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GOLDEN WINE PRODUCED FROM MIXED JUICES OF PASSION FRUIT (*PASSIFLORA EDULIS*), MANGO (*MANGIFERA INDICA*) AND PINEAPPLE (*ANANAS COMOSUS*)

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ABSTRACT

An attempt to produce yellow/golden wine was done in the laboratory using a mixture of fruits (33.3% each) as golden must extracted from *Passiflora edulis* (passion fruit), *Mangifera indica* (mango) and *Ananas comosus* (pineapple). After extraction of three juices, physical and chemical parameters were determined before and during fermentation of the must. These parameters were: wild yeast colony forming units per milliliter (CFU/ml) of fermenting must, total soluble solids (degrees Brix), pH, alcohol content, titratable acidity in percent, fermentation temperature, sugar content (g/l), and specific gravity. The fermentation of a mixture of juices was done at room temperature, i.e., at 22^{o}C, and the wild yeast used was *Saccharomyces cerevisiae*, a strain called "musanzeensis" isolated from local traditional banana wine. During substantial must fermentation, the pH decreased from 5.5 for fresh juice to 3.2 for wine, total soluble solids from 20^{o}Brix to 2^{o} Brix, titratable acidity increased from 0.68% to 1.4%, sugar content decreased from 85 g/l to 32 g/l, specific gravity decreased from 1.040 to 1.002, yeast growth increased from 3 to 18 log CFU/ml, and alcohol content increased from 0.0 to 12% alcohol by volume. After twelve days of fermentation, the color of wine remained yellow, the flavor was enhanced, sweetness diminished and the acidity (sourness) increased slightly. These chemical changes could be due to the *Saccharomyces cerevisiae* activity, which was characterized by a remarkable foam and intensive production of carbon dioxide in the fermenting wine. The mixture of the three juices from *Passiflora edulis*, *Mangifera indica* and *Ananas comosus* produced an alcoholic beverage with a wonderful flavor that was generally delicious and acceptable to 40 trained and blind panelists during sensory evaluation using as 9–point Hedonic scale. Each panelist sipped once 100ml of sample taken from wine. Thus, the obtained yellow wine should be promoted for adding value to local fruits, imported wine reduction, job creation, income generation and rural development.

Key words: *Ananas comosus*, *Mangifera indica*, *Passiflora edulis*, *Saccharomyces cerevisiae*

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INTRODUCTION

Passion fruit (*Passiflora edulis*), mango (*Mangifera indica*) and pineapple (*Ananas comosus*) are widely cultivated nutritious fruits in the Northern Province of Rwanda, especially in Gakenke and Musanze Districts where the soil is fertilized with organic manure from a mixture of cow dung and compost. The agriculture is in a green environment and the ripened fruits are harvested, sold fresh and consumed without post-harvest technology or value-added for export or income generation. Chemical composition of wines is influenced by soil fertility and climatic conditions. Physical and chemical parameters determination is important in wine processing [1]. Different imported wines are costly from developed countries and diminish local fruit and vegetable promotion [2]. Red and white wines are often expensive, but enjoyed by people during important ceremonies. Tropical fruits like ripened pineapple are good for flavored wine processing [3]. Mango fruits produce aromatic volatile compounds in fermented juices [4]. Wines produced from fruit juices like grapes are delicious and considered nutritious for humans [5]. Normally, wine is produced using pure culture of selected yeasts called *Saccharomyces spp.* [6]. Wine is obtained from fermentation of juices extracted from different comestible fruits [7, 8]. Tropical fruits have a slightly acidic pH that influences the sourness of juices [9]. The presence of organic acids in wine plays a great role in wine preservation [10]. Rwandan soil produces tasteful and delicious fruits, such us passion fruit (maracouja), mango, and pineapple suitable for post-harvest processing and exports in order to boost the national economy.

MATERIALS AND METHODS

Ripened passion, mango and pineapple fruits were collected from the local market of Musanze District in the Northern Province of Rwanda in May 2013. Cleaned fruits were peeled, crushed and pressed for juices using a blender (Philips HL1645 750-Watt 4 Jar Super Silent Vertical Mixer Grinder from France). Pure juices were sieved, filtered using tea filters and muslin cloth and pasteurized for further fermentation. The mixture of three juices was 33.3% each. *Saccharomyces cerevisiae* (dose: 2%) was isolated from local banana wine sediment and used as starter culture at room temperature (about 22^0C). Fresh juices in mixture and fermentation must were analyzed in the laboratory using AOAC (1990) methods (pH reference has AOAC official methods of analysis 960.19, 15th Ed.; alcohol content refers to AOAC official methods of analysis 920.47; and specific gravity: is instrumental and refers to AOAC official methods of analysis 988.06) [1].

Degree Brix was determined using a hand refractometer. Specific gravity was determined using a hydrometer. Temperature was determined using a thermometer. 40 panelists trained screened and blind were chosen for sensory evaluation at once (using 9-point Hedonic scale) of juices and wine obtained after fermentation. Each panelist sipped once 100ml of sample taken from wine. The description of 9 points (Hedonic values) is as follows: 9 stands for like extremely, 8 for like very much, 7 for like moderately, 6 for slightly, 5 for neither like nor dislike, 4 for dislike slightly, 3 for

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dislike moderately, 2 for dislike very much, and 1 for dislike extremely. Data analysis was carried out using Excel.

RESULTS

Table 1 shows that the wild yeast called *Saccharomyces cerevisiae*, strain "musanzeensis" isolated from local traditional banana wine was powerful to produce secondary metabolites such as alcohol during its exponential growth. The alcohol content was 12% and total soluble solids (degree Brix) were decreased by the fermentation process reflecting parallel decrease of fermentable sugar content from 85 g/l in fresh mixed juices to 32 g/l in the obtained yellow wine after 12 days of fermentation time. The multiplication of yeast in fermenting must from a mixture of juices is shown in Figure 1.

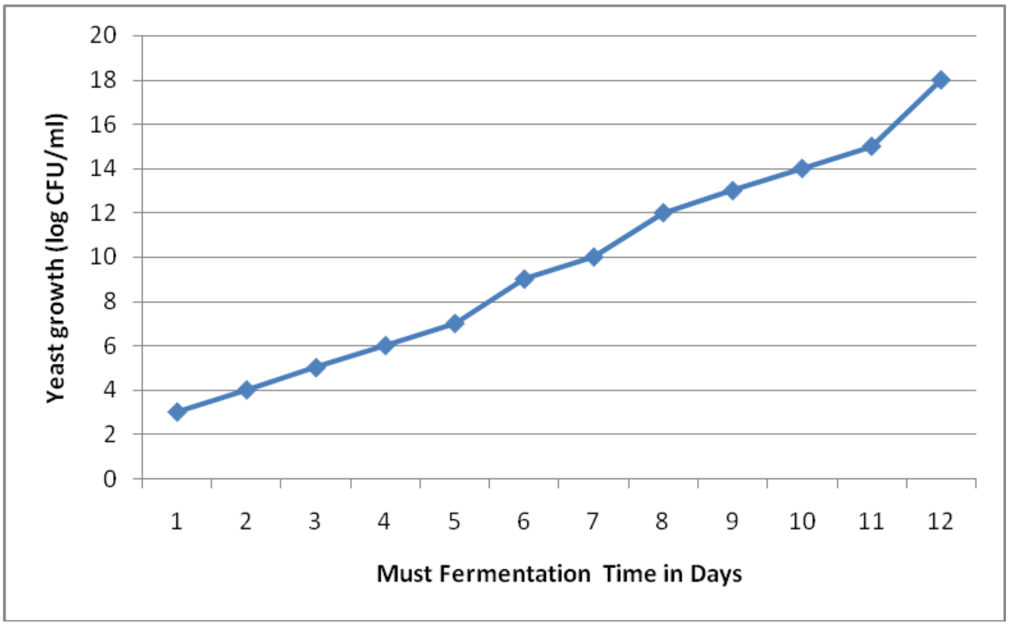

Figure 1: *Saccharomyces cerevisiae* multiplication during fermentation of must

Figure 1 shows the gradual increase of yeast biomass during must fermentation. The fermentation process was greatly reduced the total soluble solids content in degree Brix as shown in Figure 2.

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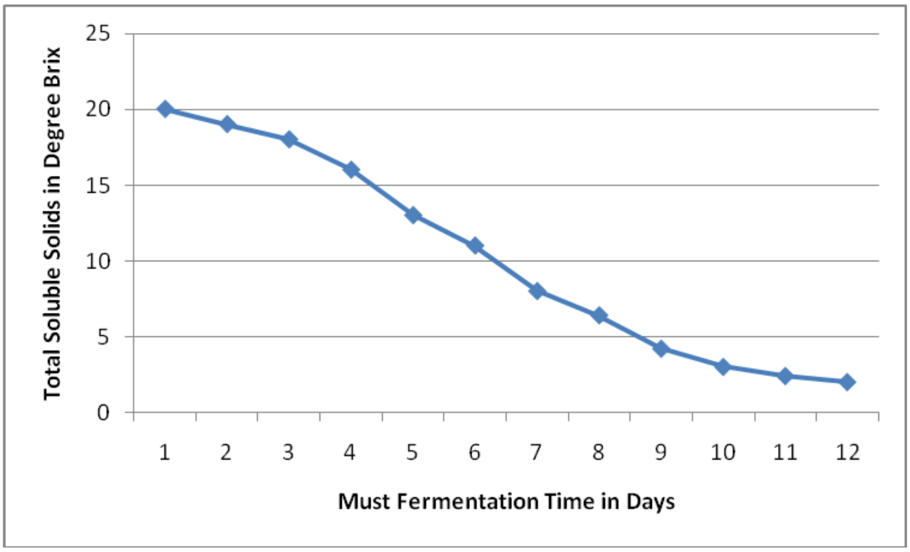

Figure 2: Total soluble solids (TSS in Degree Brix) decreased during must fermentation

Figure 2 indicates that the total soluble solids content are gradually reduced during must fermentation. The must sugar had 20^{o}Brix level and was desirable for proper fermentation and if the must sugar was below 18^{o}Brix, a wine of less than 10% alcohol would result, yet if the sugar is above 19^{o}Brix the fermentation would be retarded by long metabolic pathways of sugar degradation in must. The presence of *Saccharomyces cerevisiae* resulted in production of organic acids, ethanol and CO_2. The pH of fermenting must is a function of organic acids, such as tartaric, malic and citric acids, responsible for the sour taste of wine. The determination of pH of wine during fermentation of must was important because the metabolism of yeast is pH-dependent; the optimal pH of yeast metabolism is 4. In addition, the pH can affect the quality of wine in terms of taste, color, oxidation process, chemical stability and shelf-life. Thus, the pH variation was gradually dropped from 5.5 to 3.2 as indicated in Figure 3.

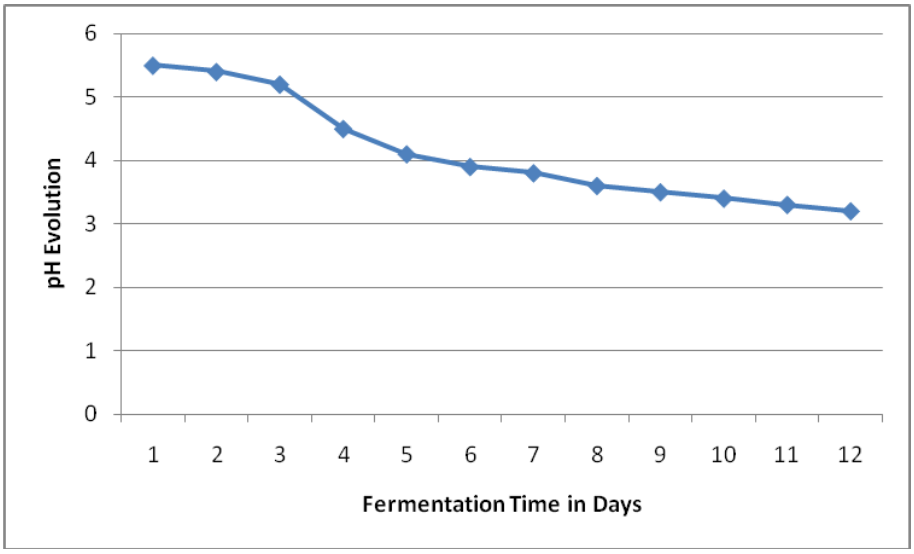

Figure 3: Gradual decrease of pH during must fermentation

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Figure 3 shows the gradually progressive reduction of pH in fermenting must over 12 days due to the fermentation process/degradation of sugars by yeast activity/metabolism leading to the increase of organic acids in produced yellow wine. The pH of fermenting must dropped from 5.5 to 3.2 leading to better sourness of produced wine. The fermentation of sugars by wild yeast isolated from local traditional banana wine leads to the sharp increase of alcohol content as given in Figure 4.

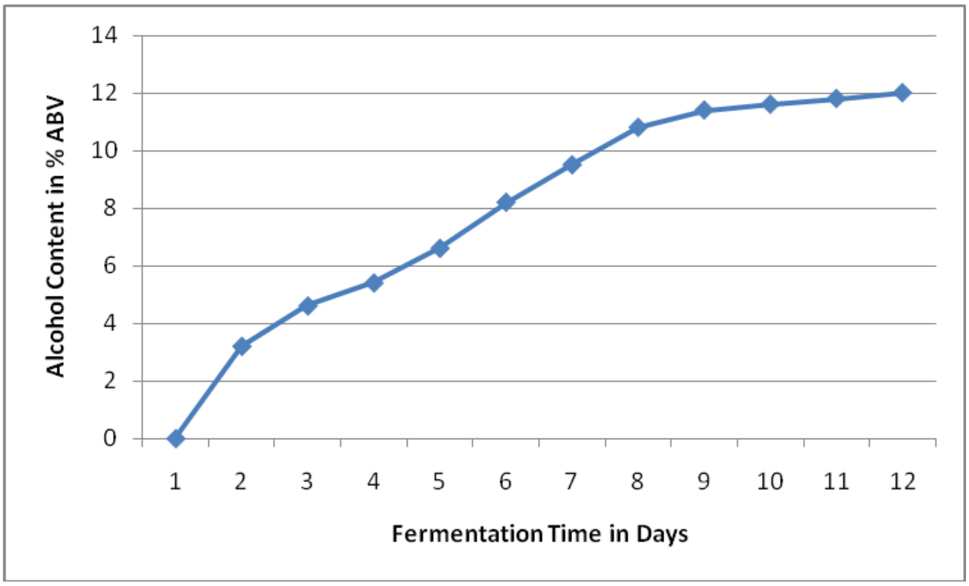

Figure 4: Alcohol content increases in fermenting mixed juices

Figure 4 shows that the alcohol by volume of fermenting must was gradually augmenting and after 12 days of fermentation of sugars, yielded 12% alcohol. The production of alcohol by wild yeast was parallel to the increase of titratable acidity in yellow wine obtained from mixed passion, mango and pineapple juices (Figure 5).

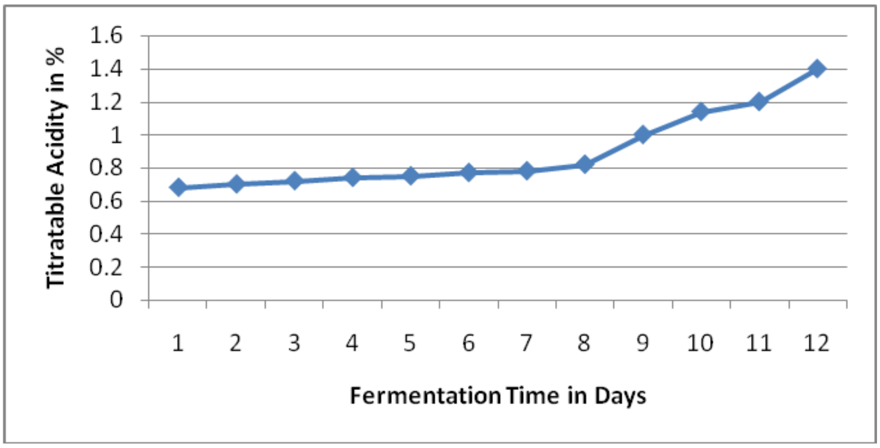

Figure 5: Determination of titratable acidity during must fermentation

Figure 5 indicates the gradual increase of titratable acidity in percent from 0.72 to 1.4 after 12 days of fermentation of mixed fruit juices. The level of cationic ions in fermenting must (juice) was high in substrate degraded by wild yeast. Different organic acids, such as tartaric, malic, lactic, citric, acetic, ascorbic, butyric, sorbic, and succinic acids, could be responsible for the high titratable acidity and lower pH in yellow wine. In ripe fruit, sugars such as fructose and glucose are in high content, but after fermentation these sugars are converted into different metabolites, such as ethanol, carbon dioxide and organic acids, leading to reduction in pH in the golden wine. In this regard, the sugar content was gradually diminished by presence of inoculated wild *Saccharomyces cerevisiae* used as starter culture in winemaking. The decrease in sugar content is shown in Figure 6.

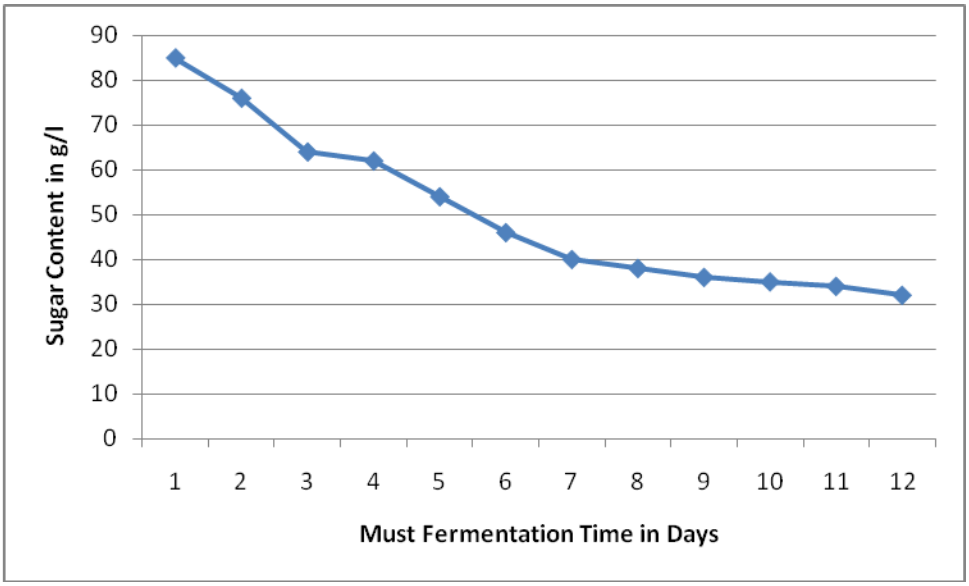

Figure 6: Decrease in sugar content during must fermentation

Figure 6 indicates the gradual decrease of sugars contained in mixed juices during fermentation activities carried out by *Saccharomyces cerevisiae* at room temperature (about 22^{0}C. The continuous decrease of specific gravity is relative to total soluble solid decomposition by inoculated wild yeast isolated from local traditional banana wine. In this regard, the decrease in specific gravity is shown in Figure 7.

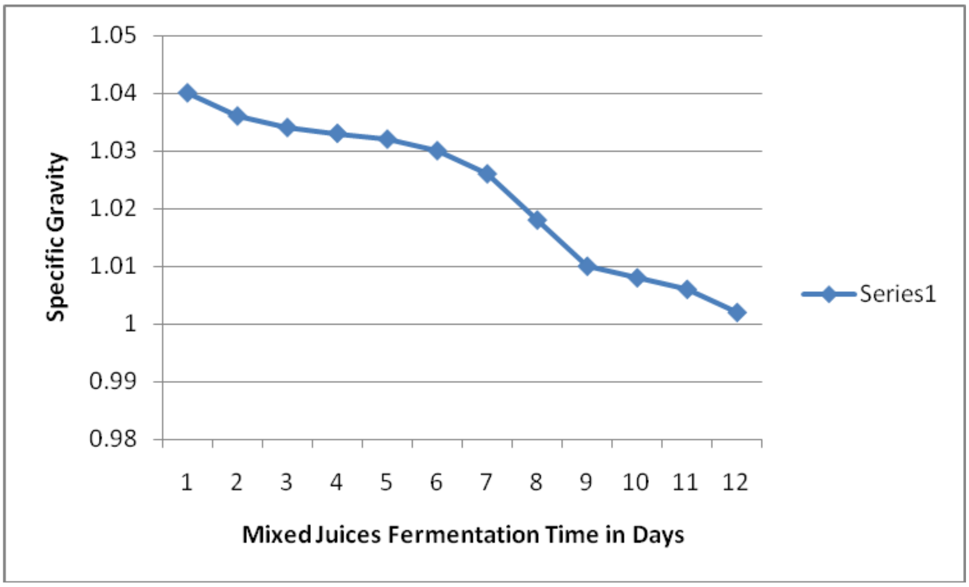

Figure 7: Variation of specific gravity in fermented mixed juices

Figure 7 shows that the gradual fluctuation of specific gravity was in descending manner and should be due to the sharp decrease of sugars and other chemical components found in mixed juices during metabolic pathways due to enzymes exerted by *Saccharomyces cerevisiae* as active starter culture found especially on local fruits harvested in Musanze area of Rwanda. Organic acids make major contributions to the composition, stability and organoleptic qualities of this golden wine. They also furnish preservative properties that enhance the stability of golden wine during the conservation period. This yellow wine was subject to alcoholic fermentation leading to best quality of end-product. In this regard, after fermentation of must, sensory evaluation was done in order to determine the yellow wine acceptance by panel members (see Table 2).

Table 2 shows that the quality of yellow wine was generally very good according to the Hedonic scale used; meaning that 40 panel members accepted the wine. The product was tasteful, and delicious with a golden color. The yellow wine gets fuller body with tropical fruit flavors and aromas of mature ripened pineapple, mango and passion fruits. These three fruits have freshness and antioxidants like vitamin C. They can be used for the production of sparkling wine if we add more carbon dioxide to golden wine. Different yeasts can be used to produce other aromas, but prominent in this process were local wild *Saccharomyces cerevisiae* found in local traditional banana wine, which gives the best alcoholic fermentation. The yeast population size is influenced by the presence of nutrients in juices and the optimal conditions of fermentation.

DISCUSSION

The mixed juices extracted from ripened mature *Passiflora edulis, Mangifera indica* and *Ananas comosus* harvested in the Northern Province of Rwanda gave a delicious,

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and acceptable golden wine more highly flavored than imported wines. The alcohol content (12% alcohol by volume) was similar to results obtained from cape gooseberry processing [6]. Total soluble solids found in fermenting must could be responsible for high alcohol content [7].

The organic acids present in wine were an important component in both winemaking and the finished product. They are present in both fresh mature and ripened fruits and even in produced wine, having direct influences on the color, balance and taste as well as the growth and vitality of yeast during fermentation and protecting the wine from pathogenic microorganisms [7, 8]. The measure of the amount of acidity in wine is known as the titratable acidity or total acidity, which refers to the test that yields the total of all acids present, while strength of acidity is measured by pH determination, with our wine having a pH between 2 and 4, which is similar to other researchers. Organic acids present in produced wine were responsible for the sourness and acidity of obtained wine [9]. Titratable acidity was an important indicator of quality of our yellow wine adding to the sour taste of the end product [9, 10]. Initially, obtained mixed juices had a weak acid (about 5.5 of pH), then after fermentation the acidity increased. This increased acidity and flavor is a normal process of secondary metabolites produced in fresh wine leading to the improvement of obtained wine [10].

Dissociable organic acid content of the mixed juices, fermenting must, and wine after 12 days yielded reliable results. Thus, organic acid content is important in the flavor, stability, sourness, body and shelf-life of the wine. This procedure determines the amount of organic acids that can be titrated with a dilute alkali solution (NaOH). Generally, the lower the pH, the higher the acidity in the wine produced. In yellow wine, the titratable acidity was 1.4% as organic acids present in produced wine. Also, in wine analysis, titratable acidity can be reported as grams per liter of tartaric acid [9, 10].

Volatile compounds are produced during fermentation for the best aroma of wine. Different volatile compounds are mainly phenol, alcohols, fatty acids, and esters. It was noted that the produced wine was accepted by panel members. Organoleptic properties were very good in fermented yellow wine produced from cape gooseberry juice fermented with *Saccharomyces cerevisiae* imported from European countries, such as Turkey (Tulip) [6]. The golden wine produced from a mixture of three ripened fruits, such as passion, mango and pineapple, had an enhanced flavor due to maturity of used fruits. The nutrients from the fruits allowed better multiplication of the wild yeast used as starter culture during fermentation.

Production of strong yellow wine, which has 11-14% alcohol by volume, was due to the high fermentable sugar content (85g/l). These monosaccharides (sugars) were converted into ethanol and carbon dioxide leading to reduction of total soluble solids in wine [9].

$C_6H_{12}O_6 \rightarrow 2\ CO_2 + 2C_2H_5OH$, this is the alcoholic fermentation formula.

This above fermentation process is an exothermic process and releases heat energy during degradation of sugar. Moreover, a lower temperature is desirable because it increases the production of esters, other aromatic compounds (flavor enhancement), and alcohol itself [9]. The total soluble solids and specific gravity gradually diminished with the degradation of sugars during the fermentation process leading to the sharp increase of alcohol, organic acids and flavoring compounds [10].

CONCLUSION

The quality of obtained yellow wine was a wholesome alcoholic and delicious beverage comparable to imported grape wines. The ideal final pH of yellow/golden wine should be between 3.0 and 3.3. The high content of fermentable sugars in mixed juice extracted from studied Rwandan fruits affects positively the alcohol content of the yellow wine. Thus, the obtained golden wine should be promoted for exports and rural development purposes.

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Table 1: Physical and chemical Parameters of mixed juices (must) during fermentation process

Time in Days	Yeasts population (Log CFU/ml): *Saccharomyces cerevisiae*	Total Soluble Solids (^{0}Brix)	pH	Alcohol Content in %(V/V)	Titratable Acidity as Citric Acid (%)	Fermentation Temperature oC	Sugar Content (g/l)	Specific Gravity
1	3	20	5.5	0.0	0.68	20	85	1.040
2	4	19	5.4	3.2	0.70	20	76	1.036
3	5	18	5.2	4.6	0.72	20	64	1.034
4	6	16	4.5	5.4	0.74	20	62	1.033
5	7	13	4.1	6.6	0.75	20	54	1.032
6	9	11	3.9	8.2	0.77	20	46	1.030
7	10	8	3.8	9.5	0.78	20	40	1.026
8	12	6.4	3.6	10.8	0.82	20	38	1.018
9	13	4.2	3.5	11.4	1.00	20	36	1.010
10	14	3	3.4	11.6	1.14	20	35	1.008
11	15	2.4	3.3	11.8	1.20	20	34	1.006
12	18	2.0	3.2	12.0	1.40	20	32	1.002

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Table 2: Sensory Evaluation of Yellow Wine obtained from Mixed Juices: Distribution of Responses on 9-point Hedonic Scale

Serial Number (S/N)	Description of 9 points of Hedonic scale	Assigned Hedonic Value	Frequency of Responses for wine sample testing by screened and trained panel members
1	Like extremely	9	11
2	Like very much	8	21
3	Like moderately	7	8
4	Like slightly	6	0
5	Neither like nor dislike	5	0
6	Dislike slightly	4	0
7	Dislike moderately	3	0
8	Dislike very much	2	0
9	Dislike	1	0
10	Total responses		40
11	Mean rating		8.08
12	Standard deviation		0.7
13	% Dislike responses		0.0
14	Observation		Wine acceptance was very good

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