CHANGES IN AROMA, PECTIN AND PHYSICAL PROPERTIES OF RIPENING COOKING BANANAS

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DECLARATION

I Namakajjo Richard Jonathan declare that this dissertation presented in for the award of a Master of Science in Food Technology has not been wholly or partially submitted in for any other degree award or professional qualification.

Signed…… Date……...

APPROVAL

This is to certify that Namakajjo Richard Jonathan carried out this research work titled "Changes in aroma, pectin and physical properties of ripening cooking bananas" under our supervision. We approve the submission of the work for the award of Master of Science in Food Technology.

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DEDICATION

I dedicate this dissertation to my father Mr. Ssebanenya Samuel Musisi and the banana industry of Uganda.

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LIST OF ABBREVIATIONS

µV	Micro-volts
ABP	Afri-Banana Products
AOAC	Association of Analytical Chemists
BP	Banana Pectin
CP	Commercial Pectin.
FAO	Food and Agriculture Organization
FREVASEMA	Fresh Vacuum Sealed Matooke
g	Grams
IPBO	International Plant Biotechnology Outreach
IPPA	International Pectin Producers Association
KARI	Kawanda Agricltural Research Insititute.
min	Minutes
mm.	Millimeters
MPO	*Mpologoma*
MUS	*Musakala*
N	Newtons
NAK	*Nakitembe*
PIBID	Presidential initiative on Banana Industry Development

RUFORUM	Regional Universities Forum for Capacity Building in Agriculture
UBOS	Uganda Bureau of Statistics

ABSTRACT

Post-harvest losses are high in the cooking banana value chain in Uganda due to the short shelf life of the green bananas (6 to 10 days). The losses give rise to large amounts of solid wastes that later decompose into greenhouse gases that contribute to global warming. Hence, there is a need to add value to waste bananas by extracting bioactive substances such as aroma and pectin. In this study, changes in pulp and peel colour, texture, fruit, pulp and peel weight, fruit, pulp and peel thickness and specific gravity were analysed during 0-12 days of ripening under ambient conditions. Pectin content was determined using acid extraction in combination with either ethanol or calcium chloride precipitation methods. Chemical properties of the pectin (ash content, methoxyl content, anhydrouronic acid and degree of esterification) were analyzed using Rangana's protocol.

Aroma compounds in the three ripening cooking banana cultivars, *Nakitembe* AAA-EA (NAK), *Mpologoma* AAA-EA (MPO) and *Musakala* AAA-EA (MUS) were analyzed using solid phase micro-extraction with gas chromatography flame ionization detection (SPME-GC-FID). Pectin extracted from the banana samples was used to formulate a pineapple jam containing different concentrations of banana pectin. The jam was evaluated for sensory acceptability using 30 untrained panelists.

Colour significantly ($p<0.05$) changed from green to yellow in all cultivars with the peel showing more observable changes compared to pulp. There was significant decrease in hardness of all the selected bananas cultivars ($p<0.05$); *Nakitembe* 84.51 N to 9.76 N (88.45 % reduction), *Musakala* 80.48 N to 11.0 N (86.33 % reduction), *Mpologoma* 76.81 to 13.46 (82.48 % reduction). During the ripening period, hardness, fruit and peel weight, as well as fruit and peel thickness decreased significantly ($p<0.05$) and the extent of decrease was specific to a given banana cultivar. There was a significant increase in pulp to peel ratio for

all the cultivars ($p<0.05$) and likewise cohesiveness significantly increased for all cultivars at the end of the ripening period.

Pectin yield was generally higher using ethanol precipitation compared with calcium chloride protocol except for stage 7 of ripening which had no significant difference ($p>0.05$) between the two methods. There was no significant difference in the chemical characteristics of pectin from pulp and peel ($p>0.05$). However, pectin from *Mpologoma* presented the highest degree of esterification, DE (91 to 94 %) compared with other cultivars. DE values of pectin extracted at different stages of ripening were not significantly different ($p>0.05$) for the same cultivar but were different ($p<0.05$) between cultivars. The sensory scores did not significantly differ ($p>0.05$) between jam treated with banana pectin and that treated with the commercial pectin (control). However, the formulation with 2.0 % banana pectin was the most preferred (6.5 out of 9.0 hedonic points).

A total of 19 compounds were detected in the banana pulp and peel based on their retention time and relative abundances. In all samples, the pulp had higher abundances compared to the peel. *Nakitembe* had more compounds in the peel and pulp compared to *Musakala* and *Mpologoma*.

This study indicated that ripened cooking bananas (*Matooke*) are prospective sources of high-quality pectin and of aroma, which can be applied in the production of fruit jam. Overall, this would valorize ripened *Matooke* that are often regarded as bio-waste and subsequently reduce on environmental degradation through pollution. There is a need to identify the ions that dominate the aroma profiles of peel and pulp detected by headspace analysis in this study. This could pave way for extraction of these volatiles for application as natural banana flavorings for other food products in a commercial context.

CHAPTER 1

INTRODUCTION

1.1 Background

Bananas (*Musa* spp.) are tropical fruits belonging to the family *Musaceae* (Nelson et al., 2006). They are the fourth most important food crop in the world after rice, wheat and corn (Arumugan and Manikandan, 2011). Bananas are an important staple food particularly in tropical countries where they contribute to food security (Kabahenda and Kapirir 2010). Fruiting in bananas begins between 8 and 13 months after planting and yields continue up to 10 years depending on cultivar and the climatic zone (Aurore et al., 2009). Continuous harvesting of bananas throughout the year provides the farmers in Uganda with a constant income source (Omulo et al., 2015).

By 2015, global production of bananas was estimated at 133.7 million tones (Lescot, 2015) up from 68.2 million tons in 2000 (FAOSTAT, 2000). It is estimated that 20 % global banana production is in East and Central Africa while Uganda's annual production is estimated at 50 % of the East Africa's production (approx. 10 million tons) which accounts for approximately 10 % of total global production and puts Uganda third after India and China (FAOSTAT, 2006; FAOSTAT, 2012).

Rubaihayo (1997) categorized bananas into four broad types according to use. These include; (i) cooking bananas that are subdivided into *Matooke* which are usually steamed before eating, (ii) plantain which are mainly used for dip frying and roasting, (iii) dessert bananas which comprise of Cavendish, *Gros michel* locally known as *Bogoya* and Apple bananas locally known as *Sukaali Ndiizi*; (iv) juice or beer bananas used for juice and alcohol production. Examples in the latter category include the *Mbidde, Kayinja* and *Kisubi*.

Most cooking banana cultivars (*Matooke*) are harvested at green maturity (about 180 days after flowering), prepared and cooked in different ways before being eaten (Gafuma et al., 2018).

Matooke belong to the triploid acuminate genome group (AAA-East African highland bananas (EAHB) and are endemic (indigenous) to the East African highlands a region recognized as a secondary centre of banana diversity (Robertshaw, 2006). These EAHB are a unique genomic group, selected over the centuries by farmers (IPBO). As many as 84 distinct cultivars are grown by farmers in the region and are classified into five clone sets (*Natikembe, Nfuuka, Musakala, Nakabululu* and *Mbidde*) according to phenotypic characteristics (Karamura 1998).

In Uganda, *Matooke* production stands at approximately 29.5 % of the world's cooking banana production placing the country as second largest producer after India (FAOSTAT, 2014; 2016). These bananas are mostly grown by smallholder farmers on 1.5 million hectares of land with an average 0.5 ha per farmer (Wanda, 2009; Ouma and Jagwe, 2010). *Matooke* production in Uganda is mainly concentrated in the western (68 %), central (23 %), eastern (8 %) regions, and less than 1 % in the northern region of the country with the central and northern regions having the highest and lowest consumption, respectively (UBOS, 2010).

In Uganda, bananas are largely cooked for food and to a small extent, its processed into juice, beer and wine (Rietveld et al., 2015). On a limited scale, dessert bananas are peeled, dried, packed and exported by FREVASEMA (U) Ltd mainly to Europe (van Asten et al., 2010). Roasting and some desert bananas are fried into crisps and chips for the local market. *Matooke* are rarely processed into other value-added products; their processing is mainly limited to peeling and vacuum packaging for export to the United States of America and Australian markets (Gafuma et al., 2018). Along the *Matooke* value chain, there are high

post-harvest losses from producer to retailer affecting 14.9 % of the produced volume, translating into 1.1 million tons per year in terms of physical or economic losses (Kikulwe et al., 2018). This represents significant food losses to consumers, equivalent to 21.3 kg per person per year, including income losses to the players that is the producers, wholesalers and retailers. The major cause of post-harvest losses during bumper harvest is ripening since there are many producers yet buyers are few in surplus seasons. Hence, storage of ripe *Matooke* leads to bruising, further ripening and overstaying which are the leading causes of physical and economic losses for banana farmers. Moreover, *Matooke* are normally stored at ambient conditions yet they are climacteric fruits, which should be consumed at green maturity, and so any form of ripening would result in rejection on the market. The high physical and economic losses coupled with minimum processing calls for intervention studies that could add value to the rejected *Matooke*. Extraction of bioactive substances such as aromatic substances and pectin could therefore be a feasible alternative. Aroma extracts from fruit materials are often utilized in the dairy industry for yoghurt production (Chandan and O'Rell, 2013) and pectin is widely used as a gelling agent, thickener, emulsifier and stabilizer in different food processing operations (Lara-Espinoza et al., 2018). Studies have also shown that banana pectin and aromatics can be utilized in the development of value-added products (Pratik et al., 2017; Castillo et al., 2015; Oleveira et al., 2015; Emaga et al., 2008b).

Banana fruit has a pleasant flavor and different researchers (Jordan et al., 2001, Bugaud and Alter 2016) have previously obtained some banana flavor extracts. Given the current public interest towards consuming natural products, application of bioactive substances extracted from ripened *Matooke* could contribute to the food-processing sector and the overall economy of Uganda through enhanced banana productivity and utilization. Characterization of the pectin and aroma extracts would establish their quality levels for appropriate application. In this study, the pulp and peel of selected *Matooke* cultivars were profiled for

their physical properties during ripening, aroma substances and pectin content. Banana pectin was then applied to a pineapple jam product and its acceptability evaluated in order to determine the prospects for commercial viability of the extract.

1.2 Problem statement

Bananas (*Musa* spp.) are a staple food for over 16 million Ugandans and many derive income from these crops and use them for cultural, medicinal and industrial applications (Kabahenda and Kapirir, 2010). Ripe cooking bananas (*Matooke*) are considered waste in Uganda since they are customarily utilized for cooking in the unripe green mature state. These pulpy fruits are prone to spoilage due to high moisture content, fragile skin and ripening which occurs at the time of harvesting, storage, marketing and processing resulting into high amounts of agricultural wastes (Arumugan and Manikandan, 2011). Due to their climacteric nature, *Matooke* are highly perishable and are normally traded in fresh form under ambient conditions which leads to high post-harvest losses ranging from 22 to 45 % (Muranga et al., 2010). The losses occur due to the changes in the physical properties (fruit texture or firmness, colour, weight, pulp to peel ratio, thickness) as well as the chemical properties of the fruit. In the ripe state, *Matooke* are often disposed off despite them containing useful bioactive substances such as pectin and aromatic compounds (Samson, 2015) which could be extracted for other applications including use as flavorings and as gelling agent in other food products, respectively.

At the farm and/or household levels, ripe *Matooke* can be given to animals as feed or chopped into pieces and left to rot in the garden to provide manure. In the urban areas where farms are less existent, the ripe *Matooke* become solid waste, which is left to rot on dumping sites giving off a foul smell as well as greenhouse gases such as methane that eventually contribute to global warming (Tock et al., 2010).

Whereas several studies revealed presence of pectin and aroma compounds in ripe bananas (Waghmare et al., 2017; Castillo et al., 2015; Oleveira et al., 2015; Emaga et al., 2008b), there is scanty information on levels and distribution (pulp vs peels) of these substances in the Ugandan cooking bananas (*Matooke*). Moreover, levels of these substances in fruits depend on the degree of ripening (Wang et al., 2018). This study aimed to investigate the spatial distribution of pectin and aromatic compounds within the pulp and peels of selected cultivars of *Matooke* grown in Uganda in order to identify cultivars with prospects for recovery of these bio-actives in a commercial context. Let alone, the quality characteristics of *Matooke* pectin in relation to the commercially available product were also investigated at various stages of ripening to determine the optimal stage for extraction of high-quality pectin.

1.3 Justification and Significance

Along the value chain, ripe and rejected bananas are in large amounts and so is the potential for availability of pectin and aromatic compounds in these materials. Undesirable changes in the physical properties of bananas act as the basis for most rejections. Pectin is an important polysaccharide with applications in foods, pharmaceuticals, and a number of other industries (Pilgim et al., 1991). Its importance in the food sector lies in its ability to act as a gelling agent in the presence of other solutes such as sucrose at low pH (Thakur et al., 1997). The current cost of pectin is high in Uganda which calls for cheaper alternative sources and ripe *Matooke* could fill this gap since there is interest on the re-use of ripe bananas from economic and environmental points of view. Ripe *Matooke* could be used as low-cost raw materials for the production of other value-added products and minimizing the pollution arising from the waste discharge (Gumisiriza et al., 2017). Pectin has a high commercial value in Uganda due to rapid growth of the food processing industry that manufactures thick juices, yoghurt and jam.

Increased product supply in Uganda has intensified competition in all food commodities and therefore product quality consistency is the key for long term profitability. As a food additive, flavors given by aromatic compounds influence the consumer choice because smell is a fundamental parameter to value the food quality and to differentiate the food types (Bagacz-Radmuska et al., 2008). Therefore, there is a need to assess ripe *Matooke* as a potential source of banana pectin or flavor production.

1.4 Objectives of the study

1.4.1 Overall objective

To profile the changes in aroma, pectin and physical properties of ripening cooking bananas

1.4.2 Specific objectives

1. To determine the changes in physical properties of selected cooking bananas (*Nakitembe, Mpologoma and Musakala*) during ripening.
2. To determine the profile of aroma active compounds in peel and pulp of the selected ripening cooking bananas.
3. To characterize pectin (yield, ash content, methoxyl content, degree of esterification, equivalent weight and anhydrouronic acid) extracted from peel and pulp of the selected ripening cooking bananas.
4. To determine the sensory acceptability of jam developed using banana pectin.

1.4.3 Hypotheses

1. The physical properties of peel and pulp do not differ among the different *Matooke* cultivars during the ripening period.

2. There is no difference in the profile of aroma compounds in the peel and pulp of different cooking banana cultivars at the different stages of ripening.

3. There is no difference in content and chemical characteristics of pectin extracted from the peel and pulp of the different cooking banana cultivars at the different stages of ripening.

4. There is no difference in the sensory acceptability of pineapple jam formulated with banana pectin compared with that from commercially available pectin.

CHAPTER 2

LITERATURE REVIEW

2.1 Banana production and consumption

Bananas (*Musa* spp.), are the largest herbaceous plant in the world, grown abundantly in many developing countries and considered to be one of the most important sources of energy for people living in the humid regions of many countries. It is also considered fourth on the list of the developing world's most important food crop after rice, wheat and corn (Arumugan and Manikandan, 2011). Bananas are an important staple food particularly in tropical countries where they contribute to food security. By 2015, global production of bananas was estimated at 117.9 million tones up from 68.2 million tons in 2000 (FAO, 2018). About 20 % of global banana production comes from East and Central Africa (Tinzaara et al., 2018). Uganda produces about 10 million tons of bananas annually, which accounts for 10 and 50 % of global and East Africa's production, respectively (Table 1; FAOSTAT, 2012). Uganda's cooking bananas specifically *Matooke* production stands at 29.5 % of the world's banana production and about 0.85 % of the world's banana production is dessert bananas (REFORUM et al., 2010). Most *Matooke* cultivars are harvested at green mature stage, peeled and consumed following different methods of cooking. Cooking bananas are mostly grown by smallholder farmers with an average of 0.5 ha per farmer or even less (Wanda, 2009; Ouma and Jagwe, 2010). Global consumption of bananas is also highest in East Africa with an average per capita consumption estimated at 440-600 kg (Kabahenda and Namumbya, 2010).

Table 1: Estimated banana production in major banana growing countries

	Total production (tonnes)	Dessert banana production (tonnes)	Cooking banana production (tonnes)
World total	133,691,965	78,860,773	54,831,192
India	27,575,000	17,075,000	10,500,000
China	12,075,238	11,506,238	569,000
Uganda	8,926,308	500,000	8,426,308
Philippines	8,645,749	5,790,091	2,855,658
Brazil	6,892,622	6,402,622	490,000
Ecuador	6,739,739	6,145,527	594,212
Colombia	5,405,365	2,587,625	2,817,740
Indonesia	5,359,115	3,289,115	2,070,000
Rwanda	3,263,462	250,000	3,013,462
Nigeria	3,222,000	315,000	2,907,000
Others	45,587,367	24,999,555	20,587,812

Source: Lescot, 2015

2.2 Evolution and diversity of bananas in Uganda

Bananas are descendants of two wild fruits (Simonds and Shepherd, 1955) that is the *Musa acuminata*, a plant originally from Malaysia that produces single sweet-pickle-size green fruits with a milky flesh and several hard peppercorn-size seeds and the *Musa balbisiana*, a plant originally from India that is larger than *M.*

acuminata and produces thousands of button-like seeds (Simonds and Shepherd, 1955). When the two cross-bred, they gave rise to several diploid, triploid and tetraploid banana genomes represented by AA, AB, AAA, AAB, ABB, AABB, AAAB, ABBB. Letters A and B represent the contributions from *M. acuminata* and *M. balbisiana*, respectively during the crossing (Nayar, 2010).

The Regional Banana Germplasm Collection Centre of Biodiversity in western Uganda has records of over 200 East African highland banana varieties from Uganda, Tanzania, Congo, and Rwanda. Currently, Uganda has more than 87 distinct indigenous cultivars (Tushemereirwe et al., 2003).

There are several types of bananas grown in Uganda and have been classified as cooking, roasting, sweet and brewing bananas based on uses (Byarugaba-Bazirake et al., 2014).

Karamura (1998) classified Ugandan cooking bananas into four clone sets: (i) *Nfuuka* which comprises of *Nakawere* and *Namande* cultivars. (ii) *Nakitembe* clone set which consists of *Nakitembe, Kibuzi* and *Nakyetengu* cultivars. (iii) *Nakabululu* clone set comprising of *Nakabululu* and *Kazirakwe* cultivars, and (iv) *Musakala* clone set containing *Musakala, Kisansa* and *Mpologoma* cultivars Non-cooking types include juice/beer clone set which is constituted of Bluggoes AAB and Ney Poovan AB genomes (*Mbidde* and *Kayinja* are typical examples), dessert varieties (AAA genome); and plantain varieties (AAB genome).

2.2.1 *Nakitembe, Musakala* and *Mpologoma* cultivars.

These are East African Highland cooking bananas commonly available in most markets of Uganda. These East African Highland bananas are clones and it has been accepted that they have diversified through distinct mutations, which are therefore clones (Trehane et al., 1995). Using phenotypic analyses, the above cultivars have been grouped into clone sets by Karamura (1999), *Nakitembe* cultivar belongs to *Nakitembe* clone set, these have got creamy brown pulp colour before maturity with no brown excretions after maturity, unripe pulp is insipid, they also have oblique compact bunches, with medium fruits 15-20 cm long. Fruit apices are intermediate (between bottle necked and blunt), and have oblique male inflorescence rachis with persistent bracts and neuter flowers. *Mpologoma* and *Musakala* cultivars belong to *Musakala* clone set. These have whitish pulp colour before maturity with no sticky brown excretions after maturity, they have unripe pulp insipid, pendulous, lax bunches and long fruits above 20 cm. Fruits have bottle necked apices and pendulous nude male inflorescence rachis.

2.3 Banana utilization in Uganda

Banana utilization in Uganda is still developing. In Uganda, Rwanda and Burundi, cooking bananas are mainly used as staple food and they are responsible for 30-60 % of calorie intake (IPBO, 2016). Beer bananas are processed into juice, beer and wine. Dessert bananas are either consumed as fruit or sometimes dried, packed and exported to European markets (van Asten et al., 2010). Plantains are either dip-fried into banana crisps and chips or roasted on open flame for the local markets.

Current efforts are exploiting other forms of banana utilization in Uganda through government aided initiatives including Presidential Initiative on Banana Industrial Development (PIBID) and Afri-Banana Products Limited (ABP). For instance, ABP have developed fresh vacuum sealed cooking bananas for export to the United States of America and Australian markets. Other products developed and are due for commercialization include vinegar, juice and wine as well as charcoal briquettes, animal feeds, art and craft materials derived from banana waste and by-products (Gafuma et al., 2018).

PIBID have invested resources in development and production of banana flour (*Tooke*). There is scant information about efforts for other strategies for commercial utilization of bananas and by-products in the development of other novel products such as pectin and aroma extracts particularly in Uganda.

2.4 Physical changes in bananas during transportation, storage and ripening

The banana fruit (cooking and desert) has exclusive physical properties such as texture, colour, shape, curvature and weight that make it different from other fruits and vegetables. Many researchers have undertaken studies about its physical and mechanical properties. Salvador et al. (2007) studied the changes in colour and texture of bananas during storage at 10^{o} C and 20^{o} C. Ahmed et al. (2001) examined the temperature effect of ripening treatment

on properties of banana fruit, whereas Kachru et al. (1995) investigated the physical and mechanical characteristics of ripening bananas. These studies found that during storage, the change in peel colour from green to yellow is gradual in the *Musa cavendish* samples, whereas the *M. paradisiacal* variety presents a different pattern, remaining green for the first days and then changing rapidly to a yellow tone from day 12 onwards. The pulp texture of the *M. Cavendish* cultivar softens more rapidly during storage, while the change is slower in the *M. paradisiacal* cultivar and little variation occurs in the texture hardness values over the storage time. As fruit ripening is a genetically programmed and highly coordinated process of organ transformation from unripe to ripe stage (Maduwanthi and Marapana, 2017), the need to investigate these changes in Ugandan cooking bananas undergoing ripening cannot be overemphasized.

2.4.1 Firmness

Firmness can be used interchangeably with hardness and measured in Newtons and decreases with passage of ripening time (Salvador et al., 2007). A set of biochemical and physical changes occur in bananas during ripening taking several pathways like degradation of starch to sugars, changes in the peel and pulp colour, thickness and weight. Texture of the banana fruit is one of the physical characteristics that changes as bananas ripen and this is manifested by the softening of the banana. Hardness of most bananas does not change significantly at early stages of maturation but it does so as ripening progresses (Newilah et al., 2009). Softening of the fruit is caused by the loss of turgor, enzymatic degradation of starch granules while textural changes are predominantly due to the changes in the chemical structure of starch granules (Finney et al., 1967). However, Kojima et al., (1996) suggested that banana pulp softening process is due to the associated processes whereby the contents of pectin, polysaccharides and starch decrease during ripening. During ripening, pectin is broken down

by the enzymes pectinase and pectin esterase, in this process the fruit becomes softer as the middle lamella breaks down and cells become separated from each other (Anisa et al., 2013)

2.4.2 Colour

Colour of the banana fruit can be described by several colour coordinate systems (Soltani et al., 2011). Some of the most popular systems are CIELAB (International Commission on Illumination of 1976) and RGB (red, green, blue). The CIELAB colour space system is based on the concept that colours can be considered as combinations of red and yellow, red and blue, green and yellow and green and blue. In determination of the exact combination of colours of bananas, coordinates of a three-dimensional colour space are assigned (Kachru et al., 1995). The L* coordinate of an object is the lightness intensity as measured on a scale from 0 to 100, where 0 represents black and 100 represents white. The a* coordinate of an object represents the position of the objects colour on a pure green and pure red scale, where -127 represents pure green and +127 represents pure red. The b* coordinate represents the position of the object's colour on a pure blue and pure yellow scale, where -127 represents pure blue and +127 represents pure yellow (Soltani et al., 2011). The peel colour changes from green to yellow during ripening of banana fruit. External changes in peel colour during ripening often reflect changes in pulp colour (Wainwright and Hughes, 1990). Chlorophylls and carotenoids are the responsible pigments for this change, the chlorophyll content decreases and becomes absent whereas carotenoids increase in the ripe fruit (Maduwanthi and Marapana 2017).

2.4.3 Peel thickness

The banana fruit is naturally wrapped up in thick skin called the peel. The peel not only protects the fruit, but also plays a crucial role in compositional changes during ripening (Sandipkumar and Shanmugasundaram, 2015). The modification of cell wall of the peel may affect firmness loss and ultimately vanishing of void spaced and hence reduction in peel

thickness (Parker et al., 2013). When banana ripening is initiated, peel thickness and cell wall number decrease continuously (Parker et al., 2013).

2.4.4 Weight

Bananas in storage lose moisture through osmotic withdrawal caused by the difference in vapor pressure. Water loss eventually causes fruit mass reduction (Patil and Shanmugasundaram, 2015); the reduction in peel thickness accelerates the weight loss of the fruit with passage of ripening period.

2.4.5 Banana Pulp to peel ratio

The pulp to peel ratio of bananas in storage increases during the ripening period (Mohapatra et al., 2016). The increase is related to accumulation of moisture in the pulp derived from carbohydrate breakdown and osmotic transfer from peel to pulp (Palmer et al., 1971). Increasing the sugar content in the pulp could allow the water to move from peel to the pulp hence increase in the pulp to peel ratio. Patil and Shanmugasundaram (2015) reported a value of 5.97 at the end of the ripening period.

2.5 Aromatic substances in banana pulp and peel

Banana fruit has a pleasant flavor and is widely consumed throughout the world (Mohapatra, 2016). For over 40 years, numerous studies have been performed on the aroma constituents of bananas. The volatile profile of bananas has been established using different extraction and analytical methods on different cultivars from Philippines and Taiwan (Liu and Yang, 2002), Costa Rica and Canary Islands (Perez et al., 1997), Japan (Shiota, 1993), Madeira (Pontes et al., 2012), Cuba (Jorge and Yanet, 2013), Guadeloupe (Brat et al., 2004), Thailand and Brazil (Heliofabia et al., 2012). Most of the studies showed that major volatile compounds are generally esters (3-methylbutyl, 2-methylpropyl, and other uncommon esters), alcohols and ketones (Zhu et al., 2007), however only some of them have been recognized as banana flavor contributors (Jorge et al., 2017). About 152 aromatic compounds have been identified

from different cultivars of fresh and processed banana products (Shiota, 1993). However, only a few of these are recognized for their sensorial relevance to the banana flavor. The commonest include pentan-2-one and isoamyl and isobutyl esters along with alcohols including pentan-2-ol, hexan-2-ol, hept-4(Z)-en- 2-ol, oct-4(Z)- en-1-ol, and oct-5(Z)-en-1-ol (Shiota, 1993).

Benzaldehyde from banana fruits imparts aromatic notes of bitter almonds at low levels and maraschino cherries at higher levels (Dan et al., 2017). Esters such as 3-methylbutyl acetate and ethyl 3- methylbutanoate are also suggested to contribute to the fermented, chemical, and medicinal notes in dessert bananas (Bugaud and Alter, 2016). This is due to interactions of these volatiles leading to difference in perception of odours and aromas. Bugaud and Alter, (2016) also attributed the pineapple aroma note to presence of non-volatile organic acids; this note reduces perception of the banana aroma, suggesting possible interactions between volatile and non-volatile compounds.

The pungent and fruity notes in banana fruits are the result of two alcohols; 3-methyl-1-butanol (isoamyl alcohol) and 2-heptanol (Jordan et al., 2001). Methyl ketones (2-pentanone) contribute the fruity and banana-like notes while 2-heptanone is responsible for the fragrant and herbaceous notes in bananas. 2-nonanone contributes to the flowery and fatty notes in the fruit (Belitz et al., 2009). Therefore, it is evident that many volatile compounds from banana fruits including esters, alcohols, ketones and aldehydes contribute to the aromatic profiles of the products made from banana fruits as reported by Batista et al. (2017).

However, not all volatile compounds can be detected by human olfactory receptors. Therefore, most of the flavor contributors go unnoticed by the olfactory system (Schieberle, 1995).

2.5.1 Analysis of flavor volatiles in bananas

One of the best ways to separate flavor volatiles from food matrices is by use of gas chromatography–olfactometry (GC-O) on serial diluted aroma distillates. Specific techniques in this regard include aroma extract dilution analysis (AEDA) (Ullrich and Grosch, 1987) and Charm analysis (Acree et al., 1984). However, it is difficult to judge the exact direct contribution of each volatile detected from GC-O. In addition, screening methods such as AEDA do not permit a study on the exact sensorial relevance of each compound on the aromatic quality of the fruit when matching the overall odour impression of the bananas. Therefore, several other analytical methods have been developed to determine the exact contribution of different aroma compounds on the aromatic quality of foods (Jorge et al., 2017). For example, Gas Chromatography Mass Spectrometry (GC-MS), Headspace solid phase micro-extraction (HS-SPME), SPME-GC-MS and GC-flame ionization detection (GC-FID). An FID typically uses a hydrogen/air flame into which the sample is passed to oxidize organic molecules and produces electrically charged particles (ions). The ions are collected and produce an electrical signal which is then measured. As common with other GC techniques, a carrier gas is required with low water and oxygen impurities since water and oxygen can interact with the stationary phase and cause significant problems such as high baseline noise and column bleed in the output gas chromatogram which both reduces the analyzer sensitivity and decreases column lifetime. FID is also extremely sensitive to hydrocarbon impurities in the hydrogen and air supply for the flame (Boudhiroua et al., 2003). Hydrocarbon impurities can cause increased baseline noise and reduce the detector sensitivity. These techniques are widely applied to determine the exact role of the volatile components in food aroma quality (Marı et al., 2001; Christophe and Pascaline, 2016; Heliofabia et al., 2012; Jorge and Yanet, 2013).

2.5.2 Effect of ripening on flavor volatiles in bananas

Depending on the storage conditions, concentrations of aromatic compounds increase progressively as the bananas ripen from stage 0 (all green) to stage 6 (all yellow). The trend changes when the bananas are left to reach stage 7 (over ripe, all yellow with dark brown specs), previous studies by Boudhrioua et al., (2003) revealed that the total area of the peaks increases from 0.5 x 10^6 AU (AU: arbitrary unit) to 2 x 10^6 AU after 7 days of storage at room temperature. Salmon et al. (1996) also observed an increase in the aromatic content of bananas during ripening. These authors suggested that the increase is due to the biosynthesis of volatile components from amino acids such as valine and leucine and to β-oxidation, which is the main degradation process of fatty acids during ripening (Salmon et al., 1996).

2.6 Pectin and its properties

Pectin is a complex mixture of polysaccharides that makes up about one third of the cell wall dry matter of higher plants (Sundar et al., 2012). The highest concentration of pectin is found in the middle lamella of cell wall, with a gradual decrease as one passes through the primary wall towards the plasma membrane (Sundar et al., 2012). Pectin provides consistence and mechanical resistance to vegetal tissues (Canteri-Schemin et al., 2005). Pectin is often associated with other cell wall components such as cellulose, hemicellulose and lignin (Harholt et al., 2010). Pectins are mainly composed of polymers rich in galacturonic acid (Figure 1), with significant amounts of rhamnose, arabinose, galactose and other sugars and are characterized by three major chains i.e. homogalacturonan (HG), rhamnogalacturonan I (RG-I) and rhamnogalacturonan II (RG-II) (Fissore et al., 2009). The main chain of pectin may or may not be esterified with methyl-ester groups in the carboxylic acid units. The extent of esterification gives it the degree of esterification (DE) which varies from low to high. Pectins are commonly classified according to their DE as high methoxyl (HM) or low methoxyl (LM) pectins (Luzio , 2013), with a DE >50 % and <50 %, respectively. HM

pectins produce a gel under acidic conditions with high sugar concentrations (Evageliou et al., 2000); whereas LM pectins form gels by the interaction of divalent cations, especially Ca^{2+} with free carboxyl groups (Cardoso et al., 2003).

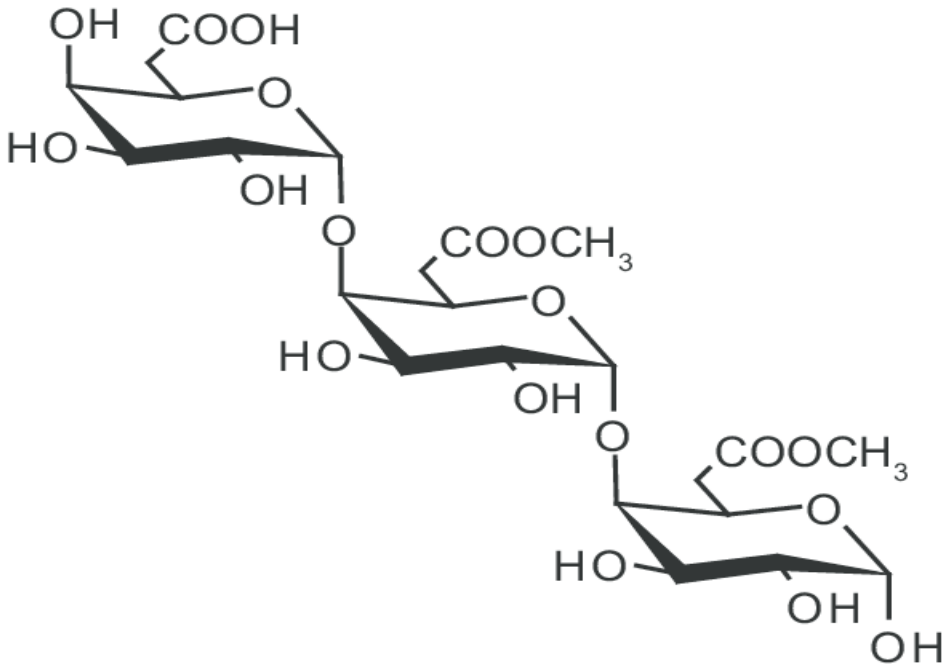

Figure 1. Structure of pectin (Assifaoui et al., 2015)

Pectin is able to form gels depending on it's molecular size, and DE, which also vary, depending on the source. Hence pectin from bananas does not form gels the same way as that from apples or oranges due to variations in molecular size and DE (Castillo et al., 2015). Pectin contains from a few hundreds to about 1000 saccharide units in a chain-like configuration; this corresponds to average molecular weights from about 50,000 to 150,000 Daltons (Sundar et al., 2012). According to the latter authors, large differences may exist between samples and between molecules within a sample and estimates may differ between methods of measurement. The amount and nature of pectin are critical for texture in fruits and vegetables during their growth, maturation, storage and processing (Jarvis et al., 2003). Pectin is also regarded as a high value functional food ingredient because of its excellent emulsifying properties and stability, which can be used as gelling agent and stabilizer

(Castillo et al., 2015). Previous researchers have found pectin to improve the sensory attributes of chips and reduce oil absorption in food products (Suyatma et al., 2015; Maity et al., 2015).

2.6.1 Pectin extraction

Extraction of pectin follows two major steps, hydrolysis of proto-pectin into pectin using acids, and subsequently precipitation by ethanol (Djilas, 2009; Pagan et al., 2001). The gravimetric method uses calcium chloride for precipitation instead of ethanol although both methods involve use of high temperatures (85 to 95^{o} C). For commercial purposes, the recovery of pectin is a crucial unit operation in order to provide adequate supply for the growing demand (Vasco-correa and Zapata, 2017).

Studies on the effects of process variables on extraction of pectin from bananas, banana peels and other materials have been investigated (Wai et al., 2010). Some studies found pectin yields from extraction of apple pomace, cocoa husks and passion fruit peels with citric acid to be similar to those obtained with hydrochloric acid (Canteri-Schemin et al., 2005; Chan and Choo, 2013). Pectin extraction yields vary with the method of extraction. Oliveira et al (2015) obtained yields of 5.2 to 12.2 % w/w dry weight from banana peels within range reported by Emaga et al. (2008b) which was from 2.4 to 21.7 %. The yield was enhanced by increasing temperatures and lowering pH. Conditions used by Emaga et al. (2008b) involved use of 90^{o} C at pH 1.5 for 240 min. and similar conditions except for the time were also adopted in this study. However, the harsher conditions reduce the quality of pectin by lowering the DE to 49 %. These authors reported time to have a positive effect on yield which was highly significant. Other studies by Garna et al. (2007) and Masmoudi et al. (2008) reported harsh conditions to favor pectin extraction yields from apple pomace and lemon by-products.

Depending on the extraction conditions, the quality of pectin also varies. Emaga et al. (2008) reported the DE values from 43.5 to 79 % when extracting pectin from banana peels using sulfuric acid. Oliveira et al. (2015) found optimum conditions of extraction using citric acid to be at 87^{o} C at pH 2.0 for 160 min. Emaga et al. (2012) and Garna et al. (2007) reported increased galacturonic acid (GA) with increasing extraction time and temperature. Chan and Choo (2013) also found that increasing extraction time resulted in higher uronic acid. Garna et al. (2007) demonstrated a very low pH (1.5) to result in higher extraction of low molecular weight compounds (non-pectic substances or degraded fractions of pectin) from apple pomace when compared to pH 2.0.

In the current study conditions of pH 2, a temperature ranging between 80^{o} C and 90^{o} C for 30 min. were adopted. Basing on the previous studies, pH 2 is the best for high quality pectin extraction and the temperature (80 to 90^{o} C) does not cause burning during extraction. In addition, care was taken to avoid long periods of direct heating which may cause thermal degradation of the pectin polymer. The extraction time was 30 min. since the pectin was already exposed by rupturing the cell walls of the bananas (peel and pulp) after crushing and blending in a laboratory blender.

Extraction of pectin using enzymes has also been applied. In this case, cell wall degrading enzymes with pectinolytic activity are used to hydrolyze non-pectin plant cell wall components (Puri et al., 2012; Fissore et al., 2009). Enzymatic extraction of pectin is environmentally safe and more effective in terms of pectin yield. Different enzymes such as polygalacturonase, hemicellulose, protease and microbial mixed enzymes, α-amylase, celluclast, alcalase, α-amylase and neutrase, xylase, cellulase, β-glucosidase, endopolygalacturonase and pectin esterase are used in pectin extraction as they have the ability to degrade pectin and modify its physicochemical properties (Yuliarti et al., 2011; Yu and Sun, 2013; Cui and Chang, 2014).

Pectin is the main polymer found in the middle lamella of tissue cells where it functions primarily as an intercellular adhesive providing mechanical support to plant tissues. Plant pectins can be water-soluble or insoluble (protopectins) and they are primarily made up of D-galacturonic acid joined by α - (1- 4) glycosidic linkages (Van Buren, 1991; Mukhiddinov, 2000) and 1, 2-D-rhamnose with D-galactose and D-arabinose side chains (Willats et al., 2001; Huisman et al.,2001). It is a complex mixture of blocks of homogalacturonic acid called 'smooth regions' mixed with blocks of homogalacturonic acid containing many neutral sugars including rhamnose, galactose, arabinose and glucose called 'hairy regions' (International Pectin Producers Association (IPPA), 2001). Neutral sugars are also present as side chains in different amounts depending on the pectin source and on the extraction method used.

Besides starch and water, pectin is the second most important component of bananas that affects their mechanical or structural and functional properties particularly in the living tissue where it causes firmness but also during cooking where it is involved in softening (Parre and Geitmann, 2005; Gafuma et al., 2018) particulalry in the presence of water where pectin undergoes solubilization. The general makeup of pectin varies with stage of maturity of the plant and it is easily brought into solution depending on the plant type. The hardness or strength of the green fruit is due to the protopectin or water insoluble pectic substances which are partially esterified polygalacturonic acid. The increased solubility of the pectic polysaccharides takes place during ripening of the fruit (Castillo et al., 2015). During ripening, pectin is broken down by the enzymes pectinase and pectin esterase, which makes the fruit, become softer. Initially, protopectin from hard green fruits is converted to pectin as the fruit matures or ripens while as it becomes over-ripe, the pectin is converted to pectic and pectinic acids.

2.6.2 Pectin characterization parameters

These are the quality indicators of pectin and of great importance; they include but not limited to equivalent weight, methoxyl content, anhydrouronic acid, degree of esterification and ash content.

2.6.2.1 Equivalent weight

Equivalent weight represents the quantity of pectin that is reactive which can undergo cross-linking reactions through polyol functional groups and is indicative of a high degree of esterification which in turn leads to a higher gelling power (Oekenfull and Scott, 1984). The equivalent weight of pectin is also the total content of free galacturonic acid that is not esterified in the molecular chains of pectin (Rangana, 1986). Pectin produced at lower pH normally has a higher equivalent weight due to the fact that low pH can cause polymerization of the pectin molecules into longer chains (Rouse, 1977; Uzma et al., 2015).

2.6.2.2 Methoxyl content

The methoxyl content is important in controlling the setting time of pectin, sensitivity to polyvalent metal cations and determines the functional properties of, and the pectin-gel texture (Constenla and Lozano, 2003). It also affects the dispersability of pectin in water where pectin with high methoxyl content is readily dispersible in water than that with less than 7.0% methoxyl content (Rouse et al., 1962).

2.6.2.3 Anhydrouronic acid (AUA)

AUA is important in determining the degree of purity of pectin, the degree of esterification and the physical properties of pectin. Pectin, which is a partly esterified polygalacturonide normally contains 10 % or more of organic materials composed of arabinose, galactose and other sugars (Pratik et al., 2017).

2.6.2.4 Degree of esterification

The degree of esterification (DE) is the ratio of the esterified galacturonic acid groups to the total galacturonic acid groups present on the pectin molecule. It is important in determining the gelling and adhesive power of pectin. The degree of esterification can be different depending on level of ripeness or maturity, part of the fruit, botanical source and method of isolation (Bonrood et al., 2005).

2.7 Fruit Jam processing

Jams are made by cooking crushed or chopped fruits with sugar until the mixture rounds up on a spoon and does not hold its shape but is spreadable (Sandra Bastin, 2004**).** Pectin is a carbohydrate found in fruits. When sugar is added, the pectin in fruit or commercial pectin precipitates out and forms insoluble fibers. An acid such as lemon juice or citric acid aids in the process. The insoluble fibers produce a mesh-like structure that traps the fruit juice or other liquid, much like a sponge absorbs water. This enables a gel to form. Slightly under-ripe fruit contains more pectin than ripe fruit. Overripe fruit may not contain enough pectin to form a gel. A general guideline is to use one-part under-ripe fruit to two parts fully ripe fruit for the best gel and flavor. The USDA canning guides recommend at least one-fourth of the fruit to be under-ripe (Bastin. 2015). Use of commercial pectin decreases cooking time, the pectin in fruit becomes water soluble when it is heated. So, for jelling to occur, the fruit must be heated. Too high of a temperature or cooking for too long can destroy the pectin, resulting in a poor gel. Too much pectin gives the jell a tough, rubbery consistency, making it difficult to spread.

Acid, The acidity level is also important for jelling. The gel does not set if there is too little acid. Too much acid causes the jell to lose liquid or weep (syneresis).

Sugar, Sugar is necessary for the gel to form. It also acts as a preserving agent and contributes flavor. When using low methoxyl pectin products, the recipes provided in the package should be the one followed (USDA canning guidelines).

CHAPTER 3

MATERIALS AND METHODS

3.1 Description of the banana samples

Three indigenous cooking banana cultivars were selected basing on their abundance on the Ugandan markets as well as their availability at Kawanda Agricultural Research Institute (KARI) farm. These included; 1.) *Musakala* (AAA-EA); the cultivar has large spacings between the fruit fingers and has a bunch which projects vertically down and the fruit fingers are about 20 cm long pointing up with a bottlenecked tip. 2.) *Mpologoma* (AAA-EA); this has got a bigger bunch with high market value although farmers do not prefer it due to its short lifespan and high sensitivity to climatic conditions, 3.) *Nakitembe*; this cultivar has bracts and floral bracts of the male flowers persisting on the rachis. The male bud is imbricated and the fruits have persistent style and stamina.

3.2 Sampling and Sample preparation

Each of the banana cultivars was harvested from KARI at green maturity (about 180 days after flowering) based on the following indices: when the ridges on the surface of the fingers had changed from angular to more round. When the dried parts of flowers at the top of the fruit would fall off easily and there was drying of the top most leaf and slight change of finger colour from dark green to pale green.

The samples were transported to Kyambogo University Food Technology laboratory and cut into clusters, washed in clean portable water and kept for ripening (Figure 2) by covering with a tarpaulin while monitoring the relative humidity (RH) and temperature (between 57 to 85 % and 24 to 31^{o} C, respectively). RH and temperature were recorded every after 8 h.

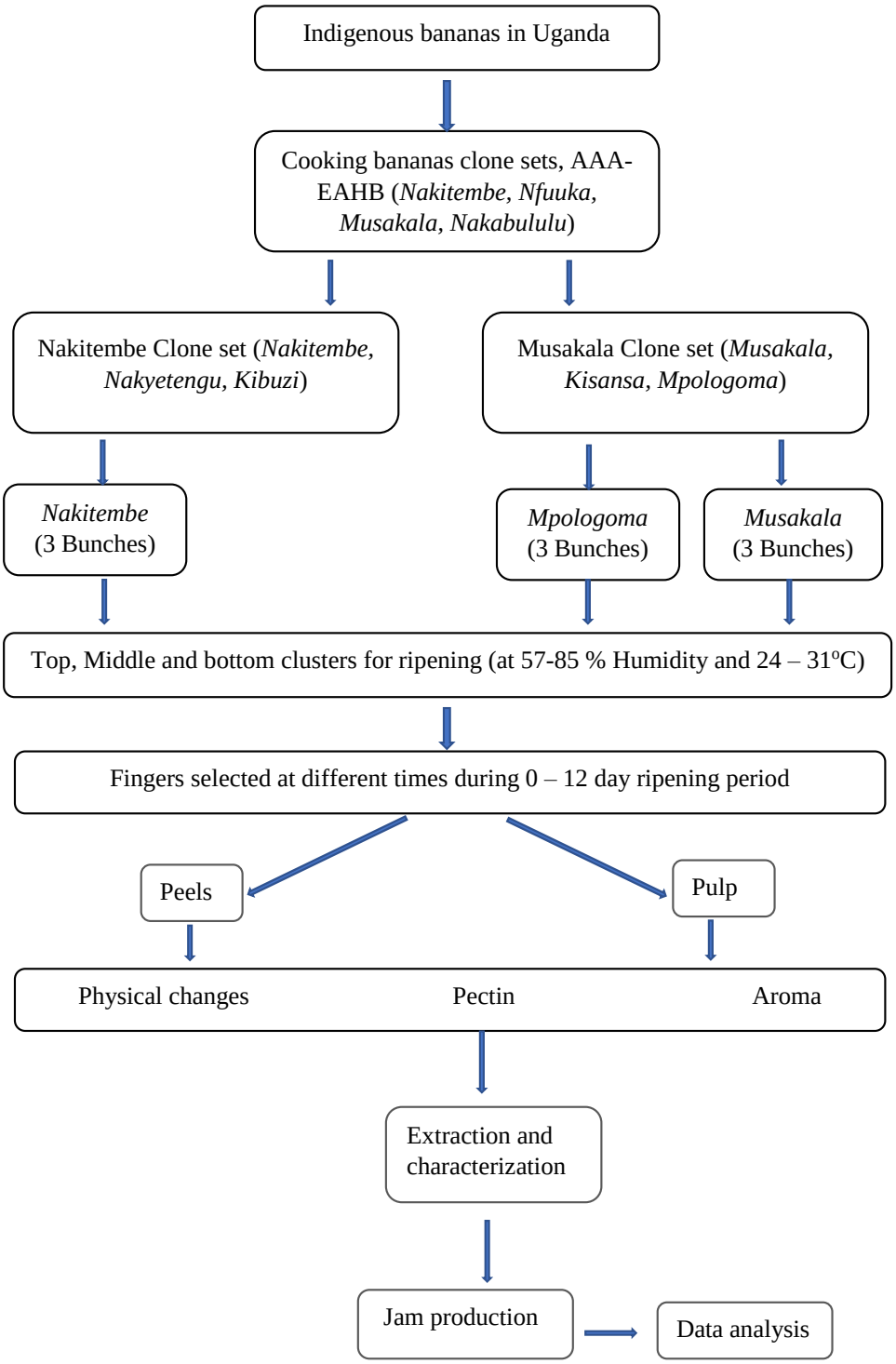

Figure 2. Sampling design

The samples that reached the desired level of ripeness based on the scale adopted from SH Prat's and Company (Luton UK) (Figure 3) were selected for further analysis and extraction of pectin and analysis of aromatic components (Figure 2).

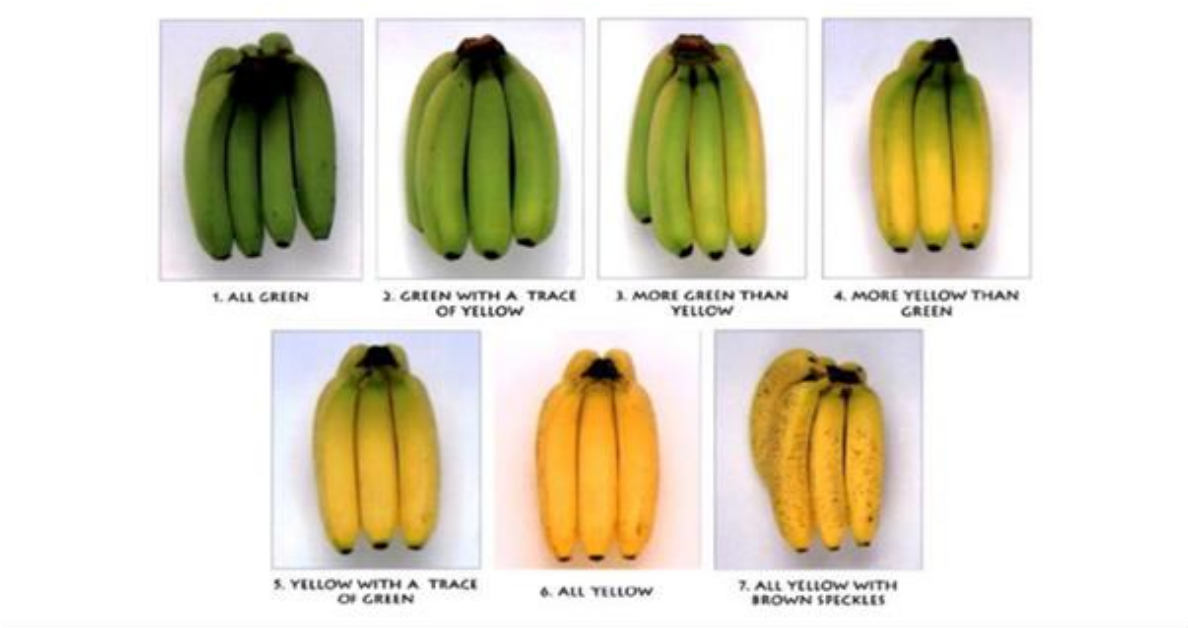

Figure 3. Colour Chart, SH Prat's & Co. (Luton, UK)

3.3 Chemicals and reagents

All chemical reagents used for sample extraction and analysis were of analytical grade and purchased from LabX Scientific Uganda Ltd. They included hydrochloric acid, sodium hydroxide, ethanol (95 %), sodium chloride, sodium metabisulphite, distilled wat er, phenolphthalein indicator, silver nitrate, glacial acetic acid, methylene chloride, cyclohexanone, anhydrous sodium sulfate and phenol red indicator.

3.4 Determination of physical properties

The physical properties including colour, texture, finger volume, finger diameter, pulp and peel thickness, fruit, pulp and peel weight were analyzed according to the method of Dadzie and Orchard (1997) with modifications. The 2nd and 3rd clusters from each cultivar were subjected to the experiment as in section 3.2. Samples were examined for the above physical parameters after every two days for a period of 12 days.

3.4.1 Measurement of peel and pulp colour

The colour of banana peel and pulp were determined using a Minolta colourimeter (Colour reader CR-10, Minolta Corp, Osaka, Japan) with a measuring range of L 10 to 100. Peel

colour was measured at three different parts- top, middle and end. To measure pulp colour, the banana fruit was cut at three different parts- top, middle and end. Readings for each cultivar were taken from the centre of the cut pulp section immediately after cutting each finger. Values of **L, a, b** were recorded in terms of International Commission on Illumination values (CIE-Lab), Where **L** represents brightness from black (0) to white (100), **a** represents green to red (-80 to +80) and **b** represents blue to yellow (-80 to +80) (Amorim et al., 2009). The colourimeter was standardized with black and white calibration tiles before taking each measurement.

3.4.2 Measurement of fruit texture

Texture measurements were carried out as hardness and cohesiveness using a TA-XT-plus Texture Analyzer (Stable M Micro Systems, Gudaiming, Surrey, UK) according to the method of Setiady et al. (2009) with modifications. Measurement was performed by penetration of the unpeeled fruit finger using a 6 mm penetration probe, in Return-to-start mode, 15 mm penetration distance, pre-test speed of 1.0 mm/s, test speed into sample of 2.0 mm/s, post-test speed of 10 mm/s, a trigger force of 0.049 N and calibrated using a 2 kg load cell. Each banana sample was positioned in the middle of the Texture Analyzer platform and start command executed. The measurements were performed in triplicate using three independent samples for each banana cultivar. The profiles of the force in form of texture curves were plotted using a computer interface and the force required to penetrate the banana sample was recorded as the first peak under the force-time curves and that indicated the hardness (N) as recommended by Jha et al. (2010a). The same analyzer recorded the resistant force (cohesiveness) as the probe retracted from the sample penetration.

3.4.3 Measurement of fruit, pulp and peel weight

Weights of the whole fruit finger, pulp and peel were separately measured using a digital top pan weighing scale (Note Book series, Japan) with pan dimensions 90 x 115 mm at operation

temperature of 25±5^{o} C with an accuracy of 0.01 g. First, the intact fruit was carefully placed on the scale on a flat surface and the stable reading recorded as the fruit weight (A). Then, the peel was carefully excised from the fruit using a stainless steel knife and its weight detrmined as before (B). The difference in weights (A-B) was regarded as the pulp weight.

The Pulp to peel ratio was obtained by dividing the pulp fresh weight by the peel fresh weight.

3.4.4 Determination of fruit specific gravity

The specific gravity was determined by using platform scale method (Patil and Shanmugasundaram, 2015). The weight of the individual fruit taken in air (A) and displaced water (B) were measured by using lab level weight balance (Note Book series, Japan), having 0.01 precision. Precaution was taken to ensure that the fruit was fully immersed in water without touching the beaker. The specific gravity was calculated using the following formula

$$\text{Specific gravity} = \frac{\text{Sample weight in air (A) x Density of water}}{\text{Weight of displaced water (B)}}$$

3.4.5 Measurement of fruit, pulp and peel thickness

The whole fruit diameter (A) was determined at the center area of each finger, perpendicular to its larger axis using a pair of Vanier calipers according to the protocol of Soltani et al. (2011). The diameter of the pulp (B) was measured accordingly after peel removal. The difference (A-B) was regarded as the peel thickness according the protocol of Dadzie and Ochard (1997). The data were expressed in millimeters.

3.5 Analysis of aroma volatiles

Ripened samples at stage 5 (Figure 3) were selected and labelled with a three-letter code (NAK for *Nakitembe*, MUS for *Musakala*, MPO for *Mpologoma*). The samples were transported to Kenya Bureau of Standards for analysis using head space gas chromatography-FID. Fresh stage 5 ripe *Matooke* from the three selected cultivars were peeled, finely sliced and then, 5 g of the fresh samples (pulp and peel) were placed into 40 ml head space vials,

and the vials were tightly sealed. The vials containing the sample were placed in the oven for 30 min. at 50^{o} C and then placed on the Headspace GC-MS instrument auto sampler tray for analysis.

3.5.1 Headspace analysis using SPME GC-FID

A 75-µm carboxen/polydimethylsiloxane SPME fibre (Supelco, Bellefonte, USA) was used for extraction of aroma volatiles from the headspace of sample vials prepared as in section 3.5. The fibre was first conditioned for 30 min. in the injection port at 250^{o} C. The SPME needle was then introduced into the septum in the lid of the vial using an auto-sampler (Teledyne tekmar versa headspace sampler, German) and the fibre exposed to the headspace for 30 min. at room temperature.

3.5.2 GC-FID analysis

The volatile compounds were detected by a GC-FID (Bruker Scion 456-GC gas chromatograph, USA) equipped with a polar column (60 m DB-WAX capillary GC column (internal diameter 0.32 mm, film thickness 0.25 m). The instrument was equipped with a split/splitless injection port operating at 250° C in splitless mode for 1 min. The oven temperature program was as follows: isothermal hold at 40° C (2 min.), constant rise to 190° C at a rate of 5° C min^{-1}, and isothermal hold at 190° C (1 min). The temperature of the FID was set at 250° C. High-purity nitrogen was used as the carrier gas with a flow rate of 1 ml min^{-1}. Make-up gas was also nitrogen, and its flow rate was 25 ml min^{-1}. Hydrogen and air were maintained at flow rates of 25 and 400 mL min^{-1}, respectively.

3.6 Extraction of pectin

Two extraction methods (ethanol versus calcium precipitation) were employed for extraction of pectin from the banana pulp and peel in order to determine the one, which optimizes yield. Ethanol precipitation was adopted from Castillo et al. (2015) whereas calcium chloride precipitation employed the method of Holt (1954) with modifications.

3.6.1 Pectin extraction using ethanol precipitation

Banana fingers from each of stages 2, 5 and 7 (Figure 3) were washed and sectioned into pulp and peel using a stainless-steel knife. The pulp and peel were analyzed separately in triplicate.

Then, 50 g of each sample was blended with 200 ml of 0.01 N HCl at pH 2. The acid (300 ml) was used to wash all the homogenized pulp from the blender into the beaker. The homogenized sample was digested with 0.01 N HCl by heating for 30 min. at 80-90^{o} C. After digestion, the solution was cooled to room temperature (~ 20 min.), filtered through a nylon cloth using cotton as a filter aid, and pressed to recover the extract. The residue was washed with 200 ml of 0.01 N HCl to further extract pectin from it. The filtrate was collected, pooled and then added with twice its volume of absolute (99 %, v/v) ethanol (VWR International, France). The precipitate was collected by filtration using a nylon cloth and oven (GenLab 12m064, UK) dried for 24 h at 50±5^{o} C. The dried pectin extract was cooled in a desiccator and then weighed. Pectin yield was calculated using the following formula;

$$\text{Pectin yield (\%)} = \frac{Ep \times 100}{Bi}$$

Where Ep = extracted pectin in grams.

Bi = Weight of banana pulp/peel in grams.

3.6.2 Pectin extraction using calcium chloride precipitation

Banana fingers from each of stages 2, 5 and 7 (Figure 3, section 3.2) were washed and sectioned into pulp and peel using a stainless-steel knife. Then, 50 g of the pulp/peel was blended and extracted with 300 ml of 0.01 N HCl for 30 min. at 80- 90^{o} C. The mixture was cooled, and filtered through a nylon cloth using cotton as a filter aid. The residue was boiled in 200 ml of 0.05 N HCl followed by 200 ml of 0.3 N HCl and then the filtrates pooled together. The filtrate was then neutralized with 1 N NaOH and added with an excess of 10 ml NaOH. The solution was left to stand overnight after stirring. Then, 185 ml of 1 M acetic acid

(Carlo Erba Reagents, France) was added and allowed to stand for 5 min. and 200 ml of 0.5 M calcium chloride (LOBA Chemie, India) solution was added and allowed to stand for 1 h and the solution boiled for 2 min. The solution was then filtered through a pre-weighed Whatman No.4 filter paper using a suction pump (Stuart RE3022C, UK). The precipitate was washed with hot distilled water until free from chloride (tested using 1 % silver nitrate). The filter paper containing calcium pectate was dried over night at 50±5^{o} C, cooled in a desiccator and weighed. Pectin yield was calculated using the following formula:

$$\text{Pectin yield (\%)} = \frac{\text{dried filter paper with calcium pectate – blank dried filter paper}}{\text{Weight of sample}}$$

3.7 Characterization of the extracted pectin

3.7.1 Equivalent weight (EQ.Wt.)

The equivalent weight (EQ. Wt.) of the extracted pectin was determined according to the method of Rangana (1986). About 0.5 g of pectin extract was weighed into a 250 ml conical flask and 1 ml ethanol was added. About 1.0 g of sodium chloride and 100 ml of distilled water were added and heated in a water bath (Stuart SWB24D, UK), for 30 min. at 45^{o} C. Finally, two drops of phenol red solution (Sigma-Aldrich, USA) were added and the solution titrated against 0.05 N NaOH to a purple colour and the titer volume recorded. Equivalent weight was calculated using the following formula:

$$\text{Equivalent weight} = \frac{\text{Weight of sample x 1000}}{\text{Ml of alkali x Normality of alkali}}$$

3.7.2 Methoxyl content (MeO)

Methoxyl content was determined using Rangana's method (1986). The neutral solution containing 0.5 g pectic substances from the equivalent weight determination (section 3.7.1) was used. To the neutral solution, 25 ml of 0.25 N sodium hydroxide solution was added. The mixed solution was stirred thoroughly and kept at room temperature for 30 min. in a flask

with a stopper. Then, 25 ml of 0.25 N HCl was added and titrated with 0.1 N NaOH to the same end point as the previous one in section 3.7.1. Methoxyl content was calculated using the following formula:

$$\text{Methoxyl content (\%)} = \frac{\text{ml of alkali} \times \text{Normality of alkali} \times 3.1}{\text{Weight of sample}}$$

3.7.3 Total anhydrouronic acid content (AUA)

Estimation of AUA was performed by making use of the titration volumes in equivalent weight and methoxyl content determinations (sections 3.7.1 and 3.7.2). Hence, total AUA was determined using the following formula adopted from Castillo-Israel et al. (2015).

$$\text{\% of AUA} = \frac{176 \times 0.1z \times 100}{W \times 1000} + \frac{176 \times 0.1y \times 100}{W \times 1000}$$

Where the molecular weight unit of AUA (1 unit) = 176 g; z = ml of NaOH from the equivalent weight determination; y = ml of NaOH from the methoxyl content determination and w = weight of the sample.

3.7.4 Degree of esterification (DE)

The DE was determined on the basis of methoxyl and AUA contents (Owen et al., 1952; Castillo-Israel et al., 2015) and calculated using the following formula:

$$\text{\% DE} = \frac{176 \times \text{\% MeO}}{31 \times \text{\% AUA}} \times 100$$

Where, % MeO = Methoxyl content % AUA =Anhydrouronic Acid content

3.7.5 Determination of ash content

The ash content of pectin extract was determined using method No. 923.3 of AOAC (2012) with modifications. One gram of the pectin extract was ground to pass a 75-micron mesh screen and placed into a tared platinum crucible then ignited in a muffle furnace (Carbolite

AAF1100, Germany) at 550^{o} C for 6 h after which the crucible containing ash was cooled in a desiccator and weighed. The ash content was calculated using the following formula:

$$\text{Ash content (\%)} = \frac{\text{Weight of ash}}{\text{Weight of pectin}} \times 100$$

3.8 Production of pineapple jam using banana pectin

To compare the extracted banana pectin with the commercially available citrus pectin as a gelling agent in fruit jam, the extracted banana pectin (*Nakitembe* stage 5) was utilized as an ingredient in pineapple jam processing.

3.8.1 Preparation of pineapple jam

Fifteen fairly sized pineapples were washed with potable water, peeled, cut into ~5 mm slices and blended into pulp. Then, 400 g of the pulp was transferred to a stainless-steel cooking saucepan and boiled for 5 min. and 600 g brown sugar added. The respective pectin ratio was added and the mixture cooked for 7 min. after which citric acid was added at a ratio of 1 citric acid: 60 fruit pulp. A set of 4 sample products was produced with different amounts of extracted banana pectin that is 0.0 %, 0.5 %, 1.0 % and 2 % of the pulp content and another set of 4 samples produced with commercial pectin in the ratios of 0.0 %, 0.5 %, 1.0 % and 2 % of the pulp were produced. Jars and their lids were sterilized by boiling in water and then filled with the prepared jam under cold water and kept upside down to cool overnight.

The jam samples were subjected to sensory evaluation and this was carried out at Uganda Industrial Research Institute (UIRI) food laboratory as outlined in section 3.8.2.

3.8.2 Sensory evaluation

Sensory acceptability attributes including colour, taste, mouth feel, flowability and overall acceptability were evaluated on a nine-point hedonic scale where 1 = dislike extremely and 9 = like extremely (Castillo et al., 2015).

Thirty untrained panelists (14 males and 16 females) of ages 20-35 participated in sensory evaluation, and all were randomly recruited at UIRI. After orientation, five digit-coded samples (Appendix 1) were given in random order to panelists along with a cup of water to cleanse their mouth between sample tasting.

3.9 Statistical analysis

All data were entered and analyzed by Excel (Excel 2013, USA) to generate the means and standard errors of the means for three sample replicates. Data were further analyzed for analysis of variance (ANOVA) and Tukey test in GraphPad (version 7, California) to separate the differences in means. The sensory evaluation results were analyzed using SPSS version 23 (SPSS Inc. Illinois, USA). P-values were set at $p<0.05$.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Physical properties of selected ripening indigenous cooking bananas

Physical parameters including peel and pulp colour, texture of the fruit finger, as well as fruit finger, pulp and peel weight were analyzed. The volume of the individual fingers and specific gravity per banana cultivar were determined and results are presented in Table 2. Changes undergone by the *Matooke* samples during ripening were evaluated to establish a basis on which ripened cooking bananas can be utilized for extraction of pectin and aroma rather than be dumped as waste.

4.1.1 Colour

The lightness (L^* values) of the pulp increased (Table 2) (from 36.2 to 40.3 for *Nakitembe*, from 36.3 to 40.4 for *Mpologoma* and from 35.7 to 41.6 for *Musakala*) through the days until the 10^{th} day. The changes in colour in terms of a^*-values increased from -9.2 to 5.3 for *Nakitembe*, from -8.7 to 5.3 for *Mpologoma* and from -9.7 to 4.8 for *Musakala* throughout the 12 days of ripening which indicated a decline in the greenness of bananas. In the same way the b^*-values (blue to yellowness) increased from 22.9 to 30.0 for *Nakitembe*, from 22.8 to 30.0 for *Mpologoma* and from 23.9 to 33.8 for *Musakala* until 10 days of ripening, and then decreased thereafter by day 12 probably due to over ripeness. The behavior for peel was different from that of the pulp (Table 2). For example, lightness (L^* values) of the peel decreased from 59.5 to 53.0 for *Nakitembe*, from 57.9 to 53.0 for *Mpologoma* and from 60.7 to 55.0 for *Musakala* through the days with some stabilization between days 6 and 10. The changes in peel colour in terms of a^*-values decreased from 5.2 to 2.8 for *Nakitembe*, from 4.2 to 2.7 for *Mpologoma* and from 2.6 to 1.6 for *Musakala* at the end of 12 days of ripening which indicated a decline in the greenness of banana peels during ripening. In the same way,

the b-values increased gradually with number of days and stabilized after 6 days of ripening representing an increase in yellowness of banana peels. Studies on different types of banana cultivars reported similar results (Yap et al., 2017, Elizagoyen et al., 2017). A suggestion from this and other previous studies (Yap et al., 2017) is that the development of darker shades and the loss of yellow tones could indicate over ripeness and loss of the banana fruit quality

Table 2. Changes in colour and texture of peel and pulp during ripening of the selected cultivars of indigenous cooking bananas

Time (days)	cultivar	Pulp colour			Peel colour			Texture (N)	
		L	A	a	L	a	B	Hardness(N)	Cohesiveness(N)
0	NAK	36.2±1.8^{cd}	-9.1±3.5^{f}	22.9±1.20^{d}	59.5±0.8^{a}	5.2±1.5^{a}	29.3±1.4^{cd}	80.47±10.07^{a}	-7.12±0.80^{a}
	MPO	36.3±1.5^{cd}	-8.7±0.7^{e}	22.8±0.7^{d}	57.9±1.6^{a}	4.2±0.9^{a}	29.8±0.6^{cd}	60.44±2.28^{a}	-5.26±1.60^{a}
	MUS	35.7±1.0^{d}	-8.8±2.4^{e}	23.9±4.9^{d}	60.7±0.3^{a}	2.6±1.8^{ab}	29.3±1.2^{cd}	76.81±3.43^{a}	-5.60±0.14^{a}
2	NAK	39.6±3.0^{c}	-9.2±2.0^{f}	23.0±2.3^{d}	58.4±1.4^{a}	2.4±0.3^{b}	29.5±1.7^{cd}	77.90±5.95^{a}	-5.93±1.04^{a}
	MPO	36.3±1.2^{cd}	-8.6±0.8^{e}	25.9±1.0^{cd}	59.5±1.0^{a}	1.1±0.3^{d}	31.2±0.7^{b}	80.48±2.61^{a}	-4.84±0.29^{a}
	MUS	36.4±1.1^{cd}	-9.7±1.9^{g}	29.9±3.2^{c}	59.3±1.3^{a}	1.0±0.2^{d}	30.9±0.9^{c}	76.81±3.43^{a}	-5.40±1.28^{ab}
4	NAK	39.6±2.9^{c}	-8.2±0.2^{e}	30.0±4.9^{c}	56.9±1.7^{b}	1.4±0.2^{cd}	29.1±0.2^{d}	84.52±3.0^{a}	-5.23±0.16^{ab}
	MPO	39.6±1.7^{c}	-8.0±1.1^{e}	27.0±1.6^{cd}	56.9±1.7^{b}	1.4±0.2^{cd}	29.1±0.2^{d}	74.36±10.16^{a}	-4.75±1.57^{a}
	MUS	39.8±1.9^{bc}	-6.8±1.3^{d}	27.3±1.6^{cd}	56.8±1.8^{b}	1.2±0.2^{d}	30.8±1.0^{c}	68.94±2.02^{a}	-4.21±0.64^{c}
6	NAK	41.9±2.7^{bc}	-7.9±1.7^{e}	32.3±4.7^{bc}	53.7±3.2^{c}	2.4±1.2^{bc}	32.2±3.6^{a}	56.09±25.12^{b}	-3.68±2.05^{bc}
	MPO	40.7±0.4^{bc}	-5.9±2.8^{d}	27.3±2.4^{cd}	53.7±3.2^{c}	2.4±1.2^{bc}	32.2±3.7^{a}	34.51±5.64^{b}	-2.68±0.45^{b}
	MUS	39.3±2.1^{cd}	-2.9±1.4^{c}	27.2±3.7^{cd}	57.9±0.4^{a}	0.7±0.3^{d}	31.6±2.7^{b}	41.38±2.77^{b}	-4.4±0.58^{bc}
8	NKT	44.2±1.2^{ab}	-4.0±0.6^{d}	32.8±1.6^{bc}	50.5±1.0^{d}	1.5±0.4^{cd}	31.9±1.5^{b}	33.19±5.51^{cd}	-3.11±1.49^{cd}
	MPO	43.5±0.4^{b}	-2.5±0.3^{c}	37.7±3.7^{ab}	53.8±1.5^{c}	2.6±0.2^{bc}	34.6±0.4^{a}	27.08±5.82^{cd}	-2.03±0.12^{b}
	MUS	45.0±2.2^{ab}	-1.1±0.4^{c}	38.2±1.9^{ab}	55.9±0.7^{b}	1.8±0.4^{cd}	34.1±0.8^{a}	23.30±0.05^{cd}	-2.18±0.32^{d}
10	NAK	47.7±2.2^{a}	-0.6±0.1^{c}	35.1±1.6^{b}	52.1±2.2^{c}	1.6±0.5^{cd}	32.9±1.9^{a}	14.35±3.06^{cd}	-1.44±0.48^{ef}
	MPO	47.4±2.4^{a}	2.8±1.2^{b}	40.4±1.6^{a}	55.7±0.7^{b}	1.3±0.1^{d}	34.4±0.5^{a}	13.36±9.56^{cd}	-2.12±0.81^{b}
	MUS	47.8±1.8^{a}	2.8±0.9^{b}	39.3±2.8^{a}	54.9±1.1^{b}	0.8±0.2^{d}	33.4±0.5^{ab}	19.71±2.15^{cd}	-2.00±0.04^{de}
12	NKT	40.3±3.8^{bc}	5.3±1.2^{a}	30.0±5.8^{c}	53.0±0.3^{cd}	2.8±0.3^{bc}	31.5±1.3^{b}	9.76±8.94^{d}	-1.02±0.34^{f}
	MPO	40.4±4.7^{bc}	5.3±1.6^{a}	30.0±6.4^{c}	53.1±2.0^{w}	2.7±0.4^{bc}	33.0±1.7^{a}	11.00±10.74^{cd}	-1.26±0.19^{b}
	MUS	41.6±1.5^{bc}	4.8±0.4^{a}	33.8±2.4^{bc}	54.9±0.9^{w}	1.6±0.3^{cd}	33.0±1.4^{a}	13.46±1.6^{cd}	-1.06±0.23^{e}

Mean values in the same column with different superscript letters are significantly different ($p<0.05$). Values are means of three independent determinations ±standard errors of the means. MUS, *Musakala*; NAK, *Nakitembe*; MPO, *Mpologoma*. *L** for the lightness from black (0) to white (100), *a** from green (−) to red (+), and *b** from blue (−) to yellow (+)

4.1.2 Texture

Texture was expressed in terms of hardness, which is the ability of the sample to resist plastic deformation by penetration, and cohesiveness, which is the stickiness of the sample to the penetration probe.

4.1.2.1 Hardness

Hardness of the selected cooking banana cultivars generally reduced with increase in the number of days of ripening (Figure 4). With *Nakitembe* having the greatest reduction of 87.87 % from 80.47 N (at zero days) to 9.76 N (after 12 days) followed by *Mpologoma*, which reduced by 86.34 % from 80.48 N (at zero days) to 11.00 N (after 12 days) and lastly *Musakala* reduced by 74.26 % from 76.81 N (at zero days) to 19.77 N (after 12 days).

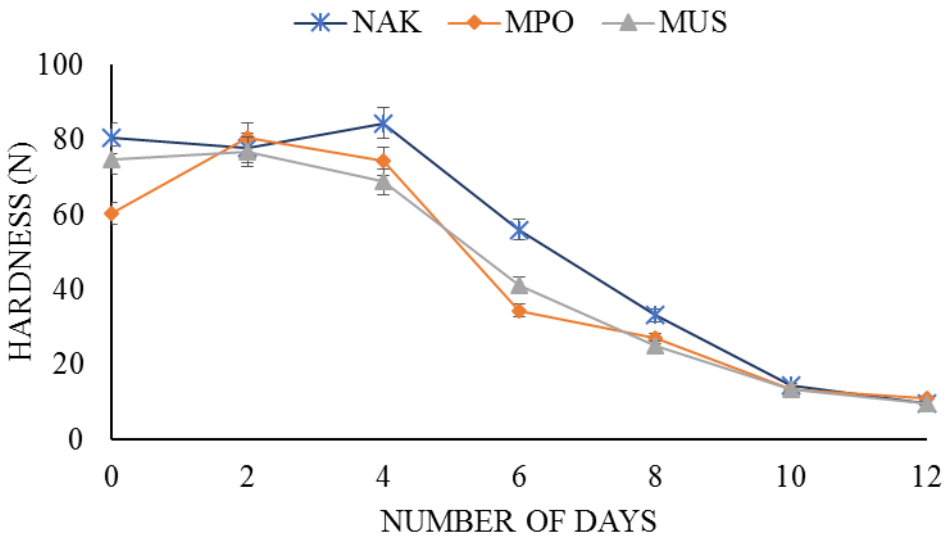

Figure 4. Changes in fruit hardness of the selected cooking banana cultivars during ripening at 57-85 % RH and 24-31° C. NAK, *Nakitembe;* MUS, *Musakala;* MPO, *Mpologoma*. Error bars are standard errors of means.

4.1.2.2 Cohesiveness

Cohesiveness of the selected banana cultivars generally increased throughout the ripening period indicating loss of firmness of the fruit as ripening progressed. At day zero, all the cultivars had cohesiveness lower than -5 N, which increased to -1.06 for *Musakala*, -1.02 for

Nakitembe and -1.26 for *Mpologoma* at day 12 (Figure 5). Overall, at the end of ripening, *Nakitembe* had the highest level of cohesiveness whereas *Mpologoma* had the lowest.

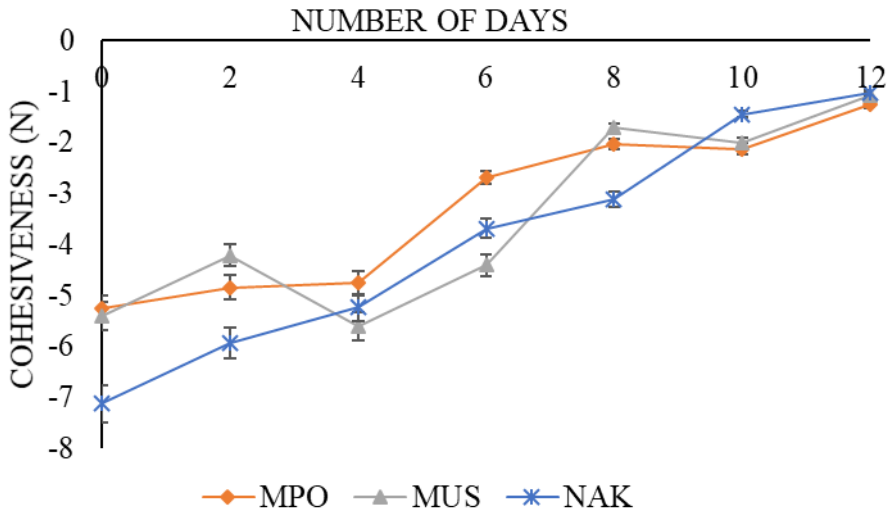

Figure 5. Changes in cohesiveness of the selected cooking banana cultivars during ripening at 57-85 % RH and 24-31^{o} C. NAK, *Nakitembe;* MUS, *Musakala;* MPO, *Mpologoma*. Error bars are standard errors of means.

According to previous studies, textural changes may be attributed to many physiological and biochemical evolutions that include degradation of pectin by enzymes (Salvador et al., 2007), water migration from the skin, hydrolysis of starch to sugars (Prabha and Lakshmi, 1998) and the degradation of starch to sugars (Cordenunsi and Lajolo, 1995; Shiga et al., 2011) during ripening. These effects can cause an increase in osmotic pressure in banana flesh, resulting in decrease of the turgor pressure accounting for softening during ripening. Overall, this results in a decrease in firmness and increase in the sweetness of the pulp. Results of the current study (Figure 4 and 5) showed that the firmness of the peel and pulp significantly reduced ($p<0.05$) from 80 N to 10 N over the ripening period.

4.2 Weight

The changes in weighting properties of the banana samples including, weights of fruit, peel and pulp, pulp to peel ratio are presented in Table 3. The average weight of fruit generally reduced throughout the 12 days of ripening with that of *Nakitembe* (NAK) from 194.29 g to 147.96 g (23.8 % reduction), *Musakala (*MUS) from 181.35 g to 166.52 g (8.2 % reduction) and *Mpologoma* (MPO) from 174.29 g to 144.53 g (17.1 % reduction). The results of this study relate to those reported by Soltani et al. (2011) for fully ripe *Cavendish* variety that is 180.56 g. Kachru et al. (1995) reported 89.69 g and 126.16 g of whole fruit for Dwarf *Scavendish* and *Nendran* varieties, respectively at the end of ripening. The average weight of fruit pulp slightly increased through the 12 days of ripening. NAK increased from 119.4 g to 124 g (3.9 % increase), MUS from 106 g to 139 g (31.1 % increase) and MPO from 92.24 g to 106 g (14.9 % increase). Soltani et al. (2011) reported an average of 115.54 g for fully ripe *Cavendish* bananas which is within the range (106-139 g) obtained in this study. On the other hand, the peel weight reduced generally across cultivars and within cultivars over the 12 days' period of ripening. Peels of NAK reduced from 74.10 g to 40.48 g (45.4 % reduction), MUS reduced from 49.69 g to 23.59 g (52.5 % reduction) and MPO reduced from 65.70 g to 34.30 g (45.8 % reduction) with *Musakala* presenting the smallest amount of peel (as seen in Table 3) and yet reduced most in the weight.

Table 3. Changes in physical parameters of the selected Ugandan indigenous cooking banana cultivars during ripening

Time (days)	cultivar	Fruit weight (g)	Pulp weight (g)	Peel weight(g)	Pulp/Peel ratio	Specific gravity	Fruit diameter (mm)	Pulp thickness (mm)	Peel thickness (mm)
0	NAK	194.29±8.0^{a}	119.4±3.85^{b}	74.1±6.57^{a}	1.61±0.59^{d}	1.03±0.3^{a}	42.0±0.5^{a}	35.0±1.0^{bc}	3.5±0.06^{ab}
	MUS	181.35±9.2^{ab}	106.16±7.53^{c}	75.29±7.0^{a}	1.41±2.70^{d}	0.94±0.6^{c}	39.13±1.6^{bc}	31.7±0.6^{d}	3.97±0.1^{a}
	MPO	157.95±8.64^{cd}	92.24±12.17^{d}	65.7±7.80^{ab}	1.40±1.56^{d}	0.96±0.7^{bc}	39.97±4.8^{ab}	32.0±1.0^{cd}	4.03±0.1^{a}
2	NAK	198.33±2.33^{a}	124.17±3.73^{b}	72.9±5.32^{a}	1.70±0.70^{cd}	0.96±0.3^{bc}	40.6±0.2^{ab}	34.2±0.3^{bc}	3.30±0.3^{ab}
	MUS	194.76±5.91^{a}	120.05±2.02^{ab}	74.1±3.74^{a}	1.62±0.42^{cd}	0.98±0.3^{b}	39.0±0.3^{b}	34.5±1.3^{bc}	3.70±0.3^{ab}
	MPO	174.29±7.38^{bc}	110.36±5.35^{ab}	61.90±4.6^{ab}	1.78±1.17^{cd}	0.99±0.3^{ab}	41.27±2.6^{a}	34.7±0.3^{bc}	3.53±0.1^{ab}
4	NAK	196.59±3.28^{a}	124.22±1.85^{ab}	68.49±2.5^{ab}	1.81±0.75^{cd}	0.96±0.2^{bc}	41.10±1.1^{a}	35.3±0.8^{ab}	3.17±0.3^{b}
	MUS	192.10±6.73^{a}	122.69±8.09^{ab}	65.96±0.2^{ab}	1.88±2.25^{cd}	0.98±1.4^{b}	40.2±0.7^{ab}	35.5±0.5^{ab}	3.13±0.2^{b}
	MPO	158.65±3.57^{cd}	97.58±4.05^{cd}	58.51±0.9^{ab}	1.67±4.71^{cd}	0.91±0.6^{cd}	39.10±0.5^{ab}	34.2±0.6^{bc}	3.30±0.3^{ab}
6	NAK	184.83±2.6^{ab}	114.15±5.42^{ab}	66.99±5.3^{ab}	1.70±1.03^{cd}	0.89±0.8^{d}	39.53±0.5^{ab}	33.2±0.7^{cd}	2.5±0.29^{bc}
	MUS	178.83±12.9^{ab}	116.69±8.36^{ab}	61.42±0.7^{ab}	1.90±1.62^{cd}	0.95±0.7^{bc}	39.7±0.3^{ab}	33.0±0.5^{cd}	2.67±0.3^{bc}
	MPO	155.11±11.5^{cd}	106.09±14.50^{ab}	48.80±2.9^{bc}	2.17±5.02^{c}	0.93±2.5^{cd}	37.07±0.4^{cd}	32.9±0.2^{c}	2.30±0.3^{bc}
8	NAK	172.1±15.2^{ab}	119.55±9.34^{bc}	56.54±6.1^{b}	2.11±1.54^{c}	0.93±0.7^{cd}	40.0±2.3^{ab}	34.7±0.3^{bc}	2.33±0.3^{bc}
	MUS	170.7±14.4^{ab}	124.83±12.16^{ab}	28.54±2.3^{cd}	2.89±4.62^{b}	0.95±0.5^{bc}	39.6±0.7^{bc}	35.2±0.8^{ab}	2.47±0.1^{bc}
	MPO	136.98±15.1^{e}	101.92±17.91^{bc}	35.18±2.4^{cd}	2.90±7.49^{b}	0.97±1.6^{bc}	39.2±0.29^{bc}	33.0±1.2^{cd}	1.90±0.3^{cd}
10	NAK	160.17±8.4^{bc}	114.22±10.43^{ab}	42.53±4.9^{bc}	2.69±2.12^{b}	0.97±4.0^{bc}	38.27±0.8^{c}	33.8±0.8^{bc}	2.07±0.1^{c}
	MUS	166.52±17.4^{b}	127.72±7.15a^{b}	26.20±1.4^{cd}	4.87±0.90^{a}	0.93±0.5^{cd}	38.9±1.7^{bc}	35.8±1.3^{ab}	1.67±0.3^{cd}
	MPO	146.13±8.42^{cd}	110.64±7.64^{bc}	35.90±2.0^{cd}	3.08±3.75^{b}	0.91±3.6^{cd}	36.63±0.3^{cd}	34.2±1.3^{bc}	1.67±0.3^{cd}
12	NAK	147.96±2.33d	105.78±2.56^{c}	40.48±0.3^{c}	2.61±7.76^{b}	0.97±1.0^{bc}	38.20±0.8^{cd}	35.5±1.3^{ab}	1.47±0.1^{cd}
	MUS	174.13±24.2^{a}	139.04±21.74^{a}	23.59±1.0^{d}	5.89±12.8^{a}	0.96±0.7^{bc}	38.3±0.7^{c}	36.9±0.7^{a}	1.03±0.1^{d}
	MPO	144.53±11.3^{cd}	106.91±7.47^{ab}	34.30±1.8^{cd}	3.12±4.24^{b}	0.95±1.1^{bc}	36.37±1.6^{d}	35.2±1.3^{ab}	1.46±0.1^{cd}

Mean values in the same column with different superscript letters are significantly different ($p<0.05$). Values are means of three independent determinations ±standard errors of the means. MUS, *Musakala*; NAK, *Nakitembe;* MPO, *Mpologoma.*

4.3 Dimensional Properties

The changes in dimensional properties including fruit diameter, pulp thickness and peel thickness of the selected banana cultivars over the 12-day ripening period are presented in Table 3. The fruit diameter reduced from 42.03 mm to 38.20 mm (9.1 % decrease) for NAK, from 39.13 mm to 38.30 mm (2.2 % reduction) for MUS and MPO from 39.97 mm to 36.37 mm (9.0 % reduction) indicating a slight decrease through the experimental period across cultivars.

The current results showed that during ripening, the peel thickness reduced with time for all the cultivars.

Nakitembe pulp thickness reduced from 3.53 mm to 1.47 mm (58.4 % reduction), *Musakala* reduced from 3.97 mm to 1.03 mm (74.1 % reduction) and from 4.03 mm to 1.47 mm (63.5 % reduction) for *Mpologoma* with *Musakala* having the greatest reduction followed by *Mpologoma* and *Nakitembe* in that order. The specific gravity of the cooking banana cultivars did not change significantly ($p>0.05$) and values for all cultivars were between 0.89 (for *Nakitembe* at day 6) to 1.03 (for *Nakitembe* at day 0) throughout the ripening period as shown in the Table 3.

4.3.1 Pulp to peel ratio

The study showed no significant difference in the pulp to peel ratio (Figure 6) among the cultivars in the first 0-6 days of ripening ($p>0.05$). However, significantly different ratios were observed in the next 6-12 days of ripening ($p<0.05$). The ratio was significantly higher in *Musakala* compared to *Mpologoma* and *Nakitembe* cultivars.

The pulp to peel ratios of all the three banana cultivars were higher than 1 (>1) (Figure 6) and ranged between 1.4 and 1.6 at zero days. This ratio then increased for all the cultivars throughout the storage days. At the end of ripening, *Musakala* had the highest ratio of 5.89. The pulp to peel ratio for *Nakitembe* increased from 1.61 to 2.61 (62.1 % increase), from 1.41

to 5.89 (317.7 % increase) for *Musakala* and from 1.40 to 3.12 (122.9 % increase) for *Mpologoma*.

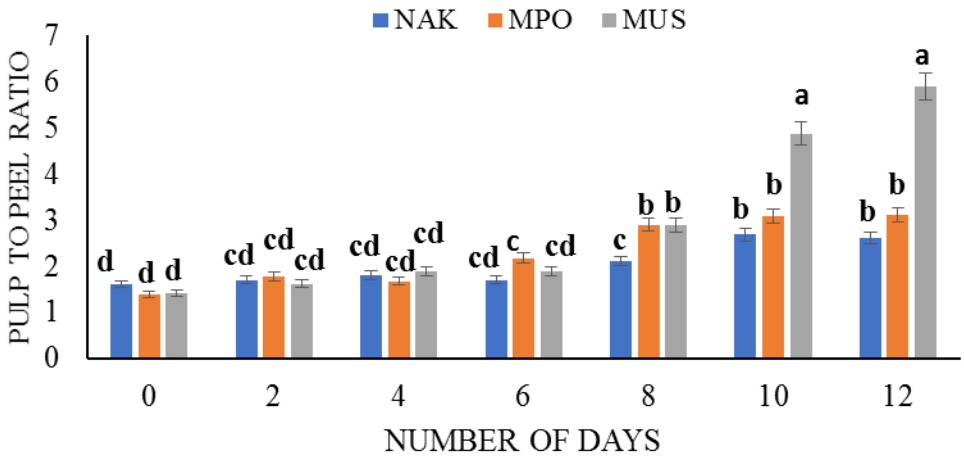

Figure 6. Changes in pulp to peel ratio of the selected indigenous cooking banana cultivars during ripening. Bars with different superscript letters are significantly different ($p<0.05$). NAK, *Nakitembe*; MUS *Musakala*; MPO, *Mpologoma.* Error bars are standard errors of means.

4.4 Aromatic compounds

In the analysis of volatile compounds present in the selected indigenous Ugandan cooking bananas, a total of 19 compounds represented by the different peaks on the chromatograms (A-Q and X) as shown Figure 7 were detected. A total of 17, 19 and 18 compounds were detected in *Mpologoma, Nakitembe* and *Musakala* pulp extracts respectively, while up to 17, 18 and 17 compounds were detected in each of their respective peel as well. In general, the relative abundance of ions from the pulp was higher than that from peel (Table 4). Some compounds were more abundant in one cultivar compared to another irrespective of pulp or peel. On average compound F (retention time 5.5 min.) had the highest abundance in all the cultivars from both the pulp and peel. Compound X (retention time 6.6 min.) was detected in *Mpologoma* and *Musakala* pulp and peel but not detected in *Nakitembe.* Likewise, compound N (retention time 12.6 min.) was detected in *Nakitembe* pulp and peel but not in *Mpologoma* and *Musakala.* Studies reported by Jordan et al. (2001) showed that the most abundant

components found in banana essence are 3-methyl-1-butanol followed by 2-pentanone, 2-pentanol acetate, and 2-methyl-1-propanol. These authors also reported that alcohols are quantitatively the most abundant components in the aromatic profile of the banana essence representing 55.6 % of the total components identified. It is therefore possible that the most abundant ions (B, C, D, E, F, G, H and K) in the pulp and peel of banana samples examined in this study comprised of esters and alcohols.

It should be noted that not all the 19 compounds detected in the current study, contribute to the overall banana flavor, therefore further research is required to identify the ions and confirm each of their aroma activity in order to determine their individual contributions to the banana flavor.

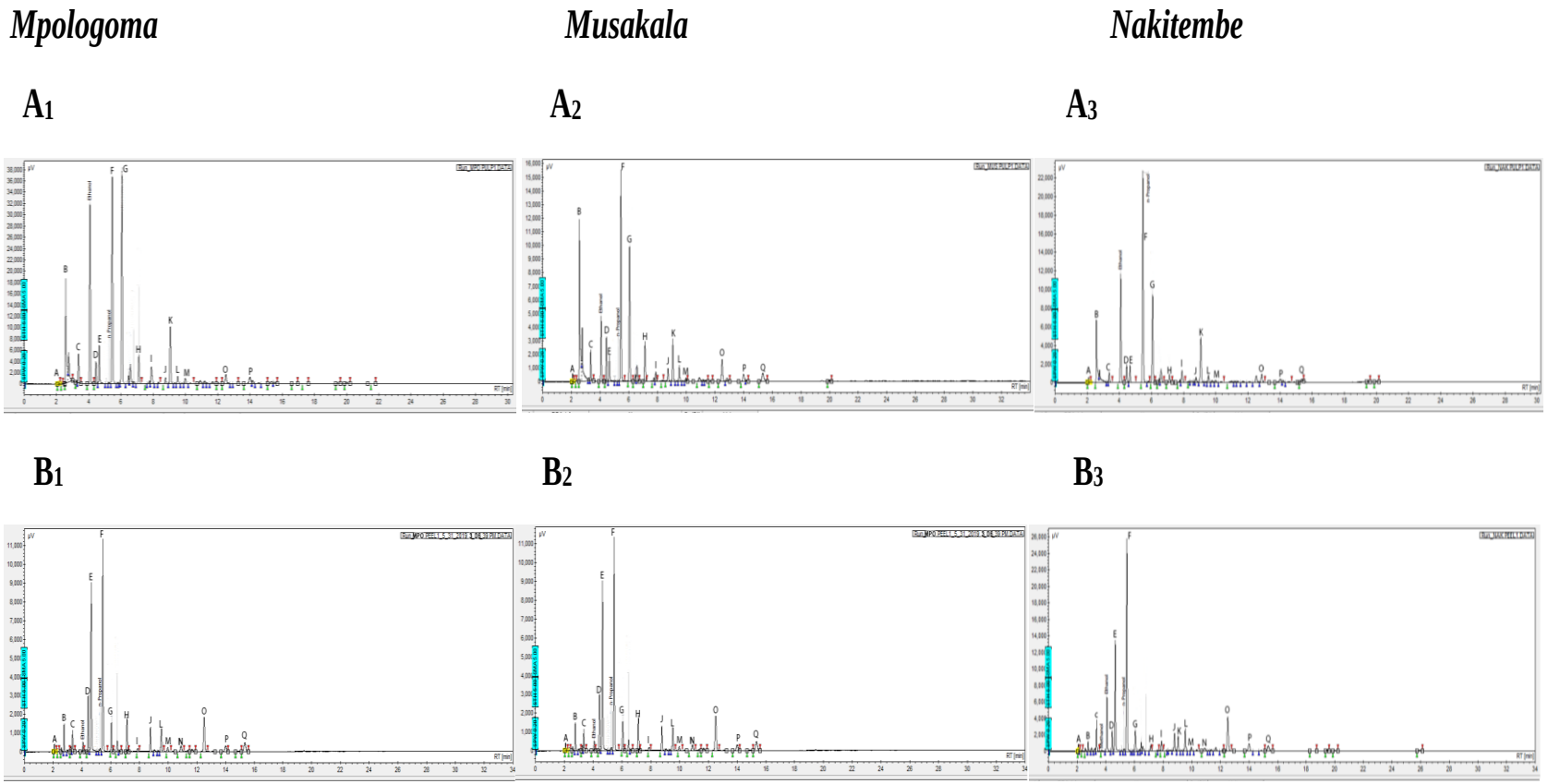

Figure 7. Spectrum of aroma ions in (**A_1 –A_3)** pulp, (**B_1-B_3)** peel of the selected indigenous cooking banana cultivars, *Mpologoma* (**A_1** and **B_1)**, *Musakala* (**A_2** and **B_2),** *Nakitembe* (**A_3** and **B_3)**

Table 4. Aromatic ions in the peel and pulp of the selected indigenous Ugandan cooking banana cultivars and their relative abundances

Compound	RT (minutes)	Relative abundances (µV)					
		Mpologoma		*Musakala*		*Nakitembe*	
		Pulp	Peel	Pulp	Peel	Pulp	Peel
A	2.0	200	200	600	600	500	800
B	2.4	18400	ND-	12000	1500	15200	1200
C	3.4	3600	4000	2400	1200	1200	3600
Ethanol	4.2	32000	1600	14800	600	11800	2800
D	4.4	4000	2800	3200	3000	2000	2400
E	4.7	6800	20000	1800	9100	2000	13400
n-Propanol	5.0	D	D	D	D	D	D
F	5.5	36800	28000	15200	11400	22000	26000
G	6.0	38000	7800	10000	1600	9600	2400
X	6.6	3200	ND	ND	800	ND	ND
H	7.2	5600	2000	3000	2000	800	800
I	8.0	3600		800	200	1250	1200
J	8.8	800		1000	1400	500	2000
K	9.0	10000		1200	ND	4800	1600
L	9.6	1200		1000	1200	800	3000
M	10.0	100		200	200	400	600
N	12.6	ND		ND	200	700	400
O	12.8	1600		1600	1800	800	4000
P	14.0	1200		600	200	400	1000
Q	15.4	ND		600	600	400	800

ND means not detected; D means detected but abundance is very low.

Letters A-Q and X represent volatile compounds.

The relative abundances were read directly from chromatograms y-axis at each specific retention time

4.5 Pectin yield

Pectin yield was evaluated using two different methods, one that uses ethanol as the precipitating agent and the second, which uses calcium chloride as the precipitating agent for the solubilized pectin.

4.5.1 Pectin yield using ethanol as the precipitating agent

The percentage yield of pectin on wet basis from the different banana cultivars is shown in Figure 8.

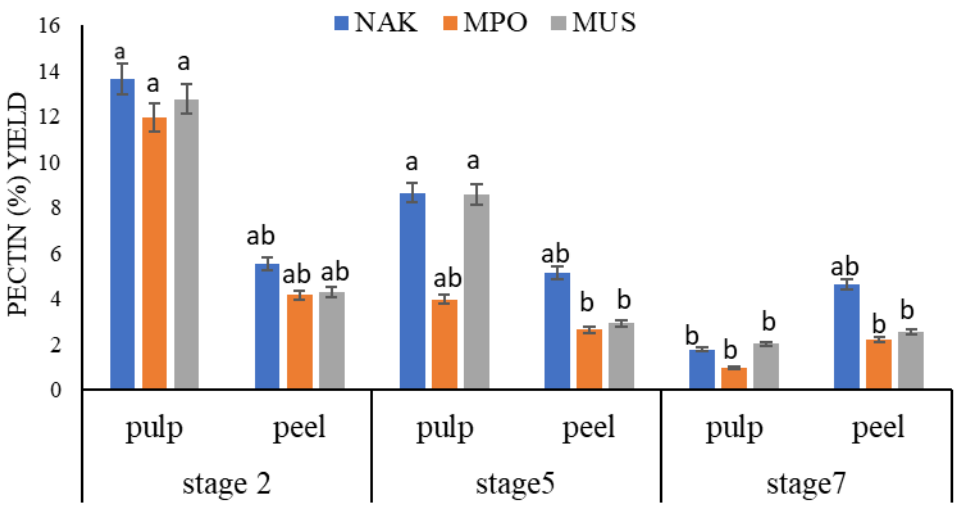

Figure 8. Pectin yield from peel and pulp of the selected indigenous Ugandan cooking bananas at stage 2, 5 and 7 of ripening using ethanol as the precipitating agent. Bars with different superscript letters are significantly different ($p<0.05$). NAK, *Nakitembe*; MUS, *Musakala*; MPO, *Mpologoma*. Error bars are standard errors of means.

There was no significant difference ($p>0.05$) in the pectin yield from *Nakitembe* peel throughout the ripening stages. Using Tukey test, it was also observed that there was a significant difference ($p<0.05$) between the yield from pulp at stage 7 and that from stages 2 and 5 except for that from *Nakitembe*.

Pectin yield is affected by the maturity and ripeness stage of the bananas since the hemicelluloses, celluloses and lignins vary as the fruit matures (Castillo et al., 2015). In this study, pectin yield was observed to decrease with increased ripening period. For instance, pectin yield from the pulp of *Nakitembe*, *Musakala* and *Mpologoma* was 13.67, 12.76 and 11.95 %, respectively at stage 2. At stage 5, the yield reduced to 8.64, 8.58 and 3.95 %, respectively which further reduced to 1.75, 2.00 and 0.96 %, respectively at stage 7. At stages 2 and 5, *Nakitembe* pulp and peel had the highest pectin yield followed by *Musakala* and *Mpologoma* having the least whereas at stage 7, the peels followed the same trend. At stage 7 still, *Musakala* had the highest pectin yield from the pulp and *Mpologoma* had the least. The results from this study could be related to those obtained in previous studies. Baker (1997) reported pectin content of fully ripe bananas to range between 0.5 to 1.28 %, whereas Kawabata and Sawayama (1974) examined bananas from three countries (Philippines, Ecuador and Taiwan), and found levels of calcium pectate to range from 0.55–0.68 %, with an average of 0.63 %. At first, pure pectin yield generally increases at the expense of protopectin as the fruit becomes more tender, making the connections between the pectin and other cellular compounds more fragile thus making pectin more fragile for extraction. However, over ripening of bananas results into a decrease in yield due to the degradation of pure pectin under enzymatic activities such as polygalacturonase, pectin methyl esterase and pectate lyase (Emaga et al., 2007).

The pectin yield from the peels of the different banana cultivars also reduced at the end of ripening. At stage 2, the peel from *Nakitembe* had the highest yield of 5.53 % followed by *Musakala* at 4.27 % and the least amount (4.15 %) was detected in *Mpologoma*. At stage 5, *Musakala* peel had the least (2.92 %) pectin content, followed by *Mpologoma* (4.15 %) and *Nakitembe* with the highest at 5.13 %. Pectin yield from the peel of *Nakitembe* was generally stable throughout the ripening stages.

It was observed that pectin yield was higher in the pulp than the peel at stages 2 and 5 for all the cultivars. However, the results were higher for the peel than pulp at stage 7 of ripeness. Overall, the results showed that pectin yield of *Mpologoma* was the lowest among the three cultivars from both peel and pulp at all ripening stages while it was highest from *Nakitembe* (Figure 8).

4.5.2. Pectin Yield from peel and pulp using calcium chloride as a precipitating agent

Pectin yield as calcium pectate was highest at stage 2 of ripeness in the pulp of *Musakala* (11.14 %) followed by *Nakitembe* (9.69 %) and *Mpologoma* (4.90 %) presenting the least pectin yield (Figure 9). In the peels at stage 2 of ripeness, pectin yield from *Mpologoma* (6.35 %) was the highest followed by *Nakitembe* (6.21 %) and *Musakala* (1.69 %). *Musakala* had a significantly ($p<0.05$) lower yield in the peels as compared to its pulp at stage 2 of ripeness. Generally, there was no significant difference ($p>0.05$) in the pectin yield from both the pulp and peel of the same cultivars at stages 5 and 7 of ripening.

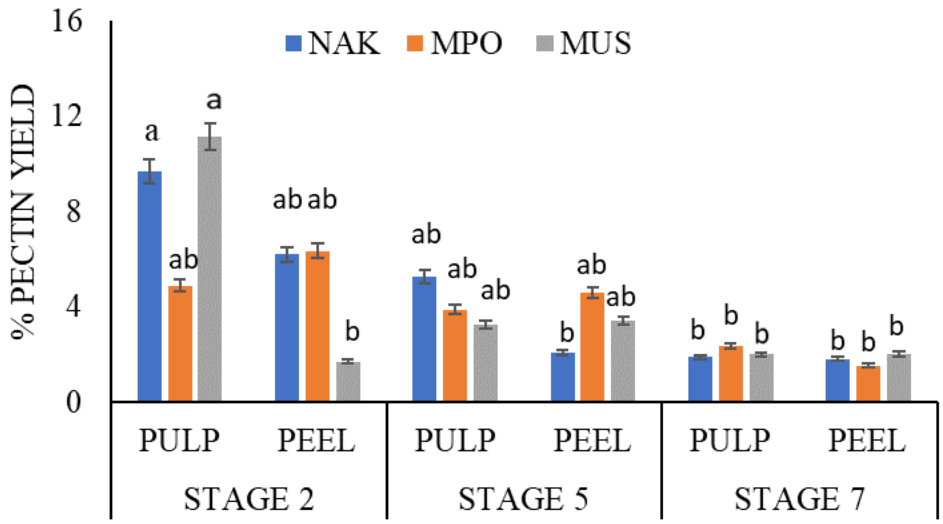

Figure 9. Pectin yield from peel and pulp of the selected indigenous cooking bananas at stage 2, 5 and 7 of ripeness using calcium chloride as the precipitating agent. Bars with different superscript letters are significantly different ($p<0.05$). NAK, *Nakitembe*; MUS, *Musakala*; MPO, *Mpologoma*. Error bars are standard errors of means.

4.5.3 Comparison of pectin yield from ethanol and calcium chloride precipitation methods

The pectin yield from the pulp and peel of the selected *Matooke* cultivars were generally higher using the ethanol as compared to the calcium chloride precipitation methods (Figure 10). The yield of pectin in *Musakala* for both of the extraction methods were the highest among all the cultivars across the ripening stages.

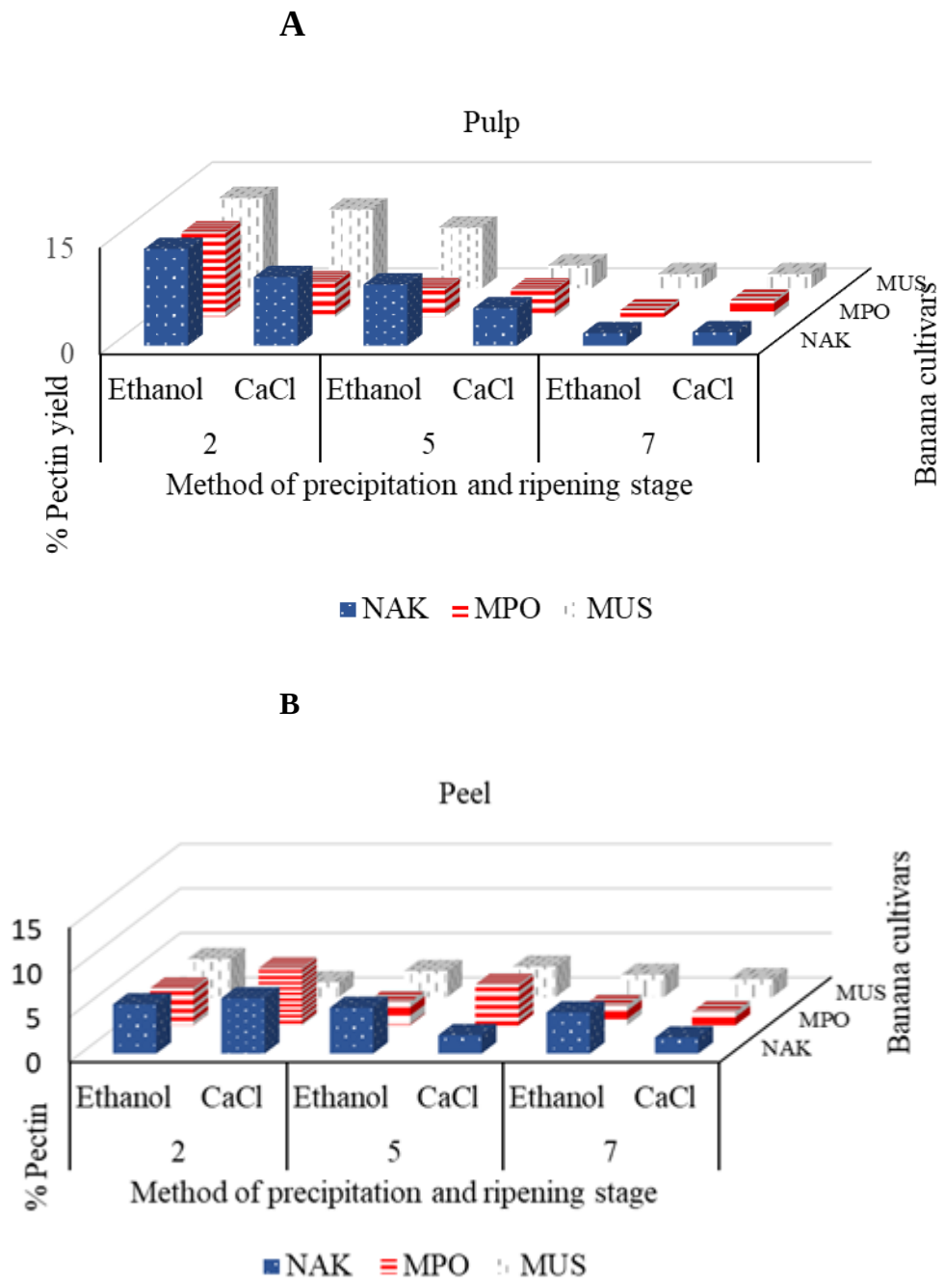

Figure 10. Comparison of pectin yield by ethanol and calcium chloride precipitation methods

These results may be due to the selectivity of only calcium pectate during precipitation using calcium chloride, which leads to low yield (Cardoso et al., 2003). This is not the case for the ethanol method, which may precipitate out other substances such as gums and phytochemicals from the bananas (Phaviphu et al., 2018). Given higher pectin yield, further

studies used banana pectin extracted using the ethanol precipitation method as described in section 3.6.1.

4.6 Characterization of the extracted banana pectin

Characterization results of pectin extracted from the pulp and peel of the selected Ugandan cooking banana cultivars are shown in Figures 11 to 15.

4.6.1 Equivalent weight (EQ. Wt.)

Generally, the EQ. Wt. of pectin extracted in this study was high with that extracted from banana pulp ranging from 1903 to 6421 and that from banana peel ranged from 1774 to 10144 g per mol (Figure 11). There was no significant difference in the EQ. Wt. of pectin from pulp and peel of *Musakala* cultivar with passage of time (>0.05).

At stage 2 of ripening, the EQ. Wt of the pectin from *Nakitembe* pulp was 1970.93, which was the lowest followed by *Musakala* with 2041.0 and *Mpologoma* the highest with 12657.5 g/mol. At stage 5, *Nakitembe* had 1903.78, *Musakala* 2051.94, and *Mpologom*a gave 6421.4 g/mol and at stage 7 *Musakala* presented the lowest EQ. Wt. of 1986.35, followed by *Nakitembe* which had 3657.5 and finally *Mpologoma* had 4500.0 g/mol. Generally, pectin from the banana pulps had a higher EQ. Wt. than that from their respective peels. *Mpologoma* had a significantly higher ($p<0.05$) EQ. Wt. in both pulp and peel across all the ripening stages. In the peel, *Mpologoma* at stage 2 gave the highest EQ. Wt value of 10144.0 g/mol followed by *Nakitembe* (4878.06) and *Musakala* (1783.0 g/mol). Stages 5 and 7 respectively gave 1780.01 and 2556.30 for *Nakitembe*, 2085.0 and 1774.50 for *Musakala* and lastly *Mpologoma* had 5804.29 and 6384.38 g/mol.

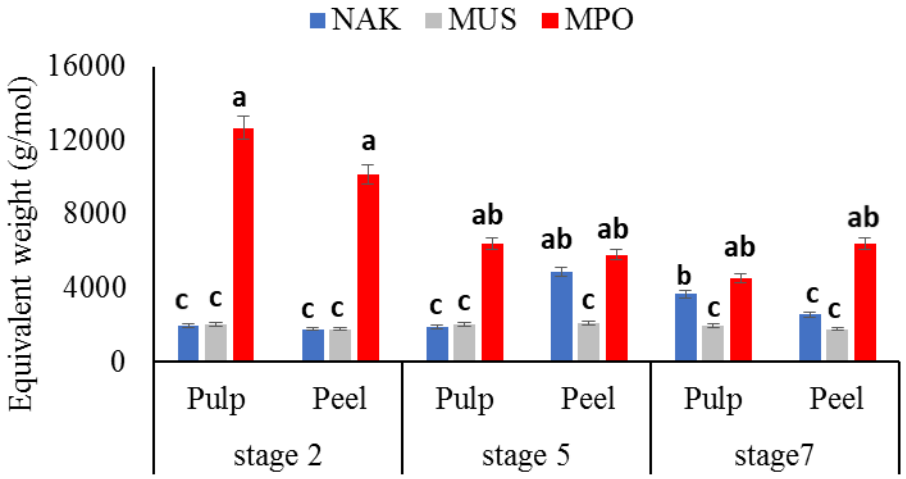

Figure 11. Equivalent weight of pectin from peel and pulp of the selected indigenous cooking bananas at stages 2, 5 and 7 of ripeness. Bars with different superscript letters are significantly different ($p<0.05$). NAK, *Nakitembe*; MUS, *Musakala*; MPO, *Mpologoma.* Error bars are standard errors of means.

Equivalent weight (EQ. Wt.) of pectin is an indicator of its jell-forming ability, with higher molecular weight pectin having better ability than lower molecular weight counterpart (Ragab et al., 2016). EQ. Wt. represents the quantity of pectin that is reactive which can undergo cross-linking reactions through polyol functional groups and is indicative of a high degree of esterification, which in turn leads to a higher gelling power (Oekenfull and Scott, 1984). Basing on the results from this study, pectin from *Mpologoma* could form better gels as compared to *Nakitembe* and *Musakala*'s.

4.6.2 Methoxy content (MeO)

There was significant difference in MeO content ($p<0.05$) of the pectin from peel and pulp across the ripening stages of the different banana cultivars (Figure 12). Methoxyl content of the banana peel and pulp pectin generally increased through the ripening stages (Figure 12). MeO ranged from 4.07 to 11.02 % and 3.67 to 8.21 % for the peel and pulp, respectively. At stage 2 of ripening, pectin from the pulp of *Nakitembe*, *Musakala* and *Mpologoma* had MeO of 5.37 %, 5.12 % and 4.07 % respectively. *Nakitembe* had the highest and *Mpologoma* had

the lowest. Further ripening to stage 5 produced pectin with MeO of 4.73 % for *Nakitembe*, 7.27 % for *Musakala* and *Mpologoma* at 5.73 %. Stage 7 of ripening had pectin with high methoxyl content for all the banana cultivars that is *Nakitembe* 11.02 %, *Musakala* 8.76 % and *Mpologoma* 7.88 %. The peel at stage 2 produced pectin of low methoxyl content (*Nakitembe* 4.02 %, *Musakala* 5.86 and *Mpologoma* 4.03 %). At stages 5 and 7, the respective MeO contents were *Nakitembe* 7.64 and 8.21 %, and *Musakala* 7.07 and 7.75 %. On the other hand, *Mpologoma* produced low methoxyl pectins throughout the ripening stages.

Within cultivars, the methoxyl content of the pectin from pulp generally increased with increase in ripeness for both pulp and peel. In addition, for all cultivars in both pulp and peel, stage 7 pectin had the highest methoxyl content except for *Mpologoma*.

The methoxyl contents of the extracted pectin vary from 0.2 to 12 % depending on the source and mode of extraction (Castillo et al., 2015). The results for methoxyl content in this study fell within the above range. The pectin extracted from peel and pulp at stage 7 for all the banana cultivars were high methoxyl pectins since their methoxyl content was higher than 7.0 % except for *Mpologoma*.

The above results could relate to the commercial pectins whose methoxyl content varies between 8 to 11 % (Castillo et al., 2015). Pectins with such MeO (8 to11 %) can form high sugar gels (>65 % sugar) whereas those with low methoxyl content (<7.0 %) can form gels with lower concentrations of sugar. Methoxyl content affects the dispersibility of pectin in water since pectin with high methoxyl content is readily dispersible in water than the low methoxyl content pectin (Rouse et al., 1962). On this note, therefore, the high methoxyl pectin extracted from banana pulp and peels in study could be suitable for industrial use particularly in jam and jelly production while the low methoxyl pectin such as that obtained

from *Mpologoma* peel at all stages of ripening could be used in yoghurt production where weaker gels are required.

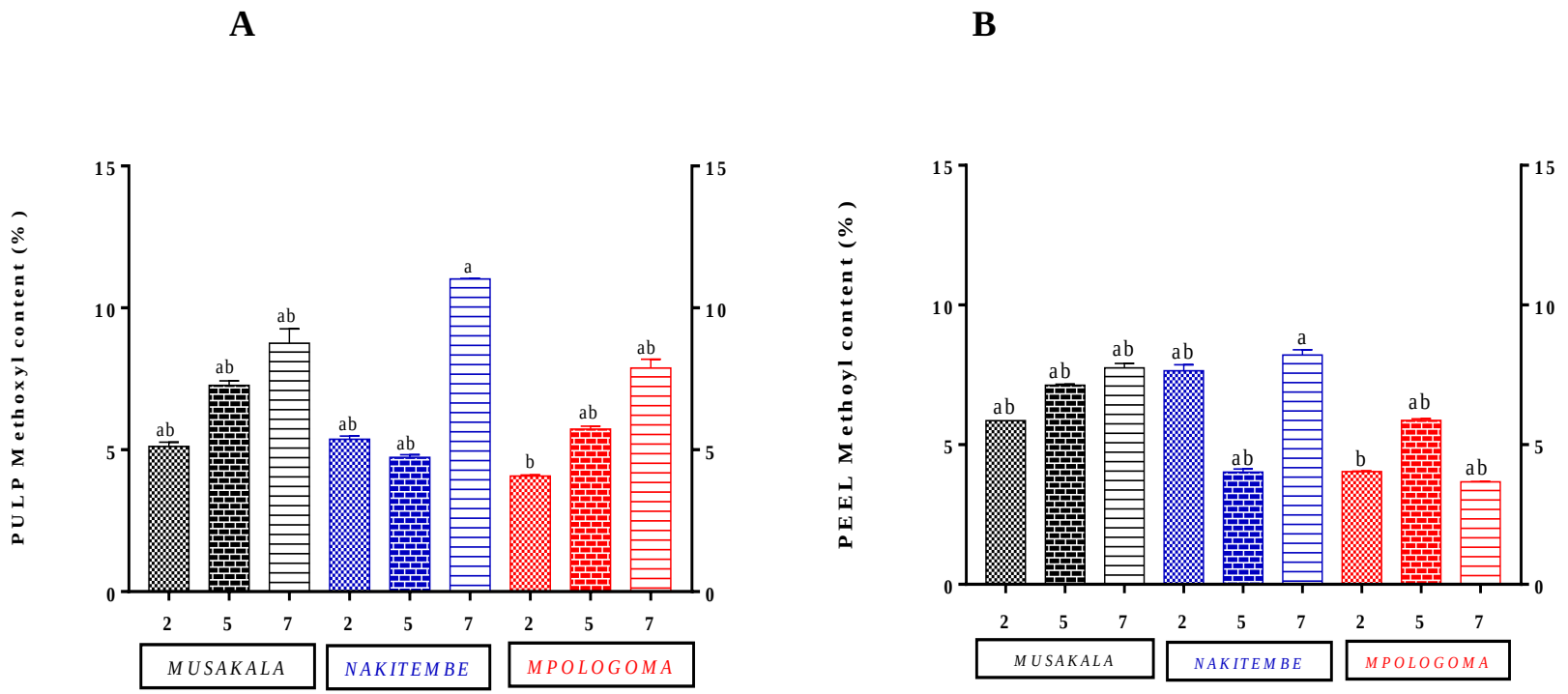

Figure 12. Methoxyl content of pectin extracted from the selected indigenous banana cultivars at different stages of ripening. The percentage MeO from the pulp (A) and peel (B). Different letters above each bar show significant difference at ($p < 0.05$).
Error bars are standard errors of means.

4.6.3 Anhydrouronic acid (AUA)

There was no significant difference in the AUA ($p>0.05$) of pectin extracted from the banana pulp as well as peel of the same cultivar across the ripening stages except for *Nakitembe* at ripening stage 7, which significantly differed ($p<0.05$) from that of *Mpologoma* at stage 2 as shown in the Figure 13.

AUA of pectin from the pulp of the selected banana cultivars increased with increase in ripening stage within the cultivars that is to say *Nakitembe* increased from 39.41 to 67.38 %, *Musakala* increased from 37.67 to 58.57 % and *Mpologoma* increased from 24.51 to 48.64 %. In the peels, AUA increased from 26.48 to 53.49 for *Nakitembe*, from 43.15 to 53.94 % for *Musakala* and finally with no increase in *Mpologoma* from 24.64 to 23.60 %. From the current results, comparing pulp and peel, showed that the pectin from pulp had higher AUA content than that from the peel.

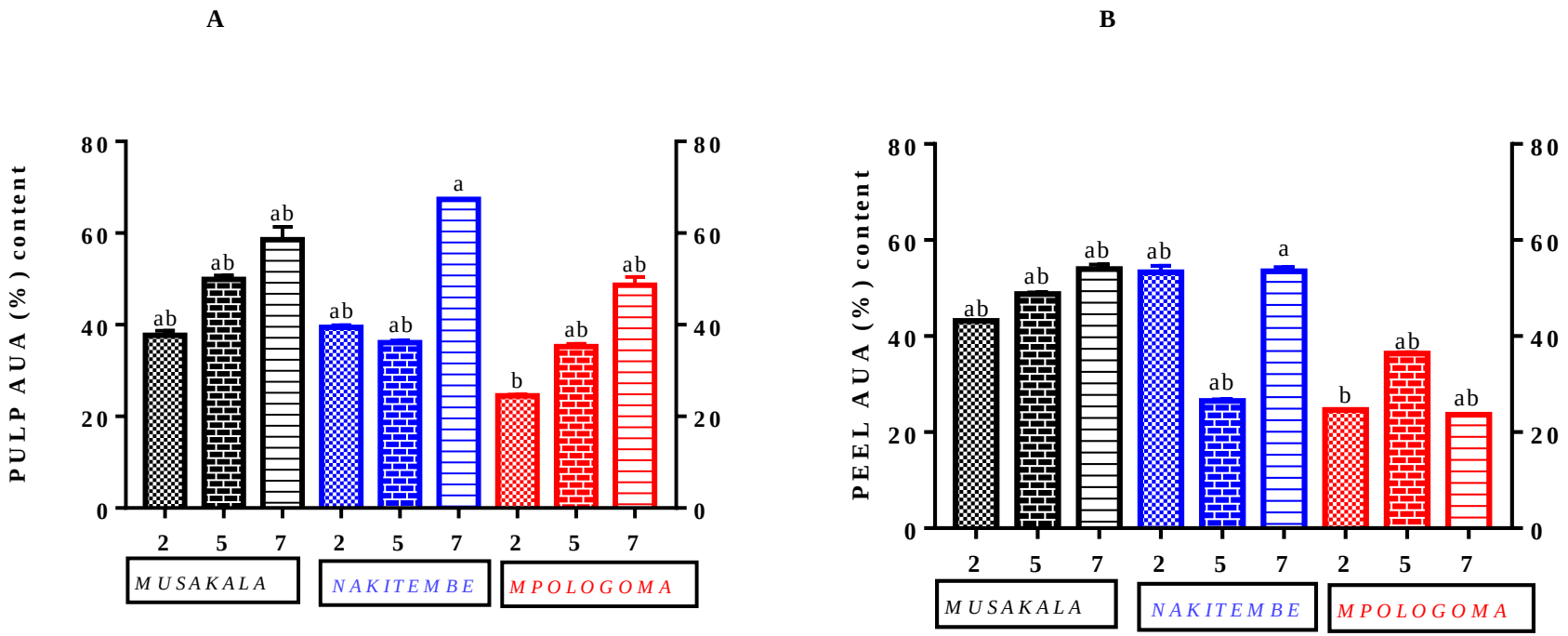

Figure 13. Anhydrouronic acid content of pectin extracted from the selected banana cultivars at different stages of ripening. The percentage AUA from the pulp (A) and peel (B). Different letters above each bar show significant difference at ($p < 0.05$).
Error bars are standard errors of means.

Pectin, is a partly esterified polygalacturonide which contains 10 % and more of organic materials composed of arabinose, galactose and other sugars (Assifaoui et al., 2015). AUA (%) is essential to determine the purity and degree of esterification plus the physical properties of pectin (Rangana, 1986). The recommended AUA (%) for extracted pectin for use in pharmaceuticals and as a food additive is not less than 65 % (May 1990; Food Chemical Codex, 1996), Pectin with AUA of less than 65 % may indicate impurities due to presence of proteins, starch and sugars in the precipitated pectin (Norazelina and Nazarrudin, 2012). This requirement limits the potential sources of commercial pectin for food and pharmaceutical purposes hence the emphasis on the studies using the hot acid extraction method for industrial extraction which is the most convenient for pectin extraction (Khamsucharit et al., 2016). Results in Figure 13 showed that generally pectin extracted at stage 7 of ripening had a higher AUA ranging between 48.6 to 67.4 % and 23.6 to 53.9 % from peel and pulp, respectively. Banana pectin extracted at stages 2 and 5 had AUA ranging between 24.5 to 49.9 % for pulp and between 24.6 to 53.3 % for peel. These results were relatively similar to those reported by Khamsucharit et al. (2016) for pectin obtained from peels of Kluai Hin (34.56 %) and Kluai Nam Wa (66.67 %) banana cultivars grown in Thailand and also could be related to those previously reported for Saba banana peel pectin (Castillo et al., 2015) which ranged from 39.68 to 57.32 %.

Therefore, based on the AUA content of pectin extracts in this study, only banana pulp pectin extracted from *Nakitembe* at stage 7 of ripeness had AUA content (67.4 %) higher than 65 % and met the criterion for commercial application hence banana pulp pectin from *Nakitembe* cultivar can be an alternative source of high methoxyl pectin.

4.6.4 Degree of esterification

There was no significant difference ($p>0.05$) in the degree of esterification (DE) of pectin in the pulp of *Musakala* and *Nakitembe* except for *Nakitembe* at stage 7. It was also observed that there was no significant difference in the DE of pectin from *Mpologoma* across all ripening stages (Figure 14). There was no significant difference ($p>0.05$) in the DE of pectin from the peel and pulp across all the stages of ripening for all the banana cultivars. These results suggested that all the selected banana cultivars contain good esterified pectin in pulp and peel across the stages of ripening and hence can be utilized for pectin production.

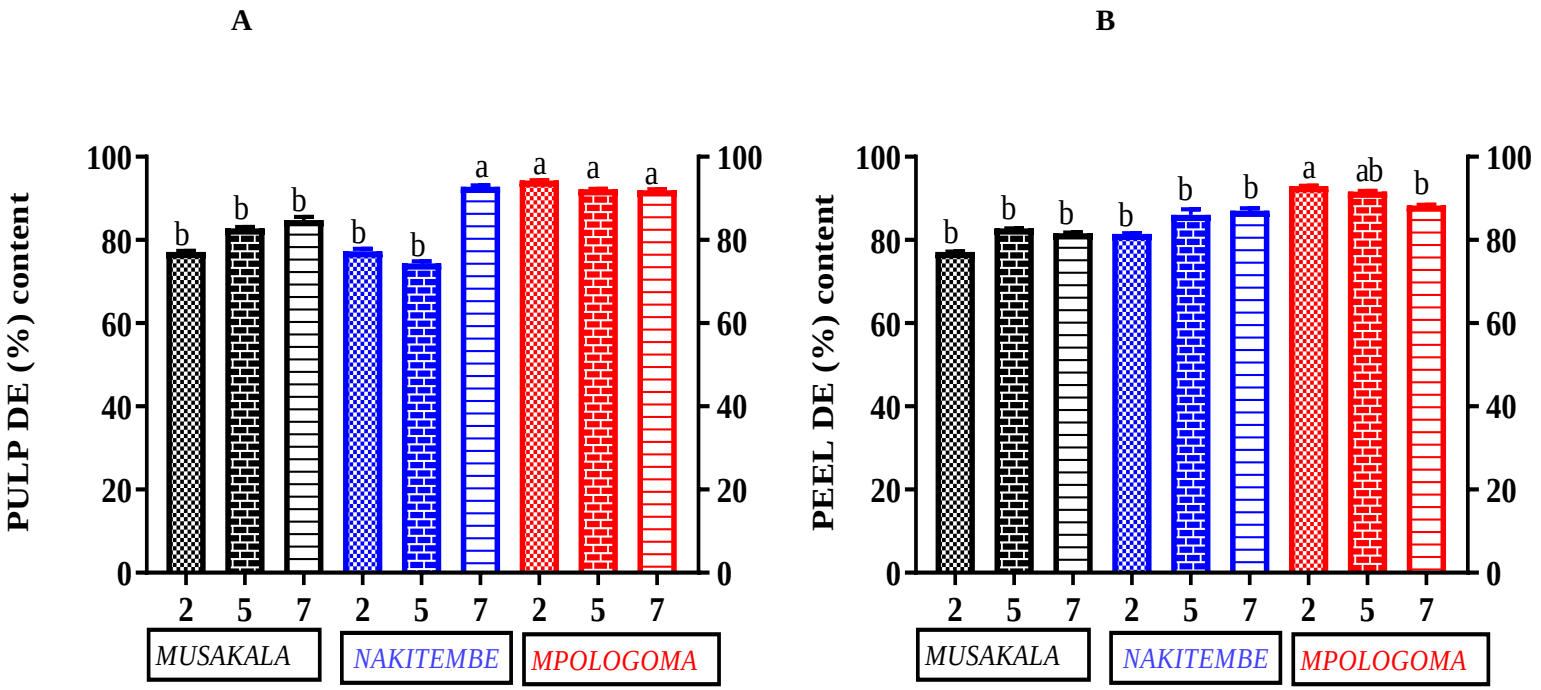

Figure 14. Degree of Esterification of pectin extracted from the selected banana cultivars at the different stages of ripening. The percentage DE from the pulp (A) and peel (B). Different letters above each bar show significant difference at ($p < 0.05$).
Error bars are standard errors of means.

Depending on the DE, pectin is divided into two groups that is high methoxyl pectin with DE higher than 50 % and low methoxyl pectin with DE less than 50 % (Khamsucharit et al., 2016). The extracted pectin can also be classified as rapid set if DE is greater than 72 % and slow set when DE is between 58 and 65 %, which describes the rate of gel formation (Shaha et al., 2013). The DE of pectin from the selected banana cultivars in the current study ranged between 77 to 94 % and 77 to 93 % for pulp and peel, respectively. These results were within the range of 60 to 90 % a level that is generally found in plant tissues (Shaha et al., 2013). The different banana varieties studied in Thailand had DE ranging between 63.15 and 72.03 % meaning that some produce rapid set pectins while others produce slow set pectins. The pectin extracted from all the banana cultivars in the current study are high methoxyl pectins and could therefore be described as rapid set pectins.

4.6.5 Ash content of the extracted pectin

There was a significant difference ($p<0.05$) between the ash content of pectin extracted from the peel and pulp across the ripening stages of the same banana cultivar except for *Nakitembe* peel whose pectin had ash content that was not significantly different ($p>0.05$) across the ripening stages (Figure 15). Comparison of the ash content at all the stages of ripening showed a significant difference at stages 2 and 7 ($p<0.05$) with *Mpologoma* having the highest and *Musakala* cultivar the lowest.

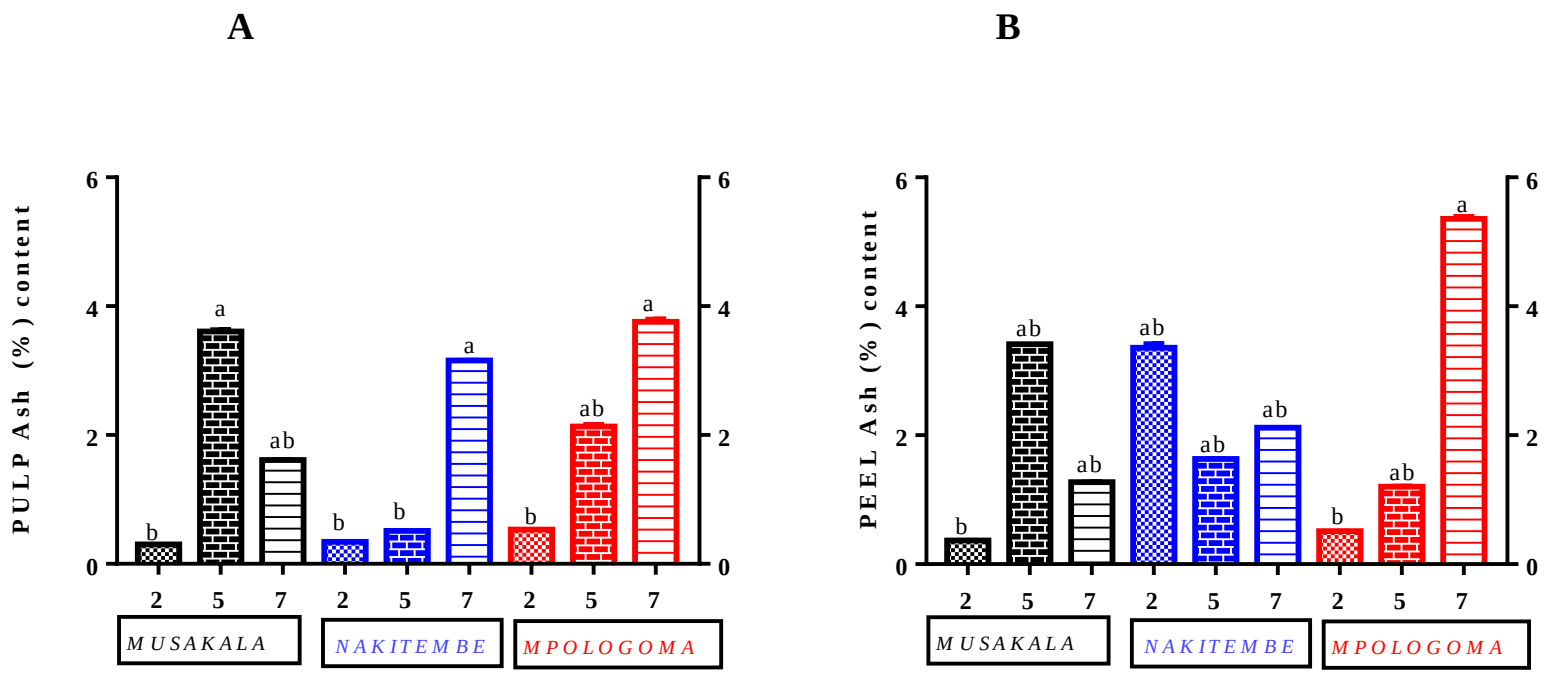

Figure 15. Ash content of pectin extracted pectin from the selected banana cultivars at different stages of ripening. The percentage ash from the pulp (A) and peel (B). Different letters above each bar show significant difference at ($p < 0.05$).
Error bars are standard errors of means.

The ash content of pectin from all the banana cultivars in peel and pulp was less than 10 %. This means that their purity is acceptable for commercial application (Khamsucharit et al., 2016; Shaha et al., 2013). To form gels of good quality, the maximum limit for ash content in the pectin used should not exceed 10 % (Norazelina et al., 2011) which is similar with the results obtained from this study. Pectin extracted at stage 2 of ripening in both peel and pulp contained the least amount of ash ranging between 0.3 to 0.52 % except for the peel of *Nakitembe* which contained 3.4 %. Overall, pectin from *Mpologoma* peel at stage 7 contained the highest amount of ash indicating that it could produce the least quality pectin among the banana cultivars.

Considering the overall quality characteristics such as high equivalent weight, which represents the quantity of pectin that is reactive and can undergo cross-linking reactions, high degree of esterification, which represents the gelling ability of pectin and basing on the purity represented by ash content, pectin extracted from *Mpologoma* pulp at stage 2 of ripeness was selected for product development studies as described in section 4.7 below.

4.7 Sensory evaluation of the banana pectin formulated Jam

Pectin was extracted from *Mpologoma* pulp at stage 2 of ripening (section 3.6.1) and used as a gelling agent at different concentrations in formulation of pineapple jam. Sensory evaluation was conducted to compare the jam produced with banana pectin with that made using commercial pectin. The scores for the different sensory attributes are presented in Table 5.

Colour of the jam without added pectin (0.0 % pectin) was significantly different ($p<0.05$) from that of the jam produced with banana pectin (BP) at the different levels of concentration (0.5 %, 1.0 %, and 2.0 % BP). However, there was no significant difference ($p>0.05$) between the colour of 0.0 % pectin jam with that of commercial pectin (CP) jam at all levels

of concentration (0.5 %, 1.0 % and 2.0 % CP). Colour of the jam with 0.5 %, and 1.0 % commercial pectin was significantly different ($p<0.05$) from the colour of the jam with 0.5 % and 1.0 % banana pectin. However, the colour of the jam with 2.0 % commercial pectin was not significantly different ($p>0.05$) from that of jam containing commercial and banana pectin.

The taste of the jam with no added pectin (0.0 % pectin) was significantly different ($p<0.05$) from all jam products with either commercial or banana pectin at all levels of concentration. The taste for 1.0 % CP was most preferred followed by 2.0 % CP, 0.5 % CP, 2.0 % BP, 1.0 % BP, 0.5 % BP and 0.0 % pectin. Comparison of the taste between banana and commercial pectin showed that there was no significant difference ($p>0.05$) meaning that both banana and commercial pectins caused similar effect to the taste of the jam.

The mouthfeel of all the jam products containing different levels of either banana or commercial pectins was not significantly different ($p>0.05$). The level of preference was high for products containing 2.0 % CP followed by 2.0 % BP, 0.5 % BP, 1.0 % CP, 0.5 % CP, 1.0 % BP and 0.0 % in that order.

The flowability was not significantly different between jam products containing banana and commercial pectins, none-the-less flowability for jam without added pectin was significantly different from that of BP and CP added jams at all levels of concentration ($p<0.05$).

Table 5. Sensory scores of pineapple jam formulated with different levels of banana and commercial pectin

Sensory attributes	Sensory scores (Mean ± SD)						
	0.0 %	0.5 %		1.0 %		2.0 %	
		BP	CP	BP	CP	BP	CP
Colour	$4.63^{a\dagger}\pm2.0^{*}$	$7.00^{b}\pm1.7^{*}$	$5.57^{a\dagger}\pm2.5^{*}$	$7.33^{b\dagger}\pm1.4^{*}$	$5.70^{a\dagger}\pm2.3^{*}$	$6.93^{b\dagger}\pm2.2^{*}$	$6.03^{a\dagger}\pm1.9^{*}$
Taste	$3.53^{x}\pm2.00$	$5.87^{y}\pm2.46$	$6.50^{y}\pm1.81$	$6.00^{y}\pm2.30$	$7.13^{y}\pm1.43$	$6.07^{y}\pm2.55$	$6.63^{y}\pm1.96$
Mouth feel	$3.77^{z}\pm2.30$	$5.97^{xy}\pm2.06$	$5.73^{xy}\pm2.30$	$5.53^{xy}\pm2.39$	$5.97^{xy}\pm2.13$	$6.10^{xy}\pm2.60$	$6.53^{xy}\pm2.00$
Flowability	$3.17^{a}\pm1.97$	$5.53^{b}\pm2.30$	$5.37^{b}\pm2.30$	$6.57^{b}\pm2.18$	$5.10^{b}\pm2.48$	$6.43^{b}\pm1.76$	$6.00^{b}\pm2.00$
Overall Acceptability	$3.78^{a}\pm0.62$	$6.22^{b}\pm0.63$	$5.79^{b}\pm0.49$	$6.36^{b}\pm0.79$	$5.98^{b}\pm0.84$	$6.38^{b}\pm0.40$	$6.30^{b}\pm0.33$

Values are means of sensory scores from 30 panelists ± standard deviations of the means

Mean values in the same row with different superscript letters are significantly different ($p<0.05$). Overall acceptability is the sum of score for colour, taste, mouthfeel and flowability divided by 4.

Sensory properties of jam have a great importance to measure consumer attitudes and their influence on food choice and acceptability (Ragab et al., 2016). From Table (5), it could be observed that the best colour of pineapple jam was obtained at 1.0 % addition of banana pectin, with the highest score (7.33), followed by banana jam of 0.5 % (7.00) and the jam with 2.0 % banana pectin (6.93). The lowest score for colour was obtained for sample prepared without either banana or commercial pectin (4.63) followed by that of 0.5 % (5.57), 1.0 % (5.70) and 2.0 % (6.03) respectively for jam that contained commercial pectin. It could be also noted that the color of tested jams increased as a result of increase in the levels of added pectin.

The score for taste of the jam samples was at its highest value (7.13) for jam with commercial pectin at 1.0 %. The taste of jam prepared with banana pectin was found to be highest (6.07) at 2.0 % concentration and lowest (5.87) at 0.5 % meaning that the formulation with 2.0% was most preferred in terms of taste.

The mouth feel score for both banana (6.10) and commercial pectin (6.63) at 2.0 % concentration indicated that they were most preferred implying that at this concentration the target for this parameter was achieved.

The highest score for flowability (6.57) was obtained for jam with banana pectin at 1.0 % concentration, followed by 2.0 % banana pectin jam (6.43). Overall acceptability of jam with banana pectin at 2.0 % concentration was the highest (6.38), followed by jam with banana pectin at 1.0 %, while the overall acceptability of jam prepared with banana pectin was found to be slightly higher than those containing commercial pectin. Generally, it could be concluded that banana pectin could be valuable and a good source for low-priced food components. Sensory evaluation showed that jam prepared using 2.0 % banana pectin characterized by its highest overall acceptability scores compared to either 0.5 % or 1.0%

addition of banana pectin. The properties imparted by 2.0 % of banana peels pectin such as, color, taste, mouth feel, flowability and overall acceptability were somewhat, not distinguishable as compared with that of commercial citrus pectin. It could be said that when applied to pineapple jam as gelling agent, at 2.0% concentration no significant differences between commercial citrus pectin in terms of sensory properties were detected therefore ripened (waste) banana can be a potential source of pectin for food application.

Based on the current results for the overall acceptability, the best formulation of jam with banana pectin in relation to that containing commercial pectin was that with pectin at 2 % concentration since it was the most preferred.

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

All physicochemical parameters analyzed during the 12 days ripening period varied according to banana cultivars and stage of ripening. Changes in these quality indices can be used to grade ripe bananas for value addition. Some parameters such as fruit firmness greatly decreased and this may affect acceptability of the bananas for their primary use by the consumers.

When ripened to stage 5, the indigenous Ugandan cooking banana cultivars examined in this study contained 19 volatile compounds which could be of aromatic value and some could contribute to the banana flavor. The peel and pulp of ripening *Matooke* contained considerable amounts of pectin which reduced across the ripening stages with *Mpologoma* presenting the best quality pectin with relatively high and stable degree of esterification across the ripening stages. This implies that pectin extracted at different ripening stages can be pooled to maximize the yield. It was also evident that the ethanol precipitation method gives higher yield of pectin in a shorter time as opposed to the calcium chloride method that yields relatively less pectin after a two days' extraction process. Overall, it was apparent from this study that pectin from *Matooke* could be utilized in a formulation of pineapple jam at 2 % concentration to give sensorially acceptable products that can be compared to those containing commercial pectin.

5.2 Recommendations

1. There is a need for further studies to identify and determine the exact flavor contributing volatile components in the *Matooke* volatile extracts.
2. For commercial pectin production, effluent from the extraction process that uses ethanol should be distilled so that ethanol is recovered for re-use.

3. Efforts through investment, should be developed that can collect all ripe rejected *Matooke* for reuse as sources of raw materials for pectin extraction a move that will empower the farmers and traders.

4. Other banana cultivars need to be studied to assess and establish their pectin content, the extracted pectin should be dried in a vacuum oven as opposed to an air oven because of high risk of oxidation and polymerization, which could lead to colour changes and interference with the pectin quality characteristics.

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Appendix 1. SENSORY EVALUATION FORM

Panelist No……...Date……...Time……

You are provided with several jam samples to carry out sensory evaluation with reference to colour, taste, mouth feel, flowability and overall acceptability. Please take sufficient time after testing one sample before the next

Rank the samples using a 9 - point hedonic scale shown below

1=Dislike Extremely
2=Dislike very much
3=Dislike Moderately
4=Dislike Slightly
5=Neither Like nor Dislike
6=Like Slightly
7=Like Moderately
8=Like Very Much
9=Like Extremely

Identify the colour of each sample and give your visual impression from light to dark.

Record your deduction by writing the values of your score in the sample score table below;

Sample code	Hedonic Parameters				
	Colour	Taste	Mouth feel	Flowability	Overall acceptability
C1010					
C051C					
B051C					
C202C					
B202B					
C101C					
B101B					

General comments…….
…….……

Which product did you like most and why?
……
……

Thanks for your time.

Appendix 2. Statistical analysis for Jam evaluation

```
ONEWAY Colour Taste Feel Flowability BY Formulation
  /STATISTICS DESCRIPTIVES HOMOGENEITY
  /PLOT MEANS
  /MISSING ANALYSIS
  /POSTHOC=TUKEY ALPHA(0.05).
```

Test of Homogeneity of Variances

	Levene Statistic	df1	df2	Sig.
Colour	2.391	6	203	.030
Taste	2.226	6	203	.042
feel	.972	6	203	.446
Flowability	1.119	6	203	.352

ANOVA

		Sum of Squares	Df	Mean Square	F	Sig.
Colour	Between Groups	167.695	6	27.949	6.884	.000
	Within Groups	824.133	203	4.060		
	Total	991.829	209			
Taste	Between Groups	240.962	6	40.160	9.051	.000
	Within Groups	900.733	203	4.437		
	Total	1141.695	209			
feel	Between Groups	142.514	6	23.752	4.733	.000
	Within Groups	1018.800	203	5.019		
	Total	1161.314	209			
Flowability	Between Groups	235.990	6	39.332	8.387	.000
	Within Groups	952.033	203	4.690		
	Total	1188.024	209			

Taste

Tukey HSD

Formulation	N	Subset for alpha = 0.05		N	1	2
		1	2			
0.0%	30	3.53				
0.5% Banana	30		5.87			
1.0% Banana	30		6.00			
2.0% Banana	30		6.07			
0.5 % Commercial	30		6.50			
2.0% Commercial	30		6.63			
1.0% Commercial	30		7.13			
Sig.		1.000	.235			

Means for groups in homogeneous subsets are displayed.

Feel

Tukey HSD

Formulation	N	Subset for alpha = 0.05		N	1	2
		1	2			
0.0%	30	3.77				
1.0% Banana	30		5.53			
0.5 % Commercial	30		5.73			
1.0% Commercial	30		5.97			
0.5% Banana	30		5.97			
2.0% Banana	30		6.10			
2.0% Commercial	30		6.53			
Sig.		1.000	.598			

Means for groups in homogeneous subsets are displayed.

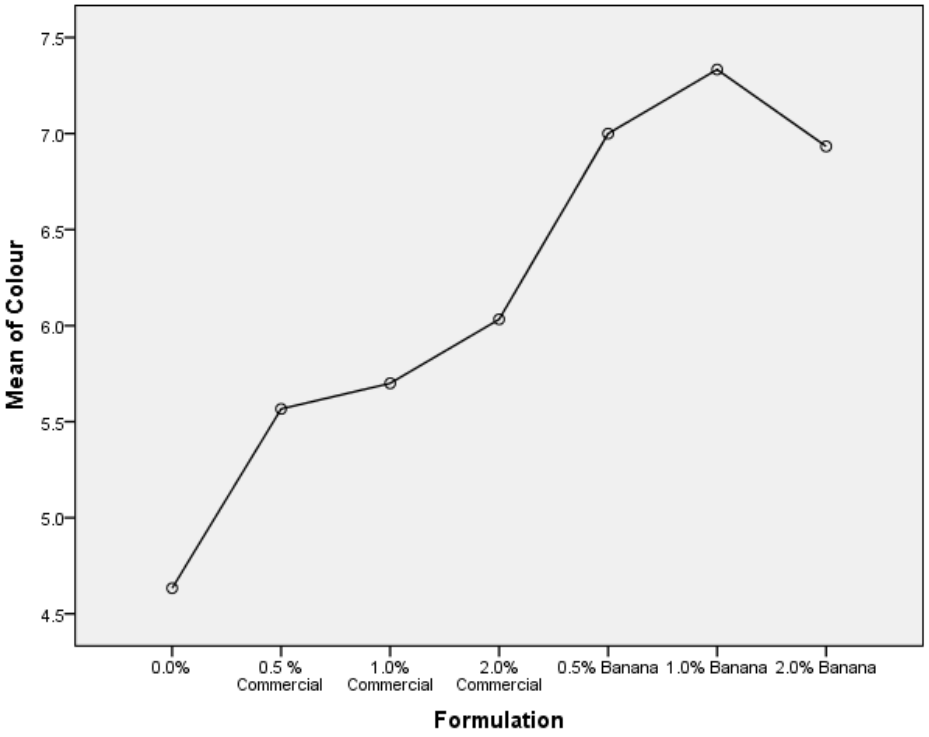
Flowability

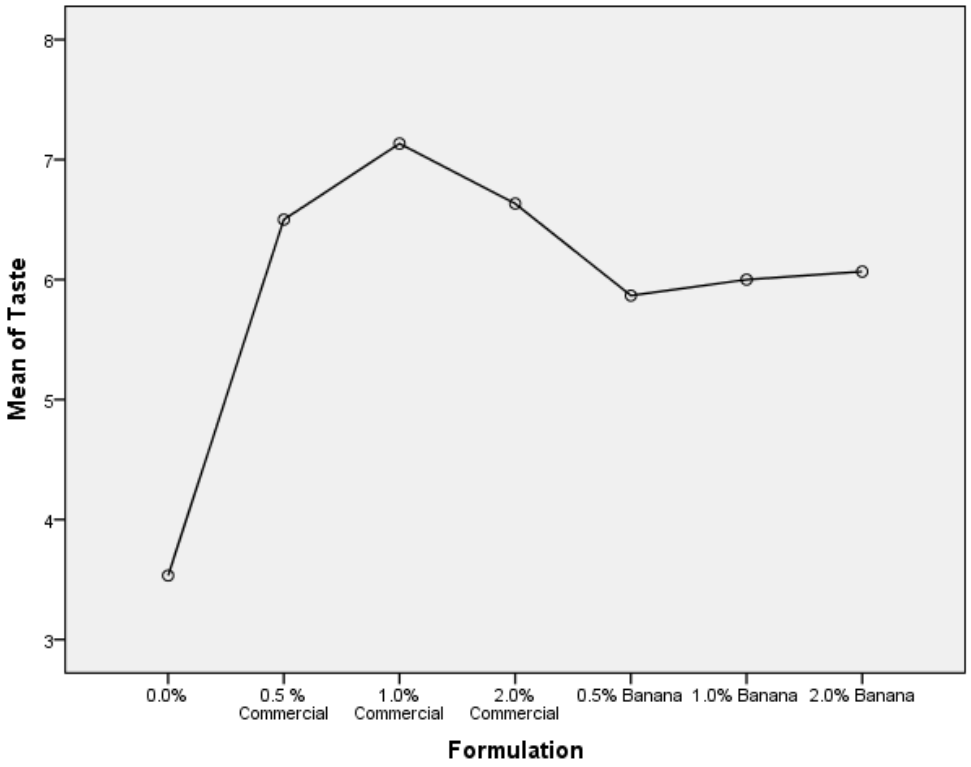
Tukey HSD

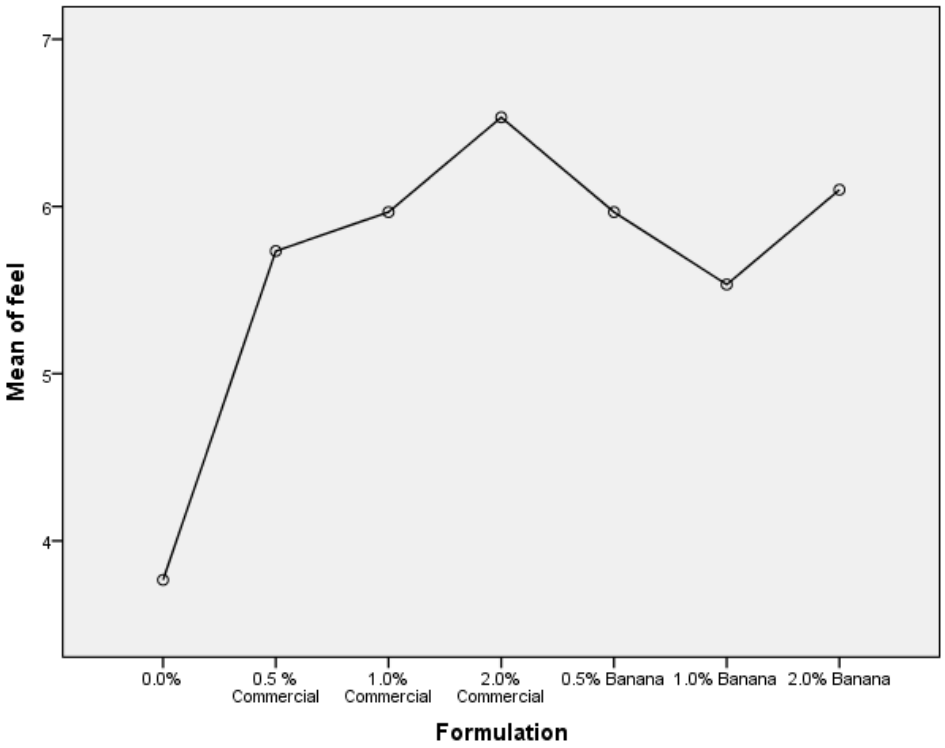
Formulation	N	Subset for alpha = 0.05		N	1	2
		1	2			
0.0%	30	3.17				
1.0% Commercial	30		5.10			
0.5 % Commercial	30		5.37			
0.5% Banana	30		5.53			
2.0% Commercial	30		6.00			
2.0% Banana	30		6.43			
1.0% Banana	30		6.57			
Sig.		1.000	.125			

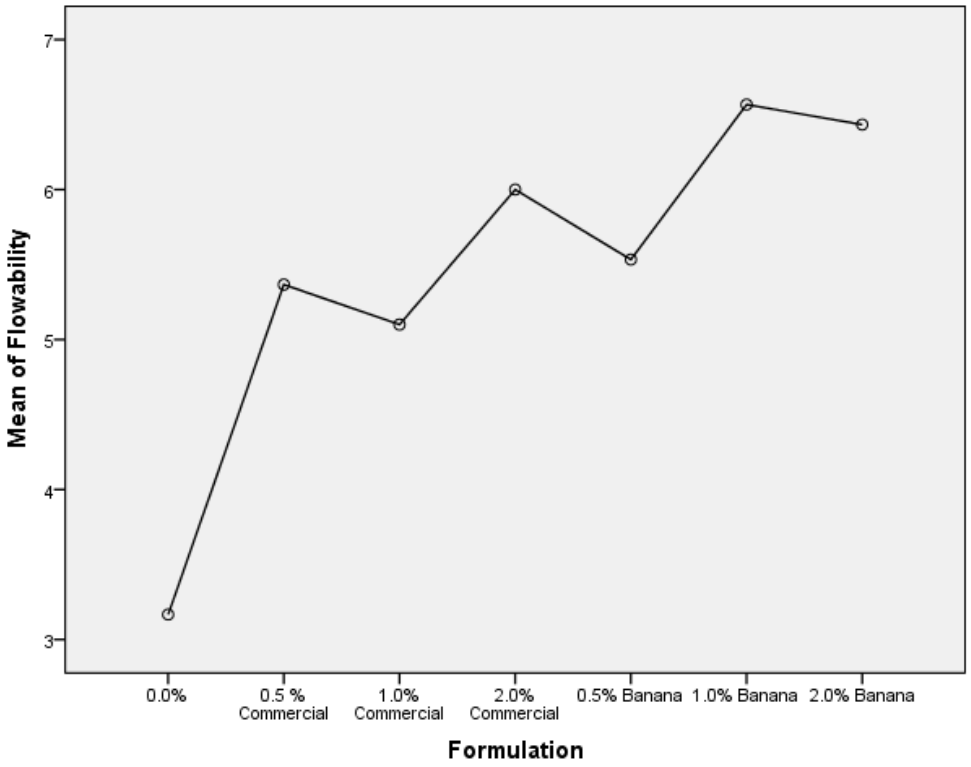
Means for groups in homogeneous subsets are displayed.

Means Plots




Mean of Taste
8
7
6
5
4
3
0.0%
0.5 % Commercial
1.0% Commercial
2.0% Commercial
0.5% Banana
1.0% Banana
2.0% Banana
Formulation


Mean of feel
7
6
5
4
0.0%
0.5 % Commercial
1.0% Commercial
2.0% Commercial
0.5% Banana
1.0% Banana
2.0% Banana
Formulation

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  /CRITERIA=ALPHA(.05)
  /DESIGN= Formulation.
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General Linear Model

Notes

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	Cases Used	Statistics are based on all cases with valid data for all variables in the model.
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[DataSet0] C:\Users\Bosco\Desktop\Richard sensory.sav

Between-Subjects Factors

		Value Label	N
Formulation	1	0.0%	30
	2	0.5 % Commercial	30
	3	1.0% Commercial	30
	4	2.0% Commercial	30
	5	0.5% Banana	30
	6	1.0% Banana	30
	7	2.0% Banana	30

Bartlett's Test of Sphericity[a]

Likelihood Ratio	.000
Approx. Chi-Square	152.441
Df	9
Sig.	.000

Tests the null hypothesis that the residual covariance matrix is proportional to an identity matrix.

a. Design: Intercept + Formulation

Multivariate Tests[c]

Effect		Value	F	Hypothesis df	Error df	Sig.
Intercept	Pillai's Trace	.940	782.897[a]	4.000	200.000	.000
	Wilks' Lambda	.060	782.897[a]	4.000	200.000	.000
	Hotelling's Trace	15.658	782.897[a]	4.000	200.000	.000
	Roy's Largest Root	15.658	782.897[a]	4.000	200.000	.000
Formulation	Pillai's Trace	.467	4.476	24.000	812.000	.000
	Wilks' Lambda	.592	4.718	24.000	698.927	.000
	Hotelling's Trace	.592	4.893	24.000	794.000	.000
	Roy's Largest Root	.357	12.072[b]	6.000	203.000	.000

a. Exact statistic

b. The statistic is an upper bound on F that yields a lower bound on the significance level.

c. Design: Intercept + Formulation

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	Df	Mean Square
Corrected Model	Colour	167.695[a]	6	27.949
	Taste	240.962[b]	6	40.160
	feel	142.514[c]	6	23.752
	Flowability	235.990[d]	6	39.332
Intercept	Colour	7998.171	1	7998.171
	Taste	7464.305	1	7464.305
	feel	6720.686	1	6720.686
	Flowability	6242.976	1	6242.976
Formulation	Colour	167.695	6	27.949
	Taste	240.962	6	40.160
	feel	142.514	6	23.752
	Flowability	235.990	6	39.332
Error	Colour	824.133	203	4.060
	Taste	900.733	203	4.437
	feel	1018.800	203	5.019
	Flowability	952.033	203	4.690
Total	Colour	8990.000	210	
	Taste	8606.000	210	
	feel	7882.000	210	
	Flowability	7431.000	210	

Corrected Total	Colour	991.829	209	
	Taste	1141.695	209	
	feel	1161.314	209	
	Flowability	1188.024	209	

Tests of Between-Subjects Effects

Source	Dependent Variable	F	Sig.
Corrected Model	Colour	6.884	.000
	Taste	9.051	.000
	feel	4.733	.000
	Flowability	8.387	.000
Intercept	Colour	1970.105	.000
	Taste	1682.245	.000
	feel	1339.124	.000
	Flowability	1331.176	.000
Formulation	Colour	6.884	.000
	Taste	9.051	.000
	feel	4.733	.000
	Flowability	8.387	.000
Error	Colour		
	Taste		
	feel		
	Flowability		
Total	Colour		
	Taste		
	feel		
	Flowability		
Corrected Total	Colour		
	Taste		
	feel		
	Flowability		

a. R Squared = .169 (Adjusted R Squared = .145)

b. R Squared = .211 (Adjusted R Squared = .188)

c. R Squared = .123 (Adjusted R Squared = .097)

d. R Squared = .199 (Adjusted R Squared = .175)

Transformation Coefficients (M Matrix)

Dependent Variable	Transformed Variable			
	Colour	Taste	feel	Flowability
Colour	1	0	0	0
Taste	0	1	0	0
feel	0	0	1	0
Flowability	0	0	0	1

Between-Subjects SSCP Matrix

			Colour	Taste	feel	Flowability
Hypothesis	Intercept	Colour	7998.171	7726.629	7331.657	7066.286
		Taste	7726.629	7464.305	7082.743	6826.381
		feel	7331.657	7082.743	6720.686	6477.429
		Flowability	7066.286	6826.381	6477.429	6242.976
	Formulation	Colour	167.695	84.305	91.343	173.014
		Taste	84.305	240.962	167.857	167.919
		feel	91.343	167.857	142.514	150.205
		Flowability	173.014	167.919	150.205	235.990
Error		Colour	824.133	224.067	280.000	345.700
		Taste	224.067	900.733	536.400	245.700
		feel	280.000	536.400	1018.800	372.367
		Flowability	345.700	245.700	372.367	952.033

Based on Type III Sum of Squares

Residual SSCP Matrix

		Colour	Taste	feel	Flowability
Sum-of-Squares and Cross-Products	Colour	824.133	224.067	280.000	345.700
	Taste	224.067	900.733	536.400	245.700
	feel	280.000	536.400	1018.800	372.367
	Flowability	345.700	245.700	372.367	952.033
Covariance	Colour	4.060	1.104	1.379	1.703
	Taste	1.104	4.437	2.642	1.210
	feel	1.379	2.642	5.019	1.834
	Flowability	1.703	1.210	1.834	4.690
Correlation	Colour	1.000	.260	.306	.390
	Taste	.260	1.000	.560	.265
	feel	.306	.560	1.000	.378

	Flowability	.390	.265	.378	1.000

Based on Type III Sum of Squares

```
ONEWAY Colour Taste Feel Flowability BY Formulation
  /POLYNOMIAL=1
  /STATISTICS DESCRIPTIVES HOMOGENEITY
  /PLOT MEANS
  /MISSING ANALYSIS
  /POSTHOC=TUKEY ALPHA(0.05).
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Oneway

Notes

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	Cases Used	Statistics for each analysis are based on cases with no missing data for any variable in the analysis.
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[DataSet0] C:\Users\Bosco\Desktop\Richard sensory.sav

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean
						Lower Bound
Colour	0.0%	30	4.63	2.008	.367	3.88
	0.5 % Commercial	30	5.57	2.459	.449	4.65
	1.0% Commercial	30	5.70	2.261	.413	4.86
	2.0% Commercial	30	6.03	1.866	.341	5.34
	0.5% Banana	30	7.00	1.702	.311	6.36
	1.0% Banana	30	7.33	1.373	.251	6.82
	2.0% Banana	30	6.93	2.227	.407	6.10
	Total	210	6.17	2.178	.150	5.88
Taste	0.0%	30	3.53	2.013	.367	2.78
	0.5 % Commercial	30	6.50	1.815	.331	5.82
	1.0% Commercial	30	7.13	1.432	.261	6.60
	2.0% Commercial	30	6.63	1.956	.357	5.90
	0.5% Banana	30	5.87	2.460	.449	4.95
	1.0% Banana	30	6.00	2.304	.421	5.14
	2.0% Banana	30	6.07	2.545	.465	5.12
	Total	210	5.96	2.337	.161	5.64
feel	0.0%	30	3.77	2.254	.412	2.92
	0.5 % Commercial	30	5.73	2.196	.401	4.91
	1.0% Commercial	30	5.97	2.125	.388	5.17
	2.0% Commercial	30	6.53	1.995	.364	5.79
	0.5% Banana	30	5.97	2.059	.376	5.20
	1.0% Banana	30	5.53	2.389	.436	4.64
	2.0% Banana	30	6.10	2.604	.475	5.13
	Total	210	5.66	2.357	.163	5.34
Flowability	0.0%	30	3.17	1.967	.359	2.43
	0.5 % Commercial	30	5.37	2.297	.419	4.51
	1.0% Commercial	30	5.10	2.482	.453	4.17
	2.0% Commercial	30	6.00	2.000	.365	5.25
	0.5% Banana	30	5.53	2.389	.436	4.64
	1.0% Banana	30	6.57	2.176	.397	5.75
	2.0% Banana	30	6.43	1.755	.321	5.78
	Total	210	5.45	2.384	.165	5.13

Test of Homogeneity of Variances

	Levene Statistic	df1	df2	Sig.
Colour	2.391	6	203	.030
Taste	2.226	6	203	.042
feel	.972	6	203	.446
Flowability	1.119	6	203	.352

ANOVA

				Sum of Squares	df
Colour	Between Groups	(Combined)		167.695	6
		Linear Term	Contrast	147.505	1
			Deviation	20.190	5
	Within Groups			824.133	203
	Total			991.829	209
Taste	Between Groups	(Combined)		240.962	6
		Linear Term	Contrast	30.476	1
			Deviation	210.486	5
	Within Groups			900.733	203
	Total			1141.695	209
feel	Between Groups	(Combined)		142.514	6
		Linear Term	Contrast	46.671	1
			Deviation	95.843	5
	Within Groups			1018.800	203
	Total			1161.314	209
Flowability	Between Groups	(Combined)		235.990	6
		Linear Term	Contrast	171.001	1
			Deviation	64.989	5
	Within Groups			952.033	203
	Total			1188.024	209

ANOVA

				Mean Square	F	Sig.
Colour	Between Groups	(Combined)		27.949	6.884	.000
		Linear Term	Contrast	147.505	36.333	.000
			Deviation	4.038	.995	.422
	Within Groups			4.060		
Taste	Between Groups	(Combined)		40.160	9.051	.000

		Linear Term	Contrast	30.476	6.868	.009
			Deviation	42.097	9.488	.000
	Within Groups			4.437		
feel	Between Groups	(Combined)		23.752	4.733	.000
		Linear Term	Contrast	46.671	9.299	.003
			Deviation	19.169	3.819	.003
	Within Groups			5.019		
Flowability	Between Groups	(Combined)		39.332	8.387	.000
		Linear Term	Contrast	171.001	36.462	.000
			Deviation	12.998	2.772	.019
	Within Groups			4.690		

Homogeneous Subsets

Colour

Tukey HSD

Formulation	N	Subset for alpha = 0.05						
		1	2	3	N	1	2	3
0.0%	30	4.63						
0.5 % Commercial	30	5.57	5.57					
1.0% Commercial	30	5.70	5.70					
2.0% Commercial	30	6.03	6.03	6.03				
2.0% Banana	30		6.93	6.93				
0.5% Banana	30		7.00	7.00				
1.0% Banana	30			7.33				
Sig.		.106	.090	.165				

Means for groups in homogeneous subsets are displayed.

Taste

Tukey HSD

Formulation	N	Subset for alpha = 0.05		N	1	2
		1	2			
0.0%	30	3.53				
0.5% Banana	30		5.87			
1.0% Banana	30		6.00			
2.0% Banana	30		6.07			
0.5 % Commercial	30		6.50			
2.0% Commercial	30		6.63			
1.0% Commercial	30		7.13			
Sig.		1.000	.235			

Means for groups in homogeneous subsets are displayed.

Feel

Tukey HSD

Formulation	N	Subset for alpha = 0.05		N	1	2
		1	2			
0.0%	30	3.77				
1.0% Banana	30		5.53			
0.5 % Commercial	30		5.73			
1.0% Commercial	30		5.97			
0.5% Banana	30		5.97			
2.0% Banana	30		6.10			
2.0% Commercial	30		6.53			
Sig.		1.000	.598			

Means for groups in homogeneous subsets are displayed.

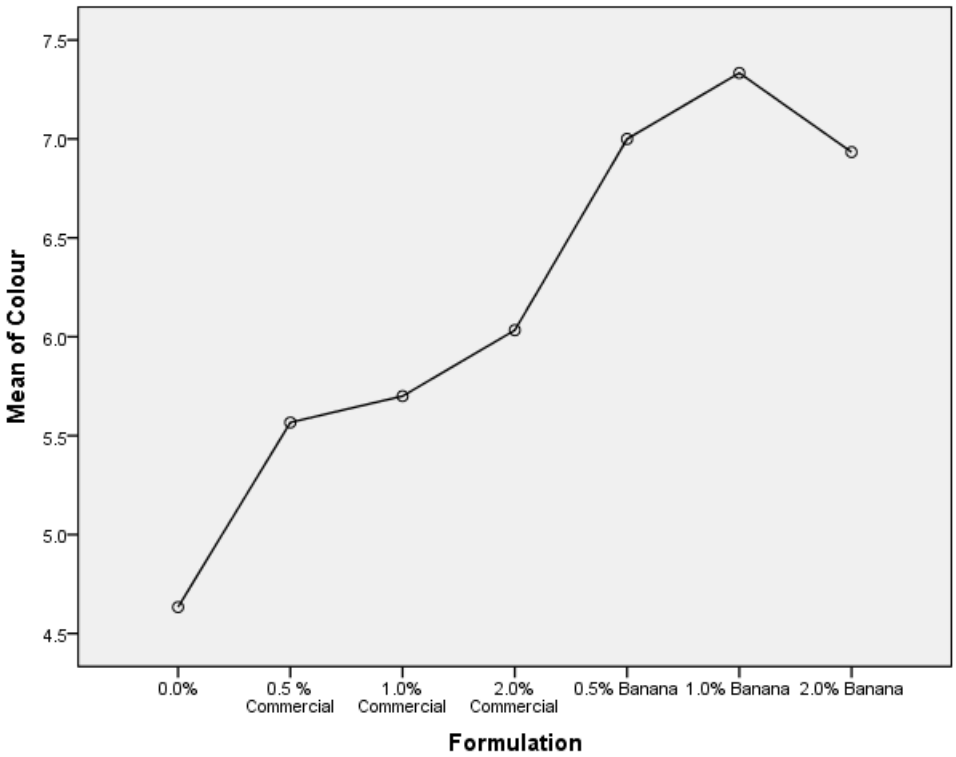
Flowability

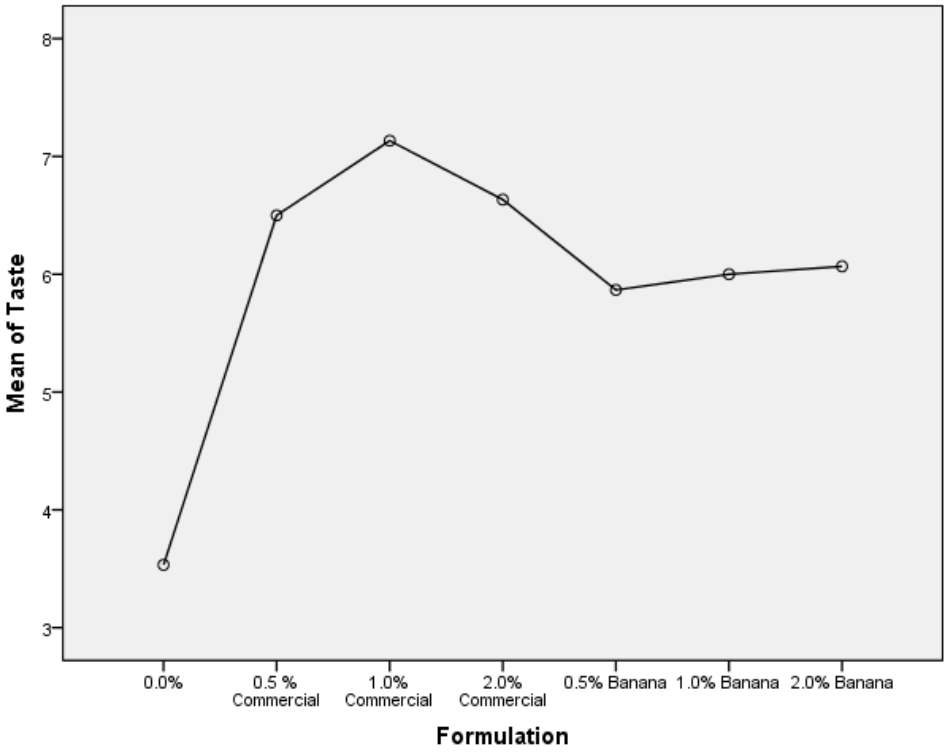
Tukey HSD

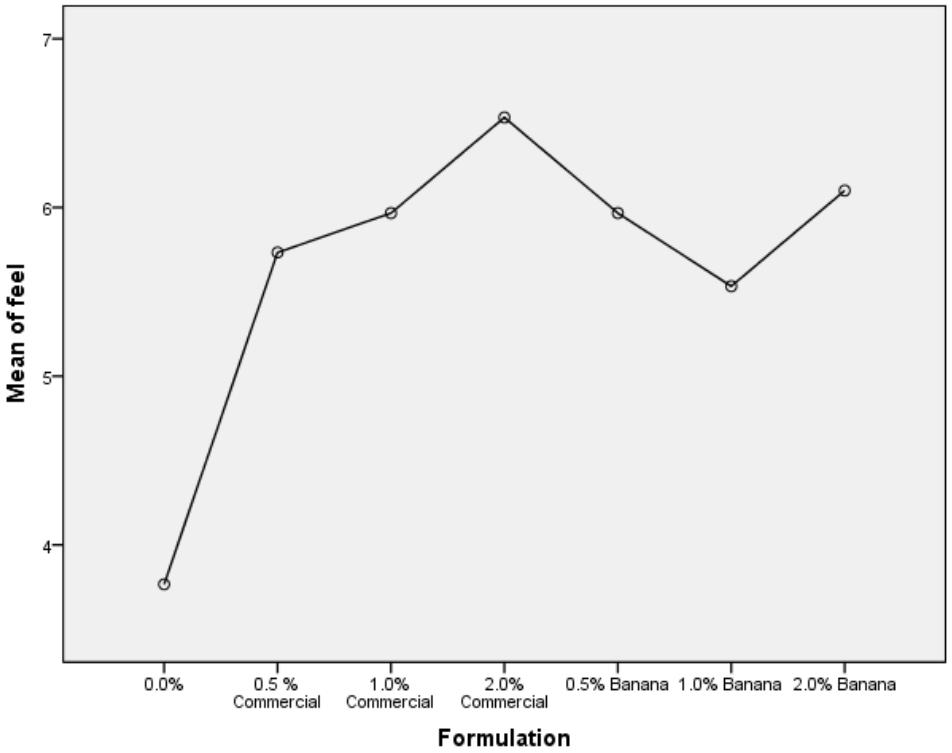
Formulation	N	Subset for alpha = 0.05				
		1	2	N	1	2
0.0%	30	3.17				
1.0% Commercial	30		5.10			
0.5 % Commercial	30		5.37			
0.5% Banana	30		5.53			
2.0% Commercial	30		6.00			
2.0% Banana	30		6.43			
1.0% Banana	30		6.57			
Sig.		1.000	.125			

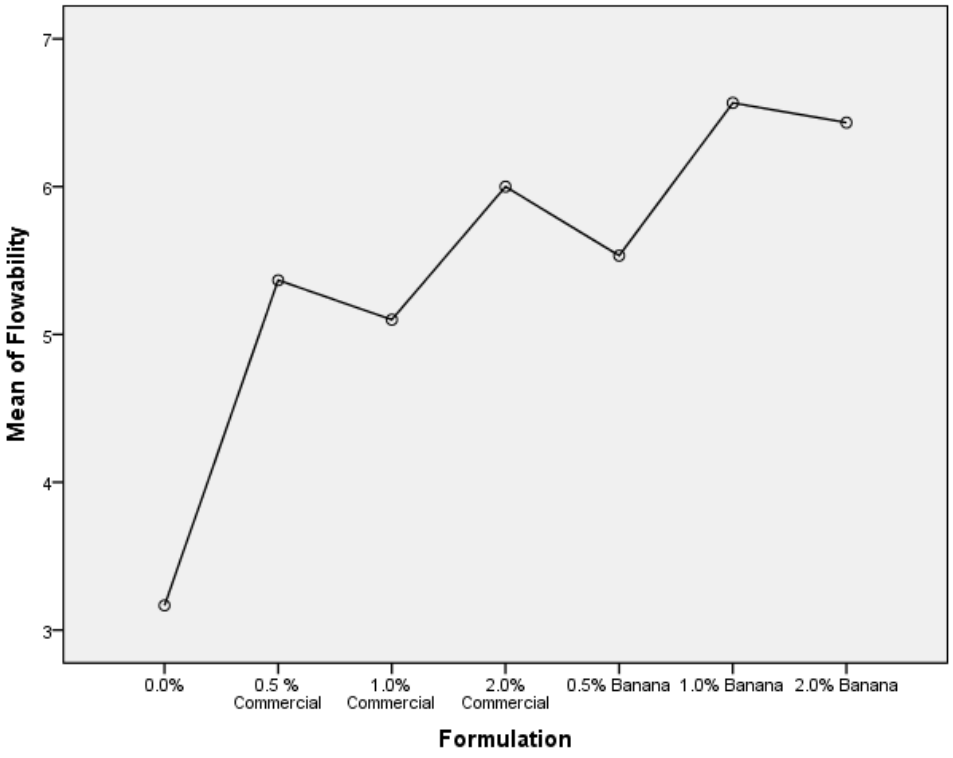
Means for groups in homogeneous subsets are displayed.

Means Plots




Mean of Taste
8
7
6
5
4
3
0.0%
0.5 % Commercial
1.0% Commercial
2.0% Commercial
0.5% Banana
1.0% Banana
2.0% Banana
Formulation


Mean of feel
7
6
5
4
0.0%
0.5 % Commercial
1.0% Commercial
2.0% Commercial
0.5% Banana
1.0% Banana
2.0% Banana
Formulation


Mean of Flowability
7
6
5
4
3
0.0%
0.5 % Commercial
1.0% Commercial
2.0% Commercial
0.5% Banana
1.0% Banana
2.0% Banana
Formulation