of a leaded fermentation with activated strain yeast inoculated at the sweetened wort of ginger with a rate feebler which is favorable for the development of the cell growing process of the active yeast.

Otherwise a value more elevated of a residual extract of the fermented wort and a feebler alcoholometric title of the distillate is obtained after the fermentation process with a rate of seeding of 26.8x106 cells of yeast/ml. That doesn’t intensify the cell rejuvenating, and as well as development of the ethylic fermentation process by the yeast. In these conditions of fermentation processes, an important population of cells of yeast identical at the parental-cell has flocculated at the starting of process and has not participated in the conversion of the sucrose into ethanol.

**Conclusion**

In order to obtain a good ethylic fermentation process, the employment of an initial quantity of sugar added on the wort of ginger must be done about 20.5°Brix of soluble extract. Overwise, a good yeasting process of this wort of ginger must be done with a pure stock of alcohogene yeast at the rate of a cell density of 13.4 x 106 cells/ml in order to obtain a good hourly yield of production in ethanol.
The juice of the pulp of the rhizome of ginger added of fermentable sugar is a good biomass that could contribute to the production of the bioethanol in exchange for yeasting by Saccharomyces cerevisiae. Table 1. Mass characteristics and physical-chemical of rhizome of ginger originating of the Niari valley.

<table>
<thead>
<tr>
<th>Statistical parameters</th>
<th>pH of soluble extract of pulp</th>
<th>Soluble extract of pulp (°Brix)</th>
<th>M : mass of rhizome tested (g)</th>
<th>V : volume of juice of pulp (ml)</th>
<th>Ratio V/M of the rhizome (ml/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>M ± δ</td>
<td>6.5 ± 0.2</td>
<td>3.75 ± 0.25</td>
<td>43.3 ± 5.1</td>
<td>22.5 ± 6.4</td>
<td>0.55 ± 0.04/1</td>
</tr>
</tbody>
</table>

n : 12 (results obtained from 12 lots of rhizome of ginger). M : mean; δ : standard deviation; M ± δ : interval of confidence.

Table 2. Valuating of alcoholmetric title and pH of fraction of 100ml of distillate.

Samples obtained order at the inoculating of the yeast, next fermenting of the sweetened wort then distillation process of the fermented wort.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean values of samples (E) of first fraction (100 ml) of ethylic distillate proceeding from the sweetened wort differently inoculated by cell density determined. M ± δ : Interval of confidence; CV: coefficient of variation.</th>
</tr>
</thead>
<tbody>
<tr>
<td>E1</td>
<td>Ethanol (°GL) = 31.75 ± 1.25</td>
</tr>
<tr>
<td>E2</td>
<td>Ethanol (°GL) = 33.5 ± 0.5</td>
</tr>
<tr>
<td>E3</td>
<td>Ethanol (°GL) = 34.83 ± 1.17</td>
</tr>
<tr>
<td>E4</td>
<td>Ethanol (°GL) = 34.6 ± 1.16</td>
</tr>
<tr>
<td>E1</td>
<td>pH = 4.1</td>
</tr>
<tr>
<td>E2</td>
<td>pH = 4.1</td>
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<tr>
<td>E3</td>
<td>pH = 4.2</td>
</tr>
<tr>
<td>E4</td>
<td>pH = 4.2</td>
</tr>
</tbody>
</table>
Ginger rhizome
  Cleaning
  Weighing
  Grinding using pestle-mortar
  Ground of ginger
  Mixing of ground and sugar
  Cooking of the mixing
  Cooling down
  Filtering
  Sweetened juice of ginger (26-29°C)
  Sweetened juice seeded
  Fermenting of wort
  Distilling of fermented wort

**Ethyllic distillate**

**Figure 1.** Diagram of production of ethyllic distillate from ginger rhizome
Figure 2. Evolution of the consumption of sugar (°Brix) in the course of fermentation of the ginger wort (26-29°C). Initial soluble extract: 31.75 °Brix for essay N°1, 21.5°Brix for N°2 and 14 °Brix for N°3.

Figure 3. Evaluation of alcohometric title of the distillate relative at the dose of initial soluble extract used for every essay tested (E1: 31.75 °Brix, E2: 21.5 °Brix etand E3:14 °Brix).
Figure 4. Evolution of the consumption of soluble extract (°Brix) during in the course of fermentation of wort of ginger in terms of rate of yeasting in M/ml (Millions of cells/ml, incubation at 26-29 °C).

Figure 5. Evolution of the pH in the course of the fermentation of ginger wort in terms of rate of yeasting: pH1 for 26x10⁶ cells/ml; pH2 for 13x10⁶ cells/ml; pH3 for 6x10⁶ cells/ml and pH4 for 1.6x10⁶ cells/ml.
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References


