



## Analysis of Sap and liquor from Nipa (*Nypa fruticans Wurmb.*) fruit stalk using adopted nipa sap closed collection vessel in Barangay Ipil and Barangay Villafranca, Gigaquit, Surigao del Norte

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**Key words:** Gigaquit Rhum, Nipa, Ethanol, pH, Salinity

### Abstract

The distilled ethanol/liquor from the naturally fermented Nipa sap (*Tuba* in local language), is locally referred to as *Laksoy*. Currently, the Gigaquit Agra Multi-Purpose Corp. of the Municipal Agriculture Office branded *Laksoy* as *Gigaquit Rhum* sold in 375ml Net Content with 80 Proof (40% Alcohol by Volume or ABV). The study analyzed Nipa sap and liquor in terms of ethanol (% ABV) and pH from the Nipa plants in the estuary of Barangay Ipil, Gigaquit, Surigao del Norte with Brackish Water and from the Nipa plants in the estuary of Barangay Villafranca, Gigaquit, Surigao del Norte with Seawater. Brackish Water is flowing in Barangay Ipil estuary. Seawater is flowing in Barangay Villafranca estuary. The adopted Nipa Sap Closed Collection Vessel was utilized preserving the components of the fresh Nipa sap such as Ethanol and pH for accurate measurement. Level of pH varies between Brackish Water and Seawater Nipa plants due to ongoing natural fermentation. After 5 days of Nipa sap fermentation, ethanol amount decreases and pH level also decreases which signifies the completion of the process of fermentation. Distilled ethanol/liquor content is higher from Nipa plants in Brackish Water with 37.35% ABV which is equivalent to 75.5 Proof than Nipa plants in Seawater. Distilled ethanol from Nipa plants in Seawater is more acidic. However, the Ethanol amount and pH levels in Gigaquit Rhum from Barangay Ipil and Villafranca does not vary statistically.

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## Introduction

Nipa palm (*Nypa fruticans* Wurmb) is a plant native to Southeast Asia. It is useful and widely distributed found in the mangrove forests, small lagoons, and estuaries (Tsuji *et al.*, 2011). It is the only member in the genus *Nypa* (Cui-Lim *et al.*, 2020). Nipa palm is spread throughout the Philippine archipelago and grows along rivers and estuaries affected by tides. It reduces the intensity of high-sea storms and high seas, maintains soil stability, and guards against erosion (Ibrahim *et al.*, 2022). Indigenous people residing in the coastal and estuarine areas used Nipa plants for various purposes. Matured leaves are used for roof thatching and wall-partitioning of dwellings. Edible parts of the plant are its young seeds and buds. The sap of the Nipa fruit stalk undergoes processes that enable sugar, vinegar, and distilled ethanol/liquor to be concentrated (Codas, 2020; Tsuji *et al.*, 2016). Nipa plant produces higher amount of ethanol than Sugar cane plant having 3,350-6,700 L/hectare/year (Tsuji *et al.*, 2011) and 14,300 L/hectare which is more than twice than that of the sugarcane (Puangpee and Chongkhong, 2016). The practice of acquiring distilled ethanol from Nipa has been present in the Philippines since the pre-colonial and Spanish colonial times (Codas, 2020). The Municipality of Gigaquit, Surigao del Norte, Caraga Administrative Region in Mindanao is popular for this practice since the locality has approximately 527 hectares of farmland for Nipa palms. The distilled ethanol/liquor from the naturally fermented Nipa sap (*Tuba* in local language), is locally referred to as *Laksoy* (Codas, 2020; Glorious Food, 2023). Currently, the Gigaquit Agra Multi-Purpose Corp. of the Municipal Agriculture Office branded *Laksoy* as *Gigaquit Rhum* sold in 375ml Net Content with 80 Proof (40% Alcohol by Volume or ABV). However, the base ingredient for making a traditional Caribbean Rhum is different from *Laksoy* although both are hard liquor/spirit beverages which have the range of 30-100 Proof, 15%-50% in ABV (Sugue, 2023). Both cannot be considered as wine beverages since liquor/spirits are higher Proof, which are classified below 15% ABV. Rhum is a sub-classification of Rum produced from freshly-pressed sugar cane (Petreycik,

2023) while the *Laksoy* is produced from Nipa sap (Codas, 2020; Glorious Food, 2023). In order to be fermented, sugarcane juice/molasses must be added with water and yeast for Rhum while there's no need to add yeast to the Nipa sap because of naturally existing yeasts and natural fermentation for *Laksoy*. Gigaquit Rhum and the traditional light rum are similar due to being colorless liquors. Light rum is sold around 80 proofs (40% ABV) in the United States (ABC Fine Wine & Spirits, 2022). However, for Gigaquit Rhum, undergo aging/maturation during the Nipa sap collection thru the use of traditional bamboo shingle collector and is produced through a single distillation process. The brand name, Gigaquit Rhum, highlights the similarities in production process between Nipa-based liquor and traditional Rhum. This resonates with consumers who are familiar with traditional Rhum but are intrigued by a unique variation made from Nipa Sap. Thus, it has a great economic potential when developed with a good strategy based on research (Ibrahim *et al.*, 2022).

Water in estuarine areas differ in salinity which can be determined in salt parts per million (PPM) and can be classified as fresh water with less than 1,000 PPM, Brackish Water also known as slightly and moderately saline water with 1,000 to 10,000 PPM, or Seawater also known as highly saline water with 30,000 to 40,000 PPM (Woodward, 2023; U.S. Geological Survey, n.d.). Middle-aged Nipa palm gives the best sap yield (Cheablum and Chanklap, 2020). Number of producing days and fruit stalk length are highly correlated with sap yield (Saka *et al.*, 2016). Madigal *et al.* (2020) developed a Nipa Sap Closed Collection Vessel that preserves the chemical properties of the sap and prevents acceleration of fermentation which yields higher amounts of ethanol. Optimization of Nipa sap for ethanol production was studied by Puangpee and Chongkhong (2016) which researchers determined that the optimum total sugar of the Nipa sap 411 g/L maximum obtained at the initial pH of 4.9 and temperature of 54 °C for 25 minutes. In the study of Limtong *et al.* (2020), the yeast present in Nipa Palm sap was determined which is *S. cerevisiae* with 100% occurrence frequency and this yeast could play

roles in the natural fermentation of the sap. *S. cerevisiae* produced higher ethanol content than *P. kudriavzevii* (Madigal, 2019). Natural fermentation consumes glucose present in the Nipa sap ranging from 3.4 – 18.8% resulting in whitish color and unpleasant smell (Mateo *et al.*, 2023; Jaraee *et al.*, 2023; Puangpee and Chongkhong, 2016). The production of Nipa Sap has a considerable impact on the consumptive water use of Nipa palms. It was found to be between 893 and 1573 L water/L ethanol, with 90% originating from farms in Nipa. Cui-Lim *et al.* (2020) studied the presence of a significant amount of distilled ethanol from a fermented Nipa palm fruit through the Litmus, Ester, Iodoform and Flammability test. Ethanol is present after 72 hours of fermentation in all of the tests previously mentioned. During the pot still distillation of the fermented Nipa sap, Ethanol gets evaporated first at 78.4 °C followed by Water at 100 °C. Cooling the alcoholic vapor results to concentrated ethanol distillates with higher alcohol concentration (The Alcohol Pharmacology Education Partnership, n.d). The analysis of Nipa Sap and Rhum from Nipa plants situated on estuarine areas in Gigaquit, Surigao del Norte with difference in salinity was not has not yet been studied.

This study analyzed Nipa sap and liquor in terms of ethanol (% ABV) and pH from the Nipa plants in the estuary of Barangay Ipil, Gigaquit, Surigao del Norte with Brackish Water and from the Nipa plants in the estuary of Barangay Villafranca, Gigaquit, Surigao del Norte with Seawater. The data from this study will be used as a basis for the standardization of the production of Gigaquit Rhum.

Explicitly, this study sought to answer the following questions.

1. What is the average salt part per million (PPM) and pH level in the estuarine Nipa plantation sampling area in Barangay Ipil and Barangay Villafranca?
2. What is the design of the Nipa Sap Closed Collection Vessel adopted from Madigal *et al.* (2020) used in the current study?
3. Is there a significant difference in the amount of Ethanol and pH level of the sap samples from Nipa plants in brackish and Seawater?
4. Is there a significant difference in the amount of Ethanol and pH level of the sap samples after 3 days (72 hours)?
5. Is there a significant difference in the amount of Ethanol and pH level of the liquor samples from Nipa plants in brackish and Seawater?
6. What is the average alcohol by volume (ABV) and proof of distilled Ethanol from the sap of Nipa plants in brackish and Seawater?

#### *Hypotheses*

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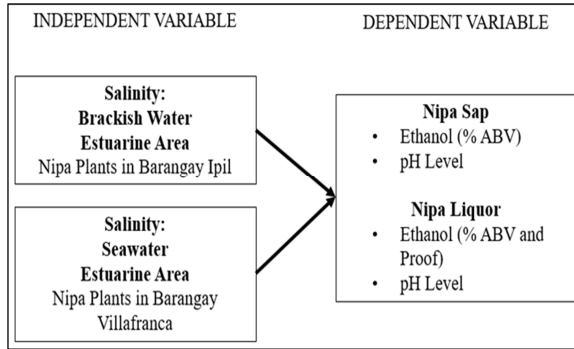
1. There is no significant difference in the amount of Ethanol and pH level of the sap samples from Nipa plants in brackish and Seawater.
2. There is no significant difference in the amount of Ethanol and pH level of the sap samples after 3 days (72 hours).
3. There is no significant difference in the amount of Ethanol and pH level of the liquor samples from Nipa plants in brackish and Seawater.

Hi:

1. There is a significant difference in the amount of Ethanol and pH level of the sap samples from Nipa plants in brackish and Seawater.
2. There is a significant difference in the amount of Ethanol and pH level of the sap samples after 3 days (72 hours).
3. There is a significant difference in the amount of Ethanol and pH content of the liquor samples from Nipa plants in brackish and Seawater.

#### *Conceptual framework*

Nipa palms are aquatic plants (CABI, 2019), wherein the sap samples are harvested from estuarine areas. It is the only palm found in a mangrove forest and is adapted to muddy soils along rivers and estuaries favoring Brackish Water environments (Nwobi, 2020; Tsuji *et al.*, 2011). According to the National Oceanic and Atmospheric Administration (2023), an estuary is a body of water where the river meets the sea affected by tides. Both Barangay Ipil with Brackish Water and Villafranca with Seawater are situated in an estuarine area. These are the independent variables of the study.



**Fig. 1.** Conceptual framework of the study

Nipa sap contains various components such as Ethanol, pH, and Temperature (Fig. 1). Ethanol is a solvent, clear colorless liquid with vinous odor and pungent taste (National Center for Biotechnology Information, 2023) which in this study was measured in Alcohol by Volume (ABV) and Proof. Proof is twice the percentage of ABV defined by policymakers in the United States (Richter, 2023) which is also used here in the Philippines. It is commonly produced by fermentation of a glucose (which is a carbohydrate). The more the total sugar content, the greater the amount of ethanol is produced (Fawad *et al.*, 2019) It was demonstrated by Theerawitaya *et al.* (2014) the Nipa pam grown in Brackish Water or Seawater tend to produce more sugar through the adaptation of the Nipa plant in a salt stress condition. The Yeasts such as *S. cerevisiae* produces ethanol as it ferments the sugar (Madigal, 2019). The pH level of a sample is a measure of the acidity or basicity of a liquid solution (McCaffrey, n.d.). During the fermentation, the Nipa sap will be acidic in the process of producing ethanol (Saka *et al.*, 2016). The Nipa sap undergoes natural fermentation wherein fermentation temperature changes with ambient temperature (Zhao and Wei, 2021). When the temperature increases, the enzymes in yeasts speed up which speeds up the rate of fermentation (Taylor, 2023). In order to distill ethanol, Pot Still method was used for distillation process. Alcohol by volume (ABV) is the measurement of the amount of alcohol in a given volume expressed in percent (%). The alcohol proof is twice the ABV. These are the dependent variables in the study.

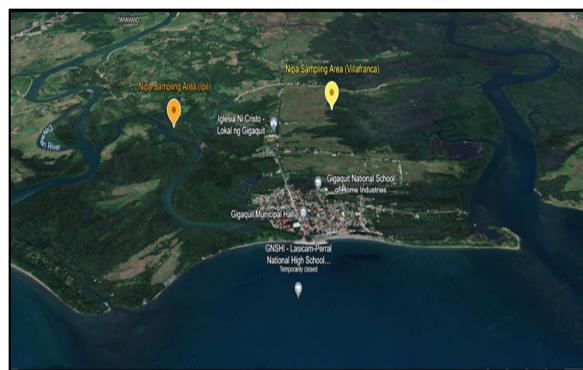
**Materials and methods**

*Research locale*

The study was conducted in the municipality of Gigaquit, Surigao del Norte. The locality has approximately 527 hectares of farmland for Nipa palms and is famous for producing Gigaquit Rhum. Some of the residents anchored their livelihood on harvesting Nipa sap, also known as *tuba*, and producing Rhum. Most of them come from estuarine areas of Barangay Ipil and Barangay Villafranca. Nipa plants are thriving along the estuarine areas of Gigaquit, Surigao del Norte. The distance between the two (2) sampling areas is 1.46 km. Sampling area in Barangay Ipil with coordinates is 9.5830420, 125.7084218 has a distance from Daywan River (Freshwater) by 1.08 km and 1.88 km from the entrance of the estuary in the coastal area of Gigaquit (Fig. 2). Sampling area (Fig. 3) in Barangay Villafranca with coordinates 9.5854956, 125.6893688 has a distance from the entrance of the estuary of 1.56 km (Google Earth, n.d.).



**Fig. 2.** Map of Barangay Ipil and Barangay Villaflor in Gigaquit, Surigao del Norte



**Fig. 3.** Location of sampling areas

#### *Research design*

The study utilized Descriptive Quantitative Research Design because the study aims analyze the difference on the amount of Nipa sap components such as Ethanol, pH and Temperature from the Nipa plants in the estuary of Barangay Ipil and Barangay Villafranca, Gigaquit, Surigao del Norte and analyze the difference on the amount of Nipa liquor components such as Ethanol, pH and Temperature.

#### *Target beneficiaries*

The findings of this study would benefit the following: *Municipal Local Government Unit (MLGU) of Gigaquit*

The result of this study will help the Municipal Agriculture Office, especially on standardizing the production of Gigaquit Rhum by optimizing the right raw materials and procedures. In this way, Gigaquit Rhum will be patented and will be an official entry of Gigaquit in One Town, One Product (OTOP) Philippines which is a program of the Department of Trade and Industries.

#### *Residents*

The people who are traditional Gigaquit Rhum makers, those who harvested Nipa sap (*Tuba*) locally called *Mananggete*, and the Gigaquit Wine Producers Association will benefit from a great economic potential of the patented Gigaquit Rhum in the future for the said product will be in demand and be renowned nationwide and overseas and increase their funds from the MLGU. The residents of the Gigaquit will have access to a premium quality of Gigaquit Rhum.

#### *Researchers*

This study may give significant contribution as a reference on their related study and give further clarification and answer their questions. Current and Future researchers will benefit from the findings of the study. The data from this study can be used for related studies on this topic.

#### *Sampling procedures*

300 milliliters (mL) water grab samples enclosed in plastic containers from sampling areas in the estuary of Barangay Ipil and Barangay Villafranca consisting of three 100ml of water samples in each area were

collected underwater during high tide (Singh, 2015). Water samples were filtered using a 40 centimeters (cm) fabric filter. Fresh Nipa sap samples were harvested from middle-aged Nipa palm fruit stalks which are randomly selected by the harvesters. Each Nipa fruit stalk undergoes cleaning for 3 days in which the outer layer of the stalk is removed and afterwards was subjected to 30 days of massage through soft blows using an improvised rubber mallet locally known as *takdug* which is made out of wood and approximately 4pcs of rubber slippers. A nipa fruit stalk received 50 to 100 soft blows from the mallet per week which was repeated 4-5 times within 30 days. Then, Nipa fruit can be cut and removed from the stalk. 2-3 thin slices are removed in the peduncle of the stalk every morning and afternoon for the sap to flow for 3 days. Then, after 3 days, the first flow Nipa sap, also known as *kutub*, which is the sweet Nipa sap which is set aside and used for making vinegar was collected by the harvester using the bamboo shingles, locally known as *sayod*, within 2 days. Afterwards, the next flow of Nipa sap, also known as *paitan*, which is a bittersweet Nipa sap used for making rhum was collected using the Nipa Sap Closed Collection Vessel adopted from Madigal et al., (2020) to reduce the chances of sap leak, foreign materials and reducing rate of fermentation in the during the sap collection and harvest. 480 Liters (L) or 120 Gallons (Gal) of Nipa sap was collected from approximately 150 randomly selected middle-aged Nipa palms which have long fruit stalks, known as *sanggutan*, and hollow Nipa fruit, known as *guwang na pungo*, along the estuary of Barangay Ipil and Barangay Villafranca. The sap collected from both sampling areas was placed in 20 plastic containers each with the capacity of 24 Liters or 6 Gallons kept at room temperature. Each container is a combination of Nipa sap from approximately 28-30 Nipa palms. 5 Nipa sap samples of 200 mL from randomly selected 5 Nipa sap containers. After the Nipa sap collection and natural fermentation, distillation followed to concentrate ethanol using the Pot Still machine at 78.4 °C resulting to 4 pcs of 24 L plastic containers filled with ethanol Nipa liquor/Gigaquit Rhum after 10 hours of distillation. 300 mL samples was collected from each container filled with distilled ethanol from both areas.

#### *Data collection procedure*

Three 100 mL water grab samples gathered from sampling area in the estuary of Barangay Ipil and Barangay Villafranca were subject to analysis using a Water Quality Multi-Functional Tester to collect data for salt PPM with  $\pm 0.01\%$  accuracy referring to the parameters for saline water by U.S. Geological Survey (n.d.) and pH level with with  $\pm 0.05\%$  accuracy referring to Water Quality Parameters and Indicators (McCaffrey, n.d.). 200 mL. Five 200 mL Nipa samples from both of sampling area were tested twice within 24 hours after a randomly selected container is full for alcohol by volume (ABV) using an Alcohol Refractometer with the scale range of 0-80% v/v (Alcohol volume percent), 1% v/v per division, in 20 °C with reference to typical proof ranges for common forms of hard alcohol (Sugue, 2023). The said samples were tested also for pH level using the Water Quality Multi-Functional Tester. Four 300mL samples of distilled ethanol from both sampling areas was tested thrice for ABV using an Alcohol Refractometer and Proof using Proof and Tralle Hydrometer for distilled beverages. The said samples were tested also for pH level using the Water Quality Multi-Functional Tester.

#### *Data analysis method*

The following statistical tools were used for the study.

##### *Measures of central tendency*

This tool was used for problem number 1 and 5 using Jamovi Software 2.4.11 under AGPL3 license (The jamovi project, 2023) to determine the average salt parts per million (PPM) and pH in the estuarine Nipa plantation sampling area in Barangay Ipil and Barangay Villafranca and to determine the average alcohol by volume (ABV) and proof of distilled Ethanol from the sap of Nipa plants in Brackish water and Seawater.

##### *Paired samples T-test*

This tool was used for problem 4 using Jamovi Software 2.4.11 under AGPL3 license (The jamovi project, 2023) to determine significant difference in the amount of Ethanol and pH level of the sap samples after 3 days (72 hours).

#### *One-way analysis of variance*

This tool was used for problem number 3, 5, and 6 using Jamovi Software 2.4.11 under AGPL3 license (The jamovi project, 2023) to determine the significant difference in the amount of Ethanol and pH content of the sap and liquor samples from Nipa plants in Brackish water and Seawater.

#### *Ethical considerations*

The researcher submitted a formal letter to the president of Gigaquit Wine Producers Association, Mrs. Helen C. Batac, and to the Municipal Local Government Unit of Gigaquit Hon. Mayor Chandru T. Bonite thru Municipal Agriculturist Mrs. Gilda Bersabal who is the in-charge of the Gigaquit Rum production in Municipal Agriculture Office for permission to conduct the study and to use the distillation machine.

### **Results and discussion**

#### *Average Salt parts per million (PPM) and pH in the Estuarine Nipa Plantation Sampling Area in Barangay Ipil and Barangay Villafranca*

Based on measuring of average Salt PPM in both sampling areas shown in Table 1, waters in the Barangay Ipil sampling area have an average of 4577 PPM. Since the parameters of salinity for Brackish Water is 1,000 to 10,000 PPM (Woodward, 2023; U.S. Geological Survey, n.d.), the Nipa plants in the sampling area of Barangay Ipil are thriving in brackish water. Waters in the sampling area in Barangay Villafranca have an average of 10986 PPM which suits the parameters of salinity for Seawater, also known as brine, ranging from 30,000 to 40,000 PPM (Woodward, 2023; U.S. Geological Survey, n.d.). As for the pH levels, Brackish Water in Barangay Ipil estuary sampling area was measured having 8.27 which is inside the range of 6.0 to 9.0 standard pH for brackish waters (Halat & Wood, 2023). The pH Level in the Barangay Villafranca is 7.77 which can be rounded up to 8.0 is also close to 8.0 to 9.0 standard pH range of Seawater (Halat & Wood, 2023). Results showed that the Seawater in the sampling area in Barangay Villafranca is more acidic than in Barangay Ipil sampling area brackish water.

**Table 1.** Measures of Central Tendency for Salt PPM and pH Level in Sampling Areas

	Sampling Area	Salt PPM	pH level
N	Barangay Ipil Estuary	3	3
	Barangay Villafranca Estuary	3	3
Mean	Barangay Ipil Estuary	4577	8.27
	Barangay Villafranca Estuary	10986	7.77
Median	Barangay Ipil Estuary	4577	8.28
	Barangay Villafranca Estuary	10916	7.80
Mode	Barangay Ipil Estuary	4478a	8.15a
	Barangay Villafranca Estuary	10847a	7.68a

<sup>a</sup>More than one mode exists, only the first is reported

*Design of the Nipa Sap Closed Collection Vessel adopted from Madigal et al. (2020)*

The study introduces the adoption of Nipa Sap Closed Collection Vessel shown in Fig. 3 adopted from Madigal *et al.* (2020) to reduce the chances of sap leak, foreign materials and reducing rate of fermentation during the sap collection and harvest. Each unit can accommodate one (1) Nipa fruit stalk peduncle. A 1.5 L PET bottle was used as the body of the vessel since it is able to contain the maximum sap yield within 24 hours. A 12 inch-sized balloon with random holes is directly placed in the bottle neck as a breather. Below the bottle neck, a hole was cut in custom depending on the diameter of the peduncle for an exact fit. In the study of Madigal *et al.* (2020), 0.865 L of Nipa Sap was the total average sap yield collected by the designed system which is 0.403 L greater than the traditional bamboo shingle collection system. The amount of ethanol during the collection of fresh nipa sap via the designed closed system has lower ethanol yield than fermented sap due to anaerobic respiration and the fermentation is not yet completely done, preserving the chemical properties of the sap. The traditional bamboo shingle collection method which is commonly used by the harvesters has a lot of disadvantages aside from the average sap yield such as spillage due to no definite size of the shingles, rodents infest drinking directly from the cut of the peduncle, and do not preserve the chemical properties of the Nipa sap. Figs 4 and 5 show the actual pictures of the Nipa Sap Closed Collection Vessels used in both sampling areas.

*Ethanol and pH Level of the Nipa Sap Samples from Nipa Plants*

Nipa sap samples were tested within 24 hours resulting in a higher amount of ethanol content of 20.60% ABV from Nipa plants thriving in Seawater as shown in Table 2.

**Table 2.** Group descriptive

	Salinity	N	Mean	Remarks
Ethanol (ABV)	Seawater	10	20.60	Higher Amount
	Brackish Water	10	19.20	Lower Amount
pH Level	Seawater	10	3.47	Less Acidic
	Brackish Water	10	3.31	More Acidic

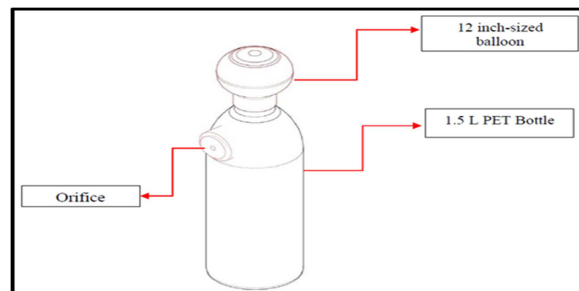
**Table 3.** Normality Test (Shapiro-Wilk)

	W	p	Remarks
Ethanol (ABV)	0.972	0.805	Significant
pH Level	0.959	0.528	Significant

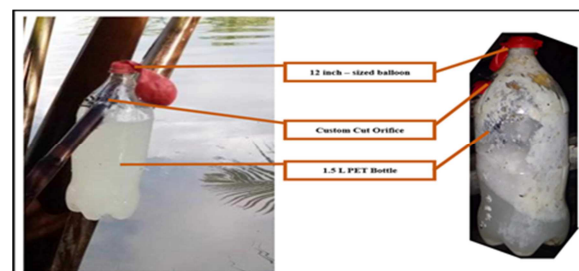
Note. A low p-value suggests a violation of the assumption of normality.

**Table 4.** Homogeneity of Variances Test (Levene's)

	F	df1	df2	p	Remarks
Ethanol (ABV)	0.475	1	18	0.500	Significant
pH Level	0.403	1	18	0.533	Significant



**Fig. 4.** Isometric View of the Sap Closed Collection Vessel of Madigal *et al.* (2020)



**Fig. 5.** Actual View of the adopted Closed Collection Vessel from Madigal *et al.* (2020) attached on Nipa Fruit Stalk peduncle in Barangay Ipil and Barangay Villafranca sampling area

19.20% ABV is the amount of ethanol of Nipa sap from Nipa plants in brackish water. In terms of pH level, Nipa sap from Nipa plants in the Brackish Water is more acidic by 3.31. During natural fermentation, yeasts convert sugars into ethanol, acetic and lactic acid which contributes to the acidity of the Nipa sap (Mateo *et al.*, 2023; Jaraee *et al.*, 2023; Puangpee and Chongkhong, 2016). Assumptions for conducting One-Way Analysis of Variance (ANOVA) were met with both variables being distributed normally and variables being compared are homogeneous (Table 3 and 4) of which Fisher's ANOVA was applied.

**Table 5.** Difference on Salinity when grouped according to Nipa Sap Components

	F	df1	df2	p	Remarks
Ethanol (ABV)	0.938	1	18	0.346	Not Significant
pH Level	5.420	1	18	0.032	Significant

Results revealed in Table 5, there was no significant difference in salinity when grouped according to ethanol (ABV) with  $F=0.938$  at  $p=0.346$  (greater than 0.05 level significance). As depicted by Tukey Post Hoc Test, ethanol's p-value is greater than 0.05 level of significance following that ethanol (ABV) in Nipa sap did not vary among Brackish Water and Seawater Nipa plants. However, there is a significant difference in salinity when grouped according to pH level, with  $F=5.420$  at  $p=0.032$ . Since p-value obtained in the Tukey Post Hoc Test is less than 0.05 level of significance, it follows that pH level of Nipa Sap varies between Brackish Water and Seawater Nipa plants. Presence of acidity in naturally fermented Nipa sap is an evidence that fermentation of the sap is ongoing (Saka *et al.*, 2016).

**Table 6.** Group descriptive

	N	Mean	Remarks
Ethanol (Day 1)	10	20.30	Higher Amount
Ethanol (Day 5)	10	16.50	Lower Amount
pH (Day 1)	10	3.40	Less Acidic
pH (Day 5)	10	3.24	More Acidic

**Table 7.** Normality Test (Shapiro-Wilk)

	W	p	Remarks
Ethanol (Day 1)- Ethanol (Day 5)	0.823	0.027	Not significant
pH (Day 1)-pH (Day 5)	0.954	0.718	Significant

Note. A low p-value suggests a violation of the assumption of normality

**Table 8.** Difference between Ethanol and pH Level on Day 1 and Day 3

	Statistic	p	Remarks
Ethanol (Day 1)- Ethanol (Day 5)	Wilcoxon W	45.0 <sup>a</sup>	0.009 Significant
pH Level (Day 1)- pH Level (Day 5)	Wilcoxon W	44.0	0.105 Not Significant

Note.  $H_a \mu$  Measure 1 - Measure 2  $\neq 0$ , <sup>a</sup> 1 pair(s) of values were tied

After 72 hours (3 days), all the samples were tested again resulting in a lower amount of ethanol (ABV) from the fermented Nipa sap of Nipa plants in both in Brackish Water estuary of Barangay Ipil and Seawater estuary of Barangay Villafranca with the mean of 16.30%. As for the pH level, it has become increasingly more acidic with the mean of 3.24 (Table 6) which signifies that the natural fermentation of Nipa sap at its completion stage wherein there is a decrease in ethanol content of fermented Nipa sap (Jaraee *et al.*, 2023) Some microorganisms present in the Nipa sap is able to metabolize ethanol back into other compounds such as acetaldehyde and Carbon dioxide through metabolic pathways (Madigal *et al.*, 2020; Alba-Lois, 2010). A low p-value suggests a violation of the assumption of normality (Table 7). Assumptions for Paired Samples T-test are not met since Ethanol (Day 1) and Ethanol (Day 5) p-value is less than 0.05 level of significance. Wilcoxon W Paired Samples T-test was applied (Table 8). There is a significant difference in Ethanol (Day 1) and Ethanol (Day 5) at  $p=0.0009$  which is less than 0.05 level of significance. However, the pH Level (Day 1) and pH Level (Day 5) of Nipa sap samples from both areas did not have significant difference at  $p=0.105$ .

**Table 9.** Group descriptive

	Salinity	N	Mean	Remarks
Ethanol (ABV)	Brackish Water	12	37.75	Higher Amount
	Seawater	12	36.75	Lower Amount
pH Level	Brackish Water	12	3.11	Less Acidic
	Seawater	12	3.08	More Acidic

*Ethanol and pH Level of the Liquor Samples from Nipa Plants*

Ethanol, locally branded as Gigaquit Rhum, was concentrated after undergoing pot still distillation of fermented Nipa sap (Codas, 2020; Tsuji *et al.*, 2016).



**Table 10.** Normality Test (Shapiro-Wilk)

	W	p	Remarks
Ethanol (ABV)	0.904	0.027	Not Significant
pH Level	0.949	0.528	Significant

Note. A low p-value suggests a violation of the assumption of normality.

**Table 11.** Homogeneity of Variances Test (Levene's)

	F	df1	df2	p	Remarks
Ethanol (ABV)	0.734	1	22	0.401	Significant
pH Level	0.271	1	22	0.608	Significant

Referring to Table 9, distilled ethanol from naturally fermented Nipa sap from Brackish Water Nipa Plants in Barangay Ipil was measured to be of higher average amount by 37.75% ABV. While 36.75% is the average ABV of distilled ethanol from naturally fermented Nipa sap from Nipa plants in Seawater estuary in Barangay Villafranca. In terms of pH Level, distilled ethanol from Seawater Nipa naturally fermented sap is more acidic than Brackish Water Nipa naturally fermented sap distilled ethanol by 3.08. The seasoned and traditional Gigaquit Rhum makers already noticed the slight difference via taste test. Assumptions for the homogeneity passed but normality was violated since Ethanol (ABV) p-value is 0.027 (Table 10) which is less than 0.05 level of significance. Kruskal-wallis was used in the One-Way ANOVA test (Table 11). There is no significant difference in Salinity when grouped according to Ethanol (ABV) at  $p=0.707$  and pH level at  $p=0.817$  (Table 12). Since the p-values are greater than 0.05 level of significance, it follows that Ethanol (ABV) and pH level did vary in different parameters of salinity.

**Table 12.** Difference between Salinity when grouped according to Nipa Sap components

	$\chi^2$	df	p	Remarks
Ethanol (ABV)	0.1418	1	0.707	Not Significant
pH Level	0.0534	1	0.817	Not Significant

*Average Alcohol by Volume (ABV) And Proof of Distilled Ethanol from the Sap of Nipa Plants in Brackish Water and Seawater*

Distilled ethanol from Nipa plants in Brackish Water have higher average ethanol content 37.35% ABV which is equivalent to 75.5 Proof than the distilled ethanol from Nipa plants in Seawater having 36.75% ABV which is equivalent to 73.5 Proof as shown in Table 13.

**Table 13.** Average Distilled Ethanol ABV and Proof

	Salinity	N	Average ABV	Average Proof
Ethanol	Brackish Water	12	37.75 %	75.5
	Seawater	12	36.75 %	73.5

Statistically proven earlier, these values have no significant difference. Gigaquit Rhum was advertised having 80 Proof but as the results shown below, the average Proof from both sampling areas didn't match as advertised. Despite of this, the Gigaquit Rhum still suits in the category in beverage as Rhum since Rum was set to be on the range of 70 to 100 proof (35% to 50% ABV).

**Conclusion**

The salinity of sampling area in Barangay Ipil and Villafranca estuary was verified by this study. Brackish Water is flowing in Barangay Ipil estuary in coordinates is 9.5830420, 125.7084218. Seawater is flowing in Barangay Villafranca estuary in coordinates 9.5854956, 125.6893688. The adopted Nipa Sap Closed Collection Vessel was utilized preserving the components of the fresh Nipa sap such as Ethanol and pH for accurate measurement. Level of pH varies between Brackish Water and Seawater Nipa plants due to ongoing natural fermentation. After 5 days of Nipa sap fermentation, ethanol amount decreases and pH level also decreases (increasing in acidity) which signifies the completion of the process of fermentation. Distilled ethanol/liquor content is higher from Nipa plants in Brackish Water with 37.35% ABV which is equivalent to 75.5 Proof than Nipa plants in Seawater. Distilled ethanol from Nipa plants in Seawater is more acidic. However, the Ethanol amount and pH levels in Gigaquit Rhum from Barangay Ipil and Villafranca does not vary statistically.

Implications of this study includes that the Municipal Agriculture Office must change the advertising of Gigaquit Rhum as having "70 to 80 Proof" than claiming to be of exactly 80 Proof, their current practice of separating the first distillate must not be separated from the rest of the distillate, their practice of mixing four 24 L of distilled alcohol in a steel storage tank with different ethanol amounts is

recommended to get the average ABV and Proof ranging from 70 to 80 Proof. Their current preference in their Nipa Sap is from Barangay Villafranca is still a good choice as they prefer to have more acidity in the Rhum. Despite the introduction of an adopted Nipa Sap Closed Collection Vessel, they can still use the Traditional Bamboo Shingle Collection System to add flavor and aroma to the Nipa sap.

Further researchers are recommended to develop a more sustainable and eco-friendly closed collection vessel for collecting the Nipa Sap, determine the amount of other components in Nipa sap collected from Traditional bamboo shingles and other sustainable / eco-friendly collector.

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