

CIDER PRODUCTION FROM GUAVA (*Psidium guajava* L.) BY USING *Saccharomyces cerevisiae* HG1.3

Doan Thi Kieu Tien, Dinh Hoang Lan Chi, Huynh Thi Ngoc Mi*, Lam Nhu Y

Can Tho University of Technology

ARTICLE INFO	ABSTRACT
Received: 13/6/2023 Revised: 05/01/2024 Published: 03/02/2024	Fermented fruit juices have become a popular beverage around the world due to its low alcoholic concentration and benefits. Besides, the natural flavour and bioactive compounds of fruits are not significantly changed after fermentation. Hence, the study on fermented beverages from fruit juices as ciders is increasing a great interest in many food industries. This study aims to optimize the conditions for the fermentation process of guava juice by using <i>Saccharomyces cerevisiae</i> HG1.3, including soluble solids concentration, pH value, yeast concentration, fermentation time, and the supplemented pectinase concentration. In order to evaluate the product, the physicochemical, sensory, and microbiological criteria were also investigated. The results show that the recovery efficiency of the product is highest (86.7%) by the addition of 0.3% enzyme pectinase. The optimal fermentation conditions include 72 hours of incubation time, 1% w/v yeast concentration, pH 4.6 and 16 °Brix. The product has an ethanol content of 5.00% v/v, a good sensory score (4.3 points), and the microbiological criteria meet Ministry of Health standards QCVN 6 - 3:2010/BYT.
KEYWORDS	
Alcoholic fruit juice Ethanol Fermentation process <i>Psidium guajava</i> L. <i>Saccharomyces cerevisiae</i> HG1.3	

NƯỚC UỐNG LÊN MEN TỪ QUẢ ỔI (*Psidium guajava* L.) SỬ DỤNG NẤM MEN *Saccharomyces cerevisiae* HG1.3

Đoàn Thị Kiều Tiên, Đinh Hoàng Lan Chi, Huỳnh Thị Ngọc Mi*, Lâm Như Ý

Trường Đại học Kỹ thuật - Công nghệ Cần Thơ

THÔNG TIN BÀI BÁO	TÓM TẮT
Ngày nhận bài: 13/6/2023 Ngày hoàn thiện: 05/01/2024 Ngày đăng: 03/02/2024	Ngày nay nước uống trái cây lên men càng được ưa chuộng do có chứa hàm lượng thấp alcohol và vẫn giữ được hương tự nhiên của trái cây, vì vậy việc nghiên cứu chế biến các loại nước uống từ trái cây lên men ngày càng được quan tâm nhiều hơn. Nghiên cứu này nhằm mục đích khảo sát các điều kiện thích hợp trong lên men nước ổi sử dụng nấm men <i>Saccharomyces cerevisiae</i> HG1.3. Các điều kiện được khảo sát bao gồm hàm lượng chất tan ban đầu, giá trị pH, tỷ lệ nấm men, thời gian lên men, và tỷ lệ bổ sung enzyme pectinase. Bên cạnh đó, các chỉ tiêu về hoá lý, vi sinh vật và cảm quan sản phẩm sau khi lên men cũng được thực hiện. Kết quả cho thấy với tỷ lệ 0,3% enzyme pectinase ban đầu cho hiệu suất thu hồi dịch quả là 86,7%. Thời gian lên men thích hợp là 72 giờ với 1% tỷ lệ nấm men, dịch trái được điều chỉnh về 16 °Brix, pH 4,6. Sản phẩm sau lên men có hàm lượng ethanol là 5,0% v/v và cảm quan rất đặc trưng về mùi, vị của nguyên liệu trái ổi. Ngoài ra, các chỉ tiêu vi sinh vật của sản phẩm cũng đạt yêu cầu theo quy định QCVN 6 - 3:2010/BYT của Bộ Y tế.
TỪ KHÓA	
Enzyme pectinase Nấm men Nước ép ổi <i>Psidium guajava</i> L. <i>Saccharomyces cerevisiae</i> HG1.3	

DOI: <https://doi.org/10.34238/tnu-jst.8115>

* Corresponding author. Email: sunny2312@cau.ac.kr

1. Introduction

Fermented fruit juices have become a common beverage as per the preferences of consumers around the globe. Partial fermentation of musts from fruits by using yeasts creates a reduced-alcohol wine or fermented beverage, which contains ethanol from 1.2 to 6.5% v/v and the natural flavor of the original juice [1], [2]. In Vietnam, fermented beverages from varieties of tropical fruits have been studied and commercially introduced such as fermented fruit juices from *Selenicereus undatus* [3], *Limonia acidissima* [4], and *Docynia indica* [5].

With pleasant flavor and taste, guava (*Psidium guajava* L.) is a potentially valuable resource for the production of fermented fruit beverages. Guava, which belongs to the *Myrtaceae* family, is a major fruit crop in tropical and subtropical regions such as Mexico, Central and South America, Europe, Africa, and Asia [6]. In Vietnam, guava has been cultivated as a meadow orchard or an intercrop among other plantation crops [7]. Considered one of the important crops in Vietnam, the guava fruit growing areas in 2021 increased up to 23,100 hectares [8]. Guavas, mangos, and mangosteens (export code: HS080450) have been exported to the EU up to 350,000 tons, which was worth more than 600 million USD during the first half of 2021 [9]. Guava fruit has a berry form, thick mesocarp, and many small hard seeds [10]. It is a rich source of essential nutrients such as calcium, phosphorous, potassium, and vitamins (A, B C) [11]. The vitamins (A, and B) of guava enhance antiviral and anticancer activity [12]. Parts of the guava tree have been used in traditional medicine, including buds, young leaves, fruits, root skin, and stem [10].

In general, species of guava are rich in nutrients and health benefits. From fresh guava fruits, secondary guava processed products help not only to exploit the food values of this crop but also to increase the farmer's income. In Vietnam, guava juice fermented with *Saccharomyces cerevisiae* STH has a good taste and quality [13]. Yeast strains have a critical impact on fermented juice profile in terms of end-product performance and sensory perception [14], [15]. Thus, to diversify fermented beverages from guava fruits, the main objective of this research is to optimize the fermentation conditions of guava cider by using *Saccharomyces cerevisiae* HG1.3.

2. Materials and Methods

2.1. Materials

Guava fruits were purchased from local markets of Phong Dien district, Can Tho city. The fruits were stored at a cold temperature for freshness and pre-treated according to Le [16]. Particularly, the guava fruits were washed under tap water and removed then the pedicels and seeds after draining with distilled water. Thereafter, the guava flesh was pressed by a juicer (Philips HR1811, Netherlands), and the physicochemical parameters were measured including pH value (pH meter, Hanna, America), the soluble solids ($^{\circ}\text{Brix}$, ATAGO, 0 – 33 $^{\circ}\text{Brix}$, France), the total sugars content by using acid dinitrosalicylic (DNS). Besides, the moisture content of guava fruits was determined by a hot air convection drying oven (Mettler, German). These analytical data were used for further experimental optimization.

Saccharomyces cerevisiae HG1.3 [14] was enriched by culturing in YPD liquid (10 g/L yeast extract, 20 g/L peptone, 20 g/L D – glucose). Enzyme pectinase was purchased from Angel Yeast (Vietnam). Acid citric, CaCO_3 , NaHSO_3 , ethanol, peptone, and D-glucose were purchased from Xilong, China; dinitrosalicylic (DNS) reagent was purchased from Merk, German; and sucrose was provided by Bien Hoa Sugar CO. LTD, Vietnam.

2.2. Methods

The guava cider fermentation procedure is as follows: primary treatment of fresh guava fruits, juice pressing, pectinase supplement, optimization of fermentation conditions, and product evaluation (Figure 1).

2.2.1. Effects of the supplemented pectinase on guava juice yield

Guava juice was supplemented with enzyme pectinase at the concentration of 0.1; 0.2; 0.3; and 0.4%, respectively, and incubated for 2 hours at 50 °C. The juice was filtered, and the yield was calculated. The sample had no enzyme pectinase treated as a control. The highest yield sample was selected for further experiments.

2.2.2. Effects of the soluble solids and pH values on guava cider fermentation

The °Brix and pH values are two essential factors for yeast growth converting sugar into ethanol [6], [17]. The pre-treated guava juice (section 2.2.1) was diluted into the water with a ratio of 1:1. Subsequently, the samples were adjusted to 14; 16; and 18 °Brix, pH 3.8; 4.2; 4.6; and 5.0, respectively, and incubated at room temperature (28-30 °C) for 72 hours.

2.2.3. Effects of yeast concentrations and incubation times on guava cider fermentation

The selected pH and °Brix in section 2.2.2 were used to determine yeast concentration and incubation time in this experiment. NaHSO₃ was added into the adjusted guava juice and left off in 2 hours. Then, the samples were inoculated with *Sac* HG1.3 (0.5, 1.0, and 1.5% of concentration, respectively) and incubated at room temperature for 48, 72, and 96 hours, respectively.

2.2.4. Evaluation of the quality of guava cider

Guava cider was analyzed ethanol content, soluble solids content, pH value, total sugars content, and sensory evaluation in the criteria of clarity and color, aroma, taste, and overall confidence according to Vietnam National Standard 3217:79. The microbiological parameters including microaerophile, the content of yeast and mold, and *Escherichia coli* were conducted by National Agro-Forestry Fisheries Quality Assurance Department - Branch 6, Vietnam.

2.2.5. Statistical analyses

The analyzed data were graphed in Excel 2016 (Microsoft Inc., USA). Statgraphics Centurion version XIX-X64 (Statpoint Technologies Inc., USA) was performed for data analysis. The results are the average of triplicate $\pm$ SD.

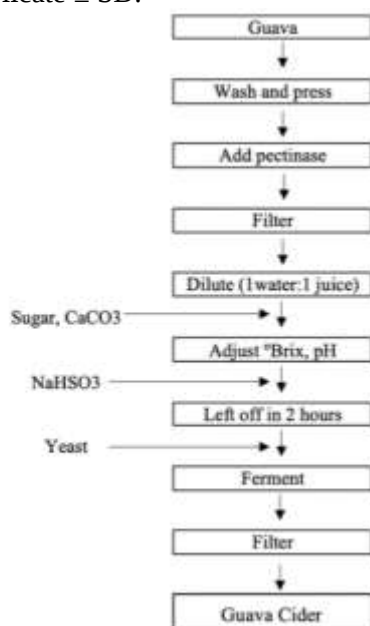


Figure 1. Flowchart of fermented guava juice procedure

3. Results and Discussion

3.1. Physicochemical characteristics of guava

The freshly purchased white guava fruits from local markets underwent pretreatment for subsequent experiments (Figure 2). It has a high moisture content of $90.73 \pm 0.11\%$ and pH of 4.36, creating a favorable environment for yeast growth. A previous study in India indicated that fresh guava contains 84.90% moisture content, pH 4.1, and 10.6% total sugars, acceptable for beverage fermentation [18]. However, the soluble solids content (8 °Brix) and total sugars (7.26%) of guava fruits in this study are quite low, therefore, the addition of sucrose is required to provide nutrition for yeast as well as the fermentation process [1].



Figure 2. *Guava (Psidium guajava L.)*

3.2. Effects of pectinase-assisted processing on guava juice extract

To increase the juicing efficiency, the fresh guava juice was pretreated with pectinase at 50 °C for 2 hours. Pectinase belongs to the polysaccharides family, a group of enzymes that breaks down pectin assisting to remove color, tannin, and soluble substrates, leading to the increase in juice yield [19]. Enzyme pectinase is extensively used in the food industry, particularly in fruit juice extraction and clarification [15], [19], [20].

The results show that the control treatment (without pectinase supplement) has a juice extraction efficiency of 62.60%, lower than that of pectinase treatments. In detail, the highest juice extraction efficiency is at 0.3% pectinase treatment induced 86.70 % juice extract, not significantly different compared to the 0.4% pectinase treatment (85.97%). According to Akesowan and Choonhahirun, 2013, the optimal pectinase of 869.36 ppm was added to obtain 85.10% guava juice in more than 2 hours [21]. Ideally, 0.3% pectinase addition is selected to enhance the extraction efficiency from guava fruits in this study. Due to the ability of pectinase to increase the clarity, stability, and quality of fruit juice extraction, it is an important step to incubate the fruit juices with pectinase for the fermentation process [19], [22].

3.3. Effects of soluble solids content and acidity conditions on sensory parameters of the fermented juice

In order to achieve the greatest quality of fermented juice, the initial soluble solids concentration and acidity were optimized for guava cider production. The effects of °Brix and pH value on the sensory parameters of the guava cider are shown in Table 1, with significant differences among sample groups at <5 % of statistics.

Guava cider, fermented at the initial conditions of 16 °Brix and pH 4.6, has the highest clarity and color point of 4.40, while 14 and 18 °Brix treatments are lesser evaluation scores for the products at 3.70, and 4.30, respectively. Moreover, the treatments at 16 °Brix, pH 4.6 produced guava cider with aroma and taste at 4.10 and 4.20 points, respectively. Meanwhile, 14 °Brix and pH 4.6 treatment resulted in clarity (3.7 points), aroma (2.9 points), and taste (2.9 points), lower than that of. The product differences among optimized conditions indicated that 16 °Brix is the greatest initial soluble solids content for yeast growth in this experiment. Additionally, the guava juice did not significantly change the natural taste, and the product has a mild taste with a low

ethanol concentration, an acceptable cider sample. These results might be explained by suitable conditions for the yeast to convert sucrose to ethanol during the fermentation process. Without optimized conditions, the high sugar concentration inhibits the yeast's fermentation ability due to high osmosis and physiological imbalance; while lower one leads to insufficient substrates for the yeast growth [19]. Regarding acidity condition, *S. cerevisiae* prefers a pH range from 4 to 6 [17]. Therefore, the pH of 4.6 along with 16 °Brix are selected for the fermentation of guava cider by Sac HG1.3 in this study.

Table 1. Effects of °Brix and pH levels on the sensory parameters

The initial °Brix	The initial pH	Sensory parameters		
		Colour and clarity	Aroma	Taste
14	3.8	3.90 ^{bcd}	2.90 ^a	2.80 ^{ab}
14	4.2	3.70 ^{abc}	2.90 ^a	2.70 ^a
14	4.6	3.70 ^{abc}	2.90 ^a	2.90 ^{ab}
14	5.0	3.50 ^a	2.80 ^a	2.70 ^a
16	3.8	3.90 ^{bcd}	3.50 ^b	3.00 ^{ab}
16	4.2	4.20 ^{def}	3.00 ^a	3.20 ^{bc}
16	4.6	4.40 ^f	4.10 ^c	4.20 ^e
16	5.0	4.00 ^{cde}	2.90 ^a	3.10 ^{abc}
18	3.8	3.60 ^{ab}	3.00 ^a	2.70 ^a
18	4.2	4.20 ^{def}	2.70 ^a	2.90 ^{ab}
18	4.6	4.30 ^{ef}	3.80 ^{bc}	3.70 ^d
18	5.0	3.60 ^{ab}	3.50 ^b	3.50 ^{cd}
F-Ratio		2.50	4.55	4.36
V-Value		0.0264	0.0004	0.0006

Note: Different letters within the column demonstrate the statistically significant difference at 5%

3.4. Effects of yeast concentrations and times on physicochemical characteristics of fermented juice

Yeast is a crucial factor in the fermentation process that decides the quality of alcoholic beverages such as fruit ciders, herb cider, wine, and so on. Guava juice was adjusted to pH 4.6, and 16 °Brix to survey the yeast concentration and fermentation time for guava cider making, the results are shown in Table 2.

At 72 hours of incubation time, the 1% yeast concentration produced 5.00% v/v ethanol, 10 °Brix, and pH 3.97 in the final product, lower than compared to three-leaf wine fermentation in the previous study (12°Brix, and pH 3.51), because of the different fermentation conditions [14]. The residual soluble solids create a mild sweetness for guava cider products. In addition, pH 3.97 is a typical acidity for fermented cider that meets the food safety demand.

Table 2. Effects of the yeast concentrations and incubation time on physicochemical contents of fermented juice

Fermentation time (hr)	Yeast concentration (%)	Parameters			
		Residual soluble solids (°Brix)	pH	Ethanol (%v/v)	Total sugars (%)
48	0.5	11.67 ^e	4.27 ⁱ	1.00 ^a	8.56 ⁱ
48	1.0	10.33 ^d	4.06 ^h	3.00 ^b	7.86 ^h
48	1.5	10.00 ^d	3.91 ^d	4.17 ^c	7.24 ^g
72	0.5	10.33 ^d	4.03 ^g	3.00 ^b	6.82 ^f
72	1.0	10.00 ^d	3.97 ^f	5.00 ^d	6.13 ^e
72	1.5	9.00 ^c	3.76 ^b	6.33 ^e	5.57 ^d
96	0.5	9.00 ^c	3.94 ^e	5.17 ^d	4.72 ^c
96	1.0	7.67 ^b	3.78 ^c	6.33 ^e	3.77 ^b
96	1.5	6.00 ^a	3.65 ^a	8.00 ^f	3.41 ^a
F-Ratio		525	29.80	4.87	3.26
V-Value		0.0056	0.0000	0.0077	0.0354

Note: Different letters within the column demonstrate the statistically significant difference at 5%

Different yeast concentrations and incubation times were studied for optimal conditions at room temperature. The juice products resulting from these treatments had diverse ethanol contents as the following: 17% v/v (48 hr, 1.5% yeast inoculum level), 5.00% v/v (72 hr, 1% yeast inoculum level), and 5.17% v/v (96 hr, 0.5% yeast inoculum level). The inoculated samples with 1% yeast and 72 hr of fermentation produced the most palatable fruit juice due to its great clarity and flavor. *Sac* HG1.3, a thermotolerant yeast (up to 37 °C), was able to produce 9.9% v/v of ethanol content in threeleaf wine at 35 °C [14]. In this study, *Sac* HG1.3 was able to ferment guava juice at room temperature which produces cider containing twice lower- ethanol content. *S. cerevisiae* is one of the typical yeasts used in apple cider fermentation. In Europe and North America, this yeast fermented pressed juice from apples at 15-25 °C, however, 25 °C is an ideal temperature [22]. These findings show that room temperature is the appropriate temperature for low ethanol juice products.

Guava fruit juice was slowly fermented for 72 hours because of its round shape, the cell surface display is rather slow which can affect nutrients absorption time and sugar transformation during the fermentation process [14]. At the condition of 72 hr and 1% yeast inoculum level, ethanol content was produced at 5% v/v, similarly to wood apple ciders (5:6% v/v) [4]. Furthermore, the soluble solids and total sugars were 10 °Brix and 6.13%, respectively, producing a specific sweetness, pleasant aroma, and acceptable beverage. In conclusion, *Sac* HG1.3 is a potential yeast strain for the production of guava cider with a low ethanol level of 5% v/v, fermented at room temperature for 72 hrs.

3.5. Effects of yeast concentration and incubation time on the sensory features of guava cider

The different conditions of yeast inoculum level and incubation time were also investigated by the sensory evaluation of the fermented products such as colors and clarity, aroma, and taste according to TCVN 3215 – 79 standards [23] (Table 3).

Table 3. Effects of yeast concentrations and time on sensory parameters

Fermentation time (hr)	Yeast concentration (%)	Sensory parameters		
		Colors and clarity	Aroma	Taste
48	0.5	3.50 ^{abc}	3.30 ^{ab}	2.90 ^{ab}
48	1.0	3.30 ^{ab}	2.90 ^a	2.90 ^{ab}
48	1.5	3.30 ^{ab}	3.10 ^{ab}	3.20 ^{bc}
72	0.5	3.40 ^{ab}	3.10 ^{ab}	3.00 ^{ab}
72	1.0	4.60 ^d	4.10 ^d	4.30 ^d
72	1.5	4.10 ^{cd}	4.00 ^{cd}	4.20 ^d
96	0.5	3.20 ^a	2.90 ^a	3.60 ^c
96	1.0	3.90 ^{bc}	3.50 ^{bc}	3.50 ^c
96	1.5	3.80 ^{abc}	3.50 ^{bc}	2.70 ^a
	F-Ratio	8.80	4.57	2.59
	V-Value	0.0000	0.0022	0.0425

Notes: Different letters within the column demonstrate the statistically significant difference at 5%

Correspondingly, treatments 0.5%; 1%; 1.5% of yeast inoculum level in 48 hours resulted in no significant changes in product quality, the statistically significant difference is at 5%. The products have low ethanol content and no difference among treatments. At 96 hours, the fermented juice has typical color, clarity, light alcoholic content, mild sweetness, sustainable taste, and naturally fresh guava aroma. Among sample groups, 1% yeast inoculum treatment produced a product with the highest points of color and clarity, aroma, and taste of 4.6, 4.1, and 4.3 points, respectively. These results repeatedly demonstrated that *Sac* HG1.3 is an acceptable yeast strain for guava cider production at the controlled conditions of room temperature, 1% yeast, and 72 hrs incubation time with the initial soluble solids and pH value is 16 °Brix and 4.6, respectively.

3.6. Sensory evaluation of guava cider

The optimal conditions for the fermented guava cider process are 16 °Brix, pH 4.6, 1% yeast concentration, and 72 hours of fermentation time at room temperature. The products were finally evaluated for physicochemical characteristics expressed by the pH values, residual soluble solids (°Brix), ethanol content, and total sugars. The fermented guava juice contains 5.00% v/v ethanol content, similar to that of wood apple cider (5: 6% v/v ethanol content) [4], fermented pitaya juice (3.19% v/v ethanol content) [3], *Docynia indica* cider (5.2% v/v alcoholic content) [5]. The total sugars, soluble solids, and pH values were $6.05 \pm 0.02\%$, 10 °Brix, and 3.95, respectively.

The Vietnamese standard methods (TCVN 3215 - 1979) were applied for the sensory analysis of guava cider. As of the results, guava cider has typical characteristics of color (4.29 points), aroma (4.43 points), and taste (4.29 points) of the fresh guava fruits (Figure 3).



Figure 3. Guava cider with 5.00% v/v ethanol content

After fermentation, the fruit juice was distilled into glass bottles and pasteurized at 75 °C for 10 minutes, then left for cold preservation. The product quality was assessed for the presence of unexpected microorganisms by using Vietnamese standards for alcoholic beverages (QCVN 6 – 3:2010/BYT) [24]. The microorganism concentration of guava cider was reported in table 4. There are passive results for *E.coli*, implicating that guava juice is safe to consume. Additionally, the total aerobic microorganisms, and the total yeast and mold content are all <1. These results demonstrate the assurance of the product's quality since it is preventable from pathogenic and spoilage microorganisms.

Table 4. Microbiological criteria of guava cider

Criteria	Methods	QCVN 6–3:2010/BYT	Results*
<i>Escherichia.coli</i> (MPN/mL)	ISO 16649–2:2001	Absence	None
Total microaerophile (CFU/mL)	ISO 4833–1:2003	10^3	<1
Total yeast và mold (CFU/mL)	ISO 21527–2:2008	10^2	<1

(*) The results were analyzed by National Agro-Forestry Fisheries Quality Assurance Department - Branch 6, Vietnam.

In general, guava juice was fermented at room temperature by *Sac* HG1.3 to produce 5% v/v ethanol and pH 3.95 in the final product meets food safety requirements as per the absence of harmful microorganisms.

4. Conclusion

This study has achieved the objective of optimal conditions for alcoholic beverages fermented from fresh guava fruits at room temperature. The optimal conditions of guava cider production include 1% yeast inoculum level, 16 °Brix, pH 4.6, and 72 hours of incubation time at room temperature. Besides, the experimental results indicated that *Sac* HG1.3 is well-adapted, effective fermentation, and a suitable yeast strain for the production of fermented fruit juices from guava. These findings, thus, provided valuable information for the exploitation and application of natural yeast strains in fermentation techniques, which helps to create a yeast profile for the fruit processing industry.

REFERENCES

- [1] G. J. Pickering, "Low- and Reduced-alcohol Wine: A Review," *Journal of Wine Research*, vol. 11, no. 2, pp. 129-144, 2000.
- [2] N. L. d. Lerma, J. J. Moreno, and R. A. Peinado, "Partial fermentation of musts from Tempranillo dried grapes," in *Microorganisms in Industry and Environment*, A. Mendez-Vilas, Singapore: World Scientific, 2010, pp. 418-421.
- [3] T. N. M. Huynh and T. K. T. Doan, "Fermentation of Pitaya (*Selenicereus undatus*) using *Saccharomyces cerevisiae* RV100," (in Vietnamese), *TNU Journal of Science and Technology*, vol. 226, pp. 137-145, 2021.
- [4] V. M. Nguyen, T. T. Tran, and R. Thach, "Study on the specific characteristics of wood apple and their fruit juice fermentation processing," (in Vietnamese), *Can Tho University Journal of Science*, vol. 11, pp. 97-104, 2009.
- [5] D. H. Nguyen, T. L. H. Hoang, T. T. M. Hoang, and V. L. Nguyen, "Study on use *Saccharomyces cerevisiae* in making cider from *Docynia indica* fruit," (in Vietnamese), *Journal of Agricultural Science in Vietnam*, vol. 8, no. 69, pp. 89-93, 2016.
- [6] C. H. Vu, *Making wine at home*, Ho Chi Minh: Agriculture publisher (in Vietnamese), 2004, p. 112.
- [7] C. H. Vu, *Fruit planting in Vietnam*, Ho Chi Minh: Agriculture Publisher (in Vietnamese), 1999.
- [8] General Statistics Office. Achievements of the agriculture industry – a year summation [Online]. Available: www.gso.gov.vn. [Accessed Oct.19, 2022]
- [9] Ministry of industry and trade. Market news of agriculture, forestry and fisheries [Online]. Available: <https://moit.gov.vn/>. [Accessed Oct.19, 2022]
- [10] T. L. Do, *Medical plants in Vietnam*, Ha Noi: Medicine Publisher (in Vietnamese), 2004.
- [11] H. C. Pham, T. P. Le, and T. H. Vo, *Guava trees (Psidium guajava)*, Ho Chi Minh: Agriculture Publisher (in Vietnamese), 2007.
- [12] A. Murphy, *Guava Cultivation, Antioxidant Properties and Health Benefits*, Newyork: Nova Science Publishers, Inc, 2017.
- [13] T. V. Pham, T. H. Vu, and T. L. Tran, "Study of affect concentration of yeast *Saccharomyces cerevisiae* (STH) to quality fermentation guava juice," (in Vietnamese), *TNU Journal of Science and Technology*, vol. 112, no. 12/2, pp. 115-118, 2013.
- [14] T. K. T. Doan, H. D. L. Bui, T. H. Y. Vien, T. T. Ha, X. P. Huynh, and T. P. D. Ngo, "Selection of thermotolerant yeasts and application in wine production from three-leaf cayratia (*Cayratia trifolia* L.) in Hau Giang," (in Vietnamese), *Can Tho University Journal of Science*, vol. 54, pp. 64-71, 2018.
- [15] I. A. Merwin, S. Valois, and O. I. Padilla-Zakour, "Cider apples and cider-making techniques in Europe and North America," in *Horticultural Reviews*, J. Janick, vol. 34, pp. 365-415, 2007.
- [16] V. V. M. Le, Q. D. Lai, T. H. Nguyen, N. M. N. Ton, and T. T. T. Tran, *Food processing technology*, Ho Chi Minh: National University Press (in Vietnamese), 2011.
- [17] D. T. Luong, *Industrial enzymes*, Ha Noi: Science and Technology Publisher (in Vietnamese), 2009.
- [18] V. S. Khandare, D. P. Waskar, B. M. Kalalbandi, and S. M. Panpatil, "Antioxidant composition of guava (*Psidium guajava* L.) beverage blended with black-carrot juice," *Horticultural Sciences*, vol. 10, pp. 112-115, 2015.
- [19] P. Alimardani-Theuil, A. Gainvors-Claisse, and F. Duchiron, "Yeasts: an attractive source of pectinases-From gene expression to potential applications: a review," *Process Biochemistry*, vol. 46, no. 8, pp. 1525-1537, 2011.
- [20] R. S. Jayani, S. Saxena, and R. Gupta, "Microbial pectinolytic enzymes: a review," *Process Biochemistry*, vol. 40, no. 9, pp. 2931-2944, 2005.
- [21] A. Akesowan and A. Choonhahirun, "Effect of enzyme treatment on guava juice production using response surface methodology," *Journal of Animal and Plant Sciences* vol. 23, pp. 114-120, 2013.
- [22] D. L. Nguyen *et al.*, *Enzyme technology*, Ho Chi Minh City: Vietnam National University Press (in Vietnamese), 2004.
- [23] Ministry of Science and Technology "Food products sensorial analysis Method by frowningmark, Vietnam National Standard 3215-79," (in Vietnamese), Ha Noi, Viet Nam, 1979.
- [24] Ministry of Health "National technical regulation for alcoholic beverages, Vietnam National Standard QCVN 6 – 3:2010/BYT," (in Vietnamese), Ha Noi, Viet Nam, 2010.