African Journal of Food Science and Technology (ISSN: 2141-5455) Vol. 3(10) pp. 277-290, December, 2012
Available Online http://www.interesjournals.org/AJFST

Full Length Research Paper

Characteristics of enzyme-treated banana juice from three cultivars of tropical and subtropical Africa

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Accepted 17 July, 2012

Enzyme treated banana juices of three cultivars from tropical and subtropical Africa were studied. The main aim in this study was to apply commercial enzymes to banana pulp and improve on juice extraction and clarification. The banana cultivars which were selected for this study were Kayinja (ABB genotype), Mbidde (AAA-EA genotype), and Bogoya (AAA genotype). Two types of banana belonging to the same genotype (Musa, AAA) grown in the tropical and sub-tropical regions of Africa, known as Williams and Gros Michel in South Africa and Uganda respectively were used for comparison of physicochemical properties. All the cultivars were purchased at full maturity and ripened to stage 8 (yellow, speckled brown) of ripeness under similar warm conditions. Different commercial enzyme preparations were added to banana pulps in the preliminary experiments at the rate of 0.03 g/L for pectin de-pectinisation and de-esterification at 15°C for 24 hours. After juice extraction four pectinolytic enzymes (Rapidase X-Press, Rapidase CB, Rapidase TF and OE-Lallzyme) were selected for further experiments. The parameters that were analysed for comparison included juice yield, total soluble solids, total acidity, pH, viscosity, specific gravity and turbidity. The juice yield was significantly improved (477% v/w) in one of the enzyme treated samples compared to the control in the Kayinja (ABB genotype). The soluble solids in the juice of the three cultivars ranged between 15° and 27°Brix. The total soluble solids of bananas of the same genotype but grown in different climates differed significantly (p<0.05). Turbidity was lower in all the enzyme-treated juices samples compared to the control samples in the 3 cultivars. The overall acceptability of the juices was assessed by a panel and scored more than six points on the nine-point hedonic scale that is at least 66.7% of the juice is acceptability in general.

Keywords: Enzymes, banana juice, Musa genotypes, juice yields and clarification.

INTRODUCTION

Banana (Musa spp.) juice is nutritionally an important beverage in the tropical and subtropical world and is rich in calories and minerals such as potassium, magnesium, phosphorus, etc (Kyamuhangireet al., 2002). With proper food hygiene under Good Manufacturing Practices (GMP), safe and quality banana juice can be processed from over-ripe rejected fruits. Bananas rejected on market because of external injury or over supply are potential sources of raw material for juice processing.

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Clear banana juice, with its widely appreciated flavour and aroma can be used in various foods and drinks (Shahadan and Abdullah, 1995).

Unfortunately, there is considerable waste of the "rejected bananas", that is, those that do not meet the quality norms for export (Viquezet al., 1981). This is due to the fact that the banana fruit is biologically active and carries out transpiration, ripening and other biochemical activities even after harvests, which deteriorate the quality of the fruit and finally make it unmarketable (Emerald and Sreenarayanan, 1999). Wastage of the banana fruit due to poor post-harvest handling or over-ripening remains a major problem today (Shahadan and Abdullah, 1995). Fruit losses can be reduced by processing over-ripe fruits into other products such as wine and vinegar (Adams, 1978). However, some tropical fruits, banana inclusive, are too pulpy and pertinacious to yield juices by mechanical methods without the expenditure of excessive amounts of energy (Kyamuhangireet al., 2002; Shahadan and Abdullah, 1995).

Banana beverages in the form of juice and traditional beer have been produced locally for several years in the banana-producing areas. Though not yet commonly available on the market, clarified banana juice has been studied by different researchers using various approaches (Viquezet al., 1981; Koffiet al., 1991).

Of the problems associated with banana juice processing, a high viscosity, problems with juice extraction (Dupaine and Delnic, 1965; Viquezet al.,1981) and browning problems (Galeazzi and Sgarbieri, 1981; Mao, 1974) seem to be the most severe (Sims and Bates, 1994). Sulphite and other chemicals have shown to inhibit banana polyphenoloxidase (Koffi et al., 1991) although safety concerns may limit its use (Sims et al., 1994). The use of commercial enzyme preparations in the fruit juice processing industry to facilitate juice release and increase juice yield is well established (Kyamuhangireet al., 2002).

The use of commercial pectinolytic enzymes as processing aids has been done with various fruits such as apple and grape to disintegrate the fruit pulps, reduce viscosity and clarify the juices (Roumbouts and Pilnik, 1978). The action of enzymes leads to degradation of and volatilisation of other insoluble materials (pectin, hemicellulose and some cellulosic materials) from the fruit pulp cell wall, resulting in increased juice yield (Kyamuhangireet al., 2002). The juice is subjected to enzyme treatment to produce the clear juice (Waldt and Mahoney, 1967). This enzymatic treatment of fruit juices has proved to be a reliable biotechnological means of producing limpid juices. Therefore, there is a need for the production of value-added products and for market development strategies (Mazk and Degner, 1994).

In this study, we investigated the effect of commercial enzymes on juice yields, clarification, total soluble solids, titratable acidity, pH, viscosity and organoleptic characteristics on juices from three different banana cultivars grown in tropical and subtropical Africa. Specific comparison of juices from 2 types of banana called Gros Michel in tropical Uganda and Williams in subtropical South Africa were made. This was chosen for comparison of physicochemical properties because the two types belong to the same genotype (Musa AAA, genotype) and raw materials for experiments were easily accessible in the two countries where the study was conducted.

MATERIALS AND METHODS

Banana cultivars used and pulp preparation

Three banana cultivars were used to study banana juices and wines. Kayinja (ABB, genotype), Mbidde (AAA-EA, genotype) and Bogoya (AAA, genotype) bananas were used in this study. Kayinja and Mbidde cultivars were purchased at the mature stage 1 of ripeness from local farmers in the Wakiso district in Uganda. "Williams" as it known in South Africa, commonly known as "Gros Michel" or Bogoya in Uganda, at stage 3 of its ripeness was always purchased from "Fruit and Veg.City" supermarket in Stellenbosch, South Africa. The bananas bought in Uganda were exported to South Africa for experiments at theInstitute of Wine Biotechnology, University of Stellenbosch. The bananas were always ripened to stage 8 (yellow speckled brown) under special warm conditions for seven to ten days at a temperature of between 28 and 33°C. This stage of ripeness before processing was desired because it is similar to the stage of the usually wasted ("rejected") bananas and the major issue was to see whether safe, stable and quality beverages can be obtained in over-ripe form. The ripened bananas were peeled manually and pulped using a motorized mixer before the pectinolytic commercial enzymes were added. Three banana pulps were weighed and separated in equal portions in their respective cultivar groupings. Then banana pulp were treated with commercial enzymes at recommended dosages and left for 24 hour reaction time before juice extraction.

Enzymes used

Various pectinolytic enzymes were purchased and used to treat the banana pulps during the preliminary experiments. However, the four enzymes that were chosen for subsequent experiments were selected based on their capacity to produce higher juice yields than the rest of the enzymes. The concentration of the enzymes that were selected after being applied as recommended by the supplier is shown in Table 3.1. The enzymes that were not chosen for further studies and the yields obtained from banana pulps are shown in Table 3.2. All the commercial enzymes were applied in three experiments in duplicates at 15^0C.

The flow chart in Figure 3.1, below, illustrates the method used for processing banana juice from commercial enzyme-treated banana pulp in this study.

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Table 3.1. The four enzymes that were selected after preliminary juice extraction

Enzyme	Description	Dose	Producer
Rapidase X-press	Pectinase activity	0.03 g/kg	Gist-brocades
Rapidase CB	Pectinases, maltodextrine, NaCl	0.03 g/kg	Gist-brocades
Rapidase TF	Pectinase, hemicellulase, cellulase activity	0.03 g/kg	Gist-brocades
OE-Lallzyme	Pectinase activity	0.03 g/kg	Lallemand

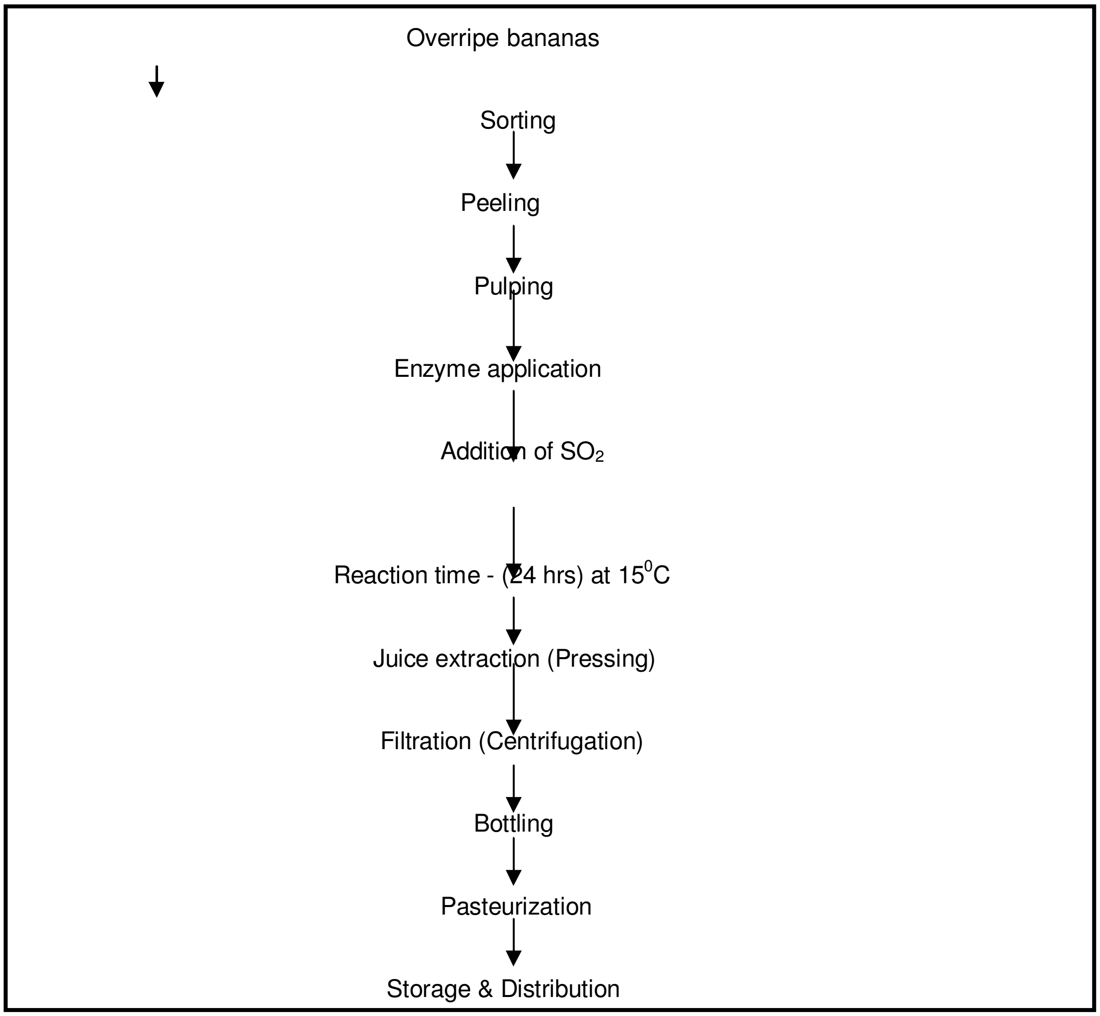

Figure 3.1: Flow chart for banana juice processing technology

Juice extraction

Juice extraction was done by treating banana pulp with pectinolytic commercial enzyme preparations and leaving the untreated (control) sample for comparison. We started this study by doing a preliminary screening of available enzymes. Juice extraction at the preliminary stage was done in three experiments in duplicates. The main strategy was to choose enzymes that would influence higher banana juice yields than the others.

Bananas were peeled manually and pulped using a blender into a rough pulp. After the pulping, the pulp was divided in equal portions for enzyme treatment reserving portions to serve as control in each cultivar. Sulphur dioxide (SO_2), an antioxidant and antimicrobial agent was applied to all pulps at a concentration of 50 ppm. Enzymes were added and the pulps were left to stand for 24 hours at 15^0C. After 24 hours, free-run juice was siphoned and measured in millilitres. The pulps were pressed, using a basket screw-press to maximise juice yields and the juices were strained through cheesecloth. Juice yield determination was presented as a ratio of juice (litres) divided by mass (kilogram) of pulp and expressed in percentages volume by weight (%v/w). All the juices extracted were compared in terms of amount yielded as influenced by the various enzymes added.The strained juice was filtered using a pressure filter in sterile conditions and bottled. Some juices were bottled and pasteurized at 85^{0}C for 5 minutes before storage. Juice samples were collected for analyses and sensory evaluation. The rest of juices obtained using the four selected enzymes were used further for banana winemaking experiments.

Physicochemical analysis

The extracted juice samples were analysed for various parameters. A refractometer (ATAGO Co, TokyoJapan) was used to determine the juice total soluble solids in °Brix. Titratable acidity and pH were automatically determined with the use of a 20-702 SM Titrino and with a glass calomel electrode pH meter(Cole Parmer, Vernon Hills ,IL, USA) respectively. Viscosity was measured, using viscometer Haake VT-02, in centipoises (cP). The specific gravity was determined with the use of a hydrometer at 20^{0}C. The turbidity of the juice was measured by a Hack turbidimeter in nephelometric turbidity units (NTU). The turbidity of the juices was determined in freshly extracted juices from both enzyme-treated and untreated banana pulps.

Statistical analysis

Analysis of variance (ANOVA) was performed on all juice yields and turbidity results (Zar, 1984). ANOVA was also performed on the viscosity of the juice. This was done using the STATISTICA 7 processing package.

Sensoryanalysis

Sensory evaluation was conducted at the Food Processing Laboratory, Department of Food Processing Technology, at Kyambogo University. Before the sensory evaluation, the panellists were subjected to a training session to familiarise them with the sensory attributes of the banana juice to be assessed. Each judge was introduced to a sensory evaluation score card and briefed on the procedures of evaluation during the training. A ready-to-drink (RTD) banana juice was presented as reference. To check on the assessor's consistency, a replicate of a juice was presented. A nine-point rating system was used. Quality evaluation was conduced by 20 trained and untrained teaching staff and students respectively. These could be considered as banana beverage connoisseurs because of their general knowledge, interest in and experience of local banana beverages. Samples were evaluated at room temperature of 25°C ±1°C under red fluorescent light and evaluation tests took place between 09:00 pm and 11:00 am. For each test, the panellist was served with 50 ml of banana juice in 100 ml colourless wine glasses. Coded juice samples that had been chilled at 7°C for 24 hours were served. Panellists were instructed to sip water and rinse their mouths between each of the juice samples provided.

The sensory attributes assessed by the panel of tasters were ranked from low to high levels on a nine–point hedonic scale. Taste was measured in terms of sweetness, whereas flavour and aroma were related to the banana flavour. The mouthfeel was assessed in terms of smoothness of the juice on the tongue. Acidity was assessed in terms of sourness of the juice in the mouth, a typical characteristic of high acetic acid. Colour of the juice was evaluated in terms of its intensity. Then, the overall acceptance was the general preference expressed by the assessors after evaluating the attributes in the sensory profile. The scores were subjected to statistical analysis at the Centre for Statistical Consultation of Stellenbosch University. To determine if there was any significant difference between the banana juices produced by application of different enzymes and the control, repeated measures of ANOVA were performed with the Bonferroni multiple testing procedure.

RESULTS AND DISCUSSION

Juice yield

The results of the banana juice yields are presented in Table 3.2 and Table 3.3. The juice yields obtained from pulps with the enzymes added in Table 3.2 were lower than those obtained in Table 3.3. The highest increase in juice yield was obtained from Kayinja (Musa, ABB genotype) pulp, which was treated with pectinolytic enzyme Rapidase X-press. The Kayinja juice yield of 57.87% v/w which was extracted from Rapidase X-press enzyme treated pulp was 477% higher than that extracted from the control sample (pulp with no commercial enzyme added) that yielded 10.03%v/w. The four selected enzymes played a significant ($p<0.05$) role in influencing extraction of banana juice. This can be deduced from the percentage of juice increases obtained. Comparing the results from all three cultivars, it can be seen that the added enzymes were most effective on juice extraction from the Kayinja cultivar. This implies that the fruit pulp of Kayinja was degraded by the pectinolytic enzymes more effectively than Bogoya and Mbidde cultivars. The huge difference in increases in yields of control and the enzyme treated samples can be attributed to the firm nature of the Kayinja fruit. Its firmness can be felt physically when the over-ripe banana fingers are pressed and that character seems to be associated with the viscosity observed in pulp (caused by the polysaccharides and proteins). This firmness of Kayinja in

Table 3.2. Juice yields influenced by enzymes excluded from further study.

Cultivar studied	Enzyme applied	Mean(%v/w)
Bogoya	Control	52.47
(Musa, genotype AAA)	EXV Lallzyme	52.47
	Rapidase filtration	52.67
	Rapidase pineapple	52. 62
	Hazyme	52.10
	Bats Liq	52.73
Mbidde	Control	57.20
(Musa, genotype AAA-EA)	EXV Lallzyme	56.86
	Rapidase filtration	57.37
	Rapidase pineapple	56.83
	Hazyme	57.80
	Bats Liq	57.27
Kayinja	Control	10.03
(Musa, genotype ABB)	EXV Lallzyme	36.87
	Rapidase filtration	35.67
	Rapidase pineapple	37.87
	Hazyme	36.23
	Bats Liq	39.47

Data are means of three experiments replicated

Table 3.3. Juice yields influenced by enzymes selected for further study.

Cultivar	Enzyme applied	Yield(%v/w)
Bogoya	Rap. X-press	67.93±1.27[a]
(Musa genotype AAA)	Rap. CB	56.73±1.00[bc]
	OE-Lallzyme	59.17± 0.61[c]
	Rap. TF	64.19±0.90[d]
	Control	52.47±0.71[e]
Mbidde	Rap. X-press	77.93±0.72[ad]
(Musa genotype AAA-EA)	Rap. CB	74.43±0.67[b]
	OE-Lallzyme	63.07±0.65[c]
	Rap. TF	77.07±0.38[d]
	Control	57.20±0.90[e]
Kayinja	Rap. X-press	57.87±1.43[a]
(Musa genotype ABB)	Rap. CB	52.27±0.97[bcd]
	OE-Lallzyme	49.60±0.82[cd]
	Rap. TF	51.17±1.31[d]
	Control	10.03±0.40[e]

Values are means ± SD of three experiments replicated.
[a]Different letters in the same column indicate significant difference at $p<0.05$.

an over-ripe state proved to be efficiently degraded by the pectinolytic enzymes and enable free-run juice release according to results of this study. Considering Bogoya and Mbidde, the highest juice yields obtained were also from the pulps treated with the enzyme Rapidase X-press, yielding 67.9% v/w and 77.9% v/w respectively. These results of fruit juice yield increases, influenced by commercial enzyme preparations with pectinolytic activities are in agreement with other research findings in various fruits and are associated with viscosity reduction in the pulp. Previous researchers (Mabesa et al., 1989; Kyamuhangireet al., 2002; Fundiraet al., 2002) had reported a similar phenomenon after adding commercial enzyme preparations to various fruit mashes at different temperatures and enzyme dosages. This juice yield is mainly influenced by the de-

Table 3.4a. Physicochemical characteristics of juices obtained from three banana cultivars.

Cultivar/Parameter	Enzymes used in juice extraction				
Bogoya (AAA)	**Control**	**Rapidase TF**	**Rapidase CB**	**Rapidase X-press**	**OE-Lallzyme**
TSS (^{0}Brix)	15.2	17.1	17.1	17.4	17.0
TA (g/L)	4.5	5.4	5.5	5.8	5.4
Specific gravity at 20^{0}C	1.13	1.10	1.11	1.11	1.10
pH	4.41	4.04	4.02	4.04	4.03
Viscosity (cP)	12.73	9.91	10.66	10.16	11.03
Turbidity (NTU)	597	531	536	532	537
Mbidde (AAA EA)					
TSS (^{0}Brix)	15.6	18.1	18.7	19.2	18.4
TA (g/L)	4.8	5.7	5.9	6.1	5.7
Specific gravity at 20^{0}C	1.12	1.11	1.10	1.10	1.11
pH	4.32	4.11	4.08	4.12	4.05
Viscosity (cP)	11.06	7.04	7.12	7.07	7.11
Turbidity (NTU)	564	495	493	490	492
Kayinja (ABB)					
TSS (^{0}Brix)	24.5	26.9	26.7	27.1	26.4
TA (g/L)	4.3	5.2	5.1	5.4	5.2
Specific gravity at 20^{0}C	1.14	1.12	1.12	1.11	1.12
pH	4.40	4.08	4.02	4.14	4.13
Viscosity (cP)	16.07	13.02	12.98	13.12	13.06
Turbidity (NTU)	683	612	614	608	614

Data are mans of three experiments replicated.

polymerisation and de-esterification effect on the complex pectin molecules, degrading cell walls of pertinacious nature and thus enabling easy release of fruit juice. The enzymes needed for pulp liquefaction are pectinases and exo-ß-glucanases (C1-cellulase, cellobiohydrolase). Exo-ß-glucanase works in conjunction with endo- ß-glucanase (C1-cellulase) which is present as Aspergillus-derived enzymes in most preparations (Nagodawithana and Reed, 1993). In our study, enzyme treatment of banana pulp gave cloudy juice. The enzymatic action leads to degradation and solubilisation of otherwise insoluble materials (pectin, hemicelluloses and some cellulosic materials) from the fruit pulp cell wall, resulting in increased juice yield (Dorreich, 1993). Because complete pectin hydrolysis is a prerequisite for successful clarification, the pectinases used in clarification contain especially high proportions of polygalacturonases and pectinesterases (Krug, 1968). The control samples from the three banana cultivars showed significantly ($p<0.05$) low juice yields, especially in Kayinja as compared to the other two cultivars. Such huge differences may be attributed to the genetic make up of the three banana cultivars with different genotypes and their natural capacity to yield juice with influence of endogenous enzymes in each cultivar especially Kayinja pulp which to a large part tended to remain colloidal (sort of slimy) not freely separating liquid-solid phases. It may also be related to high (24-27^{0}Brix) TSS in Kayinja which makes juice extraction in this particular cultivar more difficult than in the other used cultivars. Viquezet al. (1981) investigating juice yields from banana cultivar also found huge differences between control and enzyme treated juice yields. In their study, they found that the juice yield in the control (no enzyme addition) at banana grade 1 (beginning to ripen) of ripeness was 4.8% (w/w) compare to a yield of 66.5% (w/w) when the pectolytic enzyme Ultrazym 100 was added. They also tested the juice yield at grades 2 and 3 (medium ripe and very ripe respectively) of ripeness. The same tendency as the grade 1 ripeness was observed.

Experiments in which enzyme preparations (Vinozym ® Novo Nordisk Ferment) were added continuously to grape crushed mash at the rate of 2 g/hL showed the great influence of the enzyme on free-run juice, with a juice yield of 93% compared to the control, with a yield of 63% (Villettaz, 1993). Such a yield increase percentage (47.6%) of free-run juice is large and really worth enzymatic treatment of fruit mash.

The results of this study in the three banana cultivars concur with those other results previously reported (though those seem to be only on one cultivar) and confirm the significant role played by pectinolytic enzymes in influencing banana juice yields. As an innovation to discover whether Bogoya (a traditionally

Table 3.4b. Physicochemical properties of juices obtained from subtropical Williams and tropical Gros Michel types of banana (Musa, AAA genotype).

Parameter	Williams juice	Gros Michel juice
TSS (°Brix)	15.8[a]	19.5[b]
Titratable acid (g/L)	5.4[c]	5.5[c]
Specific gravity at 20°C	1.11[d]	1.12[d]
pH	4.42[cb]	4.46[cb]

Data are means of three experiments replicated.
[a] Different letters in the same row indicate significant difference at $p < 0.05$

Table 3.5. Effect of pectinolytic enzymes on the turbidity of banana (Musa, AAA genotype) juices**.**

Enzyme	Williams juice(NTU)	Gros Michel juice(NTU)
Rapidase X-press	532±0.57[a]	545±0.28[b]
Rapidase-CB	536±1.7[a]	547±0.78[b]
Rapidase-TF	531±1.20[a]	543±0.53[b]
OE-Lallzyme	537±1.69[a]	549±0.001[b]
Control	597±0.28[c]	619±0.212[d]

Values are means ± standard deviation of three experiments replicated.
[ab]Different letters in the same row indicate significant difference at $p<0.05$.
[b]The same letter in the same column indicates no significant difference at $p>0.05$.

Table 3.6. Effect of the pectinolytic enzymes on the viscosity of BogoyaMusa, genotype AAA juice measured at 20^0C.

Enzyme	Williams Juice (cP)	Gros Michel Juice (cP)
Rapidase X-press	10.16±0.014[b]	10.29±0.007[b]
Rapidase-CB	10.66±0.007[bc]	10.58±0.014[bc]
Rapidase-TF	9.91±0.014[a]	9.97±0.014[a]
OE-Lallzyme	11.03±0.07[d]	11.12±0.007[d]
Control	12.73±0.007[e]	12.89±0.007[e]

Values are means ± SD of three experiments replicated.
[a]Different letters in the same column indicate the significant difference at $p<0.05$.
[b]The same letter in the same row indicates no significant difference at $p>0.05$.

known as dessert cultivar) could yield juice, commercial enzymes that were added to its pulp influenced substantial amount of juice yield from the over-ripe fruits. The untreated mashed (pulped) Bogoya also yielded (an average of 52.5%) juice. This may create an alternative way of utilizing over-ripe Bogoya (as a beverage source) which traditionally was used as dessert and would be wasted as spoiled fruit in an over-ripe form. According to the findings in this study and those of other previous researchers (Kyamuhangireet al., 2002) enzyme treatment of banana pulp may be the best way to adopt for viscosity reduction and best way of juice extraction for banana beverages processing at large scale.

Physicochemical characteristics of juices obtained from three banana cultivars

The results of physicochemical characteristics of juices obtained from three banana cultivars are presented in Table 3.4 (a) and (b). For a comparative study of bananas of the same genotype (Musa, AAA) known as Gros Michel in Uganda and Williams in South Africa were specifically studied as representatives of bananas grown in tropical (warmer) and subtropical (cooler) climates respectively and the results are presented in Tables 3.4 (b), 3.5 and 3.6. Spider chart representations of sensory profiles for juices from the three banana cultivars are presented in Figure 3.2 (a), (b), (c) and (d).

In Table 3.4 (a), it is shown that the highest soluble solids (TSS) were 27.1^0Brix found in Kayinja juice where the pulp was treated with Rapidase X-press enzyme. The lowest TSS (15.2^0Brix) was found in the control juice sample extracted from Bogoya. Generally, TSS was higher in juices extracted from the enzyme-treated pulps than in juices extracted from the control pulps in all three banana cultivars.

Titratable acidity (TA) in juices from the three banana

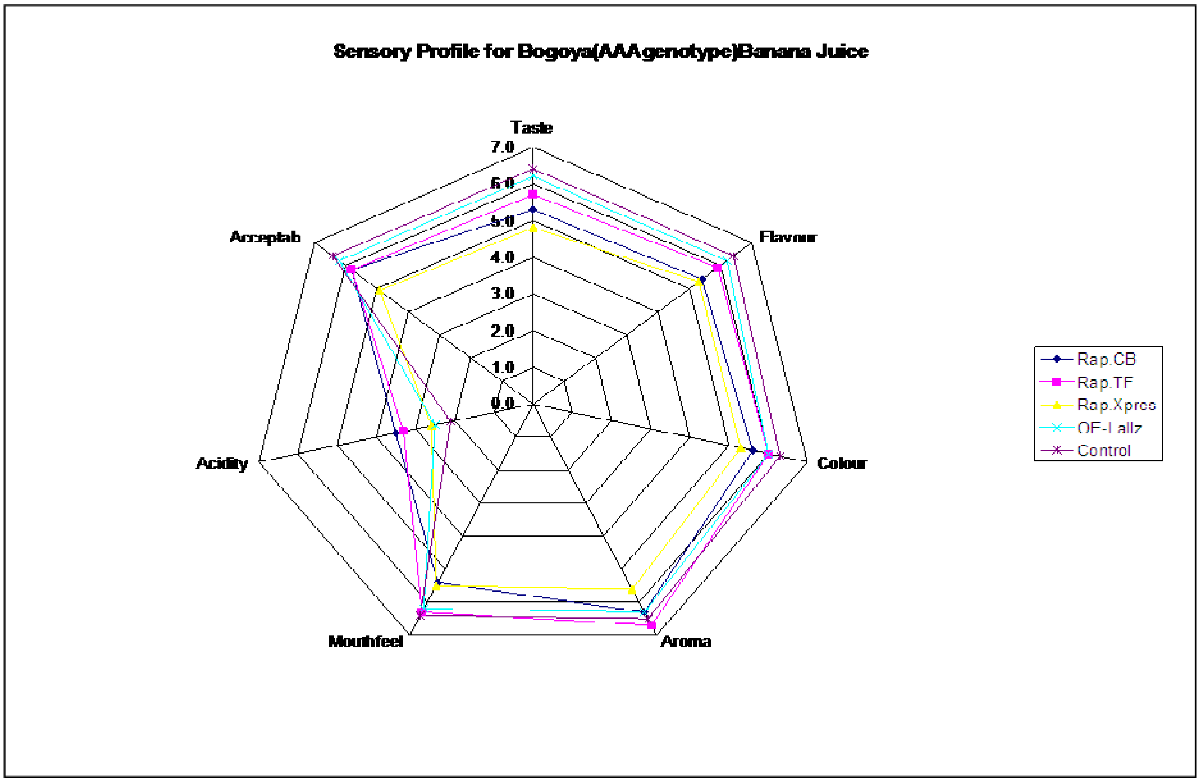

Figure 3.2a. Spider chart representation of sensory profile for Bogoya (Musa AAA genotype) banana juices.

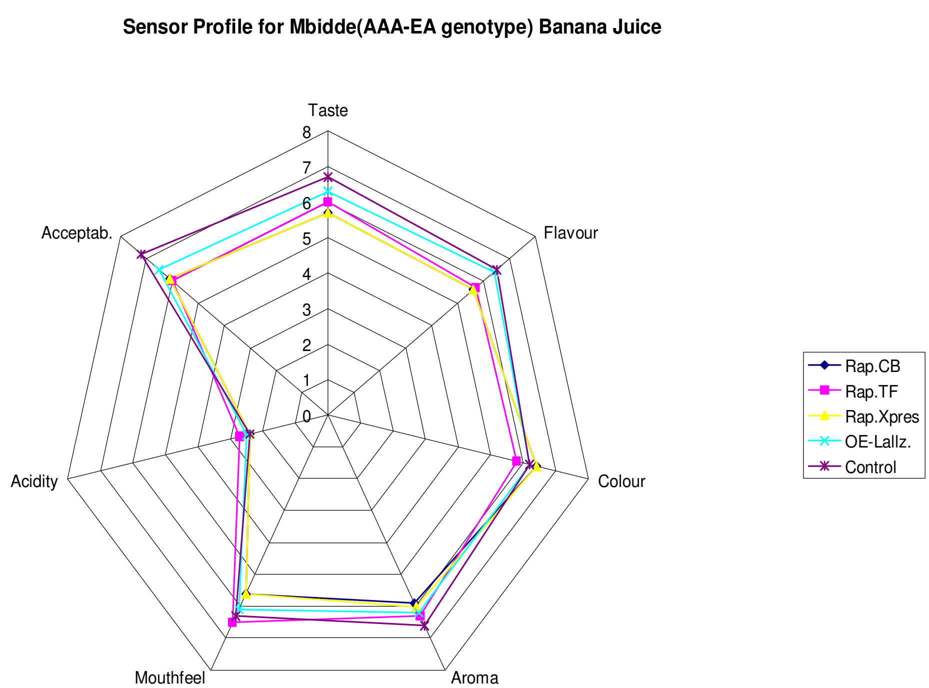

Figure 3.2b. Spider chart representation of sensory profile for Mbidde (Musa genotype AAA-EA) banana juices.

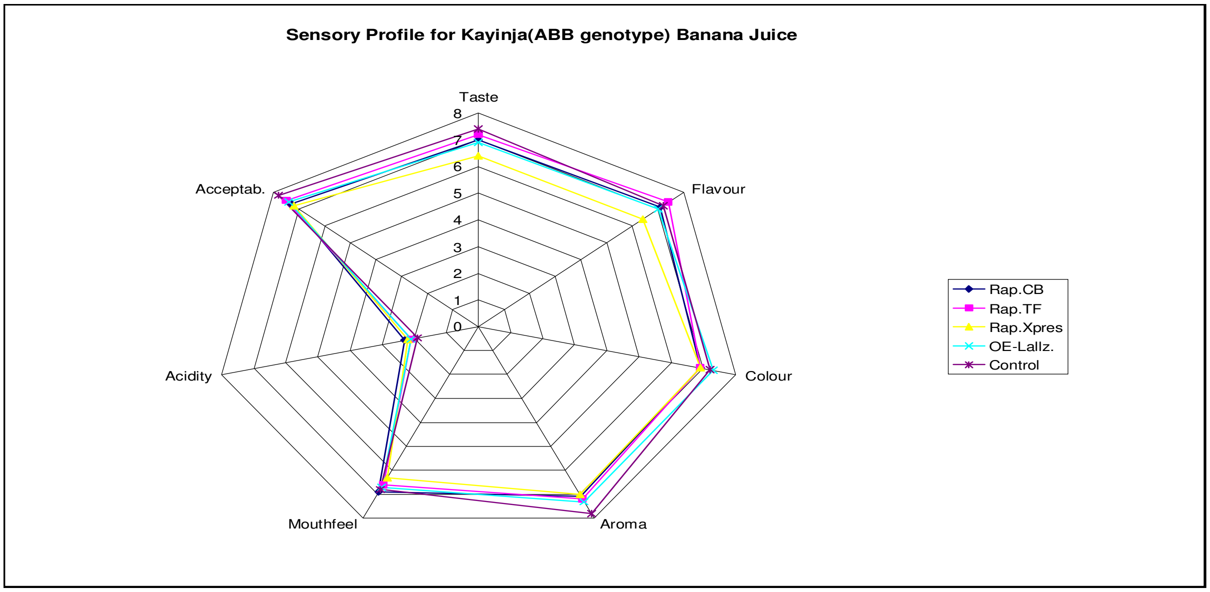

Figure 3.2c. Spider chart representation of sensory profile for Kayinja (Musa, genotype ABB) banana juices.

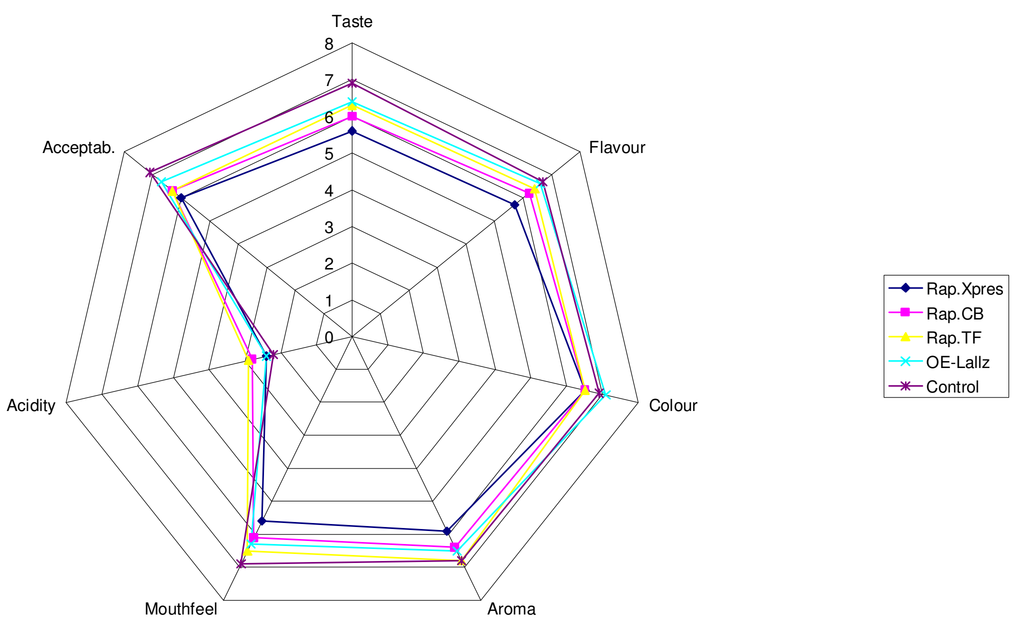

Figure 3.2d. Spider chart of the mean values of the attributes assessed in the juices obtained from all three cultivars for each enzyme used.

cultivars was also higher in the enzyme treated juice samples than in the controls. TA in the juices obtained from the three banana cultivars ranged between 4.5 and 5.8 g/L as anhydrous malic acid. The pH in banana juices ranged from 4.03 to 4.41. The pH, specific gravity, viscosity and turbidity were found higher in the control samples than in the enzymes treated juice samples in all the three cultivars.

The high total soluble solid content of enzyme-extracted juice may be explained by the degradation of the cell wall pectin, cellulose and hemicellulose, resulting in the release of some neutral sugars and leading to a Brix increase. Dorreich (1993) reported that such Brix increase can be as high as 10%. Lanzarini and Pifferi (1989) reported that the use of pectinase can considerably increase the soluble solids and reduce the viscosity of the fruit pulp.

Sandhu et al., (1989) showed that the treatment of Maharaji and Red Delicious varieties of apples with pectic enzyme resulted in an increase in TSS in the juice.

Kyamuhangireet al. (2002) found the average TSS in banana juice by the enzymatic method (34.90Brix) significantly higher ($p<0.05$) than in the mechanically extracted juice (30.70Brix). Our results agree with those previously reported on TSS when pectinolytic enzymes were added to fruit pulps.

Boultonet al. (1996) reported that typical ranges for titratable acidity in grape juices are 7 to 9 g/L as tartaric acid and pH was ranging from 3.1 to 3.4. The principal acid (TA) in bananas was investigated (Sadler and Murphy, 1998; Kyamuhangireet al., 2002) and found to be malic acid. The pH in this study comparatively showed higher levels in banana juices than in grape juices. The results obtained in this study on TSS, TA, pH and viscosities agree with those of previous researchers (Kyamuhangireet al., 2002) who had observed similar trends when they treated the banana pulp with (Pectinex Ultra SP-L), a pectinolytic enzyme.

It had been earlier reported (Rexová-Benková and Marcovic, 1976; Laing and Pretorius, 1992; Gainvorset al., 1994, Gonzalez-Candelas et al., 1995, Pretorius, 1997) that pectinases de-esterify (pectinesterases) or depolymerise (polygaracturonases, polymethylgalacturonases, pectin and pectatelyases) specific pectic substances. In addition, during the enzyme breakdown of pectin and hemicellulose, unesterifiedgalacturonic acid units are released (Doreich, 1993, Poll, 1993). This may be the reason why there is higher titratable acidity and lower pH in enzyme-treated juices than in the control juice samples. This phenomenon explains the low viscosity and turbidity levels found in the juices obtained from the enzyme-treated banana pulps. The above parameters lowered by addition of commercial enzymes to the pulps would be expected according to pectinolytic activities of enzymes applied.

The total soluble solids in Table 3.4 (b) are significantly ($p<0.05$) higher in Gros Michel juice than in Williams juice. The average pH, specific gravity and titratable acidity do not show a significant difference ($p>0.05$) in the values obtained from the two types of Musa AAA, genotype.

The high soluble solids (19.50Brix) found in the Gros Michel juice may be attributed to the longer sunny season in the tropical region being responsible for more total soluble solids (which also implies high sugar content) formation than in subtropical region. The Williams juice with 15.80Brix grows in the sub-tropical region where there occurs a relatively shorter sunny (cooler climate) season than in tropical region (warmer climate) and thus ends up with less TSS formation and distribution in the fruits. The results of TSS observed in Gros Michel and Williams in this study confirm what IITA (1993) had reported that chemical composition of bananas varies, and the variations are reported to be the result of many factors, including ecological location, nutrition, location on the bunch from which the banana fingers are sampled for analysis, and maturity of the fruit at harvest.

Regarding the relationship between enzymes and pH, every enzyme requires a specific pH for its optimal activity. This pH is of paramount importance when choosing an enzyme for industrial process. For example, for clarification of an acidic fruit juice pH<3, an enzyme exhibiting a pH range of 4-5 would show less activity at pH 3. For maximum enzyme efficiency, enzymes with optimum activity at a specific pH have to be strategically chosen (Uhlig, 1998).

Effect of enzymes on turbidity of the banana juices

The results of turbidity reduction in juices extracted after treatment of banana pulp with the pectinolytic enzymes and their controls are presented in Table 3.4 (a) and Table 3.5. The best turbidity reduction was obtained with the banana pulp treated with Rapidase X-press, measuring a turbidity of 490 NTU in the Mbidde juice. In the juices extracted from pulps treated with enzymes (Table 3.5), there was no significant difference ($p>0.05$) in turbidity levels of Williams or Gros Michel juices but there was a difference ($p<0.05$) between the turbidity levels of juices from the two types of Musa, AAA genotype. The control juices also showed a very significant difference from the juices obtained from enzyme treated banana pulps.

The freshly extracted juices generally had turbidity ranging from 490 to 614 NTU in the juices obtained from enzyme-treated pulps while in the control juices, the range was between 564 and 683 (Table 3.4 a) in all 3 cultivars.

The value of pectinases and their effectiveness at fruit pH and temperatures of up to 55oC was demonstrated by Koch (1956) not only for clarification of fruit juices but also for an improved pressing of chopped fruits, called fruit mash. Uhlig (1998) further said that current enzyme preparations used in fruit juice and wine making posses specific capabilities for degrading hydrocolloids, depending on the raw material and the product desired. The enzymes facilitate the sedimentation of colloidal particles resulting from degraded banana pectins, galactans and arabino-galactans (Colagradeet al., 1994; Haight and Gump, 1994).It was stated by Dorreich (1993) and Sole (1996) that commercial and experimental banana juice production must be based on the enzymatic method which allows ready separation of clear juice.

The juices obtained from enzyme-treated banana pulps in this study were generally less turbid than those from untreated pulps. The results obtained are in agreement with findings of the above researchers who used different commercial enzymes on various fruits in their juice clarity investigations. Pectinolytic enzymes played a significant role in turbidity reduction in the banana juices.

Effects of enzymes on viscosity of thebanana juices

The results of viscosity reduction effected by the enzymes used in this study are presented in Table 3.4 (a) and Table 3.6. The juice extracted from Mbidde pulp treated with enzyme Rapidase X-press had the lowest juice viscosity of 7.07. The viscosity of the juices remained highest in the control juices for the 3 cultivars. There was no significant difference ($p>0.05$) in juice viscosity reduction between the banana subgroups of Williams and Gros Michel treated with the same enzyme. That implied that the enzymes had the same effect on both of the Musa, AAA genotype types. The viscosity range was found between 7.07 and 13.12 cP and between 11.06 and 16.07 cP in the enzyme treated and untreated pulps respectively. The four commercial enzymes used showed a significant difference ($p<0.05$) in juice viscosity reduction compared to their respected controls. This difference may be attributed to the fact that different pectinolytic enzymes have different capacities to degrade and rapture juice cell structures by breaking the banana cellulose, hemicellulose and pectin. The role of pectolytic enzymes discussed in sections 2.7.1 and 2.7.4 shades more light on viscosity reduction in fruit processing. Previous research results on bananas have reported that ripe bananas contain approximately 3-4% total fibre (Paul and Southgate, 1978), with about 1% cellulose (Southgate, 1969), 0.5-1% pectin (Kawabata and Sawayama, 1974), and 1-2% hemicellulose (Berell, 1943). Ripe banana also may contain 1-4% starch (United Fruit Co., 1961) depending on how uniformly ripe the fruits being processed are. All the above mentioned complex substances in the banana fruit have the capacity to make juice extraction a cumbersome process. Therefore, it is of paramount importance to apply appropriate enzymes in the pulp that can degrade complex polysaccharides to enable juice release and filterability. Mabesaet al. (1989) noted that banana juice was more easily pressed from the enzyme treated pulp as compared to the untreated pulp. And they suggested that, this could be the result of the decrease in the viscocity of the juice due to the solubilisation of the pectin. Koffiet al. (1991) showed that mixtures of pectinase, cellulase and hemicellulase, when used at recommended rates, were more effective than pectinase, cellulase, alpha-amylase or galactomannanase on their own in reducing viscosity and improving the filterability of puree from both green and ripe bananas. When we compare previous results presented in Table 2.8 by Kyamuhangireet al. (2002) on pure juice yield from Kayinja (Musa, ABB genotype) and those obtained in this study, it can be seen that juice yields influenced by enzymatic action on Kayinja banana mash was different when different pectinolytic enzymes were used, though conditions (temperature, enzyme and enzyme contact time) between our experiments and Kyamuhangireet al. (2002) were not the same. Nevertheless, the juice yields influenced by the addition of enzymes to banana pulps were higher than that extracted mechanically and to the control. Future investigations may necessitate finding out the most appropriate enzyme mixtures for viscosity reduction, temperature and enzyme-mash contact time for maximum juice yields in various banana cultivars.

Sensory characteristics of banana juices

The results of sensory tests for different attributes are presented in Figures 3.2 (a), (b) , (c) and (d) and represent mean scores of the sensory characteristics on the nine-point hedonic scale. The juice attributes focused on for assessment were flavour, taste, aroma, acidity, mouth feel and overall acceptance. Flavour (a combination of taste and aroma) was related to banana flavour. The most typical banana flavour as observed by the panel was for Kayinja juice and as a result Kayinja juice scored highest in overall acceptance, with scores between 7 and 8 points on the nine-point scale. Acidity which was assessed based on sourness of juice was highest for Bogoya juice and lowest for Kayinja juice. The colour intensity of the juices from the three cultivars scored between 6 and 7.5, whereas mouth feel assessed in terms of smoothness scored between 5.5 and 7 points. Bogoya juice extracted with the aid of the enzyme Rapidase X-press, which yielded the highest juice volume, was rated the lowest in overall acceptance. Rapidase X-press treated juices scored the lowest on the nine-point hedonic scale for all the attributes tested. This may imply that whereas Rapidase X-press is capable of producing higher juice yields compared to the other enzymes, it seems somehow to affect the sensory quality of the product. The control samples of all three cultivars scored the highest in overall acceptance. This implies that the enzymes used to treat banana pulps may have slightly altered some of the attributes negatively. Overall acceptance of the juices obtained from the control pulps in the three cultivars scored 7.1 (Figure 3.2 d) on the nine-point hedonic scale and this was the highest average score. The lowest (6/9) acceptance score was for juices extracted from pulp treated with Rapidase X-press enzyme. Kyamuhangireet al. (2002) reported that the overall acceptability of enzyme-treated juices not scoring very high may be a result of a slight astringent taste as observed in their study where juices extracted by mechanical and enzymatic methods was compared. Although we did not measure astringency as one of our attributes it is tempting to speculate that the lower overall acceptability may be due to the same reason. The control samples of all three cultivars scored the highest in overall acceptance. This implies that the enzymes used to treat banana pulps may have slightly altered some of the attributes negatively.

The possibility of the enzymes used in juice extraction to slightly affect the attributes against preferences of tasters is related to what Vilanova et al., (2000) had reported (though their work was on wine not juice). They reported that with reference to wines obtained from must supplemented with commercial pectolytic enzymes, in as far as typicality was concerned, the wines display aromas that are less typical or not typical at all due to the release of terpenes and esters, a consequence of the action of some of the commercial enzymes. The results of our study seem to agree with the previous observations especially when Rapidase X-press enzyme was used. While Rapidase X-press produces the highest juice yields, it may not be the best choice for processing a ready to drink (RTD) beverage from bananas when consumer acceptance is desired. Perhaps, mixing different commercial pectinolytic enzymes or blending different batches of banana juices may improve the organoleptic qualities and general acceptance. Kyamuhangire (1990) noted that the high (29^0Brix) total soluble solids content of banana juice was an indication of high sugar content and reported that banana juice extracted from Lady-finger bananas had 25^0Brix with 21.1% total sugar. Earlier on, Wills et al. (1986) reported that bananas may contain up to 17.3-18.2% sugar. This may explain why the Kayinja juice was the most preferred juices because of the higher sugar content.

In Bogoya juice, Figure 3.2 (a), aroma was assessed highest in juice treated with Rapidase TF. The control had the highest mouthfeel. Aroma was scored lowest in Rapidase X-press treated juice whereas mouthfeel was assessed lowest in both juices treated with Rapidase X-press and Rapidase CB respectively. Colour, flavour, taste and overall acceptability were assessed highest in the control juice. On the other hand, colour, flavour, taste, aroma and acceptability were lowest in Rapidase X-press treated juices. Acidity was assessed highest and lowest in Rapidase CB and OE-Lallzyme treated juices respectively. These effects on Bogoya juices may be related to the different enzyme activities earlier described in the commercial enzyme preparations in Table 3.1.

In Mbidde juice, Figure 3.2 (b), aroma, flavour, taste and acceptability were also assessed highest in the control juice. The same attributes scored least in Rapidase X-press treated juice. Colour scored highest and lowest in the juices treated with Rapidase X-press and Rapidase TF respectively. Mouthfeel scored highest in the Rapidase TF treated juice and lowest in the Rapidase X-press treated juice. Acidity was highest and lowest in Rapidase TF treated and control juices respectively. Once again these effects on Mbidde juices may be related to the different roles played by the different enzyme activities earlier described in the commercial enzyme preparations.

In Kayinja juice, Figure 3.2 (c), mouthfeel, aroma, taste and acceptability scored most in the control juice. The same attributes scored least in Rapidase X-press treated juice. Flavour scored most in Rapidase TF treated juice and least in Rapidase X-press treated juice. Colour scored most in OE-Lallzyme treated juice and least in both Rapidase TF and Rapidase X-press treated juices. Acidity was highest and lowest in Rapidase CB treated and control juices respectively

The mean values of each attributes tested for all three banana cultivars together for each enzyme used is presented in Figure 3.2 (d), the sensory profile showed most of the attributes scoring highest and lowest in juices obtained from the untreated juices (controls) and Rapidase X-press treated sample respectively.

The lowest (5.6/9) and highest (6.9/9) acceptability scores was obtained for juices from Rapidase X-press treated and control pulps respectively. Flavour also scored lowest (5.7/9) and highest (6.7/9) in juices obtained from Rapidase X-press and control pulps respectively. Colour scored lowest in juices obtained from Rapidase X-press, Rapidase CB and Rapidase TF pulps at the same level (6.5/9) on the nine-point hedonic scale and highest (7.1/9) in juice obtained from OE-Lallzyme treated pulp. The aroma scored lowest (5.9/9) in Rapidase X-press treated pulps and highest (6.8/9) in juices obtained from Rapidase TF treated pulps and the control. Mouthfeel scored lowest (5.6/9) in juices obtained from Rapidase treated pulps and highest (6.9/9) in juices obtained from the control pulps. The acidity was assessed lowest (2.2/9) in juices obtained from the control pulps and highest (2.9/9) in juices extracted with Rapidase TF.

The overall acceptability of juices from three banana cultivars scored lowest(6.0/9) in the juices obtained from Rapidase X-press treated pulps and highest(7.1/9) in juices obtained from control(non-enzyme treated) banana pulps.

Results of this sensory evaluation showed Rapidase X-press scoring lowest in attributes (especially taste, flavour, aroma and acceptability) that would promote the acceptance of banana juice by consumers. Considering the effects of Rapidase X-press on sensory attributes affected as discussed above, it may not be advisable to encourage use of this particular pectinolytic enzyme for juice extraction if the quality of juice is to be based on the attributes tested.

Vilanova et al., (2000) had reported that with reference to wines obtained from must supplemented with commercial pectolytic enzymes, as far as typicality is concerned, these wines display aromas that are less typical or not typical at all due to the release of terpenes and esters, a consequence of the action of some of the commercial enzymes. Although the slight changes were noted by the panel in the juices in this study, it may be because of similar effects that as reported by the above researchers.

It is important to realise that the differences in the attributes assessed under different enzyme treated juices in the same banana cultivar was not statistically different.

However, there was a still a slight difference in the sensory profiles of the three banana cultivars that were studied. The banana juice from Kayinja was most preferred by panellists.

The results of this study showed that the TSS in the juices extracted from the 3 banana cultivars ranged from 15.2 to 27.1^{0}Brix. The TA ranged between 4.3 and 6.1g/L. Both TSS and TA were higher in juices obtained from the enzyme treated pulps. The pH ranged from 4.02 to 4.41 while the viscosity and turbidity ranged from 7.04 to 16.07cP and from 490 to 683 NTU respectively. The pH, viscosity and turbidity were lower in the in the juices obtained from the enzyme treated pulps compared to the control juices. The increased TSS and decreased viscosity and turbidity are of significant importance for banana beverage processing because the increase in sugar in the juice seems to make it more acceptable by the consumer and the decreased viscosity and turbidity improve processing of the product.

Therefore, the enzymatic processing used in this study seems to be a basis for future banana juice production development, following the results so far achieved. The overall acceptability could be improved on either by manipulations during juice processing such as mixing enzymes and blending different juices batches. Based on our research findings, pectinolytic enzymes may prove very useful in application of research and development (R and D) programs in the industrialisation of banana juice.

CONCLUSIONS

We can conclude that an acceptable quality banana juices can be produced from all three banana cultivar, even if the bananas are over-ripe. Results of this study showed that the commercial pectinolytic enzymes used played a significant role in increasing banana juice yields, total soluble solids and improving juice clarification. The highest juice yield was obtained from Mbidde cultivar, which is traditionally known as a “juice type” of banana because of its capacity to release free-run juice. Gros Michel and Williams proved to be reliable sources of juice, and may therefore be utilised equally well for banana juice production in the overripe form as an alternative to its traditional use as a dessert banana. The TSS of the bananas that belong to the same genotype (Musa AAA) was significantly different. Turbidity was also significantly reduced when enzymes were used.

These results form the bases for processors trying to produce high quality clarified juice from over-ripe bananas that normally are not consumed in the over-ripe state.

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