

# Volatile aroma compounds and sensory characteristics of traditional banana wine “Urwagwa” of Rwanda

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## Abstract

*Urwagwa*, produced mainly from the fermentation of banana juice, is the oldest and popular Rwandan traditional alcoholic beverage. In the present paper, the aroma profiles of *Urwagwa* wine samples collected from the districts of Rulindo and Ngoma were investigated. Headspace/ Solid-Phase Micro Extraction (HS- SPME) and gas chromatography - mass spectrometry (GC/MS) were applied for the analysis of volatile aroma compounds. Odour Active Values (OAVs) and sensory analysis were also performed to define the aromatic profile of *Urwagwa* wine. The findings showed that the aroma profiles of two types of *Urwagwa* wines analyzed were not significantly different. Forty eight volatile aroma compounds, including esters, higher alcohols, acids, terpenes, furan and phenol were identified and quantified in *Urwagwa* wines. Among them, ethyl caprylate, ethyl caproate, ethyl caprate, ethyl acetate, isoamyl acetate, ethyl acetate, ethyl butyrate, phenethyl acetate, phenethyl alcohol, caprylic acid, 1-octanol and isovaleric acid exhibited OAVs > 1, and are considered as the major contributors of aromatic character of *Urwagwa* wine; described as fruity, floral, banana, sweet and fatty notes. However, the overall aroma profiles of the investigated *Urwagwa* wines were dominated by the fruity note due to the high amount of ethyl caprylate, ethyl caprate and ethyl caproate in this Rwandan traditional banana wine.

**Keywords:** Aroma compounds; Sensory analysis; Banana wine, *Urwagwa*

## Résumé

*Urwagwa*, produite principalement à partir de la fermentation du jus de banane, est la boisson alcoolique traditionnelle rwandaise la plus ancienne et la plus populaire. Dans le présent article, les profils d'arômes d'échantillons de vin *Urwagwa* collectés dans les districts de Rulindo et de Ngoma ont été étudiés en utilisant la technique de Micro-extraction tête / espace solide (HS-SPME) et chromatographie en phase gazeuse - spectrométrie de masse (GC / MS) et valeurs actives d'odeur. Les résultats ont montré que les profils aromatiques de deux types de vins *Urwagwa* analysés n'étaient pas significativement différents. Quarante-huit composés aromatiques volatils, y compris les esters, les alcools supérieurs, les acides, les terpènes, le furane et le phénol ont été identifiés et quantifiés dans les vins *Urwagwa*. Parmi eux, le caprylate d'éthyle, le caproate d'éthyle, le caprate d'éthyle, l'acétate d'éthyle, l'acétate d'isoamyle, l'acétate d'éthyle, le butyrate d'éthyle, l'acétate de phénéthyle, le phénéthylène, l'acide caprylique, le 1-octanol et l'acide isovalérique sont les contributeurs du profil aromatique du vin *Urwagwa*; décrit comme fruité, floral, banane, notes sucrées et grasses. Cependant, le profil aromatique global de tous les vins *Urwagwa* étudiés était dominé par la note fruitée due à la grande quantité de caprylate d'éthyle, de caprate d'éthyle et de caproate d'éthyle dans ce vin de banane traditionnel rwandais.

**Mots-clés:** Composés aromatiques; Analyse sensorielle; Vin de banane, *Urwagwa*

## 1. INTRODUCTION

Aroma, one of the most important factors determining the character and quality of wine, is due to the combined effects of a great number of volatile compounds belonging to heterogenous chemical groups, including alcohols, aldehydes, esters, acids, terpenes and other minor components which already are present in the fruits or being formed during the fermentation and maturation process (Verzera *et al.*, 2008). According to legal definition, wine is the product obtained exclusively by alcoholic fermentation, total or partial, of fresh grapes, whether crushed or not, or grape must. However, in the new world, wine may refer to the fermented by-products of any fresh fruit or flower. Rwandan traditional wine, called generally *Urwagwa* (Kinyarwanda language), is produced mainly by alcoholic fermentation of juice extracted from special varieties of bananas, such as ‘*Indege*’, ‘*Inkati*’, ‘*intutu*’, ‘*kayuku*’, ‘*Gisukali*’ and ‘*Intokatoke*’ (Nsabimana and van Staden, 2007). Wine maker generally blend banana varieties in the recipe but some process single banana variety to make wine.

Volatile aroma compounds are perceived by the odour receptor sites of the smell organ, i. e. the olfactory tissue of the nasal cavity. They reach the receptors when drawn in through the nose (orthonasal detection) and via the throat after being released by chewing (retronasal detection). The concept of aroma substances, like the concept of taste substances, should be used loosely, since a compound might contribute to the typical odour or taste of one food, while in another food it might cause a faulty odour or taste, or both, resulting in an off-flavour. Hence, volatile aroma compounds are closely related to the product sensory profile which strongly impacts by the consumer’s acceptability (Vilanova, 2006; Varela and Gàmbara, 2006). Sensory analysis has defined its role in the oenological industry identifying the causes of variation of perceived quality, the corrective actions thereby becoming instrument of quality control of wines (Lawless 1995; Muñoz, 2002).

*Urwagwa* is a popular alcoholic beverage in Rwanda and play an important role in fulfilling social obligations (e.g. marriage, birth, baptism, etc.) and constitutes a significant source of income for the manufacturers and for national economy of Rwanda (Mukantwali et al., 2008). The manufacturing process of this traditional alcoholic beverage varies according to the know-how of each region of Rwanda and raw materials, thus leading to the production of different *Urwagwa* types in terms of alcohol content, colour, taste and shelf life (Munyangendo, 1983). Traditional process for the production of *Urwagwa* involves generally five mainly steps: ripening of green banana in warm pit covered with banana leaves and/or eucalyptus leaves (called *Urwina* in Rwandan language), peeling, banana juice extraction (mixing of ripe banana with spear glass, squeezing the mixture with their feet or hands), filtration of juice through grass held in calabash funnel and spontaneous fermentation. The fermentation process requires the addition of coarsely-ground, roasted sorghum malt and/or granulated sugars into the diluted banana juice, in a canoe-shaped wooden container known as “*Umuwure*” or in a clay pot (Shale et al., 2012, 2014), to improve the fermentation process, colour and flavour of final products.

Most studies conducted on Rwandan traditional banana wine have been focused to the production process, improvement of banana juice extraction methods, isolation and characterization of yeast strains involved in the fermentation (Munyangendo, 1983, Mukantwali et al., 2008, Shale et al., 2012), but the aroma profile of banana wine has not been yet fully investigated. The present work aimed to define the aroma profile of traditional banana wine “*Urwagwa*” from Rwanda. Headspace/ Solid-Phase Micro Extraction (HS-SPME) and gas chromatography - GC-mass spectrometry (GC/MS) analysis were applied to determine the volatile aroma compounds. Odour Active Values (OAVs) were assessed, already successfully used for determining the contribution of each volatile compound as wine

aroma. Sensory analyses were performed to define the overall flavor profile of *Urwagwa* wine.

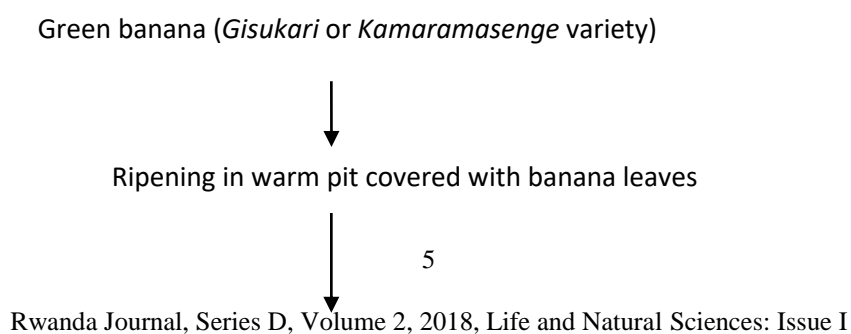
## 2. MATERIALS AND METHODS

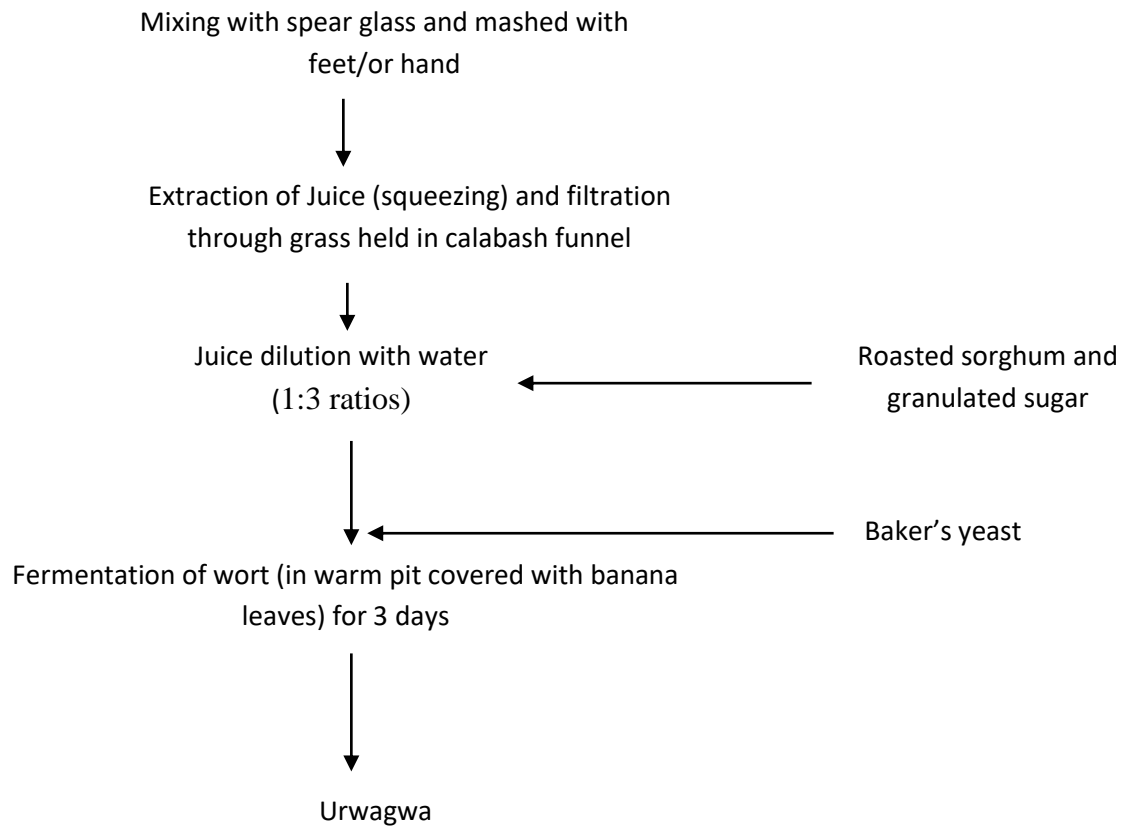
### 2.1. Samples collection

Twenty samples of traditional banana wine “*Urwagwa*” packaged in the polyethylene bottles (33 cL) were collected from Rulindo and Ngoma districts of Rwanda. The banana wines collected in the Rulindo district were made from *Gisukari* banana variety while those from Ngoma district were made from *Kamaramasenge* banana variety. The samples were stored in refrigerator at 5 °C and then analyzed after 5 days of the production. The production process of these samples is described in **Figure 1**.

### 2.2. Reagents

The pure reference compounds (Phenethyl alcohol, 1-hexanol, 1-propanol, isobutyl alcohol, isoamyl alcohol, ethyl acetate, ethyl butyrate, ethyl caproate, ethyl caprate, ethyl caprylate, caproic acid, caprylic acid and acetic acid) used in this study were purchased from Sigma-Aldrich (Belgium). 3-octanol and absolute ethanol were purchased from Acros Organics (Geel - Belgium) and Sigma-Aldrich (Belgium), respectively.





**Figure 1.** Flow diagram of production process of Rwandan traditional banana wine “*Urwagwa*” collected from Ngoma and Rulindo districts.

### 2.3. Standard chemical analysis

The samples were centrifuged at 6000 x g for 10 min, filtered through filter paper and analyzed by standard methods. The pH was measured using a pH meter 781 (Metrohm Herisau). Titratable acidity, expressed as a percentage lactic acid, was determined by

titrating the samples with 0.1 N NaOH to the phenolphthalein end point. Sugar content was measured as degree brix using a hand refractometer (ATAGO brand, Japan). Ethanol was determined by enzymatic method using the Megazyme assay Kit purchased from Sigma-Aldrich (Belgium). The free and total sulphites (SO<sub>2</sub>) were measured by titration method using Vinmetrica SO<sub>2</sub> analyser kit from Brouwland (Belgium).

#### **2.4. Volatile aroma compounds analysis**

##### *HS-SPME procedure*

Banana wine samples (10 mL) were pipetted into 20-mL round-bottomed, amber glass headspace vials, each containing 2.5 g of NaCl and 5 µL of the internal standard 3-octanol (100 mg/L in absolute ethanol), and then equilibrated at 30 °C for 10 min under agitation (Gerstel Agitator/Stirrer) at 500 rpm. After this period, the 50/30 µm Divinylbenzene / Carboxen / Polydimethylsiloxane (DVB/CAR/PDMS) fiber (Supelco Inc., Bellefonte, PA) was exposed in the headspace of the vial for 30 min with agitation at 250 rpm and the extracted analytes from fiber were automatically desorbed in injection port of the GC-MS system at 250 °C.

##### *Gas chromatography - mass spectrometry*

Analyses were carried out using an Agilent 7890 GC system equipped with a 5975C inert XL EI/CI mass selective detector (Agilent Technologies, Santa Clara, CA, USA), Thermal Desorption Unit (TDU, Gerstel), PTV inlet (CIS 4, Gerstel) and MPS 2 with headspace and DHS option (Gerstel). An HP-5 MS column (30 m x 0.25 mm ID) with a film thickness of 0.25 µm was applied to extract volatile compounds from the headspace of above-prepared glass vial. The GC was equipped with a split-splitless injector which was held at 250° C. After starting at 30 °C, the oven temperature was raised in 3 steps after 2 min: 30-70 °C at 10

°C/min followed by 1 min at 70° C, 70-220° C at 4° C/min and 220-280 °C at 20° C/min, and was finally held at 280° C for 6 min. During these programs, a constant flow rate (1.0 mL/min) of the carrier gas (Helium) was maintained. Mass spectra were obtained by electronic impact (EI) scan mode (low mass: 30.0; high mass: 500.0; threshold: 150) and temperature source (230° C) was generated.

### *Identification and quantification*

The identification of volatile aroma compounds was achieved by comparing mass spectra obtained from the sample with those from the NIST and Pal1600k.L libraries Database or from the pure standards injected in the same conditions and by comparing the Kovats index estimated for each compound on both chromatographic column with the values given in the literature (Pino *et al.*, 2010; Lyumugabe *et al.*, 2013; [www.pherobase.com/database/kovats](http://www.pherobase.com/database/kovats)).

Selective ion monitoring was used for integrations of all chromatogram peaks and the quantification was conducted according to the internal standard (3-octanol) quantification method. Quantitative data of identified compounds were obtained by using the following formula: Analyte's concentration = (Peak area of analyte / Peak area of internal standard)

x Emendation factor to internal standard

x Concentration of internal standard

The concentration of volatile aroma compounds for which there was no pure reference available was obtained by using the same emendation factor as one of the compounds with the most similar chemical structure (Perestrelo *et al.*, 2006; Li *et al.*, 2006, 2008).

### **2.5. Odour activity values (OAVs)**



Odour activity values (OAVs) were performed to evaluate the contribution of each volatile aroma compounds in banana wine. OAVs were calculated using the equation  $OAV = c/t$ , where  $c$  is the total concentration ( $\mu\text{g/L}$ ) of each compound in the wine samples, and  $t$  is the odor threshold value ( $\mu\text{g/L}$ ) of the compound in water/ethanol solution (Hellín et al., 2010); threshold values were obtained from information available in the literature (references are shown in **Table 3**).

## **2.6. Descriptive sensory analysis**

Descriptive sensory analysis of banana wine was performed by a selected panel of ten assessors trained over five sessions according to the international standards (ISO 13299, 2003). A preliminary sensory assessment of *Urwagwa* aroma was conducted on 4 wine samples from Rulindo and Ngoma districts in order to obtain a list of descriptive attributes of aroma. The attributes that were recognised by at least 50 % of the panel members were selected and put on the list. In the second phase, 16 samples of *Urwagwa* were presented in a randomized and balanced order, and the intensity of each attribute related to odour such as fruity, flora, banana, spicy, sweet, fatty and green/vegetable was quantified using a 9 points hedonic scale ranging from 1 (low intensity) to 9 (high intensity) according to the international standards (ISO 4121, 2003). Each evaluation was conducted in individual tasting booths at room temperature (20 °C) (ISO 8589, 1988) and 50 ml of each wine was served in glasses labelled with a code and covered to prevent volatile loss (ISO 3591, 1977).

## **2.7. Statistical Analysis**

The experiments were conducted in triplicate and the results were expressed as mean with standard deviation. Statistical analysis of the data was performed using SPSS Package Program. Statistical significance was taken at 95% confidence interval when  $p < 0.05$ . When

Analysis of Variance (ANOVA) revealed a significant effect ( $p < 0.05$ ), the data means were compared by the least significant difference (Duncan's Multiple Range test) test.

### 3. RESULTS AND DISCUSSION

#### 3.1. General composition of traditional banana wine "Urwagwa"

**Table 1** shows some physicochemical characteristics (ethanol content, pH, titratable acidity, total sugars expressed in brix degree, total and free  $\text{SO}_2$ ) of traditional banana "Urwagwa" wine made from *Gisukari* (collected from Rulindo district) and *Kamaramasenge* (collected from Ngoma district) banana varieties.

Ethanol, principal metabolite produced by yeast (*Sacharomyces cerevisiae*) during the fermentation of banana juice, is essential to enhance the sensory attributes of other wine components, its concentration can significantly influence the aroma and taste of the produced wine. In this study, ethanol content of *Urwagwa* wine samples from Rulindo and Ngoma vineyards was observed to be  $11.03 \pm 2.25$  (v/v) and  $7.53 \pm 1.16$  (v/v), respectively. Low ethanol content observed in *Urwagwa* from Ngoma district can probably due to *Kamaramasenge* banana variety, whose concentration of fermentable sugars is low compared to *Gisukari* banana variety (Munyangendo, 1983).

Titratable acidity of *Urwagwa* samples from *Gisukari* and *Kamaramasenge* varieties ranged respectively around  $5.76 \pm 0.21$  g/L and  $5.41 \pm 0.33$  g/L, and pH was between  $3.87 \pm 0.13$  and  $4.09 \pm 0.19$ . The principal organic acids found in must or wine are tartaric, malic; to a small extent, citric and other acids. Tartaric and malic acid account for over 90% of titratable acidity. The composition and concentration of these organic acids within the wine is influenced by many factors such as variety, climatic region, and cultural practices; their presence contributes to both a wine's flavour and to its stability (Richard *et al.*, 1988).

**Table 1.** Some physicochemical characteristics of *Urwagwa* from Rulindo (made from *Gisukari* banana variety) and Ngoma districts (made from *Kamaramasenge* banana variety)

	<b>Rulindo</b>	<b>Ngoma</b>
pH	3.87 ( $\pm 0.13$ )	4.09 ( $\pm 0.19$ )
Titrateable acidity (g/L)	5.76 ( $\pm 0.21$ )	5.41 ( $\pm 0.33$ )
Total sugar (Brix)	4.40 ( $\pm 1.00$ )	4.00 ( $\pm 0.56$ )
Ethanol (% v/v)	11.03 ( $\pm 2.25$ )	7.53 ( $\pm 1.16$ )
Total SO <sub>2</sub> (mg/L)	29.95 ( $\pm 2.11$ )	19.62 ( $\pm 1.77$ )
Free SO <sub>2</sub> (mg/L)	7.68 ( $\pm 0.57$ )	5.99 ( $\pm 0.29$ )

Due to its anti-oxidative and anti-microbial properties, sulfur dioxide plays an important role as preservative agent of fruits and wines (Alobo and Offonry, 2009). In present study, Total sulfur dioxide content ranged around 19.62  $\pm$  1.77 mg/L in *Urwagwa* from Rulindo and 29.95  $\pm$  2.11 mg/L in *Urwagwa* wine from Ngoma. Note that there is no Rwandan legislation related to sulfur dioxide levels in banana wine. Otherwise, European Union Regulation (no 1493/1999 y 1622/2000) states that sulfur dioxide should not exceed 160 mg/L as the maximum level for red wines and 210mg/l for white and rosé wines (EFSA, 2016). Based on this EU-regulation, all *Urwagwa* wine samples presented sulphite levels within the norms. However, compared to the Western wines from grape, sulfur dioxide content in *Urwagwa* wine is very lower due to the absence of sulphur additions during the *Urwagwa* winemaking. Sulfur dioxide found in banana wine derived from the yeast metabolism or as a component of finings or priming's (endogenous SO<sub>2</sub>). Yeast has the ability to produce sulfur dioxide, from the reduction of sulfate in water and grist material. SO<sub>2</sub> levels will be increased if the sulfate supply to the yeast is increased, wort clarity is increased, wort oxygenation and pitching rate are lowered and fermentation temperature is reduced. Wine yeasts can produce up to 80 mg/L of sulfites depending on the fermentation conditions and their strains (Maik et al., 2009).

### 3.2. Volatile compounds of Rwandan traditional banana wine "*Urwagwa*"

Forty eight volatile compounds (**Table 2**), including 24 esters, 11 higher alcohols, 7 acids, 4 terpenes, 1 furan and 1 phenol were identified and quantified in SPEME extracts of *Urwagwa* wines made from the banana varieties “*Gisukari* and *Kamaramasenge*” collected from Rulindo and Ngoma districts, respectively. Only eight volatile compounds (6 esters and 2 high alcohols) were not common in all *Urwagwa* samples from *Kamaramasenge* and *Gisukari* banana varieties.

Esters constituted the main class of substances in terms of the number; they represented around 50% of the total volatile compounds identified in *Urwagwa* wine (**Figure 2**). Ethyl esters of fatty acids and acetates of higher alcohols were the dominating esters in the analyzed *Urwagwa* samples. Among them, ethyl acetate, ethyl caprate, ethyl caproate, ethyl nonanoate, ethyl caprylate and isoamyl acetate represent the major esters in all analyzed *Urwagwa* wine samples. However, higher concentration of ethyl acetate (51219 µg/L) ethyl caprate (49997 µg/L) and ethyl caproate (41010 µg/L) and was found in *Urwagwa* from Rulindo district (*Gisukari* banana variety), while, isoamyl acetate (20344 µg/L), ethyl nonanoate (18421 µg/L) and ethyl caprylate (16562 µg/L) were in higher concentration in *Urwagwa* from Ngoma district (*Kamaramasenge* banana variety). Other important esters identified in all *Urwagwa* samples analyzed were 1-methylbutyl butanoate, ethyl butyrate, hexyl acetate, diethyl succinate, ethyl isovalerate and Phenethyl acetate. Isoamyl propionate, ethyl benzoate, ethyl 2-hexanoate, ethyl laurate and ethyl palmitate were detected only in *Urwagwa* samples from Rulindo. High amount of ethyl caproate, ethyl caprylate and ethyl caprate were also observed in wine prepared from Indian Cavendish banana (Ranjitha et al., 2013). These esters, formed primarily during the fermentation (Pretorius and Lambrechts, 2000; Suomalainen, 1981), are responsible of the fruity-flowery aromas in wine (Rocha et al., 2004; Verzera et al., 2008; Sánchez-Palomo et al., 2015).

**Table 2.** Concentrations of volatile aroma components ( $\mu\text{g/L}$ ) in *Urwagwa* wines from Rulindo (made from *Gisukari* banana variety) and Ngoma districts (made from *Kamaramasenge* banana variety)

Compounds	RI	<sup>a</sup> ID	Concentration ( $\mu\text{g/L}$ )	
			Ngoma	Rulindo
<b>Esters</b>				
Ethyl acetate	613 <sup>b</sup>	MS/RIL	29880	51219
Ethyl butyrate	802 <sup>b</sup>	MS/RIL	321	89
Ethyl isovalerate	854	MS	215	ND
Isoamyl acetate	879 <sup>b</sup>	MS/RIL	20344	14789
Isobutyl butanoate	898	MS	98	137
Isoamyl propionate	913	MS/RIL	ND	563
Ethyl caproate	999 <sup>b</sup>	MS/RIL	32172	41010
1-Hexyl acetate	1012	MS	535	781
1-methylbutyl butanoate	1014	MS/RIL	567	145
Ethyl 2-hexanoate	1038	MS/RIL	ND	144
3-methyl butyl butanoate	1045	MS/RIL	56	314
Ethyl benzoate	1137	MS/RIL	ND	226
Ethyl succinate	1182	MS	1056	588
Hexyl butanoate	1190	MS/RIL	715	ND
Ethyl caprylate	1198 <sup>b</sup>	MS/RIL	16562	14120
Isopentyl hexanoate	1203	MS	0.06	167
Phenetyl acetate	1262	MS	286	364
Ethyl nonanoate	1299	MS/RIL	18421	13116
Ethyl caprate	1397 <sup>b</sup>	MS/RIL	40516	49997
Ethyl undecanoate	1480	MS/RIL	333	ND
Ethyl laurate	1596	MS/RIL	ND	428
3-methylbutyl decanoate	1622	MS	77	152
Ethyl myristate	1797	MS/RIL	0.7	0.05
Ethyl palmitate	1991	MS	ND	93

**Table 2** continued

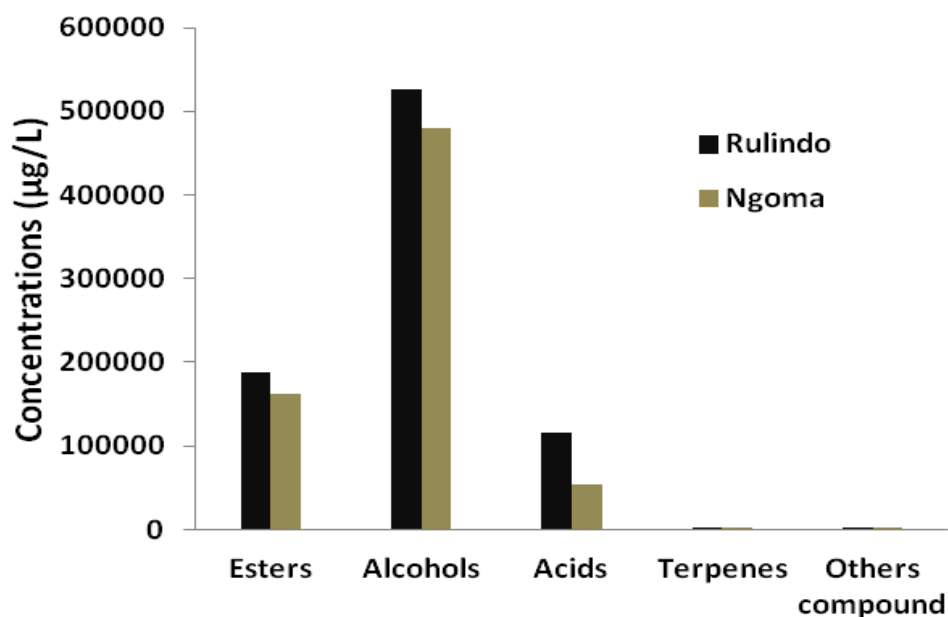
Compounds	RI	<sup>a</sup> ID	Concentration ( $\mu\text{g/L}$ )	
			Ngoma	Rulindo
<b>Alcohols</b>				
1-propanol	<600 <sup>b</sup>	MS	76582	83331
Isobutyl alcohol	<600 <sup>b</sup>	MS	20567	29808
2-pentanol	698	MS/RIL	ND	1221
Isoamyl alcohol	741 <sup>b</sup>	MS/RIL	362356	388990
2,3 butanediol	790	MS/RIL	271	195

1-hexanol	871 <sup>b</sup>	MS/RIL	1165	856
1-heptanol	935	MS/RIL	37	19
1-octanol	1082	MS/RIL	123	187
Phenethyl alcohol	1119 <sup>b</sup>	MS/RIL	18034	21867
1-nonanol	1170	MS/RIL	ND	90
<b>Acids</b>				
Acetic acid	701 <sup>b</sup>	MS	52451	112004
Isobutyric acid	773	MS	10	19
Propanoic acid	775	MS	914	610
Isovaleric acid	889	MS/RIL	412	225
Caproic acid	1066 <sup>b</sup>	MS/RIL	127	541
Caprylic acid	1188 <sup>b</sup>	MS/RIL	378	720
Capric acid	1380 <sup>b</sup>	MS/RIL	112	845
<b>Terpenes</b>				
Limonene	1019	MS/RIL	0.6	0.1
1,8-cineole	1037	MS/RIL	0.5	0.5
Dihydro-beta-Ionone	1406	MS/RIL	0.6	0.2
Trans-beta-Farnesene	1421	MS/RIL	0.1	0.3
<b>Others</b>				
2-pentyl furan	989	MS/RIL	17	75
2-methoxy 4-vinyl phenol	1315	MS/RIL	878	112

ND: No detected; RI: Retention index

<sup>a</sup> ID: identified by mass spectra (MS) and by comparison of retention index (RI on HP-5ms) calculated and retention index from literature (RIL).

<sup>b</sup> :identification confirmed by pure standard injection.



**Figure 2.** Concentration of different groups of volatile compounds in *Urwagwa* wines made from *Kamaramasenge* (Ngoma) and *Gisukari* (Rulindo) banana varieties.

Alcohols are quantitatively the largest group of the volatile compounds in *Urwagawa* wine of Rwanda (**Figure 2**). This group is composed of 1- propanol, 2-methyl-1-propanol, 2-methyl-1-butanol, 2,3 butanediol, 1-hexanol, 1-heptanol, 1-octanol, 1-nonanol, and phenyl alcohol. 2-pentanol and 1-nonanol were found solely in *Urwagwa* samples from Rulindo district. However, 1-propanol, 2-methyl-1-propanol, 3-methyl-1-butanol and phenyl alcohol are major higher alcohol found in all *Urwagwa* samples from Rwanda. These fermentation-derived products contribute to the alcoholic, floral and green grass aroma of the wine (Sanchez-Palomo et al., 2015); they are also known to be important as precursors of corresponding esters which contribute most significantly to the wine aroma.

Fatty acids constituted also the abundant group in the aromatic components of wines. In the present study, acetic acid was the major fatty acid found in all *Urwagwa* wine samples analyzed. However, the highest concentration was observed in *Urwagwa* made from *Gisukari* banana variety. At low level, acetic acid contributes to the complexity of the wine bouquet, but, they can have negative effect on wine aroma when above their thresholds (Swiegers and Pretorius, 2005). Caprylic, caproic, capric, propanoic, isobutyric and isovaleric acids were also detected in all analyzed *Urwagwa* wine samples. These fatty acids are generally produced by yeast metabolism during fermentation and accumulate in wine (Swiegers and Pretorius, 2005). The amount of medium-chain fatty acids (caprylic and capric acid) released into the fermentation medium depending to the yeast strain, medium composition and fermentation conditions (Krauss and Forch, 1975; Jones et al., 1981). Although they are do not associated with wine quality, these fatty acids have a characteristic odor and are interesting as flavor factors in wine. However, caprylic, caproic and capric acids can affect the wine aroma negatively when their sum is at the level beyond 20 mg/L (Shinohara, 1985).

Four terpenes were identified in *Urwagwa* wine made from *Kamaramasenge* and *Gisukari* banana varieties, including limonene, 1-8 cineole, trans-beta-Farnesene and dihydro-beta-ionone. The terpenes can originate from the raw material (Peña-Alvarez et al., 2004) or liberated by alpha-glycosidases from yeasts during the fermentation process (King and Dickinson, 2000). Despite their generally low concentrations, the terpenes are regarded to be a positive quality factor of a wine because terpenes contribute to its aroma, serving as indicators to differentiate the wine, and may contribute to floral note of the wine (Falqué et al., 2001; Calleja and Falqué, 2005). In the present work, the terpenes content does not allow distinguish the *Urwagwa* wine made from *Kamaramasenge* banana (Ngoma district) variety to those from *Gisukari* banana variety (Rulindo district). However, Limonene and 1,8-cineole were also found in Western wine where it serves as indicator to distinguish the varieties and quality of grape (Zhang et al., 2007). Capone et al. (2012) reported that the presence of Eucalyptus leaves and to a lesser extent grape vine leaves and stems is the major contributor to the 1,8-cineole (Eucalyptol) concentration in grape wine. The presence of 1,8-cineole in *Urwagwa* wine can also be due to the presence of Eucalyptus leaves during the traditional process of green bananas ripening and juice extraction.

Other compounds detected in *Urwagwa* wine made from *Kamarasenge* and *Gisukari* banana varieties were 2-pentylfuran and 2-methoxy 4-vinyl phenol. 2-pentylfuran has always been associated to the Maillard reaction as it is one of the main pathways generating furan. The presence of 2-Pentylfuran was also reported in Brazilian merlot wine (Welke et al., 2012) and sorghum beer (Lyumugabe et al., 2013). 2-methoxy 4-vinyl phenol is a major odour compound in many white wines, and aroma of the pure compound is described as wine-like aroma (Comuzzo et al, 2006). Ranjitha et al. (2013) reported that origin of this compound in banana wine lies in the fermentation process because it was absent in banana juice.



### 3.3. Odour activity values (OAVs) and Sensory characteristics of *Urwagwa*

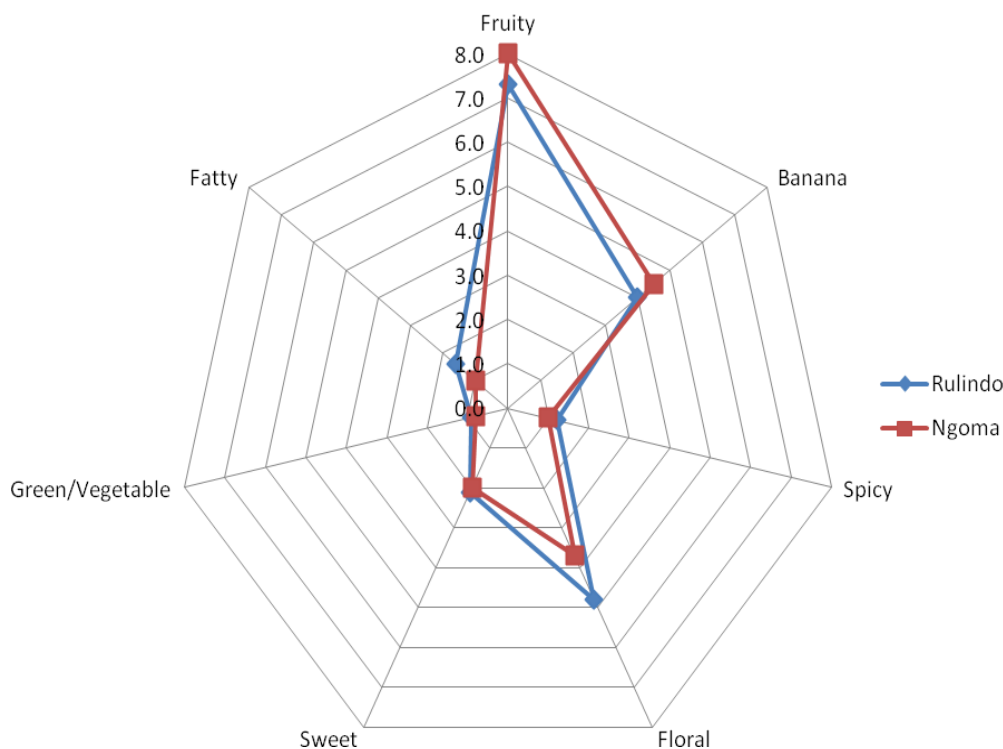
The characterization of potentially most important volatile aroma compound of *Urwagwa* wine was determined by odour activity values (OAVs), i.e. the ratio of the concentration of the compound to the odour threshold in wine (Sanchez-palmos et al., 2015). **Table 3** lists the odour activity values for 15 volatile aroma compounds with OAV > 1 in *Urwagwa* wine. Compounds that exhibited OAVs higher than 1 were considered to contribute individually to the *Urwagwa* aroma and were designated would be the characteristic aroma compounds for *Urwagwa*. The results showed that ethyl caprylate, ethyl caproate, ethyl caprate, ethyl acetate, isoamyl acetate, ethyl acetate, ethyl butyrate, phenetyl acetate, 3-methyl-1-butanol, phenetyl alcohol, caprylic acid, 1-octanol and isovaleric acid exhibited odour activity values higher than 1 for all *Urwagwa* wines studied. These compounds can contribute directly to the aroma profile of *Urwagwa* wine. However, as odour threshold is affected by additive, synergic and antagonistic effects of the volatile compounds in a matrix, the identification of the most powerful odorants only on the basic of their OAVs should be considered provisional (Sanchez-palomo et al., 2015). At present, this property can only be verified by means of

**Table 3.** OAVs of the aroma compounds of *Urwagwa* from Ngoma and Rulindo districts

Compounds	Odour threshold (µg/L)	Odor description	Ngoma	Rulindo
Ethyl acetate	7500 <sup>a</sup>	Fruity, sweet	4.0	6.8
Ethyl butyrate	20 <sup>b</sup>	Fruity	16.1	4.5
Ethyl caproate	5 <sup>a</sup>	Fruity, anise	6434	8202
Ethyl caprylate	2 <sup>a</sup>	Fruity,, floral	8182	7060
Ethyl caprate	200 <sup>a</sup>	Fruity, fatty, pleasant	206.6	259.0
Isoamyl acetate	30 <sup>a</sup>	Banana	678.1	493.1
Phenetyl acetate	250 <sup>a</sup>	Floral, pleasant,	1.2	1.5
Isoamyl alcohol	30000 <sup>a</sup>	Cheese	12.1	10.3
1-octanol	120 <sup>a</sup>	Intense citrus , roses	1.0	1.5
Phenethyl alcohol	14000 <sup>a</sup>	Rose, pollen, perfume	1.9	1.7

Isovaleric acid	33 <sup>c</sup>	Fatty, rancid	12.5	3.8
Caprylic acid	500 <sup>a</sup>	cheese, fatty acid, rancid	<1	1.4

<sup>a</sup> Guth (1997). <sup>b</sup> Dragone et al. (2009). <sup>c</sup> Ferreira et al. (2000).



**Figure 3.** Descriptive sensory analysis of the analyzed *Urwagwa* wine samples

sensory tests, although an approximation can be obtained by considering the variability in geometric terms of concentration or of concentrations normalised by their thresholds (López et al., 2003).

To define the overall aroma profile of *Urwagwa* wine, the descriptive sensory analysis was performed by a selected panel of ten assessors trained over five sessions. The aroma descriptors used during sensory analysis were defined in preliminary session and represent the main constituents of the aroma profile of the wine: fruity, floral, banana, green/fresh, sweet, spice and fatty. The results of sensory analysis (**Figure 3**) showed that all *Urwagwa* wines have the similar aroma profile. The highest score obtained was those of fruity note (7.3

for *Urwagwa* wines from Rulindo, and 8.0 for those from Ngoma), followed by floral note (3.7 for *Urwagwa* wines from Ngoma, and 4.8 for those from Rulindo) and banana note (4.0 *Urwagwa* wines from Rulindo, and 4.5 for those from Ngoma). These results show that the overall aroma profile of all *Urwagwa* wines studied is dominated by fruity note. Based on the OAVs, the fruity character of *Urwagwa* wines can be due to the high amount of ethyl and acetate esters, namely, ethyl caprylate, ethyl caprate, ethyl caproate, ethyl butyrate and ethyl acetate; while floral and banana notes can be correlated to the isoamyl acetate, phenethyl acetate, ethyl caprylate, phenethyl alcohol and 1-octanol. However, other compounds identified in *Urwagwa* wine (OAV less than 1) may also have contributed to enhance the intensity of some notes already present because of the synergistic effects with other odorous compounds in matrix of the wine.

#### **4. CONCLUSION**

The aroma profiles of *Urwagwa* wine samples collected from the districts of Rulindo (made from *Gisukari* banana variety) and Ngoma (made from *Kamaramasenge* banana variety) were investigated. The results showed that the aroma profiles of these two types of *Urwagwa* wine were not significantly different. Forty eight volatile aroma compounds, including esters, higher alcohols, acids, terpenes, furan and phenol were identified and quantified in *Urwagwa* wine. Among them, ethyl caprylate, ethyl caproate, ethyl caprate, ethyl acetate, isoamyl acetate, ethyl acetate, ethyl butyrate, phenethyl acetate, phenethyl alcohol, caprylic acid, 1-octanol and isovaleric acid exhibited OAVs > 1, and are considered as the major contributors of aromatic character of *Urwagwa* wine; described as fruity, floral banana, sweet and fatty notes. However, the overall aroma profile of all *Urwagwa* wines studied was dominated by the fruity note due to the high amount of ethyl caprylate, ethyl caprate and ethyl caproate in this Rwandan traditional banana wine.

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