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**Development of nanoparticle-enriched edible coatings to improve fruit quality  
and extend the shelf life of Cavendish bananas**

By

**ODETAYO TEMITAYO**

DISSERTATION

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**MASTER OF SCIENCE**

in

**BOTANY**

at the  
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NOVEMBER 2021

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## SUPERVISOR'S APPROVAL

The research reported in this dissertation/thesis was completed by the candidate while based in the Botany and Plant Biotechnology Department, Faculty of Science, University of Johannesburg, Auckland Park campus, South Africa. This work is based on the research supported by the National Research Foundation (grant number: 121968). I have approved that this dissertation be submitted for evaluation.

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## ABSTRACT

Bananas have a very short shelf life due to their climacteric nature, which means they continue to ripen even after harvest. As a result, a novel postharvest management strategy is required to extend the shelf life of the fruit while decreasing wastage and losses. The application of edible coatings to extend the shelf life of bananas and maintain postharvest quality is being considered as a potential option. To our knowledge, little research has been published on the application of *Aloe vera* and moringa coatings enriched with chitosan nanoparticles to enhance the shelf life of banana fruit at room storage. This study was conducted to investigate the effect of adding chitosan nanoparticles to the edible coatings to maintain the post-harvest quality and prolong the shelf life of bananas considering the preservative and antioxidant properties of *Aloe vera* and moringa leaf extracts. *Aloe vera* and moringa plant extracts were applied as a coating on the surface of the bananas to test the effect of the treatment on firmness, weight loss, total soluble solids (TSS), mineral content, protein content, total phenolics, and shelf life over a 24-day storage period at  $24 \pm 1^\circ\text{C}$ . When *Aloe vera* and moringa plant extracts were tested, the results showed that *Aloe vera* coating had significant ( $p < 0.05$ ) effect, reduced TSS value, retained fruit firmness, and had a greater overall phenolic content compared to the control and the moringa coatings during storage. However, moringa coatings significantly ( $p < 0.05$ ) retained the mineral and protein content of banana fruits. Both coatings extended the shelf life by up to 4 days compared to the control. Furthermore, chitosan nanoparticles (CN) mixed with *Aloe vera* and moringa basal coatings were used as postharvest coating treatments to improve their barrier properties and extend the shelf life of Cavendish bananas. When compared to basal *Aloe vera* (AV) and *Moringa oleifera* (MO) coatings, the coatings enhanced with chitosan preserved the quality of bananas and extended shelf life by 3 and 4 days, respectively. *Moringa oleifera* plus chitosan nanoparticle (MO+CN) and *Aloe vera* plus chitosan nanoparticle (AV+CN) were then tested on ethylene treated and non-ethylene treated Cavendish bananas. MO+CN had a better performance and was also consistent in slowing ripening processes. The delayed weight loss, retained fruit firmness, and reduced disease incidences in ethylene treated banana was achieved by coating with MO+CN. MO+CN had a significant ( $p < 0.05$ ) effect on ethylene treated compared to non-ethylene treated bananas. The MO+CN treatment received the highest score in a consumer acceptance

test. This study proves that edible coatings containing chitosan nanoparticles could be used for maintaining the postharvest quality and extending the shelf life of bananas. These findings highlighted the edible coatings' capacity to operate as a gas barrier, altering the internal fruit environment. According to the findings of this study, adding chitosan nanoparticles to aloe and moringa edible coatings improved the quality of banana fruit during storage at room temperatures. The MO+CN coating was the most effective for quality preservation of banana fruit. As a result, MO+CN is recommended for coating banana fruit to improve postharvest quality and shelf life.



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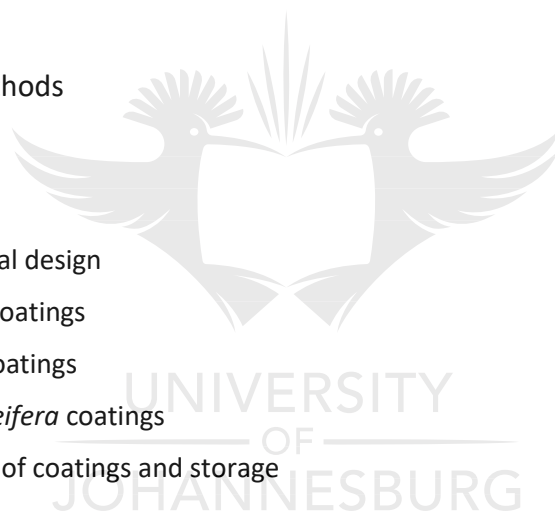
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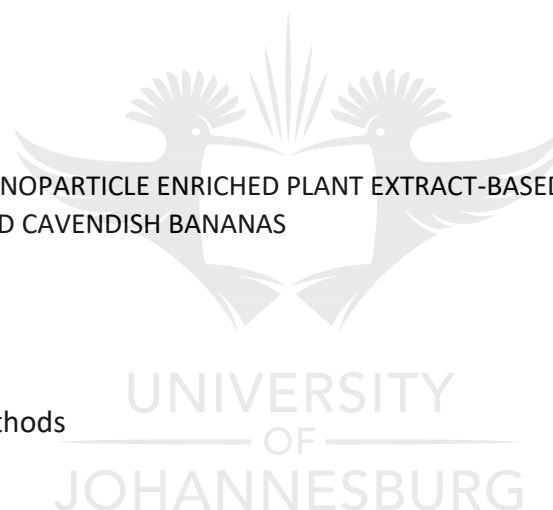
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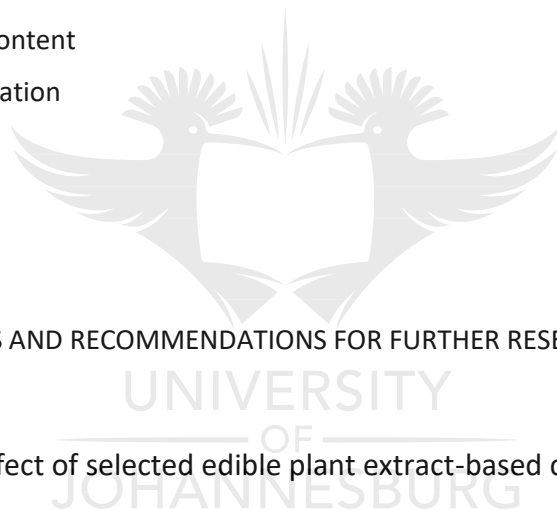


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## CHAPTER 1: INTRODUCTION

### 1.1 Rationale for the research (nature and scope)

Bananas are among the most important sub-tropical fruits grown in South Africa and are planted for sale in the local market or self-consumption and only a fraction of all bananas are sold in the world market. According to FAO (2018), the annual production of bananas in South Africa is 444879 tonnes from an area of 7482 ha. In South Africa, Mpumalanga, KwaZulu-Natal, and Limpopo provinces are the leading producers of bananas. In some parts of Africa like Ethiopia and Rwanda, post-harvest loss of banana was estimated at around 26.5% and 80% respectively (WFLO 2010; Mabratie et al., 2015) but in South Africa, it is difficult to give proper estimation due to limited investigation in the field. Bananas, on the other hand, as a respiratory climacteric fruit, are known for their quick deterioration due to physiological aging, biochemical alterations, and microbiological infections during transportation and storage (Soradech et al., 2017). These concerns frequently limit their edible quality and shelf life, potentially resulting in huge annual losses of fresh fruits. For this reason, technologies such as controlled atmosphere (Ahmed et al., 2006), modified atmosphere packaging (Workneh et al., 2009), nitrous oxide (Palomer et al., 2005), and 1-Methylcyclopropene treatment (De Martino et al., 2007), hot water treatment (Hassan et al., 2004), low-temperature storage (Zhang et al., 2010; Kudachikar et al., 2011), polyethylene bags (Hailu et al., 2012) were studied for their ability to maintain fruit quality and extend shelf life of banana. However, their application is cost-intensive, and they sometimes affect the nutritional and sensory properties of fruits (Guo et al., 2016). On the other hand, the use of natural extracts such as edible coatings is gaining much attention because they are relatively low cost, easy to prepare, safe for human consumption, environmentally friendly and can reduce the use of synthetic packaging materials (Bourtoom, 2008; Abdelrazek et al., 2019; Hafez et al., 2019). Most edible coatings are dipped or sprayed directly onto the surfaces of fresh food, then air-dried, and are usually applied in liquid form (Galus & Kadzińska, 2015; Tahir et al., 2019). The potential of using leaf extracts of *Aloe vera* and *Moringa oleifera* to improve the shelf and storage life of fresh food has sparked a lot of attention. By lowering ethylene production, respiration rate, and internal metabolic processes linked with fruit softening, colour development, enzymatic browning, and decay, both coatings play a

critical role in delaying fruit ripening. Due to their antifungal qualities, both coatings minimize microbial decomposition while also preserving visual appearance, firmness, and phenolic content, resulting in improved eating quality. Nanoparticles, when combined with an edible coating can increase mechanical and barrier properties while also acting as an antibacterial agent, as in the case of chitosan, resulting in improved food quality and safety (Belbekhouche et al., 2011; Martelli et al., 2013). Chitosan nanoparticles have been demonstrated to have great selective permeability, preventing weight loss and preserving fruit firmness for longer periods (Eshghi et al., 2014; CorreaPacheco et al., 2017). Fruits such as banana continue to respire and transpire after harvest and during storage, which leads to accelerated loss of postharvest quality. This study seeks to address the necessity for finding more cost-effective and affordable alternatives that could improve or complement the current techniques used in fresh produce markets for extending banana shelf life.

## **1.2 Justification**

In the tropical and subtropical regions of the world, bananas are the most extensively farmed and consumed fruit, and they are a major staple food crop for millions of people (Deka & Choudhury, 2006). In comparison to many other fruits, bananas have a more balanced nutritional profile. They include almost all the vital nutrients, such as minerals and vitamins (Islam et al., 2001). Bananas are usually harvested when they are ripe green and stored at room temperature or at a low temperature. Bananas are sensitive to diseases caused by bacteria due to their high nutritional value. Bananas are similarly susceptible to low-temperature storage (Malmiri et al., 2011). All the constraints limit the banana's handling, storage, distribution, and marketing possibilities. Banana fruit's spoiling was ascribed to negative physiological changes, such as weight loss due to respiration and transpiration, loss of flesh hardness, and resistance to microbial assault (Soradech et al., 2017). This type of spoiling can happen during shipment or at the market, leading to significant financial losses for the farmer, importer, and retailer. Furthermore, no known technique for extending the shelf life of bananas exists among South African growers/traders. As a result, due to the perishable nature of bananas, a significant amount of them go to waste. Therefore, reducing postharvest losses of banana fruit will cut the cost of production and distribution, lowering consumer prices, and improving farmer income. However, more

than 100 million tons of bananas are harvested and sold globally. As a result, increasing the shelf life of postharvest bananas has received a lot of attention (La et al., 2021). Cold, modified atmosphere, and controlled atmosphere storage technologies are regarded as expensive means of preserving bananas and may cause physical harm to fruits, resulting in chilling injury and poor ripening. Edible coatings are cost-effective and safe ways for extending the shelf life of postharvest bananas (Thakur et al., 2019). The mechanism of edible coatings protecting perishable fruits from degradation is to decrease respiration, improve textural quality, maintain volatile flavour, minimize microbiological development, and delay dehydration (Debeaufort et al., 1998). Many additives, such as antibacterial and antioxidant agents, taste compounds, and other nutrients, are widely added to edible coating systems to improve their performance (Nair et al., 2018). Antibacterial agents like chitosan have been used extensively in the development of edible coatings to improve food quality and safety. Most recently chitosan nanoparticles have been studied in banana and the results showed that the shelf life of coated banana was extended for 11 days compared to 9 days in the control (Lustriane et al., 2018). Nanoparticles were formulated from raw chitosan components. Because of their unique biological activities due to their small size and quantum size impact, chitosan nanoparticles have been employed as a medication carrier to date (Divya & Jisha, 2018). The permeability of the surface to water vapour and gases can be reduced by reducing the chitosan size while keeping the desired qualities of the chitosan coating (Divya et al., 2018). The application of edible coatings of *Aloe vera* gel, *Moringa oleifera* leaf extract, and chitosan nanoparticles in the banana industry in South Africa is a novel technology that has not been investigated before. As a result, this work could serve as a baseline for further research into the impact of nanoparticle-enriched coatings on the biochemical and molecular features of fruits as they ripen. It is thought that the findings of this study would be significant in determining how to increase the shelf life of not only bananas, but also other climacteric fruits including mango, avocado, papaya, and other similar fruits.

### **1.3 Aim**

This research aims to develop nanoparticle-enriched edible coatings to control ripening and extend the shelf life of Cavendish bananas

### **1.4 Objectives**

1. To evaluate the effect of selected edible plant extract-based coatings on shelf life of Cavendish bananas
2. To evaluate the effect of nanoparticle enriched plant extract-based coatings on shelf life of Cavendish bananas
3. To compare the efficacy and consistency of nanoparticle enriched plant extract-based coatings on shelf life of ethylene treated and non-ethylene treated Cavendish bananas

### **1.5 Outline of dissertation**

#### **Chapter 1: Introduction**

This chapter provides an overview of the issues experienced during the postharvest storage of bananas and provides background information on the technology that has been employed to increase shelf life, highlighting limitations and potential solutions. The research goal, justification and study objectives are also highlighted in this chapter.

#### **Chapter 2: Literature review**

This chapter provides a comprehensive assessment of literature on the use of edible coatings in climacteric fruits, nanoparticles, and their impact on quality parameters and fruit shelf life. The chapter also identifies the present knowledge gap in bananas that needs to be addressed and that aligns with the study's aim, as well as the safety and concerns associated with nanoparticle use.

#### **Chapter 3: Effect of selected edible plant extract-based coatings on shelf life of Cavendish bananas**

This chapter provides a detailed report on the effect of selected edible plant extract of *Aloe vera* and *Moringa oleifera* coatings on the shelf life of Cavendish bananas.

#### **Chapter 4: Effect of nanoparticle enriched plant extract-based coatings on shelf life of Cavendish bananas**

This chapter reports on the effect of chitosan nanoparticle-enriched with *Aloe vera* and *Moringa oleifera* plant extract coatings on the shelf life of Cavendish bananas. The chapter aims to address if adding chitosan nanoparticle to edible coatings will further extend the shelf life of banana and more quality postharvest parameters are assessed.

#### **Chapter 5: Effect of nanoparticle enriched plant extract-based coatings on shelf life of ethylene treated Cavendish bananas**

This chapter reports on the effect of nanoparticle enriched plant extract-based coatings on the shelf life of ethylene treated and non-ethylene treated Cavendish bananas. This chapter aims to address how well chitosan nanoparticle enriched edible coatings may work well with ethylene treated banana (industry standard) and compared with whether efficacy of coating is consistent for ethylene treated and non-ethylene treated bananas.

#### **Chapter 6: Conclusion and recommendations for further research**

This chapter re-states the research goal and objective by offering an overall conclusion from the experimental work done in Chapters 3–5, as well as how the coating developed can benefit the banana fruit sector. This chapter also includes suggestions for future applications and how the technology can help the fresh produce business.

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## CHAPTER 2<sup>1</sup>: NANOTECHNOLOGY-ENHANCED EDIBLE COATING APPLICATION ON CLIMACTERIC FRUITS

### Abstract

Climacteric fruits continue to ripen after harvest and produce ethylene, coupled with an increase in respiration rate, which contributes to more rapid perishability. Inhibition of ethylene biosynthesis has been shown to be an efficient way to delay the onset of ripening and lengthen shelf life. The use of edible materials as coatings presents an efficient approach in preserving the quality of fruits. Edible coatings have many benefits, such as affordability, easy application and use of natural ingredients. Nanotechnology provides interesting approaches to the management of fruit shelf life after harvest. Nanotechnology has the capacity of producing new materials by minimizing the size of components to a nanometric level. These kinds of nanomaterials possess distinct and improved properties for delaying fruit ripening and decay. The main goal of adding nanoparticles to edible coatings is to enhance the biopolymer's mechanical properties and water vapour barrier. Nanoparticles also contain biopolymer-like features and are thought to have superior antibacterial, antifungal, and antiviral properties than edible coatings. This review is aimed at summarizing recent findings on the application of edible coatings in the form of nanoparticles, and their effect on quality parameters and shelf life extension. Peer-reviewed articles were obtained by using Scopus and Science Direct. The current materials widely used for coating climacteric fruits are zinc, silver, and chitosan nanoparticles. Zinc nanoparticles have been shown to be more effective in delaying ripening significantly by reducing weight and moisture loss and ensuring retention of fruit firmness. Further research is needed to understand their effect on other physicochemical properties of fruits.

**Keywords:** *plant extracts, nanoparticles, postharvest, shelf life*

### 2.1 Introduction

Technologies such as ozone treatment (Alencar et al., 2013; Ali et al., 2014), modified atmosphere (Lalel et al., 2005; Lanka et al., 2011; Prange et al., 2013), 1-

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Methylcyclopropene (Mir et al., 2004; De Martino et al., 2007; Razzaq et al., 2016), controlled atmosphere storage (Hailu & Worku, 2017), and low-temperature storage (Kudachikar et al., 2011) have been used in controlling ripening in climacteric fruits. For several years up to date, low-temperature storage is used commercially in the supply and marketing chain from precooling at the farm to using refrigerated trucks set at a certain temperature for transportation to cold rooms at the fresh produce market (Zhang et al., 2016). Although low-temperature storage has been successful in controlling ripening in climacteric fruits, however storing fruit at suboptimal temperatures can cause chilling injury leading to internal browning and emergence of black spots (Sivankalyani et al., 2015). Other post-harvest technologies have also been successful, but some have certain disadvantages, such as a reduction in the volatile esters in fruit and a negative impact on the nutritional properties of fruit (Skog & Chu, 2001). Furthermore, the application of controlled and modified atmosphere is expensive, and it involves more labour, thereby limiting its practical application. Use of 1-Methylcyclopropene on climacteric fruit, might induce chilling injury and require additional time to ripen adequately, limiting its economic applicability. (Harris et al., 2000; Mir et al., 2004; De Martino et al., 2007; Rassak et al., 2016).

Edible coating is another type of technology becoming popular for controlling the ripening of climacteric fruit because it is simple to prepare, widely available, relatively low cost and mostly do not need the use of sophisticated atmosphere and temperature control technologies (Suhaget al., 2020; Jodhani & Nataraj, 2021). Edible coatings are thin layers of edible material added on the surface of climacteric fruits to enhance the appearance, maintain quality, prevent microbial growth, reduce respiration, slow maturation, minimize water loss, and extend shelf life (Murmu, & Mishra, 2018). Besides, the use of edible coatings in fresh produce has been approved by the US Food and Drug Administration and is considered to meet the GRAS (generally regarded as safe) requirements (Martín-Belloso et al., 2009; Dhall, 2013). To comply with export protocol, all the processes involved in the application of edible coatings must follow the high hygiene requirements and the quantity used must not exceed the amount necessary to achieve the desired physical and nutritional impact on the fruit (Nussinovitch & Nussinovitch, 2003). However, some edible coatings can pose some disadvantages when used on fruits, for instance, edible coating formulated from polysaccharides and proteins exhibit weak water vapour barrier properties while lipids

and waxes exhibit poor mechanical characteristics, thus new alternative methods have been developed to solve these drawbacks.

Nanotechnology is a novel approach for creating new edible coating components for the post-harvest management of fresh produce by reducing the size of the edible coating particles to a nanometric scale ranging from 1-100 nanometres (Parisi et al., 2015). The application of nanotechnology has proven to be one of the best strategies for extending the shelf life of fresh fruits, and nanoparticle used for edible coatings also possess unique properties such as antibacterial and antifungal (Lloret et al., 2012; Bajpai et al., 2018; Singh et al., 2020). Recently, there have been studies on the effects of the addition of nanoparticles to edible coatings and these studies have revealed that the addition of nanoparticles to edible coatings has opened new opportunities not only for enhancing higher antimicrobial, antifungal, and antiviral properties, but also to improve the cost-effectiveness of edible coatings (Sorrentino et al., 2007). The reviews by Nor and Ding (2020), Dhaka and Upadhyah (2018), and Ncama et al. (2018) have compiled all possible coatings that are functional for tropical fruits and horticultural produce, safe concentration and use of commonly consumed plant extracts which have potential to be used on fresh produce. The above reviews cover the use of polysaccharides, proteins, and lipids as edible coatings on major tropical fruits. The reviews also show that edible coatings are a promising method for extending the postharvest shelf life of tropical fruits. The current review talks specifically to the use of nanoparticle materials in addition to edible coatings on climacteric fruits. Recent review studies have reported the successful application of nanotechnology in food packaging industry, agricultural sector, agri-food sectors, and postharvest disease management of fruits and vegetables, where nanotechnology involves the application of nanoparticles to the edible coatings (Ruffo Roberto et al., 2019; Al-Tayyar et al., 2020; Mahela et al., 2020; Chawla et al., 2021; Lang et al., 2021; Wahab et al., 2021). As a result of all these studies, the application of various kinds of nanoparticles to the different edible coatings in order to maintain the quality of food appears to be a viable consideration to be pursued further. However, there is still limited information about the effectiveness of nanoparticle-edible coating material in regard to their application to climacteric fruits. Therefore, this review aims to highlight the most relevant and recent information on the use of edible coatings enriched with chitosan, silver, and

zinc nanoparticles in extending postharvest storage life and the overall preservation of the quality of popular climacteric fruit.

## 2.2 Problems faced by climacteric fruits

Based on their respiration behaviour and ethylene production, fruits such as banana, mango, guava, apricot, pear, papaya, apple, avocado, tomato, and plantain are referred to as climacteric fruits (Atkinson et al., 2011). Climacteric fruits are usually harvested at physiological maturity and remain firm without major changes in peel colour, texture, or composition before the beginning of maturation (Aguilera et al., 2003). After harvesting, the fruits undergo progressive deterioration, resulting in a relatively short post-harvest life, increased respiratory rate, autocatalytic ethylene development, increase in susceptibility to various pathogenic infections, and sensory changes such as colour, taste, and texture change such as softening (Palapol et al., 2009; Paul et al., 2012).

In post-harvest fruit management, the perishability of fruit after harvest is a major challenge faced by the industry influencing produce marketability, especially for international trade (Singh et al., 2014). Globally, a massive quantity of fruit is wasted before the commodity reaches consumers, with about 50% of those food losses being valuable fruits. The U.S.D.A. Economic Research Service reports that over 34.6% of the loss is directly related to unwanted climacteric maturation, resulting in subsequent spoilage, degradation, mechanical injuries, and physiological disorders in produce (Kader, 2004; Barth et al., 2009). The perishability of the fruits is mainly attributable to adverse physicochemical changes, such as loss of weight due to respiration, softening of the flesh, deterioration of quality due to microbial attack, and changes in the content of sugar and acid. Another big threat to the global fruit supply chain is contamination by fungal pathogens. Postharvest fruit loss due to phytopathogen fungi now accounts for more than half of all agricultural fruit production (Zhang et al., 2017). The most important factors causing postharvest losses are intrinsic physiological senescence and invasion by fungal pathogens. Anthracnose caused by *Colletotrichum* spp is one of the common fungal diseases that can result in serious economic and extensive post-harvest losses during transportation and storage of climacteric fruits (Bautista-Baños et al., 2013; Pavitra Kumari & Singh, 2017). Symptoms of post-harvest anthracnose are sunken black spots that occur on the fruit surface during ripening (Tian et al.,

2016). Mango anthracnose, caused by *Colletotrichum gloeosporioides*, is a significant threat to farmers around the world, as it results in massive pre-and post-harvest losses to mango crops (Lima et al., 2013; Pavitra Kumari & Singh, 2017). During postharvest preservation and following export, papaya deteriorates, primarily due to anthracnose caused by *Colletotrichum fructicola* and *Colletotrichum gloeosporioides* Penz (Madani et al., 2014; Vilaplana et al., 2020). The most damaging postharvest disease of bananas is anthracnose, which is caused by the fungus *Colletotrichum musae* leading to substantial economic loss (Khaliq et al., 2019). It degrades the fruit's quality and nutritional value and makes it unfit for marketing and consumption, resulting in significant losses for farmers and traders.

The use of synthetic chemical fungicides has resulted in issues with postharvest disease control, including decreased efficacy, increased plant pathogen resistance to active ingredients, environmental damage, and a serious negative impact on human health (Dubey et al., 2007; Kumar & Kudachikar, 2018). Synthetic chemicals have been used to reduce fungi attacks in postharvest storage of fruits but there are concerns against their safety due to the toxicity of chemicals thus making it an urgent need to find alternative environmentally friendly technologies. With the above-mentioned problems facing climacteric fruits, one of the most preferred solution includes the application of edible coatings (Romanazzi et al., 2018). Many studies have shown that edible coatings made from a variety of biopolymers can effectively preserve the nutritional properties and extend the shelf life of climacteric fruits. To suppress decay, improve fruit quality, prolong the shelf life of climacteric fruits during postharvest storage, edible coatings such as chitosan, *Aloe vera gel*, and gum arabic are widely used (Maqbool et al., 2010; Berumen-Varela et al., 2015; Khaliq et al., 2019).

### **2.3 Formulation of edible coatings**

Generally, films from edible coatings are prepared from polymers such as hydrocolloids (polysaccharides and protein), lipids, a combination of both (referred to as composite coatings), and the addition of plasticizers (Dhall, 2013). Edible coatings can be applied over the food product in liquid form by spraying, extrusion, solvent casting, brushing, or dipping to achieve a thin protective layer (Yousuf et al., 2018; Thakur et al., 2019). A review by Nor and Ding (2020) compiles all possible coatings

that are functional for tropical fruit. The review also covers the fundamentals of coating attributes, materials, and processes, which include the following: the effectiveness of various coating materials such as polysaccharide, protein, lipid, and composite based coatings have been highlighted, and various application methods, and coating protection. Dhaka and Upadhyay (2018) wrote a simple review on edible films and coatings, as well as recent innovations in the field. This analysis provided a detailed overview of various aspects of edible films and coatings, as well as a discussion of current trends and innovations. Furthermore, edible coating can be formulated using polymeric materials derived from both plant and animal sources. The application of animal extracts in the production of edible coatings has been limited due to allergies from animal foods and the waxy nature of coatings thereby limiting food product acceptability compared to plant extracts with medicinal benefits (Oms-oliu et al., 2010; Flores-López et al., 2016). Plant extract derived edible coatings can delay ripening, improve aesthetic appearance by shinning the produce and covering minor scars (Murmu & Mishra, 2018). Also, they are an inexpensive means for maintaining the quality of fresh produce. The use of edible coatings from plant extracts has been proposed to decrease the usage of non-biodegradable storage polyethylene plastic films and containers, thereby reducing pollution to the environment (Bourtoom, 2008).

Plant extracts with high antioxidant properties can also improve the nutritional qualities of fruits. The effect of plant edible coatings on the quality attributes and nutritional characteristics of various climacteric fruits such as banana, apple, mango, and papaya has been studied. A review by Ncama et al. (2018) gave a comprehensive report on the use of plant extract derived edible coatings for both climacteric and non-climacteric fruits. Some of the plants whose extracts are used as edible coatings for climacteric fruits include moringa leaf extract (Tesfay & Magwaza, 2017), corn starch and rice starch (Razak & Lazim, 2015; Thakur et al., 2019), *Aloe vera* (Khaliq et al., 2019), and gum arabic (Maqbool et al., 2011). The use of natural edible coating extracts is one of the most promising technologies to enhance the protection and quality of fruits because it is considered as being environmentally friendly and acceptable for consumers (Janisiewicz & Korsten, 2002).

### **2. 3.1 Unique features of edible coatings**



The edible coating serves as a barrier to control moisture loss and gas exchange ( $\text{CO}_2$  and  $\text{O}_2$ ) between the fruit and their surrounding environment thereby slowing down the rate of respiration, retarding the physiological ripening process, and preventing the loss of natural volatile flavour compounds (Rojas-Graü et al., 2009; Pratiwi et al., 2015; Khatri et al., 2020). Furthermore, edible coatings can safely be consumed as part of the product and contain health benefits because they are made of food-grade products (Shit & Shah, 2014). Other advantages of coatings include their edibility and biodegradability, as well as the avoidance of waste and the commercialization of food without preservatives (Tavassoli-Kafrani et al., 2016; Kumari et al., 2017). The coating enhances post-harvest shelf life by delaying physicochemical changes and preventing the development of physiological disorders (Kumar et al., 2017). Likewise, edible coatings have a high propensity to provide active compounds such as antioxidants (ascorbic acid, citric acid, oxalic acid), antimicrobials (potassium sorbate, essential oil), texture enhancers (calcium chloride, calcium lactate, calcium gluconate), and nutrients (vitamin E), which can improve resistance to fungal pathogens plus the dietary and organoleptic characteristics of fruits (Martín-Belloso et al., 2009; Dhall, 2013; Arnon-Rips et al., 2019). To choose a coating for fruits, it is important to understand the properties of the coatings and how well they interact with the fruit surface and surroundings during storage. Edible coatings need to have a flawless adhesion capacity, high microbial protection, moisture exchange properties, appealing aesthetic appearance, and above all availability at an affordable price (Poverenov et al., 2013). It is advisable to use coatings that have been shown to remove respiration peaks efficiently and reduce the output of ethylene to a minimal level.

#### **2.4 The effect of edible coatings on quality attributes in climacteric fruits**

The ripening process of climacteric fruit shows a dramatic increase in ethylene production and respiration rate at the onset of ripening. Various parameters such as weight loss, firmness, total soluble solids, total phenol, and antioxidant activity, decay rate, and shelf life have been used to assess quality in climacteric fruit (Hudina et al., 2012). The efficacy of using edible coatings has been demonstrated by increasing evidence from numerous studies. In a study that investigated the effect of shellac and gelatin composite coatings for extension of shelf life of a banana, Soradech et al.

(2017) observed 60% of shellac and 40% gelatin act as an effective physical barrier around the fruit, resulting in a slow decrease in weight loss, and softening. The quality was maintained for more than 30 days compared to uncoated banana fruit. A report by Jaiswal et al. (2018) indicated that the incorporation of citric acid and neem extract improved the effectiveness of *Aloe vera* by maintaining the firmness, colour, sensory attribute, and market value of tomato fruit. *Aloe vera* 40% plus citric acid gave the best result compared to other concentrations (20%, 60%, and 80%). Recently, a study by Kubheka et al. (2020) showed the effect of gum arabic- and carboxymethylcellulose-containing moringa leaf extract on maintaining quality and control of *C. gloeosporioides* on Maluma avocado at cold storage for 21 days. Based on the results, 15% gum arabic plus moringa followed by 10% gum arabic and moringa and 1% carboxymethylcellulose plus moringa were the most effective in reducing weight and firmness loss. The coatings also delayed color change and inhibited the growth of *C. gloeosporioides* compared to the control.

Edible coatings containing antioxidants and antimicrobials have been shown to improve the nutrient value and protect against pathogens and spoilage (Dhall, 2013; Pranoto et al., 2015; Arnon-Rips et al., 2019;). A report by Yang et al. (2019) evaluated the efficacy of gum arabic enriched with white and red roselle extracts on the postharvest quality of blueberry fruit stored at refrigerator temperature. Gum arabic was found to have antioxidant and antimicrobial effects, owing to polyphenol compounds in the gum. The existence of more bioactive compounds, such as phenols and flavonoids, which could inhibit certain microorganisms, could explain the red roselle extract's higher antimicrobial capacity against all tested microorganisms. Due to the synergistic effects of natural antimicrobial compounds in gum arabic and roselle extracts, this finding showed the least decay. Gum arabic combined with roselle extract served as a barrier between the blueberries and their surroundings, controlling gas and water vapor exchange and delaying weight loss during storage. The slower loss of firmness of coated fruits may be linked to a well-maintained cellular membrane as a result of gum arabic, gum arabic + white roselle extracts, gum arabic + red roselle extract coatings inhibiting PPO enzyme activity, reducing the softening process of the fruit, therefore, the addition of white and red roselle to gum arabic showed a better performance in maintaining firmness, reducing weight loss, and decay percentage compared to gum arabic alone and the control. Edible coating containing gum ghatti

enriched with clove oil was used on banana and papaya fruits to extend shelf life. Gum ghatti 3% plus clove oil 0.1% retained the ascorbic acid, total phenol, and antioxidant activity in both fruits. In bananas, shelf life was extended by 3 days in both fruits relative to the control (Joshi et al., 2017; Joshi et al., 2018).

A report by Abd El-Razek et al. (2019) showed that moringa and green tea extract act as an antioxidant coating and was effective in reducing vitamin C loss, total soluble solids, total phenol, and antioxidant activity, and a decrease in weight loss was observed in mango fruits at 2,4 and 6 weeks during two consecutive seasons. Moringa leaf extracts are also rich in antioxidants and have antibacterial effects against a range of microorganisms. The high phytochemical constituents of moringa plant extracts, which include phenols, alkaloids, and tannins among a few others, have been attributed to the inhibitory effect of plant extract on the mycelial growth of various pathogens. Furthermore, tea leaves are high in polyphenolic compounds, which have a high antioxidant potential and antimicrobial activity in general, hence, the properties of moringa and green tea make them suitable as coating materials. Natural substances present in moringa and green tea extracts, which are high in antioxidants, serve as electron donors, creating free radicals that minimize normal respiration and transpiration, as well as stomata closure. The reduction of fruit decay caused by the coating of moringa, and green tea extracts is linked to a reduction in the activity of cell wall degrading enzymes, which prolongs the postharvest period and delays fruit ripening. Because of its low oxygen permeability, which decreased enzyme activity and prevented oxidation of vitamin C, moringa and green tea extracts as antioxidant coating treatments were successful in reducing vitamin C loss in mango fruits during all storage periods. The best result to achieve a high value of storability and quality was shown in applying 10% moringa leaf extract followed by 5% green tea extract under refrigerated storage.

Shah and Hashmi (2020) investigated the impact of chitosan in combination with *Aloe vera* gel on the storage life of mango fruits. He found that adding chitosan to *Aloe vera* lowered weight loss, respiration rate, and ethylene generation more effectively than using chitosan alone or control samples. Furthermore, the combination treatment preserved fruit quality metrics such as titratable acidity, total soluble solids, fruit firmness, ascorbic acid, and peel colour. This study shows that combined application of chitosan

and *Aloe vera* synergistically improve the phenolic content of mango fruit, sustaining high ascorbic acid, total phenolic content, and antioxidant activity of mango fruit during storage. This suggests that addition of *Aloe vera* may enhance the barrier of chitosan coating thereby improves antimicrobial properties and decreases permeability to water and gaseous exchange. Jodhani and Nataraj (2021) focus their research to study how *Aloe vera* gel, lemon peel extract, and their combination as edible coating treatment affect the banana postharvest quality and shelf life when stored at room temperature. The consistency of bananas in treated fruits at the end of the storage period indicated that the lemon peel extract concentrate in the edible coating treatment prevents microbial contamination and protects the fruits from pathogenic fungi deterioration. When compared to the control banana, the coated banana had less weight loss and good firmness, as well as no extreme infection, which is the most important factor that decides the banana's storage life and consistency. Aloe gel and LPE coating application dramatically reduced fruit respiration slowed ripening and delayed the emergence of visual indicators of consistency loss, all of which are undesirable to consumers. The aloe gel and LPE coating effectively reduced the rate of water loss from bananas during storage, according to these findings.

Daisy et al. (2019) observed the effect of gum arabic on the shelf life and quality of mango fruit during storage at room temperature. Gum arabic coatings were shown to have gas and water vapour barrier properties, allowing mango to last longer while preserving quality. Gum arabic changed the environment around the fruits by creating a semi-permeable film that prevented moisture and gas loss migration through the coat thereby delaying ripening and this helped gum arabic coated mango to have a shelf life of 15 days compared to less than 10 days in control fruits. The efficacy of various plant edible coatings including gum arabic, sodium caseinate with the addition of lemongrass, and cinnamon essential oil was evaluated by Murmu and Mishra (2018) at varying concentrations on the post-harvest quality attribute of guava fruit. The fruit with coating exhibited a slower rise in total and reducing sugar, lowest browning rate, higher retention of ascorbic acid, phenol and flavonoid content, overall sensory score, and extended shelf life by 33 days compared to the control. The addition of lemongrass oil and cinnamon oil helped retain higher membrane integrity thereby preventing disease occurrence and sustained metabolic rate.

Unfortunately, the application of edible coatings still faces limitations: for example, they may be unattractive to the consumers as some exhibit their colour or require undesirably high dose applications to be effective. Studies have shown nanomaterials with a unique character such as small size and quantum size have been explored to produce nanoparticles which can improve the efficacy of coatings (He & Hwang, 2016).

## **2.5 Nanotechnology**

Nanotechnology is another form of innovation that offers countless post-harvest management approaches capable of producing new materials by reducing particle sizes to a nanometric scale (at least one-dimensional ranges of 1-100 nanometres) giving materials with distinct and improved properties compared to larger ones (Magnuson et al., 2011; Parisi et al., 2015). Nanoparticles (NPs) (nanoscale structures with sizes ranging from 1-100 nm) have emerged as one of the outstanding nanotechnology discoveries designed to solve the day-to-day problems of the world today. Based on fundamental characteristics, nanoparticles exhibit an entirely new or enhanced properties, such as size, large ratio of surface area to volume distribution, and morphology. It is important to remember that only a small amount need be added to form a strong interfacial contact with the edible coat polymer. The addition of nanoparticles to the edible coating results in a significant extension of fruit shelf life compared to the effect of the pure polymer without nanoparticles (Gad & Zag Zog, 2017). Nanoparticles, when added to edible coatings can significantly improve the mechanical and barrier properties, and increase thermal stability (De-Moura et al., 2009; Shankar & Rhim, 2015). Nanoparticles can be classified into two groups namely organic NPs and inorganic NPs. More emphasis is placed on inorganic nanoparticles because of their stability compared to organic ones that are heat labile compounds. Inorganic nanoparticles consist of metal or metal oxides, such as gold (Au), silver (Ag), iron oxide ( $\text{Fe}_3\text{O}_4$ ), titanium oxide ( $\text{TiO}_2$ ), copper oxide (CuO), zinc oxide (ZnO), aluminium oxides, cerium dioxide hydroxides, calcium carbonate, and carbon-based materials (Bouwmeester et al., 2014; He & Hwang, 2016). According to numerous reports, using nanotechnology to extend the shelf life of fruits is one of the most effective techniques (Lloret et al., 2012; Flores-López et al., 2016; Ruffo Roberto et

al., 2019; Ijaz et al., 2020; Bhusare & Kadam, 2020). The most used method for the application of nanoparticles is the dipping method (Table 2.1), by immersing the fruit into the coating solution forming a thin layer on the surface of fruits and subjecting it to cold or ambient temperature storage. A review by Rajat Suhag et al. (2020) reporting on film formation and deposition methods of edible coating on food products suggested that the dipping method was found to be the cheapest and easiest to use on the surface of food products among other edible coating methods.



Table 2.1: Nanoparticle-enriched edible coatings applied on popular climacteric fruits

Fruit type	Nanoparticle components	Other ingredients	Coating method	Effect	References
Apple	Chitosan	Acetic acid	Dipping	The coating, improved consistency of colour quality, slowed down fruit softening and decreased weight loss by up to 2.5 times over 9 weeks of storage	Gardesh et al. (2016)
	Silver/Zinc oxide	Gelatin/Chitosan	Dipping	The fruit quality was preserved, and the shelf life was extended by 42 days	Bakhy et al. (2018)
Tomato	Gum Arabic	Tween & NaCl	Dipping	It maintained overall quality and extended the storage life by 14 days.	Paladugu, and Gunasekaran, (2017)
	Silver	Silver nanoparticles from Chinese tea	Dipping	Specifically found to reduce weight loss in fruit, and extended shelf life of fruit by 18days of storage at room temperature	Gao et al. (2017)
	Silver/Zinc	Gelatin/Chitosan	Dipping	The fruit quality was maintained, and the shelf life of coated fruit was extended by 63 days	Bakhy et al. (2018)
	Zinc	Carboxymethylcellulose	Dipping	The combination showed a beneficial effect in improving quality parameter compared to control and effectively delayed the disease severity during 15 days of storage	Saekow et al. (2019)
Banana	Zinc oxide	Soybean proteins isolate & cinnamaldehyde	Dipping	It effectively delayed ripening and improve the shelf life of banana by maintaining the nutrient content and hinder the loss of water during seven days of storage	Li et al. (2019)
	Chitosan	Chitosan	Dipping	The coatings maintained the sensory quality and extended the shelf life of banana for several days	Lustriane et al. (2018)
	Chitosan	Acetic acid	Spraying	The ripening was delayed by showing a slower rate of skin discoloration as compared to control during six days of storage	Esyanti et al. (2019)
	Zinc	Chitosan/gum Arabic	Dipping	The consistency of the bananas was retained for a slightly longer period and shelf life was prolonged after more than 17days in storage	La et al. (2021)

Fresh produce	Nanoparticle components	Other ingredients	Coating method	Effect	References
Banana	Silver	Neem and Ajwain	Spraying	Control Anthracnose disease in banana	Jagana et al. (2017)
Mangoes	Calcium	Ascorbic acid	Dipping	It alleviates internal browning, maintains the phenolic compound of mangoes during cold storage	Lo'ay and Ameer (2019)
	Zinc	Carrageenan	Dipping	Increase antimicrobial properties and maintain the shelf life of whole mango fruit	Maindrawan et al. (2018)
	Silver	Chitosan and Tween 80	Dipping	The combination minimized postharvest decay by inhibiting anthracnose incidence on mango during seven days of storage	Chowdappa et al. (2014)
	Zinc	<i>Aloe vera</i> gel & glycerol	dipping	It improves quality parameter during 9days of storage	Dubey et al. (2019)
Fig	Zinc	Cassava starch & stearic acid	Dipping	It was effective in reducing weight loss, delayed microbial growth, and improve the shelf life of fresh-cut mango during storage at 8 °C, for 12 days.	Luliani et al. (2018)
	Zinc	Chitosan & acetic acid	Dipping	Coating delayed the ripening of fruits, and keep quality during storage	Lakshmi et al. (2018)
Guava	Chitosan	Xanthum gum & tween	Dipping	It enhances overall quality during cold storage and shelf-life periods	Gad and ZagZog, 2017
Papaya	Silver	hydroxypropyl methylcellulose	Dipping	Silver nanoparticle was effective against <i>Colletotrichum gloeosporioides</i> , preserved postharvest quality, and shelf life was extended by 14 days during storage	Vieira et al. (2020)
Apricot	Silver	Glycerol	Dipping	It significantly reduces weight loss, decay percentage and kept the quality for 24 days at 6 °C	Shahat et al. (2020)



### **2.5.1 Synthesis of nanoparticles**

Currently, various physical and chemical methods are widely used to synthesize nanoparticles, enabling particles with the necessary characteristics to be obtained. These manufacturing methods can present several drawbacks such as the use of non-biodegradable stabilizing agents, labour-intensive, usage of toxic chemicals, and are potentially detrimental to the environment and living organisms (Phanjom & Ahmed, 2015; MubarakAli et al., 2015). Therefore, to minimize hazards to the environment, green/biochemical synthesis of nanoparticles offers an appealing means for nanoparticle synthesis and promises to help overcome these physical and chemical disadvantages (Shankar et al., 2004; Nayak, 2015). This is due to low synthesis costs, short development time, easy accessibility, eco-friendliness, economic considerations (have the potential to generate high production volumes) and use of plant-based materials (Kavitha et al., 2013; Akintelu & Folorunso, 2019). The green synthesis technique involves the use of naturally existing resources where an extract of plants acts as a reducing and stabilizing agent (Sharma et al., 2016; Jamdagni et al., 2018).

The green synthesis of different nanoparticles based on plant extracts has been extensively studied since the last decade as shown in Figure 2.1 (Esa & Sapawe, 2020). Many studies on the production of silver nanoparticles from plant extracts have been reported by (Srikar et al., 2016; Rajeshkumar & Bharath, 2017; Ahmad et al., 2019). A thorough examination of the green synthesis and characterization methods for ZnO NPs derived from various biological sources was studied and has thus become a major research subject (Agarwal et al., 2017, Ahmed et al., 2017; Bandeira et al., 2020). Alternatively, nanoparticles can be synthesized with the use of natural polymers from marine (chitin and chitosan) or agricultural waste (cellulose, gums, starch, and pectin) which has the added advantage owing to their stability, small size, edibility, and non-toxic nature (He & Hwang, 2016).

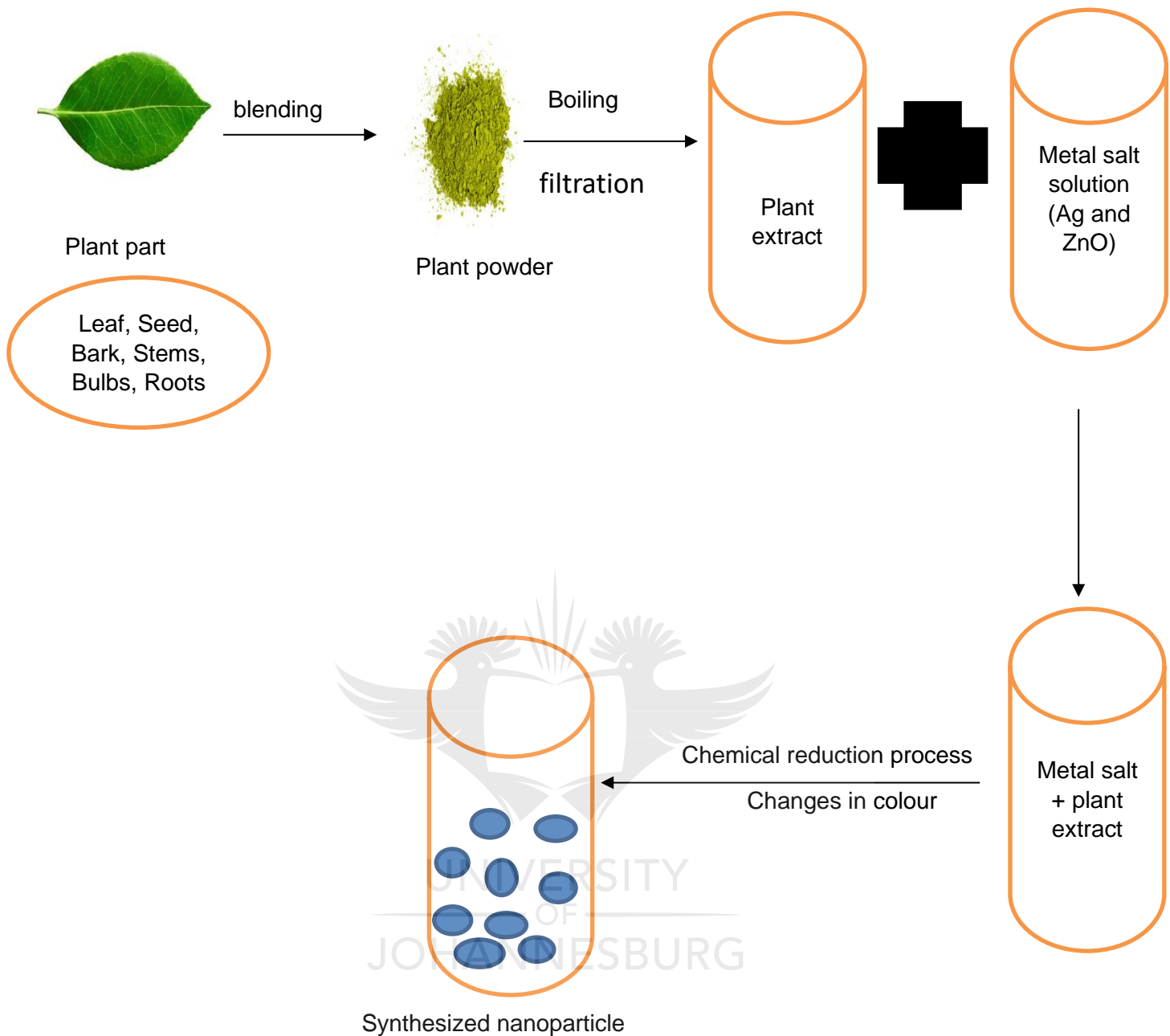


Figure 2.1. Schematic representation of nanoparticle synthesis using green method (Esa & Sapawe, 2020).

## 2.6 Nanoparticles commonly used in climacteric fruit

Several nanoparticles have been used in fruits. The most explored nanoparticles in climacteric fruits are zinc oxide, silver and chitosan considering their higher antimicrobial activity and stability. Nevertheless, other nanoparticle such as iron,

titanium dioxide, cerium oxide, and copper have been used in various fields of the food sector. Titanium dioxide nanoparticle was reported used with chitosan coating to form a film on the surface of mango fruit. It was effective in reducing losses caused by decay, delay respiration and maintain the firmness of fruits (Xing et al., 2020). Copper nanoparticles were sprayed on tomatoes and showed an increase in the content of bioactive compound and maintenance of quality (López-Vargas et al., 2018). Also, cerium oxide was blended with ascorbic acid exhibiting a significant reduction of internal browning of mango fruit during cold storage. In addition, nanoparticles have been used in food packaging and additives but there are limited studies on their application on climacteric fruits therefore there is a need to explore the use in shelf life extension of fruits (Kumar et al., 2020).

### **2.6.1 Chitosan nanoparticle**

Chitosan (CS) is one of the promising biopolymers that has been studied as a nanoparticle because of its film-forming capability, biodegradability, biocompatibility, and antimicrobial activity, non-toxic to humans, ease of alteration, and flexible physical and chemical properties (Jianglian & Shaoying, 2013; Divya & Jisha, 2018). Chitosan derived from the deacetylation of chitin in an alkaline medium is obtained from the waste products of the shellfish industry (Xu et al., 2005; Suseno et al., 2014). Chitosan is considered generally regarded as safe (GRAS) and recently approved by the United States Food and Drug Administration (USFDA) [Katiyar et al., 2014; Hu et al., 2019]. Chitosan nanoparticles (CS-NPs) are a derivative of CS with excellent physicochemical properties (Divya et al., 2017; Kassem et al., 2019). The chitosan nanoparticles outperformed the chitosan edible coatings in terms of antioxidant and antibacterial activity. CSNPs have a smaller particle size and a larger contact area than CS, which contributes to their high biological activity, the CSNPs can move through biofilms and destroy pathogenic bacteria because of the same reasons (Qi et al., 2004; Shrestha et al., 2010; Huang & Li, 2014). The potential advantages of chitosan nanoparticles over conventional chitosan are distinct as they strengthen the barrier properties and functionality of edible coatings because of their increased surface area (Eshghi et al., 2014). Chitosan nanoparticles have the distinctive characteristics of chitosan biopolymer and are considered to have higher antimicrobial activities and barrier properties (Kalaivani et al., 2020). In previous research, a

nanochitosan-based coating was successfully applied on apples and bananas (Gardesh et al., 2016; Lustriane et al., 2018; Esyanti et al., 2019). This form of coating greatly enhanced sensory efficiency, increased storage life, and retained the fruit's bioactive components.

### **2.6.2 Zinc oxide nanoparticle (ZnONPs)**

Zinc oxide nanoparticles have gained the attention of many researchers for their unusual peculiar optical and chemical behaviours among the metal oxide nanoparticles, which can be easily tuned by adjusting the morphology. Within the broad family of metal oxide nanoparticles, zinc oxide nanoparticles have been used in various advanced numerous cutting-edge applications, such as electronics, communications, sensors, cosmetics, environmental protection, biology, and the medical industry, and their food safety (GRAS-Generally Recognized as Safe) has been properly approved by the US Food and Drug Administration (FDA) (Rasmussen et al., 2010; Noshirvani et al., 2017). Due to its excellent mechanical properties, barrier capacities, biocompatibility, and antimicrobial broad-spectrum performances, the zinc oxide nanoparticle (ZnONP) has gained considerable interest in sciences (Yusof & Zain, 2019). The antimicrobial properties of ZnO particles are due to the reactive oxygen species that form on their surface. In addition, recent scientific studies have shown that zinc is a promising coating material due to its being a relatively potent antimicrobial agent with high stability as a comparison to natural-based coating and there is no possible risk to human health from its use (Sun et al., 2018). Furthermore, as stated elsewhere, ZnONP's addition to polysaccharides, lipids, and protein-based biopolymers can effectively improve the mechanical properties, barrier capacities, and physicochemical properties of edible coatings (Muraleedaran & Mujeeb, 2015; Wu et al., 2019). ZnONPs have highly effective antibacterial activity and are considered as a possible additive to replace hazardous chemicals and physical antibacterial materials (Awwad et al., 2020).

### **2. 6.3 Silver nanoparticles (AgNPs)**

Among nanoparticles, silver nanoparticles (AgNPs) are one of the most studied as they have shown to be efficient against different microorganisms and are safe for humans (Aadil et al., 2018). Around the same time, silver has been adopted as an antimicrobial material that is relatively free of adverse effects. A wide variety of antibacterial, antifungal, and antiviral properties are found in silver nanoparticles. Due to its biocidal activity against a wide range of Gram-positive and Gram-negative microorganisms, yeast, molds, and viruses, silver is currently the most researched antibacterial nanoparticle. The release of Ag<sup>+</sup> ions, which bind to electron donor groups in molecules containing sulfur, oxygen, or nitrogen, is primarily responsible for the antimicrobial activity of silver nanoparticles. Additionally, AgNPs outperformed metallic silver in antimicrobial properties due to their incredibly large surface area, which allows for better contact with microorganisms (Toker et al., 2013). The safety limit of silver declared by EU safety regulations for foods is 0.05 mg/kg (Fernández et al., 2009). It is proved that a silver concentration of 0.06 mg/L is acceptable for coating fruits and vegetables (An et al., 2008). The use of silver nanoparticles, which include a wide variety of compounds that can be used in the formulation of edible coatings, is the most recent breakthrough advancement in the application and development of edible coatings for fresh fruit.

## **2.7 Application of nanoparticle-enhanced edible coatings on climacteric fruits**

The application of nanoparticle-enhanced edible coatings has been explored in postharvest shelf life research and can be effective in improving colour quality, firmness, increase antimicrobial properties, control enzymatic activity, and reducing weight loss of fruits (Table 2.1). The incorporation of silver nanoparticle into sodium alginate inhibits the growth of the microbial diseases in pear because after coating, the silver nanoparticle incorporated sodium alginate coatings maintains its antibacterial activity against Gram-positive and Gram-negative bacteria. The coated fruit was found to be suitable for up to 10 days in storage as judged by the colour and appearance, texture, and aftertaste compared to sodium alginate-coated and uncoated fruit (Mohammed Fayaz et al., 2009). Arroyo et al. (2020) investigated the effect of chitosan plus alginate plus zinc nanoparticles on the postharvest life of guava. The coating was able to prevent rot appearance, retarded physiochemical changes

related to maturation. Zinc nanoparticles combined with 90 or 100% chitosan, and 10% alginate extended the shelf life by 13 days compared to the control in guava.

The utilization of nanoparticles shows improvement in the storage quality of fruits. Joshy et al. (2020) applied novel zinc oxide nanoparticle-reinforced xanthan gum on apple and tomato fruit. It preserved the fruits from deterioration and water loss. Based on these findings, the manufactured edible coatings may serve as a novel antimicrobial agent to safeguard the fruit from microbial contamination. Malek (2020) observed the effect of storage temperatures on the shelf life of golden lily mangoes treated with zinc oxide nanoparticle and tapioca starch for 7 days of storage at 32°C, 27°C, and 5°C. The fruit firmness was reduced at 32°C but storage life was 2 days maximum while storage temperature of 5°C was found to be most suitable for delaying textural changes and maintaining the storage life of mango by 7 days. The study's most striking finding is that thanks to zinc oxide's relatively strong antimicrobial properties and starch's good mechanical properties, the ZnO-starch coating's reduced anthracnose disease growth, delayed texture changes and maintained the shelf life of mango at lower storage temperatures.

Chandirika et al. (2018) studied the effect of silver nanoparticles on the quality attributes of tomato fruit at room temperature. Their results indicated that the application of silver nanoparticles showed an extended shelf-life period from 16 to 21 days and sensory quality was maintained when compared with control (non-coating). Li et al. (2019) successfully developed zinc nanoparticles with soybean protein isolate and cinnamaldehyde as a coating for banana stored at room temperature. Results concluded that the coatings not only delayed ripening and extended shelf life up to 7 days in storage at room temperature but also inhibited fruit fungus spoilage through the oxidative stress-directed manner. Table 2.1 shows more studies done on the application of nanoparticles to edible coating resulting in keeping the quality of climacteric fruits.

## 2.8 Advantages and disadvantages of adding nanoparticles to edible coatings

The benefits and drawbacks of the addition of nanoparticles to edible coatings have been studied. The addition of nanoparticles to edible coatings has provided various benefits, such as increased antimicrobial activity, and formation of stronger coating homogeneity on fruit surface (Severino et al., 2014; Acevedo-Fani et al., 2017). Chitosan, zinc oxide, and silver are the most widely used nanoparticles in climacteric fruits that have shown promising effects when applied to edible polysaccharide and protein materials. It has been stated that zinc has better compatibility and heat resistance in climacteric fruits (Table 2.1). The main goal of adding nanoparticles to edible coatings is to enhance the biopolymer's mechanical properties and water vapor barrier.

Firstly, ZnO enhanced xanthan hybrid method provides greater health benefits considering zinc requirement in the human body and is healthy in blood compatibility and toxicity tests (Joshy et al., 2020). Zinc oxide metal has been shown to have antimicrobial properties, demonstrating strong effectiveness in inhibiting the growth of pathogenic microorganisms, even when added in small amounts such as 0.1-0.5% (w / v) (Esparza-González et al., 2016).

Recently, Meindrawan et al. (2018) found that the addition of zinc nanoparticle to carrageenan effectively decreases weight loss and total acidity, preserve firmness and delay discoloration and decay of mango fruit compared to carrageenan alone. This is because zinc can improve the gas barrier of the coating as compared to carrageenan alone which tends to be hydrophilic. Similarly, zinc significantly improved the quality of cherry tomatoes by suppressing their respiration and water evaporation thus ensuring a better preservative effect at room temperature storage. The addition of zinc to carboxymethylcellulose and cinnamaldehyde not only reduced weight loss and ensured fruit firmness for a longer period but significantly inhibited the tested fungi showing greater antimicrobial activity compared to non-coated or pure carboxymethylcellulose with cinnamaldehyde (Guo et al., 2020). It is possible to attribute this effect to the synergistic antifungal effect between ZnONPs and carboxymethylcellulose. Gad and Zag Zog (2017) tested xanthan gum mixed with 0.2% and 0.4% chitosan nanoparticles against the uncoated and xanthum alone on

guava fruit. Xanthan gum mixed with 0.2% nanochitosan decreased decay, colour change, maintained fruit firmness, vitamin C, and good taste compared with xanthan gum or a high concentration of chitosan nanoparticles (0.4%). It also enhances the overall quality and extended shelf life at cold storage. This recent utilization of nanoparticle has encouraged the use of a lower concentration of coating administered in the form of nanoparticles to enhance quality and extend the shelf life of fruits.

Chitosan nanoparticles have demonstrated significant effects as a postharvest treatment in terms of antioxidant, antibacterial, and antifungal activities compared to chitosan (Divya et al., 2017; Avelelas et al., 2019). Compared to the use of chitosan, chitosan nanoparticles are more active and perform better, which is due to smaller particle size and increased nanoparticle contact area (Qi et al., 2004; Orellano et al., 2019). The size reduction of chitosan to a nanoscale can improve the functionality and properties at lower concentrations (Eshghi et al., 2014). The effective concentration of chitosan decreased significantly to 0.5% when used in nanoparticle form as compared to the higher amount suggested in previous studies for coating fruits (Esyanti et al., 2019). Chitosan alone or with other polymers has been used at a concentration as high as 2% on fruit to effectively preserve the quality of fruit but with the introduction of nanoparticles, a lower amount is required to effectively preserve the quality of fruits (Suseno et al., 2014; Khatri et al., 2020).

As the penetration and absorption of chitosan increase dramatically in the form of nanoparticles, the effective amount of chitosan used for coating fruits can be substantially or greatly reduced (Zahid et al., 2012). Jagana et al. (2017) studied the impact of nanosilver concentration with plant extract of neem on anthracnose diseases in banana fruit. The nanoparticle, applied using a spraying method, was able to control anthracnose disease in banana even at a low concentration of 0.2%. This was because nanoparticles were able to penetrate microbial cells effectively showing complete inhibition of spore germination of *Colletotrichum musae*. Edible coatings such as plant extracts may possess strong odour and flavor and may have a strong negative effect on the sensory properties of fruits thereby limiting their application. Silver nanoparticles, on the other hand, do not adversely affect the sensory characteristics of fruits and are more acceptable to consumers (Chandirika et al., 2018).



Consequently, the higher concentration of nanoparticles can cause physiological damage thereby changing the internal atmosphere of fruits, and increased chlorophyll degradation by enhancing fruit ripening (Zambrano-Zaragoza et al., 2013). Developing a nanoparticle can cost a lot of money, thereby it is advisable to synthesis the material via a green method by using plant extracts, which have a lower cost. Edible coatings such as alginate display low viscosity fluid when used with zinc nanoparticles leading to an inability of gas exchange between fruit and the environment. Chitosan on the other hand has shown synergy relations, emulsifying and crosslinking abilities when used with zinc (Arroyo et al., 2020). Therefore, some nanoparticles do not form a synergy effect with edible coatings, which might limit their application in postharvest treatment.

## **2.9 Safety concern and legislation for use of nanoparticle-enhanced edible coatings**

Nanoparticles are used in the fruit industry for a variety of reasons, one of which is their unique properties, which are associated with their small size. Small particles, for example, are digested faster, have a greater surface reactivity, and can more efficiently penetrate biological barriers than larger particles. Currently, there is insufficient legislation regarding the use of nanoparticles in fruits, and consumers view emerging innovations as posing a danger to their health and the environment. Legislative barriers and uncertainty about the effectiveness of such systems, as well as their economic and environmental impact, may be the primary reasons for this. While legislation is still in its early stages, it must discuss all aspects of nanotechnology's use in the fruit industry around the world. Only the European Commission (EU) member states of Sweden, France, Denmark, Belgium, and Switzerland have adopted their regulations for nanomaterials or nano-enabled goods at this time (Arts et al., 2015). Recent EU regulations mandated that any food ingredient derived from nanotechnological applications be subjected to a safety evaluation before being approved for use (Cubadda et al., 2013). Only a few nano-form substances have made it to commercial applications thus far, particularly in the EU. The US Food and Drug Administration (FDA) has approved the use of many forms of nanoparticles, including Ag and titanium NPs, in commercially available products like antibacterial

skin lotions and sunscreens. Nanoparticles have been used in the food industry for edible foods, providing some confidence that they can be used in fruits with a high degree of protection (Table 2.2). FresherLonger™, BagsFresherLonger™, Anson Nano Silver Fresh Containers, Nano Silver Food Container, Fresh Box Nano Silver Food Container, Miracle Food Storage, and Anson Nano Freshness-Keeping Film are silver nanoparticles commercially available for use in countries such as the United States, China, and South Korea, and they are used in packaging of food products. With the ever-increasing usage of nanoparticles on fruits on a commercial scale, it is critical to stress the value of performing short and long-term toxicity studies, both for the environment and for humans, to ensure consumer protection. Multiple factors influence the toxicity of NPs, including form, scale, surface charge, composition, and NP stability. The key danger is that nanoparticles used directly on fruit could migrate into the fruit product. More research demonstrating that the nano material does not migrate into the food matrix may help with regulatory and consumer acceptance. Also, the consequences of long-term ingestion of low yet regular concentrations or doses of nanoparticles on fruits, on the other hand, have yet to be investigated. Risk assessments, biosafety, and legislation for inorganic nanoparticles are still a work in progress that necessitates further study. This reality motivates researchers to continue their research and development efforts in the field of nanotechnology in order to secure a more accurate understanding of the materials' applications, risks, and benefits in fruits during the postharvest stage.

Future research should focus on potential human health consequences, as certain materials, such as TiO<sub>2</sub>, have been linked to colon cancer. Jing Deng et al. (2021) explores recent developments in the production of food nanoparticles as well as the possible threats and found that they pose a possible threat to the human gastrointestinal tract. To effectively avoid the potential risks of nanoparticle applications in the food industry, it is important to include a specific description that encompasses the unique properties of nanocomponents as well as the required application or limitations of nanomaterials for the related products. Finally, before nanomaterials can be commercialized, definitive and conclusive studies on their safety and environmental impact are needed (de Azevedo et al., 2018).

Table 2.2: Application of nanoparticles in the food industry

Nanoparticle	Application in the food industry	References
Zinc oxide	Active packaging for fresh orange juice	Emamifar et al. (2010)
Zinc oxide	Food additive	Perez et al. (2012)
Zinc oxide	Antimicrobial food packaging	Suo et al. (2017); Beak et al. (2017)
Zinc oxide	Antimicrobial agent	(Kim et al., 2020; McClements & Xiao, 2017).
Zinc oxide	Food packaging material	Espitia et al. (2016)
Zinc oxide	Food lining in packaging	Silvestre et al. (2011)
Silver	Antimicrobial agent in food packaging	Medina-Reyes et al. (2020)
Silver	Surface coatings for sweets	Medina-Reyes et al. (2020)
Silver	Antimicrobials in marine shrimp farming	Camacho-Jiménez et al. (2020)
Silver zeolite	Food preservation	Kawahara et al. (2000)
Silver	Antimicrobial packaging	Chaudhry et al. (2008)
Silver	Commercial food containers	Artiaga et al. (2015)
Silver	Commercial containers and bags	Ozaki et al. (2015)
Silver	Food storage and food packaging materials	
TiO <sub>2</sub>	Food colorant	Chen et al. (2017)
TiO <sub>2</sub>	Food additives	(EFSA, 2000; Weir et al., 2012)
Zinc oxide and silver	Nanocomposite packaging for chicken	Panea et al. (2014)
Chitosan	Cheese, meat, and fermented sausage production	Wang et al. (2004)
Chitosan	Glazing material for frozen shrimp	Solval et al. (2014)
Gold	Food additives	(EFSA Panel on Food Additives and Nutrient Sources added to Food, 2016)
Silicon dioxide	Anticaking and antifoaming agent in foodstuffs	(JECFA, 2016)

## 2.10 Conclusion and future trends

Climacteric fruits are a central component of the human diet, supplying important minerals and vitamins for human health. Efforts have been made to improve storage conditions of the fruits, monitor their susceptibility to disease after harvest, and

preserve their freshness to meet consumer demands. The acceptability of fruits by consumers depends on quality parameters such as colour, texture, absence of decay, and most importantly, the nutritional and health benefit it provides. Edible coatings driven by their low cost and non-toxic nature are among the well-studied natural polymers and their application has proven to be promising for fruit preservation. The application of nanoparticles appears to be highly promising in the field of postharvest storage for extending the shelf life of climacteric fruits. The current materials widely used for coating climacteric fruits are zinc oxide, silver, and chitosan nanoparticles because they show promising results in preserving the postharvest quality of fruits. This review has summarized that nanoparticle-enhanced edible coating applied to climacteric fruits can effectively improve their physical and sensory properties, inhibit the growth of microbes, and prolong the shelf life of fruits. When used singly, some edible coatings have shown unsatisfactory results in practical application, hence, their combination with nanoparticles helps to improve their physicochemical and biological properties.

There is a great potential to extend the use of other nanoparticles such as copper, cerium oxide, and titanium oxide as coating materials as they are less toxic. Also, food-grade nanomaterials such as starch, cellulose, and gums are edible and non-toxic and hence, present promising prospects for use in fruit coating. The combination of nanoparticle-enriched edible coating with the use of existing technologies such as low-temperature storage and controlled atmosphere storage is another great field of research. To understand the method of applying nanoparticle-enhanced edible coating, and their effect on sensory and nutritional properties of climacteric fruits, further research is crucial. Nonetheless, despite research emphasis on improving the appearance of fruit, there is still a lot to be done towards improving the organoleptic properties and nutritive values through the reasonable application of food-grade materials to synthesize nanoparticles. Again, further investigation is necessary regarding the behaviour of these materials after ingestion and maximum allowable amounts of the nanoparticles that may be present in fruits to create a healthy nanoparticle that could be used for commercial products.

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## 2.12 Declaration of competing interest

Conflict of Interest: The authors declare that they do not have any conflict of interest.

Ethical Review: This study does not involve any human or animal testing.

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## CHAPTER 3: EFFECT OF SELECTED EDIBLE PLANT EXTRACT-BASED COATINGS ON THE SHELF LIFE OF CAVENDISH BANANAS

### 3.1 Abstract

The primary purpose of postharvest management is to lengthen the shelf life of fruits while retaining their quality. The use of an edible coating to extend the shelf life of fruit is a technique that can be applied. This study evaluated the effect of edible coatings of moringa and *Aloe vera* extracts on the total soluble solids (TSS), weight loss, firmness, decay rate, total phenolics, mineral content and shelf life of bananas. The bananas were immersed in a coating solution containing *Aloe vera* 50% (AV) and *Moringa oleifera* 10% (MO). A 24-day postharvest experiment with 4-day sampling intervals was conducted to examine the effect of *Aloe vera* and moringa coatings on the storability of banana fruits at room temperature ( $24 \pm 1^\circ\text{C}$ ). Both coatings had a significant ( $p < 0.05$ ) impact on firmness, total soluble solids, total phenolic and mineral analysis but a non-significant effect on weight loss during storage. *Aloe vera* coatings slowed weight loss, total phenolic loss, firmness loss, and TSS rise more than moringa coated and control samples. Moringa coated fruits also had a significantly higher mineral content value (Zn, Cu, Mn, Mg, Al, Ca, and K) than *Aloe vera* coated fruits and control group. Furthermore, the *Aloe vera* and moringa coatings increased the shelf life of bananas by up to 24 days whereas uncoated fruit had shelf life of 20 days. *Aloe vera* and moringa coatings, based on these findings, might be an efficient way to improve the shelf life of bananas.

**Keywords:** *Aloe vera*, *moringa*, *total phenolics*, *postharvest quality*

### 3.2 Introduction

Banana (*Musa* spp.) belonging to the *Musaceae* family is widely grown in tropical and subtropical regions of Africa, Latin America, and Asia (Pratiwi et al., 2015). Banana is among the most important tropical fruits grown in South Africa. In the world, the annual production of the crop is around 114 million metric tons from an area of 5.6 million ha (FAO, 2018). It is the world's fifth most important agricultural food crop in terms of global trade, after coffee, cereals, sugar, and cacao. Aside from grapes, citrus fruits, and apples, it is one of the world most important fruit crop (Aurore et al., 2009). It is a

good source of minerals such as potassium, calcium, phosphorus, and magnesium (Foster et al., 2003). Bananas are a key source of health-promoting phytochemicals, according to several studies (Someya et al., 2002; Davey et al., 2007; Singh et al., 2016). They have a short shelf life at room temperature and are extremely vulnerable to low temperatures (below  $10 \pm 1^\circ\text{C}$ ), which leads to high postharvest loss (Al-Qurashi et al. 2017). The banana fruit has a quick ripening period, which possess a post-harvest challenge for the banana industry around the world, particularly in areas where refrigeration is not available (Peroni-Okita et al., 2013). Banana fruits are usually harvested at physiological ripe stage and remain fresh without major changes in peel colour, texture, or composition before the beginning of ripening (Aguilera et al., 2003; Prabha & Kumar, 2015). After harvesting, the fruits undergo progressive deterioration, resulting in a relatively short post-harvest life, increased respiratory rate, autocatalytic ethylene development, increase in susceptibility to various pathogenic infections, and sensory changes such as colour, taste, and softening (Palapol et al., 2009; Paul et al., 2012). The occurrence of postharvest changes is a natural ripening process. However, this ripening can be slowed down within certain limits by harvesting at an appropriate maturity stage, controlling storage temperatures, and through ethylene regulation. Several technologies have been tested to slow down banana ripening and extend shelf life. These include storage in a controlled atmosphere (Ahmad et al., 2006), low temperature (Facundo et al., 2015), modified atmosphere packaging (Workneh et al., 2009), as well as nitrous oxide (Palomer et al., 2005), and 1-methylcyclopropene treatment (De Martino et al., 2007). In certain cases, problems like chilling injury, excessive ripening, and the high capital costs of certain improved storage technologies restrict the implementation of these applications.

On the other hand, the use of natural extracts such as edible coatings have been gaining popularity in the fruit industry due to their increased efficacy, reduced cost, ease of application, and environmental benefits (Malmiri et al., 2011). Secondary metabolites in natural plant extracts, such as phenols, flavonoids, alkaloids, and terpenoids, have been used to prolong postharvest life and thus extend the shelf life of fruits (Daniel & Krishnakumari, 2015). There are few studies on the use of edible coatings in bananas, most of which have been linked to postharvest uses and shelf-life extension (Maqbool et al., 2011; Razak & Lazim, 2015; Alali et al., 2018; Majeed et al., 2019). The edible coatings consist of a thin layer of edible material that serves

as a barrier to control moisture loss and gas exchange (CO<sub>2</sub> and O<sub>2</sub>) between the fruit and their surrounding environment, thereby slowing down the rate of respiration, retarding the physiological ripening process, and preventing the loss of natural volatile flavour compounds (Rojas-Grau et al., 2009; Pratiwi et al., 2015). Edible coatings are prepared from polymers such as polysaccharides, proteins, and lipids (Fakhouri et al., 2015). Edible coatings can be applied over the food product in liquid form by spraying, extrusion, solvent casting, brushing, or dipping to achieve a thin protective layer (Yousuf & Singh, 2018; Thakur et al., 2019). The edible coating can be formulated from extracts derived from both plant and animal sources. The use of animal extracts has been limited due to allergies associated with the consumption of animal products, religious concerns, and the formation of impermeable layers like wax. Plant edible coatings are usually obtained from components regarded as safe for human consumption such as fruits, herbs, or spices with high medicinal benefits (Flores-López et al., 2016). A review by Ncama et al. (2018) gave a comprehensive report on the use of such coatings on fresh produce. Plant extracts containing high antioxidant properties, such as those derived from *Moringa oleifera* (MO), gum arabic, *Aloe vera*, and green tea, have gained popularity in maintaining the shelf life quality of fruits. Corn starch (Razak & Lazim, 2015), *Aloe vera* (Khaliq et al., 2019; Majeed et al., 2019), gum arabic (Maqbool et al., 2011), and gum ghatti (Joshi et al., 2018) have been tested on banana and showed promising results.

*Aloe vera* has been gaining more attention in the banana industry due to its gel-forming abilities, high stability, and biochemical properties (Misir et al., 2014). *Aloe vera* gel is rich in bioactive compounds and can form a protective barrier against moisture loss, reducing respiration rate, weight loss, softening, colour changes, and ethylene generation, due to its antimicrobial and antioxidant properties (Marpudi et al., 2011; Vieira et al., 2016; Mendy et al., 2019). *Aloe vera* does not pose any known risks to human health; it is cost-effective and readily available in bulk quantities. Overall, the use of *Aloe vera* coating resulted in the preservation of quality and the extension of shelf-life of several fruits such as kiwi fruit (Benitez et al., 2013), tomato (Chauhan et al., 2015), mango (Chauhan et al., 2014), raspberry (Hassanpour 2015), and strawberry (Sogvar et al. 2016). Moringa plant extracts, including from leaf, bark, sap, root, flower, and seed, have antibacterial and antioxidant properties due to high levels of phenolics, vitamins, and carotenoids (Tsfay et al., 2011; Busani et al.,

2012; Tesfay et al., 2016). Moringa has long been known to help preserve perishable foods by preventing dehydration, inhibiting respiration, enhancing textural quality, retaining volatile taste components, and inhibiting microbiological development (Yousef et al., 2015). When compared to aqueous extracts from MO seeds and fruits, MO leaves contain a larger amount of phenolics and total flavonoids, as well as strong inhibitory action (Singh et al. 2009). Furthermore, prior research has found that many MO leaf extracts contain a wealth of phytochemicals such as tannins, saponins, alkaloids, terpenoids, and carotenoids, all of which play an important role in bacterial eradication (Doughari et al., 2007; Nweze & Nwafor 2014; Abdulkadir et al. 2015). Moringa leaf extract as an edible coating has been shown to improve the postharvest quality of fruits such as avocado, mango, and tomato (Tesfay et al., 2017; Abd El-Razek et al., 2019; Kator et al., 2019).

Although the benefits of plant extract coatings have been reported, there is little information on how they might be used to coat fruits to extend their shelf life and improve the postharvest quality of bananas. However, to the best of our knowledge, there is limited information on moringa coating application on Cavendish bananas. To date, little has been known about moringa extract as a fruit coating on Cavendish banana. The application and effectiveness of an edible coating produced from aloe gel and moringa on banana fruits to improve their shelf life and marketability have been described in this paper. According to the literature survey by Odetayo et al. (2021), no studies evaluating the effects of *Aloe vera* and moringa as an edible covering on the shelf life and quality of banana fruit have been conducted. Therefore, this study aimed to evaluate the effect of *Aloe vera* and moringa extract on the postharvest life of banana fruits.

### **3.3 Materials and Methods**

#### **3.3.1 Materials**

A total of 10 boxes of green banana (Cavendish subgroup) fruit were purchased from farmers at Dortannion farm Mbombela, Mpumalanga 25°28'12.4"S 30°58'41.1"E. The fruit was transported to the Botany Laboratory at the University of Johannesburg. The selected bananas (1080 fingers) were small to medium in size with an average weight ranging from 70 to 140 g. Any bruised or diseased fruit were discarded, and the remaining fruits were cleaned using a moist soft paper towel to get rid of dirt particles

on the surface. The bananas used in the experiment were at ripening stage A1 according to the colour chart index in Fig 1 (Freshmark, South Africa). They were stored at room temperature for three days until they reached stage A2 banana (light green with a tinge of yellow) before the coating was applied (Freshmark, South Africa). Fresh leaves of *Aloe vera* were harvested from Johannesburg Botanical Garden (26°08'60.00"S 28°00'0.00"E). *Moringa oleifera* leaves were harvested from the Agricultural Research Council in Mbombela (25°27'04.06"S 30°58'09.01"E). All chemicals were purchased from Sigma-Aldrich (South Africa).

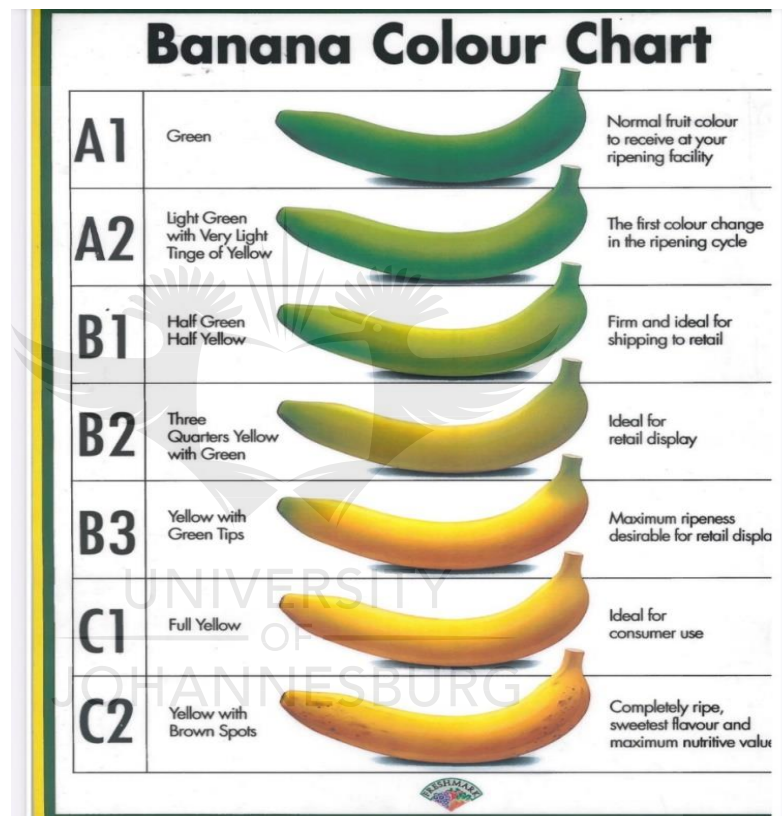


Figure 3.1: Banana colour chart (Freshmark, South Africa)

### 3.3.2 Methods

#### 3.3.2.1 Experimental design

Having attained stage A2, the banana fruits were then assigned to three postharvest treatments: control (untreated), *Aloe vera* (50% v/v) and moringa (10% v/v). Each treatment comprised four replicates, each with 90 fruits. The fruit was dipped in treatment solutions for 1 min before air-drying for 10–15 min on a laboratory bench at room temperature (24 ± 1°C). The concentration of *Aloe vera* and moringa were

selected based on previous studies by (Abd El-Gawad et al., 2019; Ali et al 2019; Abd El-Razek et al., 2019) where the authors used 50% *Aloe vera* and 10% moringa leaf extracts on fruits such as mango and litchi.

### **3.3.3 Preparation of coatings**

#### **3.3.3.1 *Aloe vera* coatings**

A mechanical procedure was used to obtain the pulp (gel) and liquid fractions according to Khaliq et al. (2019) with the modifications of adding ascorbic and citric acid. Firstly, the leaves of *Aloe vera* were washed with distilled water to remove dirt. Then, aloin (a yellow-coloured liquid) was extracted by cutting the base of the leaves. The skin was carefully separated from the colourless parenchyma gel using a knife, and the gel was then separated from the epidermis using a spoon. The gel was homogenized with a blender for 6 min. The mixture obtained was filtered using a muslin cloth to remove fibrous fractions. Ascorbic acid (1% v/v) and citric acid (1% v/v) were added to prevent browning and to maintain the pH. *Aloe vera* gel was diluted with distilled water to make (50:50 v/v) and 1% (v/v) glycerol was added as a plasticizer.

#### **3.3.3.2 *Moringa oleifera* coatings**

A mechanical procedure was used to obtain the liquid extract according to Hafez et al. (2019) with the modification of adding glycerol. The leaves of *Moringa oleifera* were removed from the stalk and stem. Firstly, the leaves were washed with distilled water to remove dirt and, afterward, spread on a clean paper towel. The yellow and black coloured leaves were discarded, and green leaves were air-dried on the laboratory bench. After drying they were placed in a zip bag and stored in a freezer. The leaves were freeze-dried for 24 h and later ground to a powder using a coffee grinder. After grinding, 50 g of leaf powder was diluted with 500 mL of distilled water in a beaker. The mixture was heated to about 80°C in a water bath for 10 min. The mixture was then cooled to room temperature. A sieve (100 µm) was used to filter the extract. The moringa extract was stored at 4°C. The extract was diluted with distilled water (10:90 v/v) to produce the edible banana coating. The moringa coating concentration (10% v/v) was selected according to previous studies and 1% (v/v) glycerol was added as a plasticizer.

### 3.3.3.3 Application of coatings and storage

Three treatments were used in the experiment, namely *Aloe vera* gel 50% (v/v), *Moringa oleifera* 10% (v/v), and the control (untreated). Glycerol 1% (v/v) was added to both treatments, except the control, as a plasticizer. Banana fingers were dipped in a coating solution for 1 min and allowed to air dry for 10-15 min at room temperature to allow a thin layer to be formed on the surface of the fruit. Storage temperature and relative humidity were set at room temperature ( $24 \pm 1^\circ\text{C}$ ) and 54-56%, respectively, in a storage chamber. The effectiveness of the coating on bananas was determined by measuring weight loss, decay rate, soluble solids, and firmness, among other quality parameters. Experimental data were recorded every 4 days for up to 24 days of storage. Temperature and relative humidity were collected with a Hobo data logger (Thermochron, USA) during shelf-life investigations.

### 3.3.4 Quality parameters

#### 3.3.4.1 Weight loss

At the start of the experiment, ten fruits from each replicate were marked and kept separate for periodic weighing using a digital balance (Delta range, Switzerland). Following treatment (day 0), and at various intervals (days 4, 8, 12, 16, 20, and 24) during storage, the weight of individual lots was recorded. The percentage loss of initial weight was used to calculate cumulative weight losses. Total weight loss was calculated as the difference between the initial and final fruit weights after 24 days of storage (Sharmin et al., 2015). Weight loss was determined by taking the initial and final weights differences and expressed as a percentage

$$\text{Percentage weight loss} = \left( \frac{W_i - W_f}{W_i} \times 100 \right)$$

Where  $W_i$  = initial fruit weight

$W_f$  = final weight loss

#### 3.3.4.2 Firmness

Fruit flesh firmness was measured with an 8 mm round stainless-steel probe, attached to a fruit pressure tester (Mod.el FT 327, Effegi, Italy). With the aid of a cutting blade, a thin section of the epicarp was removed (the thickness did not exceed 1 mm). The

measurements were taken on the pulp of the bananas at two different locations in their equatorial region. The mean of the measurements from each fruit sample was used to calculate the values in kilograms (kg), which were then converted to Newtons (N).

#### **3.3.4.3 Total soluble solids (TSS)**

The refractive index of fruit juice was measured at 20°C with a hand refractometer (Atago, Japan) to determine the soluble solid. The refractometer prism was washed and calibrated with distilled water to give a 0% reading before each sample was analyzed for the estimation of sucrose expressed in TSS. The banana pulp (50 g) with 150 mL of distilled water was homogenized using a blender to get a filtrate. To obtain the % TSS reading directly from the instrument, a drop of banana juice squeezed from the fruit pulp of each fruit of the three replications of each treatment was mounted on the prism glass of the refractometer. Brix was used to represent the findings (AOAC, 2016; Lopez-Palestina, 2018).

#### **3.3.4.4 Determination of total phenolic content**

Total phenolics (TP) were analyzed according to Murmu and Mishra (2018) using the Folin–Ciocalteu reagent (FCR) method with slight modification. Freeze dried banana samples (0.5 g) were homogenized in a glass tube with 10 mL of 80% methanol using a magnetic stirrer. They were centrifuged at 25°C for 30 min at 2500 g. The supernatant was collected and used for the analysis of total phenols. The methanol extracts (0.5 mL) were mixed with 5 mL of deionized water and 0.5 mL of the Folin–Ciocalteu reagent (Sigma Aldrich, USA) was added. After 10 min, 1.5 mL of saturated 20%(w/v) aqueous sodium carbonate solution was added to the mixture. The mixture was mixed rigorously and allowed to stand for 30 min at room temperature. The absorbance was measured at 760 nm of the reaction mixtures against the blank (methanol) using a spectrophotometer (WPA, Biochrom England). The content of phenolics in the extracts was expressed as mg of gallic acid equivalent per 100 g of the dry weight (mg GAE/g DW).

#### **3.3.4.5 Mineral analysis and protein content**

The mineral composition was determined according to Ngobese & Workneh, (2018) with slight modifications. For analyses, 0.5 g sample aliquots were burnt to ash in a furnace overnight at 450°C and placed in 100 mL pre-weighed glass beakers. The ash



was moistened with a few drops of distilled water before adding 2 mL of concentrated HCl to each sample. The samples were evaporated slowly to dryness on a water bath in the fume cupboard with the extractor fan on. Thereafter, 25 mL of freshly prepared 1:9 HCl solution and distilled water was added into each sample and the mixtures were filtered using Advantec 5B:90 mm diameter filter paper. The filtrates were diluted with deionized water to a 5:20 dilution ratio and analyzed for phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sodium (Na), zinc (Zn), copper (Cu), manganese (Mn), iron (Fe), nitrogen (N) and aluminum (Al), using an inductively coupled plasma optical emission spectrometer Vista-MPX 2004 (Varian, Mulgrave, Australia). Analysis was done against known standard solutions of the elements (Separations, South Africa) and a blank of deionized water. The mineral element content was expressed as mg 100 g<sup>-1</sup> of DM. The crude protein composition was calculated by multiplying the total nitrogen composition (as measured by ICP–OES mineral analysis) by the conversion factor (6.25) to get the protein fraction.

#### **3.3.4.6 Shelf life**

Shelf life was assessed by conducting a visual observation. In this present study, the shelf life of bananas was graded daily on a scale of 0 to 100% for visual yellowing of peel to brown colour. When bananas showed more than 80% browning, they were deemed to have reached the end of their shelf life (Hassan & Mahfouz, 2010)

#### **3.3.4.7 Statistical analysis**

To establish the significant difference between the treated and control fruits on the same day of storage, a one-way analysis of variance (ANOVA) and Pearson correlation coefficient between the parameters and treatment was done using IBM SPSS statistical software (SPSS v 27). At  $p < 0.05$ , multiple comparisons were done using the Turkey Post hoc test to separate the means.

### **3.4 Results and Discussion**

#### **3.4.1 Weight loss**

Weight loss after harvest affects the quality and value of fresh produce, lowering its quality and value (Silva et al., 2017). The amount of weight loss is a good measure of how effective the coatings were at preserving bananas. Weight loss began after

storage and continued during the storage period. Weight loss was observed in both coated and control bananas in the current report (Fig 3.2), with coated fruits losing more weight than the control from 4 to 16 days. Control fruits only started showing more weight loss than coated fruits from storage at day 20 to 24. Weight loss continued during the storage period, with the control fruit losing 23% of its weight at day 20, which was 1.39% more than the coated fruit for the same time (Fig 3.2), indicating a loss of marketability. *Aloe vera* coated fruit (25%) showed less loss of weight compared to moringa coated fruit (26%) and the control (27%) after 24 days of storage. This might be because edible coating creates a barrier on the fruit surface that reduces moisture loss from fresh products, preserving the quality and fresh appearance of fruits according to Silva et al. (2017). At the end of day 24, the coated fruit and uncoated had turned brown and lost a considerable amount of marketability, so storage day effects on the fruit were not investigated further. Other fruits, such as guava (Silva et al., 2018) and avocado (Tesfay et al., 2017) have also been shown to benefit from edible coatings in terms of weight loss reduction as compared to the control.

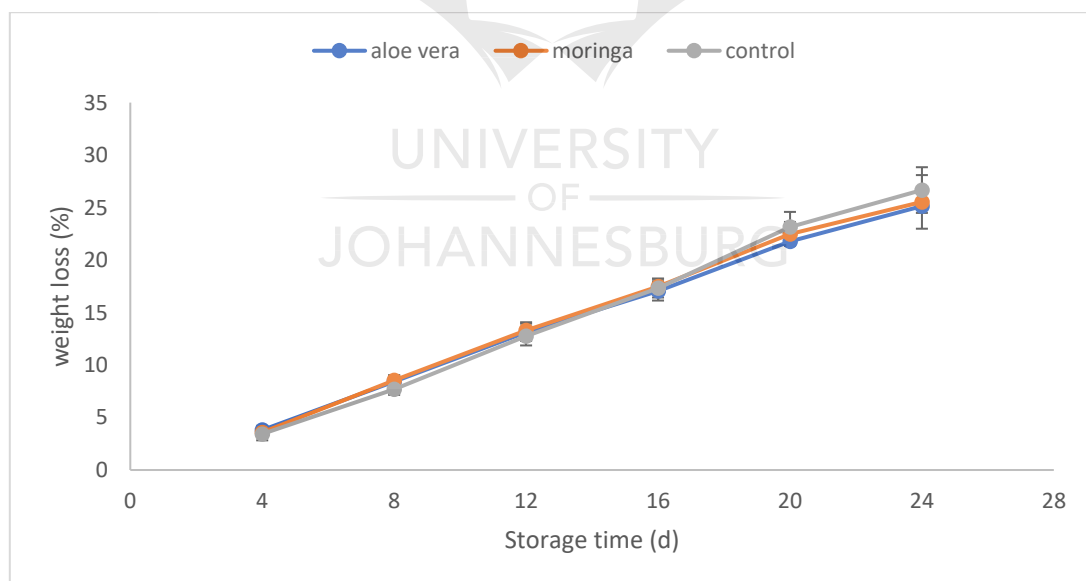


Figure 3.2: The effect of *Aloe vera* gel, moringa leaf extract and control on weight loss of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.2 Firmness

Fruit firmness is an important factor in determining the fruit's postharvest life and quality. Bananas soften during storage, contributing significantly to pathogen invasion, dehydration due to water loss, and cell wall degradation (Deng et al., 2017). In line with previous studies, the firmness of banana fruits decreased during storage (Stanley et al., 2013; Fan et al., 2018). During the ripening process, both the control and coated fruit lost firmness consistently and gradually as shown in Fig 3.3. The firmness of the uncoated banana decreased significantly ( $p < 0.05$ ) from 47 N on day 0 to less than 35 N on day 24 while the coated banana with *Aloe vera* (50%) and moringa (10%) coatings decreased significantly ( $p < 0.05$ ) from 52 N to 46 N and from 49 N to 45N respectively, after 24 days of storage. The findings in Fig 3.3 show that treating bananas with *Aloe vera* and moringa plant extracts reduced the firmness loss of the fruits significantly ( $p < 0.05$ ) from day 16-24 days. The results revealed that *Aloe vera* gel and moringa coating made a significant positive difference ( $p < 0.05$ ) compared to the control on the firmness of the fruits at day 4, 12, 16, 20 and 24 days, with *Aloe vera* showing a significant higher firmness ( $p < 0.05$ ) from moringa at day 8, 20, and 24. The softening of banana flesh for control fruits was connected to the action of cell wall degrading enzymes, which hydrolyse starch to soluble sugars and protopectin to water-soluble pectin. Better firmness preservation in coated fruit suggested that *Aloe vera* and moringa coating were successful in slowing down metabolic and enzymatic activities in fruit, which resulted in slower pulp tissue degradation. The findings in the study were similar to those obtained by Hazrati et al. (2017), Khaliq, et al. (2019), Thakur et al. (2019), who treated papaya and bananas fruit with plant extract coatings, and they found better firmness consistency than those that were not.

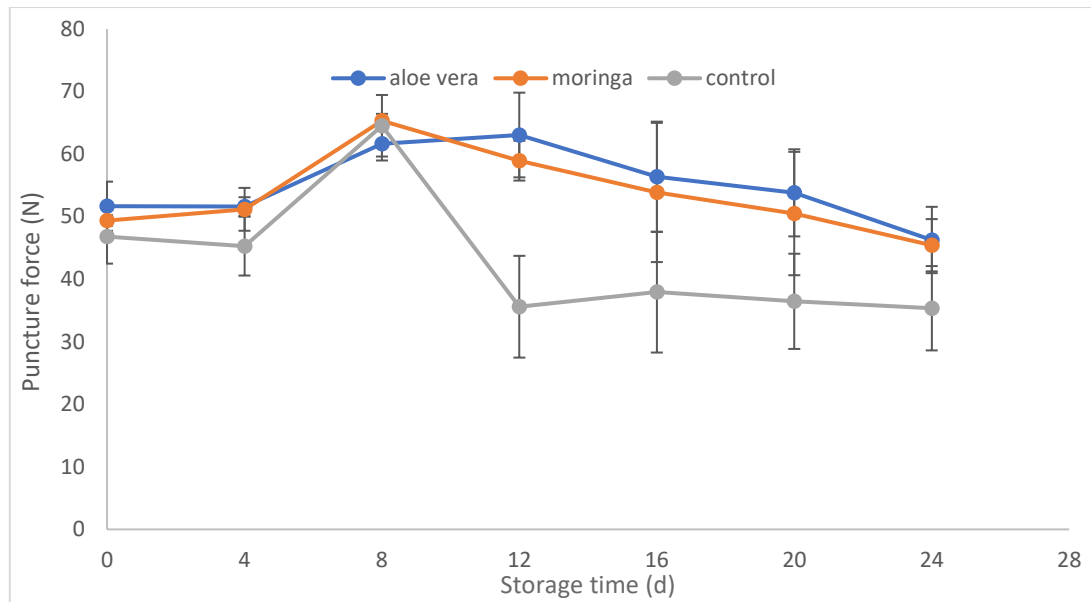


Figure 3.3: The effect of *Aloe vera* gel, moringa leaf extract and control on firmness of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.3 Total soluble solids (TSS)

Total soluble solids are an important quality metric that indicates a fruit's maturity level. Because the amount of TSS in the fruit rises as it matures and ripens, the soluble solids content of the fruit can be used as a useful indicator of the maturity or ripeness stage (Ngnambala, 2013). Between day 0 and day 24, the soluble solid content of untreated and coated banana fruits increased considerably (Fig 3.4). During the 24 days of storage, the soluble solid content of the bananas was measured. The analyses were carried out on day 0 of the experiment, as well as days 4, 8, 12, 16, 20, and 24 of storage. TSS concentrations gradually rose as storage time progressed. Significant changes ( $p < 0.05$ ) were also detected in connection to the TSS of banana fruits at different days of storage due to the different treatments in the current study. In comparison to treated banana fruits, the largest increase in TSS was observed in the control treatment from days 8 to 24 (Fig 3.4), while the coated sample revealed only a lower increase in total soluble solid content. The ripening and senescence of the fruit cause an increase in soluble solid content. Thus, the rise in TSS concentration in control fruits could be related to the breakdown of starch into soluble sugar components (Arthey & Philip, 2005; Ali et al., 2010). Furthermore, the findings of Dave

et al. (2017) for pear fruit provided a similar reason for the increase in TSS during storage. After 24 days of storage, *Aloe vera* coating (15 °Brix) significantly ( $p < 0.05$ ) reduced the increase in TSS concentration compared to moringa coated fruits (17 °Brix) and the control (23 °Brix) among the three treatments. Edible coatings are thought to protect fruits from drastic changes in TSS by limiting gas exchange, decreasing respiration, and inhibiting metabolic processes in the coated fruits (Dong & Wang, 2018). The results of the various treatments on total soluble solids revealed that *Aloe vera* gel and moringa coatings had a noticeable effect on TSS shifts, with the fruits coated with *Aloe vera* gel and moringa showing a small decrease in TSS compared to the control treatment (Fig 3.4). The lower metabolic activities of the coated fruits could have been due to regulated gaseous exchange, which may have caused the TSS delay in coated fruit. Previous research on banana and papaya fruit coated with plant extracts such as *Aloe vera* found similar findings in terms of a lower rise in TSS compared to the control (Soradech et al., 2017; Khaliq et al., 2019; Parven et al., 2020).

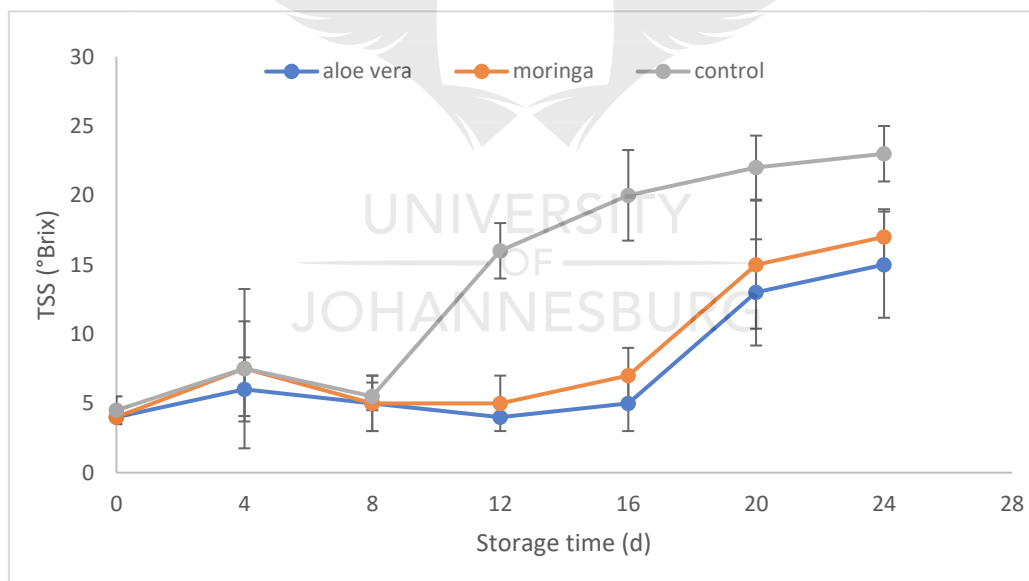


Figure 3.4: The effect of *Aloe vera* gel, moringa leaf extract and control on total soluble solid of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

#### 3.4.4 Total phenolic content (TPC)

The phenol level of the fruit gradually decreased over the first eight days of storage. After that, it dropped sharply, reaching a minimum at the end of the storage period. The drop in phenolic compounds at the end of storage could be attributable to the disintegration of cell structure as a result of fruit senescence (Macheix et al., 1990; Palafox-Carlos et al., 2012). There was no significant difference between coated and uncoated fruits at the start of the storage period ( $p > 0.05$ ). However, when compared to uncoated and moringa coated fruits, *Aloe vera* maintained significantly ( $p < 0.05$ ) higher TPC at day 16 and 24 days of storage. Fig 3.5 shows the total phenolic content of banana fruit as a function of coatings and storage period. The total phenolic content of the coated and control bananas reduced over the storage period, while the moringa coated fruits exhibited a significant ( $P < 0.05$ ) larger drop or decrease. During storage for 24 days, fruit coated with *Aloe vera* (188 mg GAE/g DW) had higher phenolic content than moringa (164 mg GAE/g DW) and control fruit (180 mg GAE/g DW). Wang and Gao (2013) found similar outcomes in strawberry fruit treated with chitosan coating. However, *Aloe vera* had greater total phenol levels overall, but MO had no discernible impacts. These results could be attributable to the edible coating's general preservation effects, which acted as a barrier to gaseous exchange, reducing oxidation processes and delaying fruit senescence (Maqbool et al., 2011; Alali et al., 2018). These findings largely corroborate those of Alali et al. (2018), who found that gum arabic treatments retained higher levels of total phenols during aging in bananas.

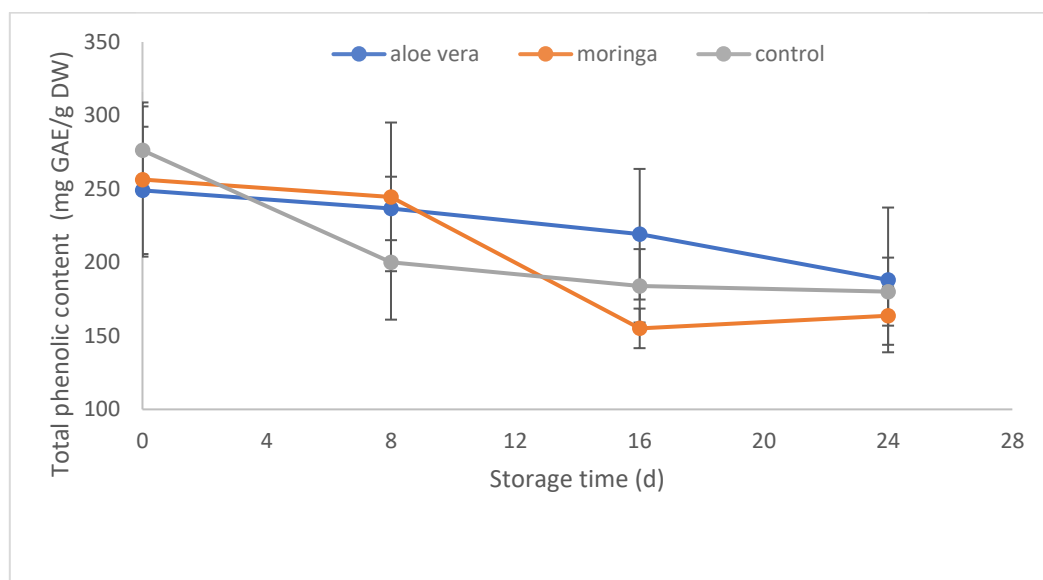
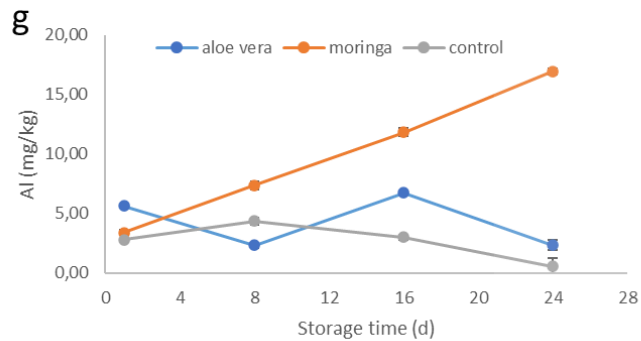
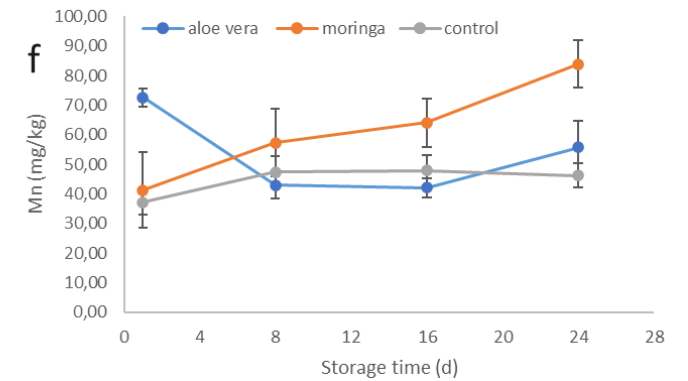
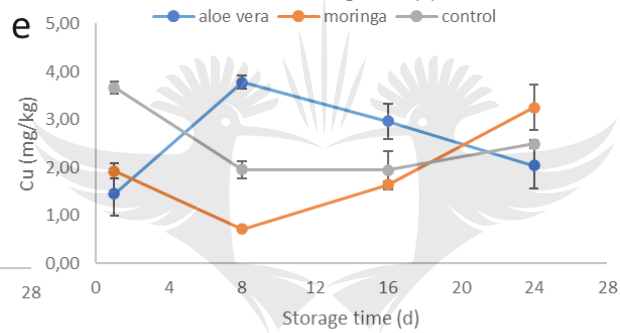
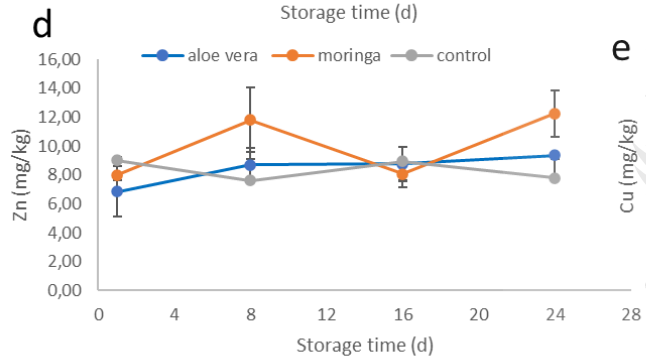
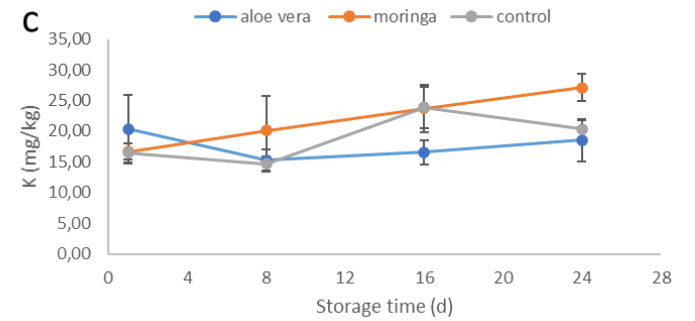
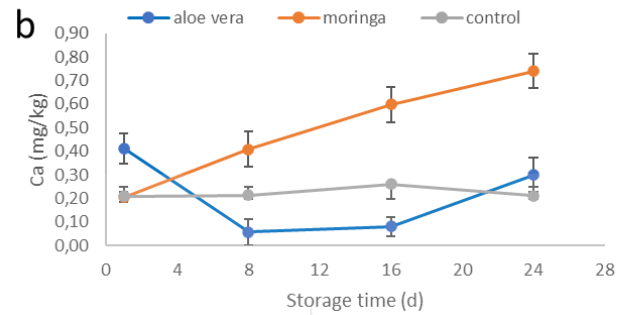
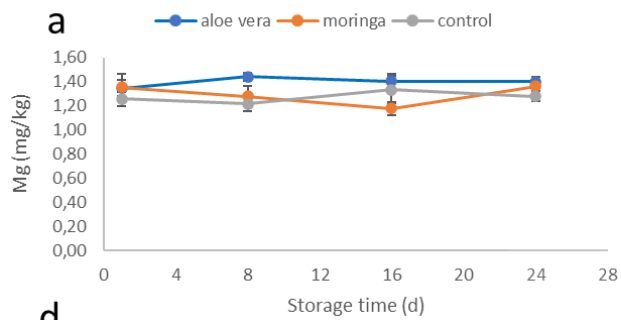


Figure 3.5: The effect of *Aloe vera* gel, moringa leaf extract and control on total phenolic content of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.5 Mineral analysis

Bananas are high in mineral content and are a healthy source in the diet (Qamar & Shaikh, 2018). Banana cultivars can supply macro and micronutrients in the diet (Wall, 2006). Different coating treatments had a significant effect ( $p < 0.05$ ) on the mineral composition (Ca, Cu, Mg, Mn, Zn, Al, and K) of banana pulp (Fig 3.6). The use of *Aloe vera* and moringa plant extracts significantly ( $p < 0.05$ ) retained the mineral content during storage at room temperature. Moringa was significantly greater in Zn, Cu, Mn, Mg Al, Ca, and K mineral content compared to *Aloe vera* and moringa at days 1,8, 16 and 24 of storage. The mineral content of moringa was Zn (12.25 mg/kg), Cu(3.25 mg/kg) , Mn (83.90 mg/kg) Al (16.95 mg/kg), Mg (1.4 mg/kg), Ca (0.74 mg/kg), and K (27.1 mg/kg) compared to *Aloe vera* at Zn (9.37 mg/kg), Cu (2.04 mg/kg) , Mn (55.75 mg/kg) , Al(2.34 mg/kg), Mg (1.4 mg/kg), Ca (0.30 mg/kg), and K (18.5 mg/kg) and control was Zn (7.78 mg/kg), Cu (2.49 mg/kg) , Mn (46.17 mg/kg), Al(0.56mg/kg), Mg (1.3 mg/kg), Ca (0.21 mg/kg), and K (20.4 mg/kg ) at 24 days. Overall, moringa extract was able to retain the mineral content of the banana after 24 days of storage. Banana treated with moringa extract coatings were rich in Mn, followed by, Al, and Zn according to this study.



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Figure 3.6: The effect of *Aloe vera* gel, moringa leaf extract and control on mineral content of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.6 Protein content

*Aloe vera* and moringa coatings had a significant effect on changes in the protein content of the bananas. Protein content for the control and *Aloe vera* coated samples decreased from 5.39% to 5.15% and 5.71% to 5.06% from day 1 to 24 respectively. Protein content of moringa coated fruits increased from 5.69% to 6.15% from day 1 to 24 of storage. At the end of storage day 24, moringa coated fruit has a significantly ( $p < 0.05$ ) higher protein content compared to *Aloe vera* coated samples and the control (Fig 3.7). Ariyo et al. (2021) recorded higher protein content of Cavendish banana of 3.78% when the banana was left to ripen naturally compared to when it was ripened artificially. Our findings were similar to Islam et al. (2018) who used edible coating of chitosan and guava leaf extract on banana, carambola, and tomato at room temperature. The crude protein content of the samples was however lower when compared to other protein-rich foods including beans, gourd seeds, and soybeans, which have crude protein levels ranging from 23 to 34% (Comai et al., 2011). This shows that to compensate for the protein shortage in bananas, consumers should take protein-rich meals.

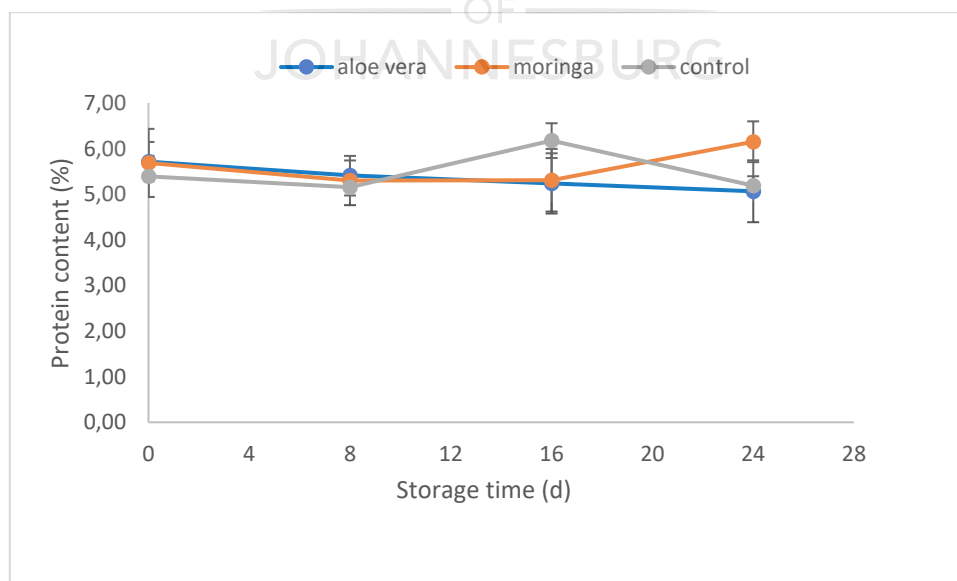


Figure 3.7: The effect of *Aloe vera* gel, moringa leaf extract and control on protein content of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.7 Shelf life

A basic parameter used to assess fruit quality is shelf life, which begins with the harvesting of fruit and ends with the beginning of fruit decay. Banana coated with *Aloe vera* gel and moringa extracts had the longest shelf life of 24 days, while the control had the shortest shelf life (20 days). From the results in Fig 3.8, it is obvious that coated fruits were preserved better in storage than the control fruits. The results showed that the use of plant edible coatings extended the shelf life of a coated banana. The result of this study agrees with those of Parven et al. (2020) who found that *Aloe vera*-coated papaya fruits had a longer shelf life than control fruits. The current findings also matched those of Eshetu et al. (2019), who found that applying an edible coating to mango fruits increased their shelf life.

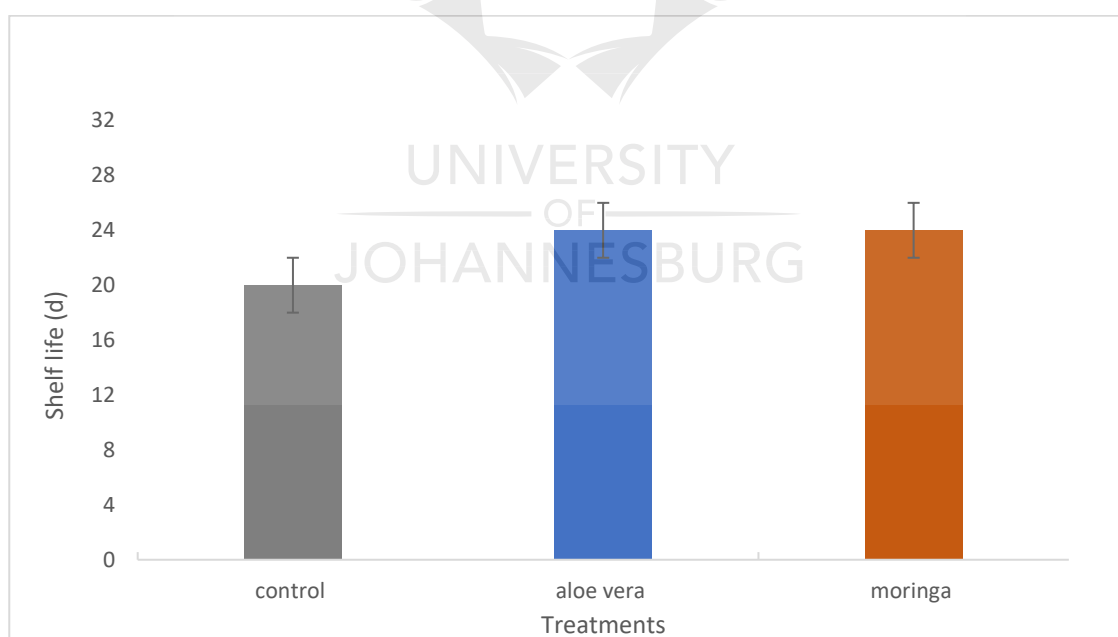


Figure 3.8: The effect of *Aloe vera* gel, moringa leaf extract and control on the shelf life of bananas stored at  $24 \pm 1^\circ\text{C}$  for 24 days. Error bars represent standard error (SE) at  $n = 4$

### 3.4.8 Pearson correlation

Table 3.1 shows the correlation coefficients that describe the relationship between treatment (*Aloe vera*, moringa and control) and quality parameters such as weight loss, firmness, and total soluble solids. *Aloe vera* coated fruit showed a strong correlation ( $r = 0.792$ ) between firmness and total soluble solids while moringa coated fruit showed a strong correlation value ( $r = 0.719$ ) between weight loss and total soluble solids. Regarding the control fruits, there was a strong correlation ( $r = 0.905$ ) between firmness and weight loss. Higher retention of firmness in coated samples could be due to the greater effect of hydrophobic properties of the barrier to moisture loss and outstanding antimicrobial activity in limiting the cell wall degrading enzymes (Qu, et al., 2020). This result was also consistent with the negative strong correlation ( $r = -0.781$ ,  $r = -0.905$ ) between firmness and weight loss for both moringa and *Aloe vera* coated bananas (Table 3.1). This finding is in line with Mohammadi et al. (2020) and Kubheka et al. (2019), who found correlation between peach and avocado fruit weight loss and firmness.

Table 3.1 Correlation coefficients between quality parameters of bananas stored at  $24 \pm 1^\circ\text{C}$

		WL	FM	TSS
<i>Aloe vera</i>	WL	1		
	FM	-0.781	1	
	TSS	-0.712	0.792	1
Moringa	WL	1		
	FM	-0.905	1	
	TSS	0.719	-0.472	1
Control	WL	1		
	FM	0.905	1	
	TSS	0.114	-0.038	1

WL = Weight loss, FM = Firmness, TSS = Total soluble solids.

### 3.5 Conclusion

The use of *Aloe vera* and moringa as coating agents on Cavendish bananas could help to slow down the ripening process and maintain quality. The application of *Aloe vera* and moringa coatings to banana fruits has a shelf life of up to 24 days compared to the control which had shelf life of 20 days. This means the coating only extended the shelf life of banana fruits by 4 days. According to the findings, *Aloe vera* 50% was the

most efficient treatment for decreasing weight loss, firmness loss, total phenolic and TSS increase. Moringa coatings, on the other hand, showed promise in retaining the protein content and mineral content of banana fruit. Overall, plant extract coatings containing 50% *Aloe vera* were shown to be the most effective coating for maintaining banana quality throughout storage. This research suggests that *Aloe vera* and moringa edible coatings might be beneficial in the banana sector for preserving postharvest quality and increasing shelf-life. However, more research is needed to improve the barrier properties of *Aloe vera* and moringa edible coatings before they can be commercialized to be used in the banana industry.

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## CHAPTER 4: EFFECT OF NANOPARTICLE-ENRICHED COATINGS ON THE SHELF LIFE OF CAVENDISH BANANAS

### 4.1 Abstract

The short shelf-life of bananas at room temperature is a serious problem since the temperature accelerates ripening-related processes like tissue softening, senescence, and fungal diseases. The study aimed to investigate the effect of enriching *Aloe vera* and *Moringa oleifera* plant extract edible coatings with chitosan nanoparticles on the postharvest quality of 'Cavendish' bananas. Banana fruits were dipped into the following treatments: *Aloe vera* 50% (AV), *Moringa oleifera* 10% (MO), *Aloe vera* 50% + chitosan nanoparticle 2% (AV+CN), and *Moringa oleifera* 10% + chitosan nanoparticle 2% (MO+CN). Fruits were stored at room temperature ( $18 \pm 1^\circ\text{C}$ ) for up to 30 days. Sampling was done on a five-day interval and sensory evaluation was done after 25 days in storage. The results showed that adding chitosan nanoparticles to edible coating had a significant ( $p < 0.05$ ) impact on the firmness, ethylene rate, respiration, and total phenolic content during storage. MO+CN coating reduced weight loss (23%), respiration rate (18 mg/kg/h), and ethylene generation (144  $\mu\text{L/kg/h}$ ) to a larger extent followed by AV+CN, MO, and AV. In addition, the combination treatment preserved fruit quality parameters such as total soluble solids, fruit firmness, and peel colour. Furthermore, MO+CN had the highest score for colour (6 score), texture (7 score), odour (7 score), and overall acceptability (6 score) for the consumer analysis using a 9-point hedonic scale. As a result, adding chitosan nanoparticles to plant extract coatings, especially *Moringa oleifera*, improved their efficiency and can be utilized to increase the storage life of banana fruit.

**Keywords:** *chitosan, sensory quality, plant extract, mineral analysis*

### 4.2 Introduction

The Cavendish subgroup is one of the most popular cultivated bananas in the world (Lobo & Rojas 2020). It is also an important international trading commodity that is widely dispersed around the globe (Kuan et al., 2015). In terms of nutrition, banana is one of the world's most important food crops, containing minerals, vitamins, carbohydrates, flavonoids, phenolic compounds, and they supply a large quantity of energy to the body (100 Cal/100 g) (Aurore et al., 2009; Ahmed et al. 2017). It is both

cost-effective and easily available to individuals from all walks of life, alleviating food insecurity issues in many countries. Bananas also contain antioxidants like ascorbic acid, tocopherol, beta-carotene, phenolic groups, dopamine, and gallic acid (Shian & Abdullah 2012). Bananas, on the other hand, are climacteric fruits making them very perishable, and thus prone to postharvest weight loss, texture softening, starch to sugar conversion, chlorophyll degradation, and decay thereby limiting their postharvest life. As a result, the problem of postharvest loss of banana fruits needs to be addressed to satisfy supply and demand for this fruit in both the domestic and export markets. Several post-harvest treatments, such as modified atmosphere packaging (Siriwardana et al. 2017), edible coating (Maqbool et al. 2011; Andrade et al. 2015; Soradech et al. 2017), and essential oils (EOs) (Siriwardana et al. 2017; Vilaplana et al. 2018), have been evaluated to improve postharvest sensory quality, microbiological safety, and nutritional value of bananas. Although cold storage has been shown to extend shelf life, it is costly to adopt and requires infrastructure and postharvest facilities. Furthermore, banana fruit has a strong sensitivity to low temperatures and viruses, making cold storage difficult (Cano et al., 1997). Banana fruit has been observed to have a longer shelf life when stored in a modified or regulated environment (Yousaf et al. 2006; Bhande et al., 2008). The sensitivity of bananas to CO<sub>2</sub> damage and the creation of off-flavour because of anaerobic respiration and ethanol synthesis limit the adoption of various storage technologies (Yousaf et al. 2006; Bhande et al. 2008). This has prompted researchers to investigate other postharvest treatments that might be feasible. The application of edible coatings appears to be a suitable option for market growth because it extends the time for fruit delivery to internal and external markets. Film-forming polymers, such as natural carbohydrates, lipids, and proteins, are the major components of edible coatings (Zaritzky, 2010). These polymers can be utilized on their own or alongside other natural polymers. Because of their edibility, appealing appearance, selective permeability to gases (CO<sub>2</sub> and O<sub>2</sub>), superior mechanical qualities, non-toxicity, non-polluting properties and low cost, edible coatings in fresh produce offer several advantages (Azevedo et al., 2014). They can boost the commercial value of fruits by improving their appearance, safety, and quality, while also acting as carriers of useful substances including antioxidants, antimicrobials, and nutraceuticals (Tavassoli-Kafrani et al., 2020). Both plant and animal sources can be used to formulate the edible

coating. Plant extracts are especially popular because of the medicinal benefits they provide (Oms-oliu et al., 2010). corn starch (Razak & Lazim, 2015), *Aloe vera* (Khaliq et al., 2019; Majeed et al., 2019), gum arabic (Maqbool et al., 2011), and gum ghatti (Joshi et al., 2018) have been tested on banana and showed promising results. Specifically, *Aloe vera* and moringa leaf extracts are high in antioxidants and have antibacterial activities against a wide range of microbes and microorganisms, making them appealing as coating materials (Rao et al., 2001; Dang et al., 2008; Misir et al., 2014). They have also been demonstrated to boost the quality of fresh produce after harvesting. Antibacterial agents have been used extensively in the development of edible coatings to improve food quality and safety (Nguyen et al., 2020). The addition of nanoparticles to edible coatings increases their functional, nutritional, organoleptic, and mechanical qualities, and might be regarded as a novel way to maintain product quality. Previous studies have also shown that the benefits derived from adding nanoparticles to edible coatings include improving the effectiveness of edible coatings in preserving the quality of fresh produce, as well as acting as an antibacterial agent, as in the case of chitosan, and increasing food safety during storage (Belbekhouche et al., 2011; Martelli et al., 2013). Chitosan a natural material is one of the promising biopolymers that has been explored as a nanoparticle due to its film-forming ability, biodegradability, biocompatibility, non-toxic to humans, ease of modification, and versatile physical and chemical properties (Jianglian & Shaoying, 2013; Muxika et al., 2017; Divya et al., 2018). Previously, chitosan coating was successfully used to increase the storage life of a variety of fruits, including citrus (Arnon et al. 2014), papaya (Dotto et al. 2015), and mango (Silva et al. 2017). Chitosan nanoparticles have superior physicochemical characteristics and more effective antimicrobial properties than chitosan coatings. This is owing to their large surface area and charge density, increasing the efficiency of its interaction with bacteria's negatively charged surfaces (Arora & Padua, 2010; Yien et al., 2012; Osheba et al., 2013; Pilon et al., 2015). According to Lustriane et al. (2018) and Esyanti et al. (2019), chitosan nanoparticles and chitosan have a positive impact on banana postharvest quality, including positive effect on shelf life, starch content, weight loss, pulp to peel ratio, TSS, and sensory quality. The use of chitosan nanoparticles on bananas could extend their shelf life up to 11 days in storage. Melo et al. (2018) found that edible chitosan nanoparticle coatings were efficient in retaining the physicochemical, sensory, and microbiological



characteristics of table grapes after harvest. Chitosan nanoparticles slowed the ripening of the grapes, resulting in less weight loss, soluble solids, and sugar content reduction, as well as improved moisture retention and the maintenance of titratable acidity levels compared to control. The coating composition determines the efficiency of edible coatings. As a result, adding nanoparticles to plant extract coatings might increase their effectiveness as edible coating. There is, however, a dearth published reports on the use of chitosan nanoparticles in combination with *Aloe vera* gel and moringa on bananas. Similarly, the impact of chitosan nanoparticles in edible coatings on important banana parameters such as respiration rate, ethylene production, phenolic content, and sensory characteristics is yet to be investigated. Given the beneficial potential of chitosan nanoparticles, the goal of this research is to see how the addition of chitosan nanoparticles to the edible coating of *Aloe vera*, and moringa plant extracts coatings affect postharvest quality measures and sensory quality of banana fruit during room storage.

### **4.3 Materials and methods**

#### **4.3.1 Materials**

Cavendish bananas at maturity stage A1 (Freshmark South Africa) were purchased from farmers at Novasun Limitada, Komatipoort, Mpumalanga, South Africa. The fruits were transported to the Botany Laboratory at the University of Johannesburg. Any bruised or diseased fruit was discarded. The green bananas (900 fingers) were small to medium in size with an average weight ranging from 79 to 140 g and the bananas were stored at room temperature for seven days until they reached ripening stage A2 (light green with a tinge of yellow) (Fig 3.1) before the edible coating was applied. Fresh leaves of *Aloe vera* were harvested from Johannesburg Botanical Garden (26°08'60.00"S 28°00'0.00"E). *Moringa oleifera* leaves were harvested from the Agricultural Research Council in Mbombela (25°27'04.06"S 30°58'09.001"E). All chemicals were purchased from Sigma-Aldrich (South Africa).

#### **4.3.2 Methods**

##### **4.3.2.1 Experimental design**

The banana fruits were assigned to four postharvest treatments: *Aloe vera* 50% (AV), *Moringa oleifera* 10% (MO), *Aloe vera* 50% + chitosan nanoparticle 2% (AV+CN), and

*Moringa oleifera* 10% + chitosan nanoparticle 2% (MO+CN). Each treatment comprised three replicates, each with 60 fruits. The fruit was dipped in treatment solutions for 1 min before air drying for 10-15 min on a laboratory bench at room temperature.

### **4.3.3 Preparation of coatings**

#### **4.3.3.1 *Aloe vera* coatings**

A mechanical procedure was used to obtain the pulp (gel) and liquid fractions according to Khaliq et al. (2019) with some modifications that included adding ascorbic acid and citric acid. Firstly, the leaves of *Aloe vera* were washed with distilled water to remove dirt. Then, aloin (a yellow-coloured liquid) was extracted by cutting the base of the leaves. The skin was carefully separated from the colourless parenchyma gel using a knife, and the gel was then separated from the epidermis using a spoon. The gel was homogenized in a blender for 6 min. The mixture obtained was filtered using a muslin cloth to remove fibrous fractions. Ascorbic acid 4% (v/v) was added as an antioxidant. *Aloe vera* gel was diluted with distilled water to give a (50:50 v/v mixture) and 2% (v/v) glycerol was added as a plasticizer.

#### **4.3.3.2 *Moringa oleifera* coatings**

A mechanical procedure was used to obtain the liquid extract according to Hafez et al. (2019) with modifications such as adding glycerol. The leaves of *M. oleifera* were removed from the stalk and stem. Firstly, the leaves were washed with distilled water to remove dirt and afterwards spread out on clean tissue paper. The yellow and black coloured leaves were discarded, and the green leaves were air-dried on the laboratory bench. After drying, they were placed in a zip bag and stored in the freezer. The leaves were dried on an open bench for 72 h and later ground to a powder using a coffee grinder. Fifty grams of leaf powder was diluted with 500 mL of distilled water in a beaker. The mixture was heated to about 80°C in a water bath for 10 min. The solution was then allowed to cool to room temperature. A sieve (100 µm) was used to filter the extract. The stock moringa extract was stored at 4°C. The extract was diluted with distilled water to give a (10:90 v/v required mixture) for the edible coating for the banana. The 10% moringa coating concentration was selected according to previous work done in the laboratory and (2% v/v) glycerol was added as a plasticizer.

#### **4.3.3.3 Chitosan nanoparticle preparation**

Based on earlier work with modifications, chitosan nanoparticles were chemically (ionic gelation) created Esyanti et al. (2019). At room temperature, chitosan (0.2% w/v) was dissolved in acetic acid (0.5% v/v) and homogenized with a magnetic stirrer. To increase the wettability of the solution, Tween 80 (0.1% v/v) was added. The sodium tripolyphosphate (TPP) solution is produced by dissolving it in deionized water (1 mg/mL). The nanoparticles solution was made by swirling the chitosan solutions which contained the Tween surfactant while adding TPP droplets at a volume ratio of 5:1, in a glass beaker at room temperature for 30-60 min. After adding TPP to the chitosan solutions, nanoparticles developed spontaneously.

#### **4.3.3.4 Preparation of edible coating enriched with chitosan nanoparticle solution**

Chitosan nanoparticle coating solutions containing *Aloe vera* gel 50% (v/v) and moringa 10% (v/v) were prepared by adding chitosan nanoparticle (CN) % (v/v) individually to each solution while mixing on a magnetic stirrer. The remaining volume was made up by adding distilled water to AV+CN and MO+CN and homogenizing all the ingredients together to obtain a homogeneous and smooth mixture of coating solutions. Each banana batch required around 8 L of the coating solution. The mixtures were stirred at room temperature for 2 h to form emulsions for coating the bananas (Hadian et al. 2017; Shahbazi, 2018).

#### **4.3.3.5 Application of coatings and storage**

Four treatments were used in the experiment: *Aloe vera* 50% (AV), *M. oleifera* 1% (MO), *Aloe vera* 50% + chitosan nanoparticle 2% (AV+CN) and *M. oleifera* 10% + chitosan nanoparticle 2% (MO+CN). Banana fingers were dipped in a coating solution for 1 min and then air-dried for 15 to 20 min at room temperature to allow a thin layer of edible coating to be formed on the surface of the fruit. After coating treatment, the bananas were moved to a dark storage chamber. Storage temperature and relative humidity were  $18 \pm 1^\circ\text{C}$  and 35-45%, respectively. The effectiveness of the coating on bananas was determined by measuring weight loss, decay rate, soluble solids, and firmness, among other postharvest parameters. Experimental data were recorded 5

days intervals for up to 30 days of storage. Temperature and relative humidity were recorded with a Hobo data logger (Thermochron, USA) during shelf-life investigations.

#### **4.4 Postharvest quality parameter**

##### **4.4.1 Colour**

A colour reader CR-10 plus (Konica Minolta, Japan) was used to determine the colour of the banana fruit's skin which included L\*, a\*, and b\*. Four fruits were randomly picked in each replicate at each phase for the colour measurements, and readings were taken from three separate spots on the fruit. The colour index: L\* (white-black), a\* (red-green), and b\* (yellow-blue), were then measured and reported (Murmu & Mishra, 2017) from the CIELAB scale.

##### **4.4.2 Weight loss**

At the start of the experiment, four fruits from each replicate were marked and kept separate for periodic weighing using a digital balance (Delta range, Switzerland). Following treatment (day 0), and at various intervals (days 5, 10, 15, 20, 25, and 30) during storage, the weight of individual lots was recorded. The percentage weight loss from the initial weight was used to calculate cumulative weight losses. Total weight loss was calculated as the difference between the initial and final fruit weights after 24 d of storage (Sharmin et al., 2015). Weight loss was determined by taking their initial and final weights differences and expressed as a percentage

$$\text{Percentage weight loss} = \left( \frac{W_i - W_f}{W_i} \times 100 \right)$$

Where  $W_i$  = initial fruit weight

$W_f$  = final weight loss

##### **4.4.3 Firmness**

Fruit flesh firmness was measured with an 8 mm round stainless-steel probe, attached to a Fruit Pressure Tester (Mod. FT 327, Effegi, Italy). With the aid of a cutting blade, a thin section of the epicarp was removed (the thickness did not exceed 1 mm). The measurements were taken for the firmness of the pulp of the bananas at two different locations in their equatorial region. The mean of the measurements from each fruit

sample was used to calculate the values in kilograms (kg), which were then converted to Newtons (N).

#### **4.4.4 Total soluble solids (TSS)**

The refractive index of fruit juice was measured at 20°C with a hand refractometer (Atago, Japan) to determine the soluble solid. The refractometer prism was washed and calibrated with distilled water to give a 0% reading before each sample was analysed for the estimation of sucrose expressed in TSS. The banana pulp (50 g) in 150 mL of distilled water was homogenized using a blender to get a filtrate. To obtain the % TSS reading directly from the instrument, a drop of banana juice squeezed from the fruit pulp of each fruit of the three replications of each treatment was mounted on the prism glass of the refractometer. Brix was used to represent the findings (AOAC, 2016; Lopez-Palestina, 2018).

#### **4.4.5 Respiration and ethylene rate**

The banana samples (n=2) were put in a 1000 mL airtight plastic container with a rubber septum in the lid at room temperature according to Nasrin et al. (2017). A F-950 Three Gas Analyzer (Felix instrument, America) with a syringe was placed through the rubber septum into the container to measure the amount of CO<sub>2</sub> produced in percentage and ethylene rate in ppm after covering for 2 h. The respiration rate and ethylene rate were converted to mg CO<sub>2</sub> emitted per kg per h and µL C<sub>2</sub>H<sub>4</sub> emitted per kg per h, respectively. The respiration rate and ethylene were then determined using the following equation:

$$\text{Respiration rate} = \frac{\text{Amount of respiration}}{\text{Weight of fruit}} \times \text{incubation time}$$

$$\text{Ethylene rate} = \frac{\text{Amount of ethylene}}{\text{Weight of fruit}} \times \text{incubation time}$$

#### **4.4.6 Disease incidence**

Disease incidence means the percentage of banana infected with diseases. In this study, the apparent black patches and visible symptoms were regarded as disease. The disease incidence of bananas was calculated by using the following formula (Hossain & Iqbal, 2016):

$$\text{Disease incidence (\%)} = \frac{\text{Number of bananas infected}}{\text{Total number of bananas}} \times 100$$

#### **4.4.7 Determination of total phenolic content**

Total phenolics (TP) were analysed according to Murmu and Mishra (2018) using the Folin–Ciocalteu reagent (FCR) method with slight modification. Pulp tissue (3 g) was homogenized in a glass tube with 30 mL of methanol using a hand blender. The homogenates were centrifuged at 25°C for 30 min at 2500 g. The supernatant was collected and used for the analysis of total phenol. The methanol extracts 0.5 mL were mixed with 5 mL of deionized water and 0.5 mL of the Folin-Ciocalteu reagent (FCR) (Sigma Aldrich, USA) was added. After 10 min, 1.5 mL of saturated 20 % (w/v) aqueous sodium carbonate solution was added to the mixture. The mixture was mixed vigorously and allowed to stand for 30 min at room temperature. The reaction mixture absorbance was measured at 760 nm against the blank (methanol) using a spectrophotometer (WPA, Biochrom, England). The content of phenolics in the extracts was expressed as mg of gallic acid equivalents per 100 g of the fresh weight (mg GAE/g FW).

#### **4.4.8 Mineral analysis and protein content**

The mineral composition was determined according to Ngobese & Workneh, (2018) with slight modifications. For analyses, 0.5 g milled sample were burnt to ash in a furnace overnight at 450°C and placed in 100 mL pre-weighed glass beakers. The ash was moistened with a few drops of distilled water before adding 2 mL of conc HCl to each sample. The samples were evaporated slowly to dryness on a water bath in the fume cupboard with the extractor fan on. Thereafter, 25 mL of freshly prepared 1:9 HCl solution and distilled water was added into each sample and the mixtures were filtered using Advantec 5B:90 mm diameter filter paper. The filtrates were diluted with deionized water to a 5:20 dilution ratio and analysed for phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sodium (Na), zinc (Zn), copper (Cu), manganese (Mn), iron (Fe), nitrogen (N) and aluminium (Al), using an inductively coupled plasma optical emission spectrometer Vista-MPX 2004 (Varian, Mulgrave, Australia). Analysis was done against known standard solutions of the elements (Separations, South Africa) and a blank of deionized water. The mineral element content was expressed as mg 100 g<sup>-1</sup> of DM. The crude protein composition was

calculated by multiplying the total nitrogen composition (as measured by ICP–OES mineral analysis) by the conversion factor (6.25) to get the protein fraction.

#### **4.4.9 Consumer evaluation**

On the 26th day of storage, a sensory evaluation was conducted by 57 untrained panellists ranging in age from 20 to 40 years old. They were made up of University of Johannesburg students and staff. On a standard 9-point hedonic scale (dislike extremely = 1; dislike very much = 2; dislike moderately = 3; dislike slightly = 4; neither like nor dislike = 5; like slightly = 6; like moderately = 7; like very much = 8; like extremely = 9), participants were asked to score the bananas from each treatment for colour, texture, odour, and overall acceptability. The panellists were served all the samples together, which were coded with random three-digit numbers (Shirani & Ganesharanee, 2009). A consent form was given out at the start of the experiment, and the procedure was explained. Three samples corresponding to the treatments were examined by each panellist. Ethical approval and Institutional permission were obtained from the Faculty of Sciences Ethics Committee with reference 2020-07-13/Ngobese\_odetayo before panellists from the university were recruited for participation.

#### **4.4.10 Shelf life**

Shelf life was assessed by conducting a visual observation. In this present study, the shelf life of bananas was graded on a scale of 0 to 100% for visual yellowing of peel to brown colour daily. When bananas showed more than 80% browning, they were deemed to have reached the end of their shelf life (Hassan & Mahfouz, 2010; Taduri et al., 2017).

#### **4.4.11 Statistical analysis**

To establish the significant difference between the treated and control fruits on the same day of storage, a one-way analysis of variance (ANOVA) was done using IBM SPSS statistical software (SPSS v 27). At  $p < 0.05$ , multiple comparisons were made using the Turkey Post hoc test to separate the means.

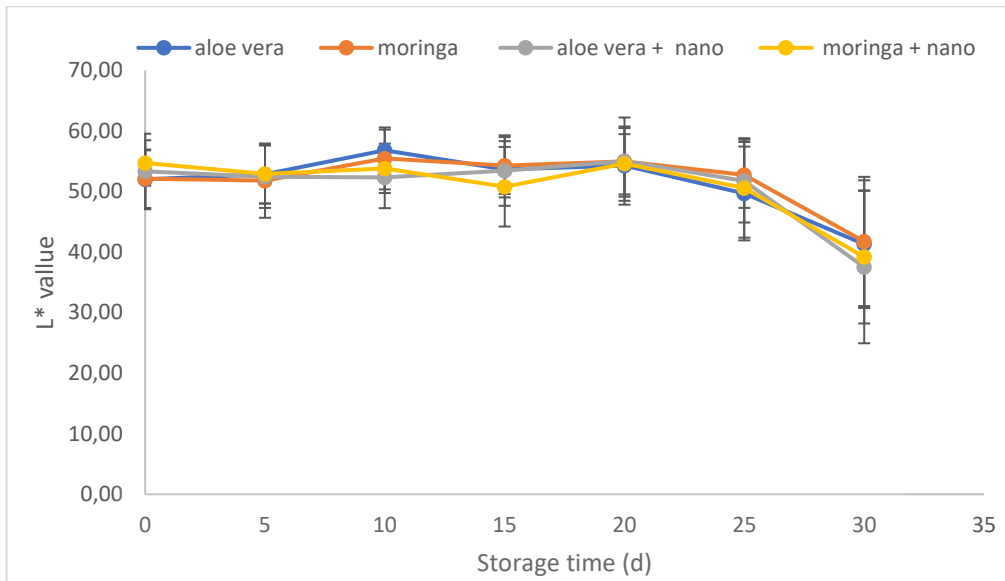
### **4.5 Result and Discussion**

#### **4.5.1 Colour**

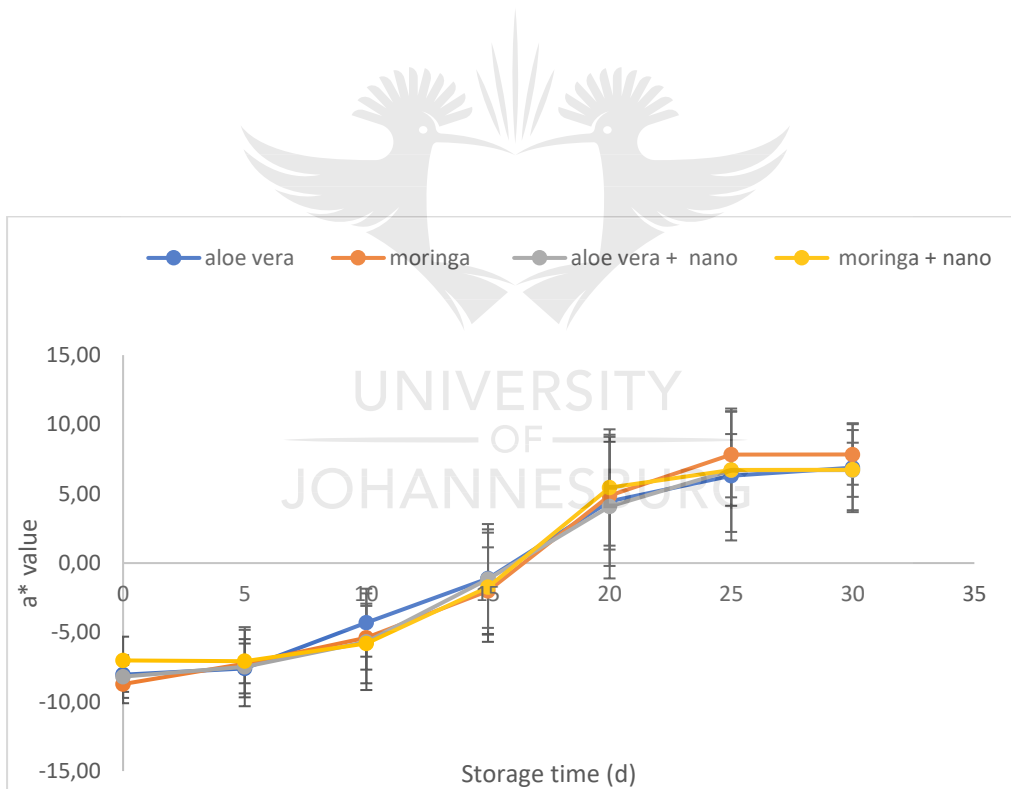
Fruit colour is the most important and often used criterion for assessing the quality of fresh produce. The colour of the surface of the banana changes from green to yellow as it ripens, making it one of the crucial factors in determining the quality and marketability of the fruit. It has something to do with the breakdown of the green pigment chlorophyll and the replacement of it with new pigments like carotenoids (Ahmed & Palta 2016; Lo'ay & Dawood 2017). Fig 4.1 depicts changes in the L\*, a\*, and b\* values of banana skin colour after 30 days of storage. Regardless of treatment, the L\* value decreased as the storage period progressed. In this study, the findings revealed that a gradual decrease occurred in L\* value in all coated fruit. Compared to AV (41.31) and MO (41.74) alone, fruits treated with AV+CN and MO+CN had lower L\* values (37.51 and 39.19, respectively) at the end of storage (Fig 4.1A). All the treatments showed a negative a\* value from day 0 to 15 and started showing positive value from day 20 meaning that the banana was turning from a green colour to a red colour (Fig 4.1B). Negative a\* values reflect the green colour of fruits, while positive a\* values portray the red colour which indicates the initiation of fruit ripening (Basulto et al., 2009). Fruit treated with AV+CN (6.71) and MO+CN (6.73) had the lowest a\* index at the end of the storage period, compared to MO (7.84) and AV (6.88), respectively. This result supports the findings of Adjouman et al. (2018), who found that polysaccharide coatings slowed the increase in a\* value in tomatoes when compared to the control. The addition of chitosan nanoparticles to *Aloe vera* and moringa treatments could have delayed chlorophyll degradation of the fruit peel, according to the findings. This can be linked to the edible coating treatment, which acted as a gas exchange barrier (Olivas et al., 2008). During storage from day 0 to 20, all the coated bananas showed a slight increase in b\* value, indicating an increase in yellowness intensity. Overall, AV+CN (20.63), and MO+CN (20.94) have lower b\* values compared to AV (22.82) and MO (21.60) at day 30 which implies that the addition of nanoparticle to edible coating help delay banana yellowing (Fig 4.1C). However, from day 25 to 30 there was a decrease in b\* value which shows bananas are turning from yellow to brown (Fig 4.2). The statistical analysis for this study shows MO+CN show significant difference for L value at day 10 and 15 days of storage but no significant for a\* and b\* value. This result was similar to the study by Maftoonazad et al. (2007) where avocado showed reduced b\* values indicating a decrease in yellowness and an increase in darker chroma.



A



B



C

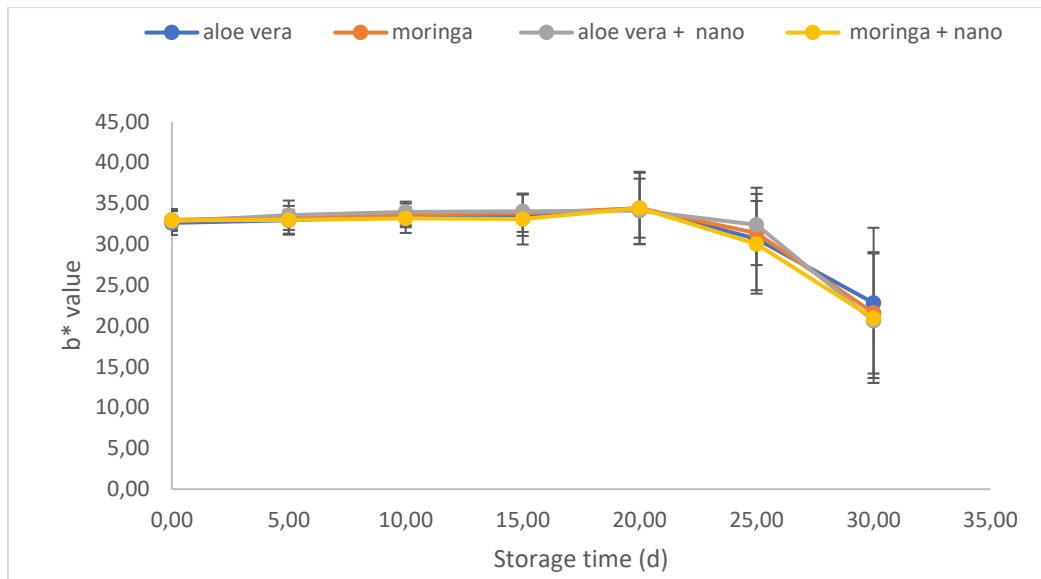


Fig 4.1. The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on colour (L, a, b value) of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$ . A = L value, B = a\* value and C = b\* value

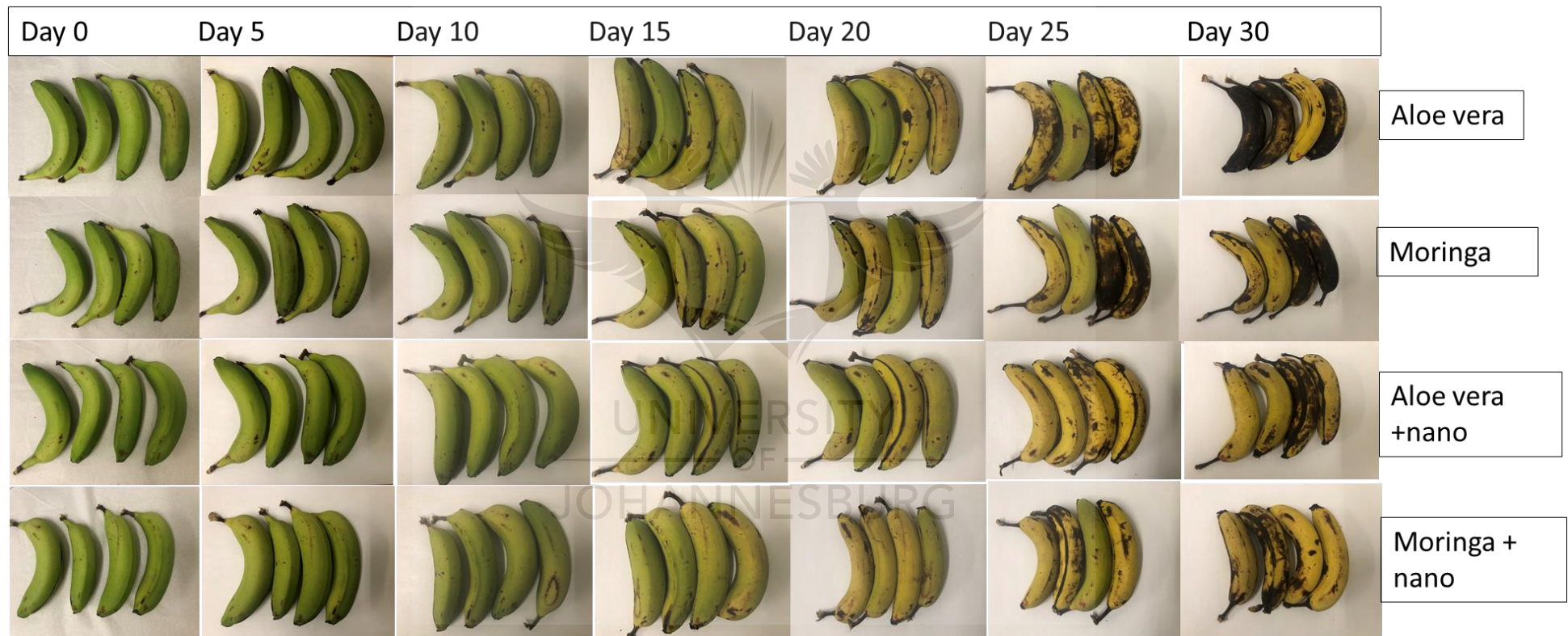


Fig 4.2. The effect of *Aloe vera*, moringa, *Aloe vera* + nano and moringa + nano coatings on changes in ripening stages of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days

#### 4.5.2 Weight loss

Fruit weight loss was another factor in predicting fruit quality. The main reason for fruit weight loss is water loss owing to the respiration process (Eshghi et al. 2014). Fig 4.3 depicts the effect of various treatments on total weight during the storage period. At day 5 of storage, MO+CN had weight loss of 2.74% compared to AV+CN (3.79%), AV (3.99%) and MO (3.57%) respectively. After 30 days of storage, the most effective treatment in terms of preventing weight loss was MO+CN (23%), followed by AV+CN (26%), MO (28%), and AL (31%). The difference in thickness and rheological qualities of coatings could explain the observed variation in weight loss for edible coatings alone. MO+CN and AV+CN outperformed MO and AV throughout the research, as evidenced by our findings where MO+CN reduced weight loss in all the days during storage but there was no significant different between coatings during storage. The antimicrobial properties of the chitosan nanoparticle, which limit the loss of carbon atoms generated by the respiration process, may have contributed to the weight loss reduction following the addition of chitosan nanoparticles to the MO/AV coating. According to Silva et al. (2017), the coating technique creates a barrier on the fruit surface that reduces moisture loss from the fresh produce, allowing fruits to retain their freshness and quality. Because water plays an important role in preserving the shelf life, quality, and market price of fruits (Silva et al., 2017), reducing water loss during storage is critical. Lustriane et al. (2018), showed similar weight loss effects in banana fruit coated with chitosan nanoparticles and chitosan in a prior investigation. Weight loss was reduced using chitosan nanoparticles in apple Gardesh et al. (2016), composite coating of gum arabic with chitosan in banana (Maqbool et al, 2011), and chitosan mixed with pectin in mango (Medeiros et al., 2012) in previous research. Therefore, the addition of chitosan nanoparticles to edible coatings effectively reduced the rate of water loss from bananas during storage, which is vital to the maintenance of shelf life, quality, and market price (Silva et al., 2017). Although previous studies show *Aloe vera* coatings were able to reduce weight loss in bananas, moringa leaf extract was able to establish a modified atmosphere around the perishable fruit, allowing for shelf-life extension through gas exchange regulation and weight loss prevention (Ali et al., 2015). Moringa leaf extract coating was able to reduce weight

loss in tomatoes according to Kator et al. (2019). Similarly, the findings of Vieira et al. (2016), who found weight retention in blueberry fruit treated with *Aloe vera* gel embedded in the chitosan coatings support our result. Therefore, according to this finding, the addition of chitosan nanoparticles improves the efficacy of *Aloe vera* and moringa extract coatings and greatly minimizes the weight loss in banana fruit during storage.

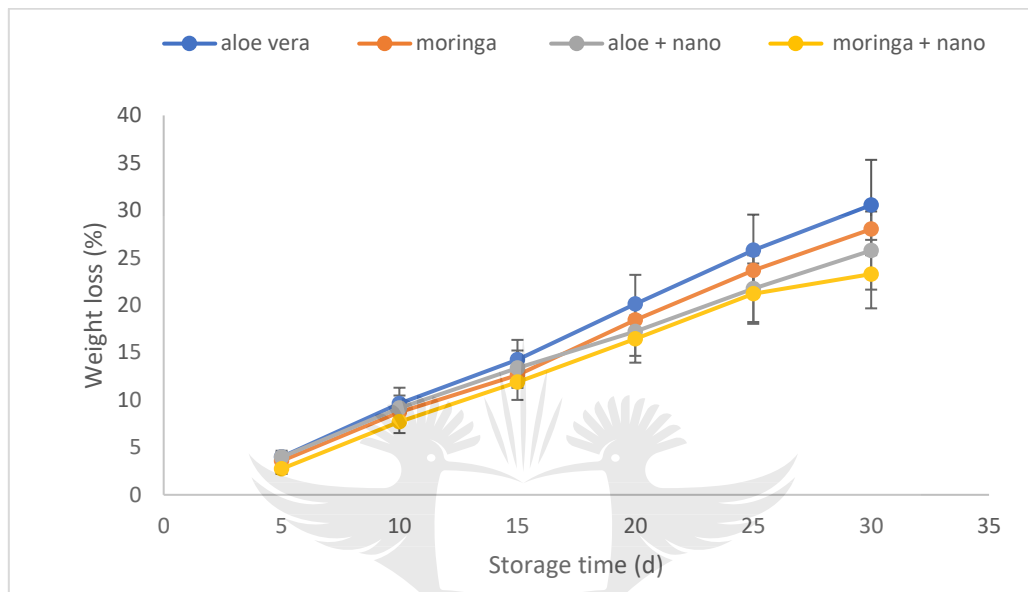


Fig 4.3. The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the weight loss of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 5 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.3 Firmness

Firmness, along with colour and aroma, is one of the factors that consumers appreciate when purchasing some varieties of fruit according to Misir et al. (2014). Starch is a bulk carbohydrate found in banana fruit and its metabolism causes significant fruit softening. Amylase activity which hydrolyses 1,4-glycosidic bonds is thought to be responsible for starch degradation (Zhang et al., 2010). Starch dissolves into soluble sugar when the banana ripens. Banana pulp has a higher sugar concentration than the peel, causing water to flow from the peel to the pulp (Hailu et al., 2014), making the pulp softer as it ripens. During storage, all bananas softened (Fig 4.4). The treatments caused a significant difference ( $p < 0.05$ ) in fruit firmness

after storage. The firmness of the fruits decreased as the storage period progressed, with the change occurring faster in the AV and MO fruits than in the MO+CN and AV+CN coated treatments. The rapid alterations seen in AV and MO coated fruits could be due to faster ripening, which leads to early softness. The fruit coated with an edible coating enriched with chitosan nanoparticles, on the other hand, softened to a smaller degree. Additionally, MO+CN and AV+CN coated fruit were found to be significantly firmer ( $p < 0.05$ ) than MO and AV treated fruit. From day 0 to 30 days of storage, the firmness of the banana coated with edible coating alone dropped dramatically from 61 N to 40 N for aloe and 56 N to 41 N for moringa. This softening is primarily caused by hydrolytic enzyme activity on carbohydrate polymers, which causes degradation of the middle lamella of the cortical parenchyma cell wall (Al Eryani-Raqeeb et al., 2009; Azene et al., 2014). The firmness of the banana increased even more after the addition of the chitosan nanoparticles, with the greatest value of roughly 44 N for AV+CN and 46 N for MO+CN, respectively. Better firmness retention in MO+CN fruit suggested that adding chitosan nanoparticles to edible coating helped slow down metabolic and enzymatic activity in the fruit, resulting in delayed pulp tissue degradation. The greater antifungal activity and coverage of the cuticle and lenticels in bananas coated with MO+CN and AV+CN coatings may reduce microbial infection, respiration, and other ripening processes during storage, resulting in the maintenance of fruit firmness (Rao et al., 2001; Dang et al., 2008; Ali, et al., 2011; Misir et al., 2014). The results from this study show that treating bananas with edible coatings enriched with a chitosan nanoparticle system significantly increased their firmness. Our findings are therefore in agreement with Tesfay and Magwaza (2017), who stated that avocado coated with moringa, and CMC had the highest firmness compared to control and other treatments. Similarly, Thakur et al. (2019) findings are consistent with this study, which developed a rice starch edible covering combined with sucrose esters and found that it improved banana firmness retention.

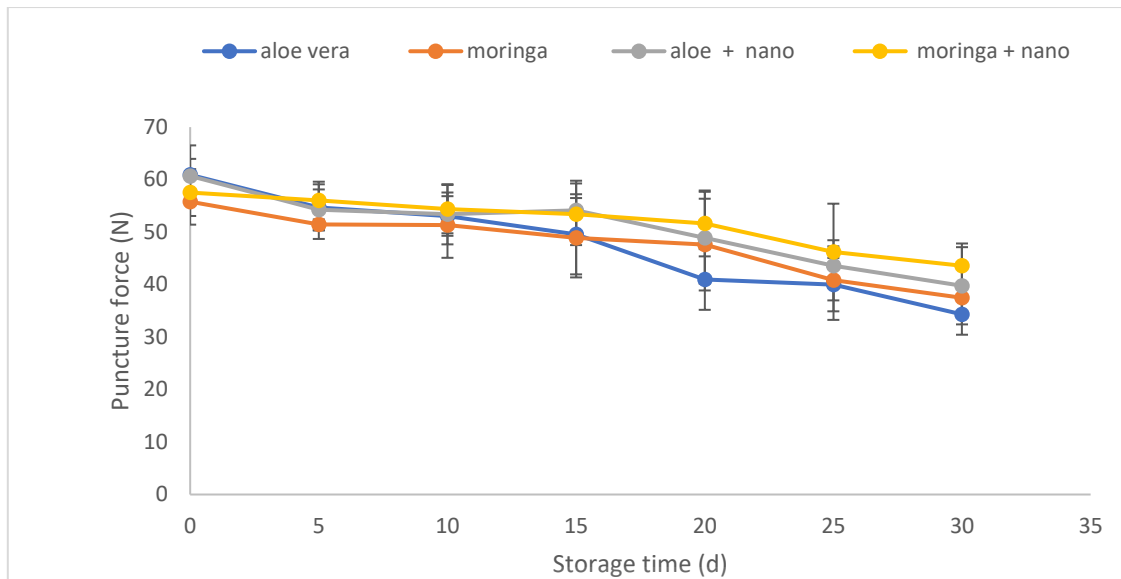


Fig 4.4. The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the firmness of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.4 Total soluble solids (TSS)

In banana pulp, sugars such as glucose, sucrose, and fructose make up most of the total soluble solids. As noted by Xiao et al. (2018), starch is hydrolysed into sugars during ripening, resulting in a rise in sugar content and a decrease in starch content. However, throughout the storage period, all analysed samples showed a gradual increase in soluble solids concentration (Fig 4.5). The TSS of all banana samples increased from 4 °Brix, 4 °Brix, 4 °Brix, and 4 °Brix at day 1 to 38.67 °Brix, 34 °Brix, 33.33 °Brix, and 32 °Brix at day 30 for MO, AV, AV+CN and MO+CN respectively. The loss of water, the degradation of starch to simple sugars, and the disintegration of cell wall polysaccharides could all contribute to the increase in the TSS of the fruit during storage (Sun et al., 2013). The increase in soluble solids concentration reported in this investigation was similar to that previously seen in mango and banana (Gutiérrez-Martnez et al. 2015; Khaliq et al. 2016; Khaliq et al., 2019). Moreover, a similar explanation for the increase in TSS during storage has been provided by Thakur et al. (2019) who studied the effect of rice starch edible coating blended with sucrose esters on controlling the postharvest physiological activity of Cavendish banana stored at  $20 \pm 2^\circ\text{C}$ . Compared to other treatments, MO+CN and AV+CN slowed the increase in

TSS of banana fruits but no statistically significant difference, according to the findings reported here. This is because the coatings can create a semipermeable layer or barrier surrounding the fruit's surface, altering the interior atmosphere by lowering oxygen and/or increasing carbon dioxide levels, lowering the fruit's respiration and metabolic activity (Ali et al., 2010; Gol et al., 2015). As a result, the rate of ripening and senescence of the fruit were slowed down (Vargas et al., 2008). The slower metabolic activities of fruit occur due to regulated gaseous exchange, which may have caused the increase in TSS to be delayed in MO+CN and AV+CN fruit. A prior investigation on fruit coated with biopolymer coatings reported similar results in terms of a slower increase in TSS (Al-Qurashi et al., 2017; Soradech et al., 2017). Furthermore, chitosan nanoparticles may cause a decrease in the soluble solids content of bananas by decreasing respiration and metabolic activity in the fruit, causing the ripening process to be delayed (Lustriane et al., 2018). When chitosan was present in the form of nanoparticles within the edible coatings, this feature improved or strengthened the effectiveness of the coatings, as evidenced by the reduced concentration of soluble solids in bananas coated with edible coatings containing nanoparticles. A recent study on bananas coated with chitosan edible-coating enhanced with citrus lemon peel extracts and *Ocimum tenuiflorum* leaf extracts demonstrated similar outcomes in terms of the slower increase in TSS (Deb Majumder & Ganguly, 2020).

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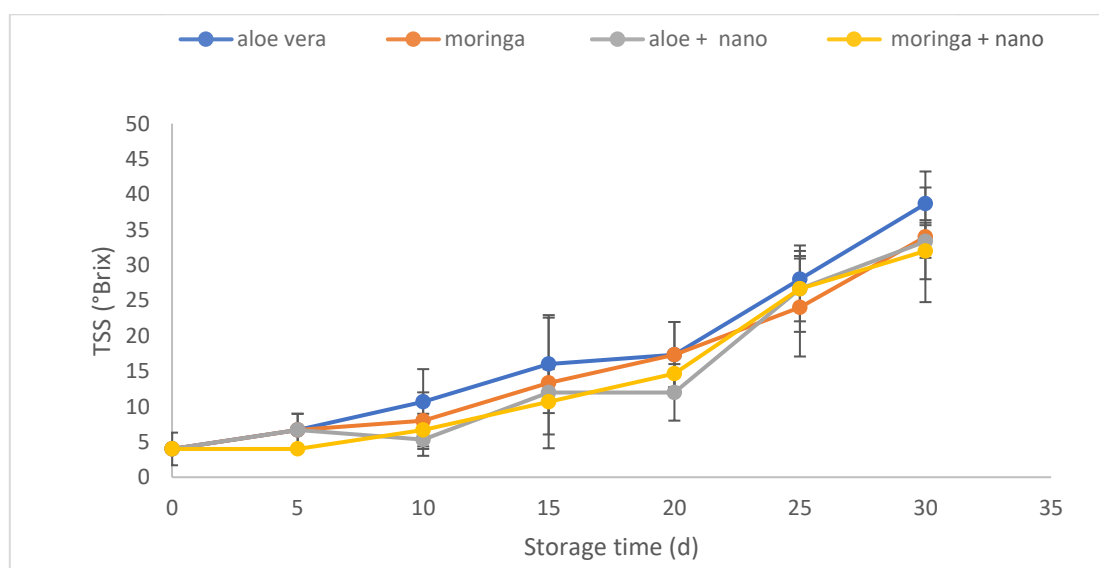




Figure 4.5: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the TSS of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-4% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.5 Respiration rate (RR)

Cavendish bananas which follow a climacteric ripening pattern during postharvest storage have lower storability because high rates of weight loss, softening, acidity losses, and colour changes are associated with increased respiration rates (Ding & Darduri, 2009). The changes in the respiration rate ( $\text{CO}_2$  gas) of bananas subjected to various storage treatments are depicted in Fig 4.6. The initial RR values for all treatments were similar and did not differ until day 5 of storage. After the tenth day, there was an increase in the RR in the coated banana fruits which increased dramatically by day 20. At day 20, all the treatments showed a peak in respiration, with MO (66 mg/kg/h) displaying a higher rate than MO+CN (55 mg/kg/h), AV (50 mg/kg/h), and AV+CN (45 mg/kg/h) coated fruits. The limited storage life of climacteric fruits can be attributed to an increase in respiration during the ripening of banana fruits (Khaliq et al. 2015). MO (44 mg/kg/h) and AV treated fruit (33 mg/kg/h) had the highest respiration rates at day 30. Fruit senescence could be linked to the increase in respiration rates in AV and MO treated fruit. Fruit treated with MO+CN (18 mg/kg/h) and AV+CN (30 mg/kg/h), on the other hand, had the lowest respiration rates at day 30. Addition of chitosan nanoparticle to MO significantly ( $p < 0.05$ ) reduce respiration rate at day 30 compared to using MO alone. The addition of chitosan nanoparticles to edible coatings slowed the rate of decomposition (Fig 4.2), which could explain why the respiration rate was reduced (Paladines et al., 2014). In other words, the decrease in respiration rate in MO+CN and AV+CN fruit was caused by a decrease in stomal gas permeability, which results in a decrease in  $\text{O}_2$  and an increase in  $\text{CO}_2$  in the internal gas atmosphere (Paladines et al., 2014). This effect of chitosan nanoparticles could be related to their antibacterial capabilities, which improved the coating's barrier properties and limited gas transport. Various investigations are based on the application of chitosan nanoparticle coating, such as in the case of apple (Gardesh et al., 2016), strawberry (Eshghi et al., 2014), and table grapes (Gao et al., 2013), support the conclusions of the current study. As a result, the coatings may improve

the quality of the fruit by increasing CO<sub>2</sub> levels to substantial amounts, modifying the internal atmosphere. Mandarin coated with *Fircus hirta* fruit extract enriched with alginate (Chen et al., 2016) and avocado coated with moringa extract enriched with carboxymethyl cellulose, as well as chitosan (Tesfay & Magwaza, 2017) both, had a similar effect on respiration rate retardation. The results showed that the addition of chitosan nanoparticles to edible coatings could delay the rise in RR of banana fruit in this study.

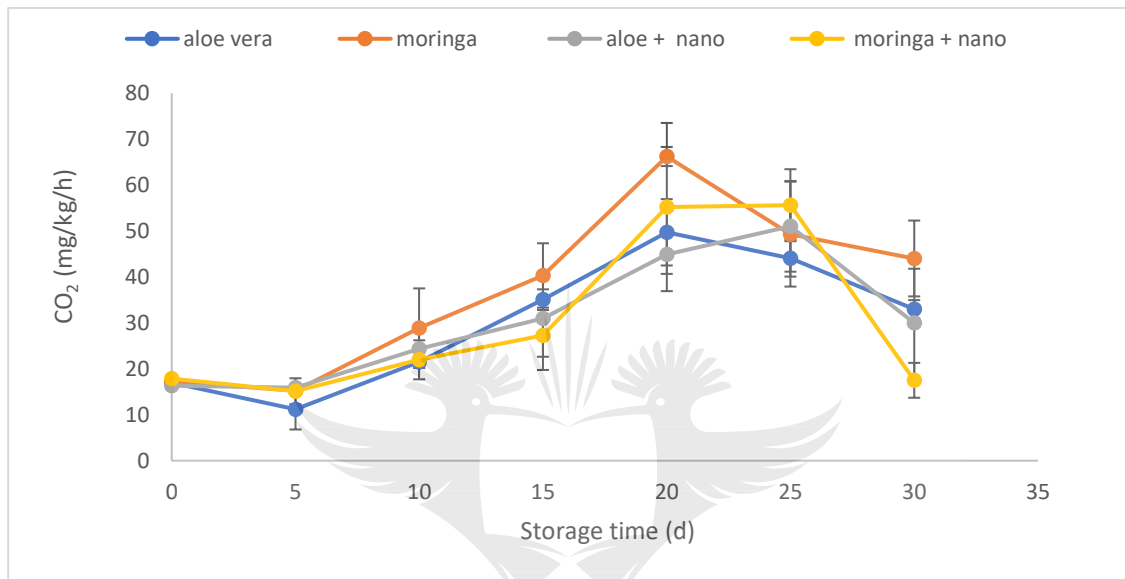


Figure 4.6: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the CO<sub>2</sub> of bananas stored at 18 ± 1°C and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at n = 3

#### 4.5.6 Ethylene rate

Ethylene plays an important role in the ripening of climacteric fruits, and its biosynthesis increases as the fruit ripen. Fig 4.7 depicts changes in ethylene production in banana fruit coated with different treatments. The rise in ethylene rate from day 0 to day 20 was slower for all treated fruits, as seen in (Fig 4.7). From day 25 to day 30, there was a greater significant difference ( $p < 0.05$ ) in the ethylene rate between edible coated fruits alone and edible coated fruits enriched with nanoparticles, with MO (446  $\mu\text{L/kg h}$ ) showing the highest ethylene rate compared to AV (299  $\mu\text{L/kg h}$ ), followed by AV+CN (227  $\mu\text{L/kg h}$ ) and MO+CN (144  $\mu\text{L/kg h}$ ) at day

30. The banana is a climacteric fruit, and a rise in ethylene production during ripening is a normal physiological process. Thakur et al. (2019) found a comparable increase in ethylene in Cavendish banana coated with rice starch blended with sucrose ester at the end of storage at room temperature. However, ethylene generation was reduced when chitosan nanoparticle was added to the edible coating. This reduction could be achieved by constructing a semipermeable membrane over the fruits, which alters the internal atmosphere, delaying metabolic activity and potentially reducing ethylene production (Khaliq et al., 2016). MO+CN treatment was significantly ( $p < 0.05$ ) lower than AV+CN for ethylene rate in this study at day 30. Chitosan, operating as a barrier film, created an altered internal atmosphere and a selective membrane permeability for ethylene diffusion in and out of the fruit, as well as lowered ethylene generation by the fruit, according to previous research (Ali et al., 2011). Chauchan et al. (2015) found that a composite coating of shellac and *Aloe vera* gel inhibited ethylene generation in tomato fruits when compared to those coated with shellac alone and uncoated control samples. Similarly, Mthembu et al. (2018) investigated the influence of moringa-based edible coatings on papaya quality and shelf life. Their findings revealed that papaya fruit treated with moringa-based edible coatings had significantly lower ethylene production.

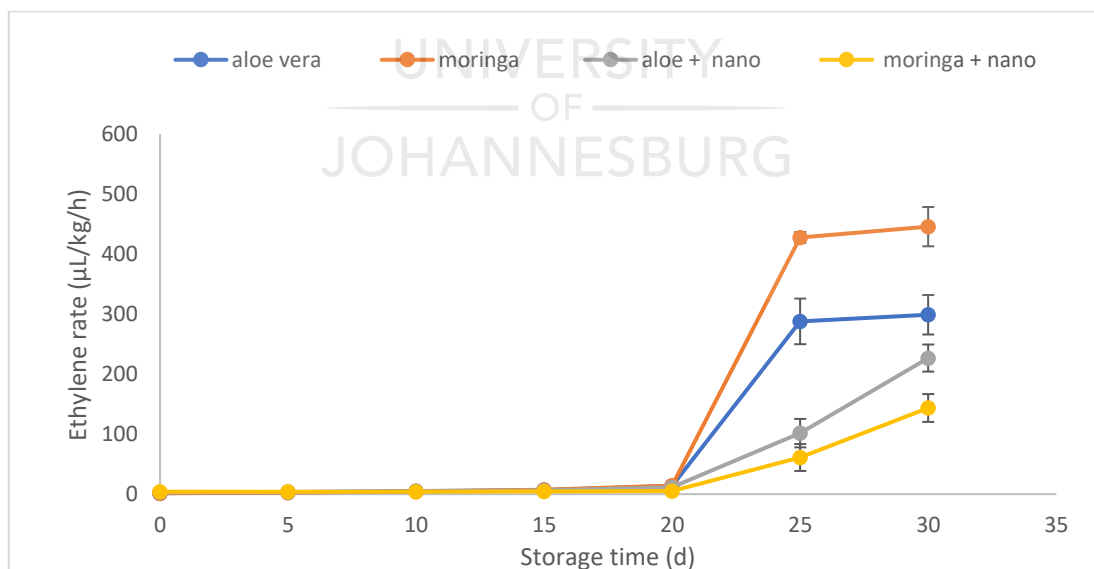


Figure 4.7. The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the ethylene rate of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.7 Disease incidence

One of the most significant causes of economic losses in the global sector is postharvest deterioration caused by fungal diseases (Ranasinghe et al., 2003; Palou et al., 2016). The application of natural antibacterial substances in the form of edible coatings may work synergistically to reduce anthracnose diseases in bananas (Maqbool et al., 2010). Over the years, researchers have been particularly interested in selecting appropriate antibacterial formulations and application dosages. The coating could help the cell wall maintain its integrity against fungal attack and postpone pathogenic infection, according to Abebe and Mohammed (2017). The effect of the AV, MO, MO+CN, and AV+CN on disease incidence in banana samples is depicted in (Fig 4.8). On day 15, AV and MO coated samples began to show indications of disease, however, MO+CN and AV+CN samples did not show disease incidence until day 20 of storage. At the end of storage day 30, the samples treated with MO+CN (58%) and AV+CN (67%) had the lowest disease incidence, whereas AV and MO had the highest disease incidence of 83% and 75%, respectively. According to this research, adding chitosan to the edible coating of *Aloe vera* and moringa plant extract helps minimize disease incidence, with MO+CN having the lowest disease incidence but no significant difference among treatments. In agreement with this finding, Tabassum et al. (2018) and Hossain and Iqbal (2016) both found a decrease in disease incidence in banana samples, with clear evidence of treated samples being more acceptable than control samples. However, other researchers combined *Aloe vera* gel with chitosan to create composite coatings that extended the shelf life of fruits and vegetables more effectively (Khoshgozaran-Abras et al., 2012; Adetunji et al., 2014; Vieira et al., 2016). For example, Vieira et al. (2016) tested the performance of chitosan alone and chitosan - *Aloe vera* combined for coating blueberries. In terms of preventing fungal development during storage, the chitosan - *Aloe vera* coatings outperformed the chitosan coatings. The shelf life was increased to roughly 5 days due to *Aloe vera*'s antifungal properties. Elsabee and Abdou (2013) found that

chitosan has antibacterial and antifungal activities, which helps to prevent postharvest fruit diseases.

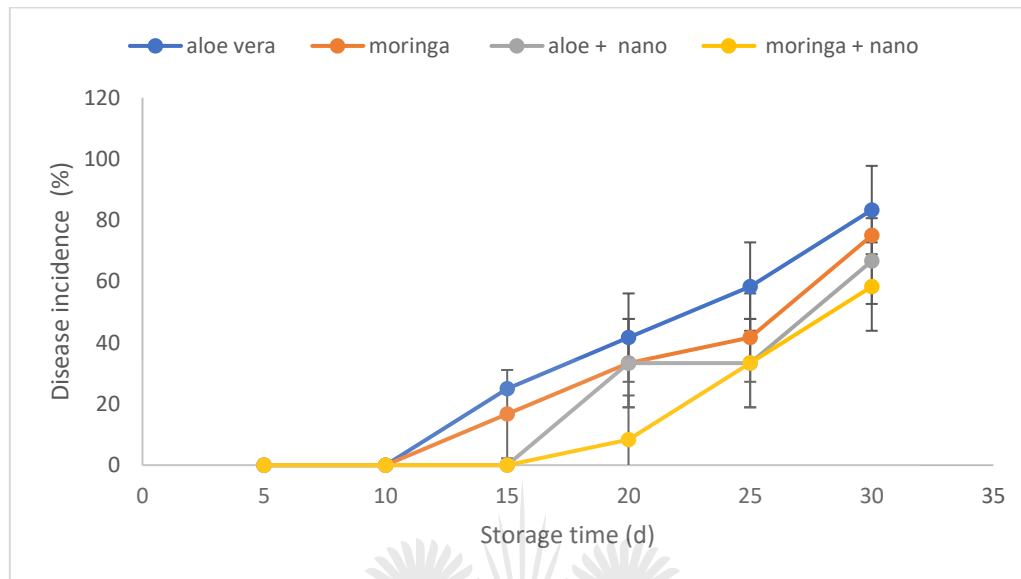


Figure 4.8: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the disease incidence of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 d. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.8 Total phenolic content

The concentration of phenolics increases with ripening and then decreases because of oxidation reactions and senescence. The concentration of phenolics might also vary based on the variety of fruit. Overall, it is well-known that the concentration of phenolics is reduced as a result of oxidation (Ali et al., 2019, 2020). With prolonged storage time, total phenolic content increased initially and then declined (Fig 4.9). The decline in phenol concentration in fruit during ripening could be attributed to cell structural breakdown caused by senescence during storage (Macheix et al. 1990). Overall, the maximum and minimum total phenolic content were observed in MO coated fruit (84 mg GAE/g FW) on day 20 and (18 mg GAE/g FW) on day 30. With AV and AV+CN coated fruits a decrease in total phenolic content occurred throughout storage from (63 mg GAE/g FW) to (30 mg GAE/g FW) and (64 mg GAE/g FW) to (19 mg GAE/g FW) respectively, as shown in (Fig 4.9). Whereas MO and MO+CN coated fruits, an increase in total phenolic content occurred during the first 20 days but dropped on the

last day of storage. The decrease in phenol consumption in coated fruits is attributable to a decline in polyphenol enzyme activity, according to Lo'ay and Taher (2018). On day 30, *Aloe vera* (30 mg GAE/g FW) had a significant ( $p < 0.05$ ) higher phenolic value, followed by MO+CN (22 mg GAE/g FW), AV+CN (19 mg GAE/g FW), and MO (18 mg GAE/g FW). However, AV had greater total phenol levels overall, but other treatments had no discernible impacts. These findings largely corroborate those of Alali et al. (2018), who found that gum arabic treatments with and without salicylic acid retained higher levels of total phenols during ripening in bananas. Such results could be attributable to the edible coating's general preservation effects, which acted as a barrier to gaseous exchange, reducing oxidation processes and delaying fruit senescence (Maqbool et al., 2011; Alali et al., 2018).

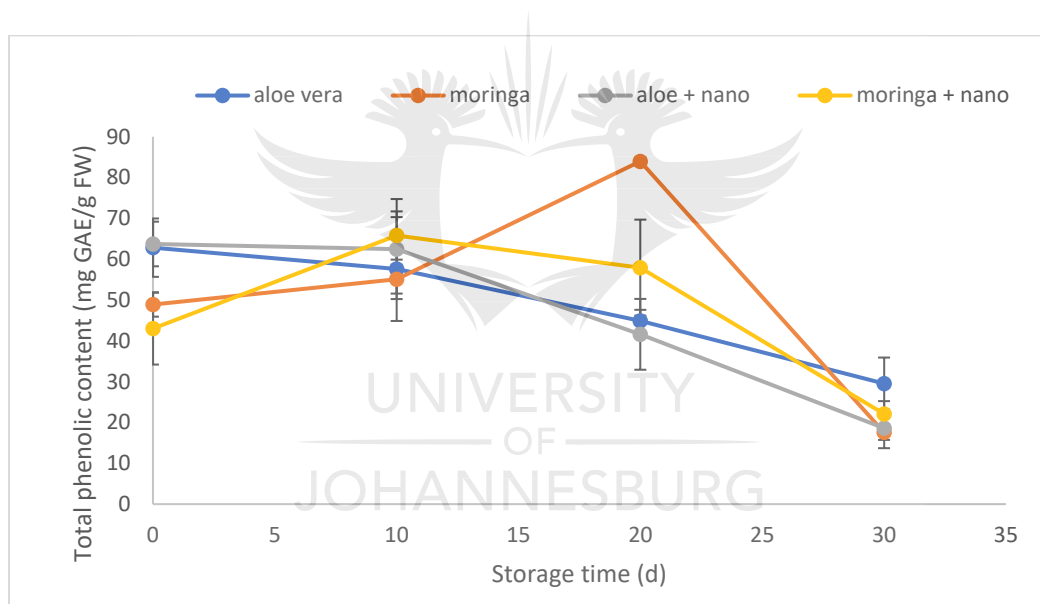


Figure 4.9: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the total phenolic of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.9 Mineral analysis

The sodium (Na), potassium (K), manganese (Mg), calcium (Ca), copper (Cu), aluminium (Al), magnesium (Mn), phosphorus (P), iron (Fe), and zinc (Zn) contents were investigated, and the findings are shown in (Fig 4.10). Except for Fe, there was a significant ( $p < 0.05$ ) change in mineral content in all coated fruits. At day 30 of

storage, sodium was discovered to be the most abundant element in all coated fruits, with a mean value of 479.50 mg/kg for AV coated fruits, followed by MO (102.77 mg/kg), AV+CN (102.58 mg/kg), and MO+CN (97.84 mg/kg). Al content was significantly ( $P < 0.05$ ) higher for AV (78.37 mg/kg) coated banana compared to MO+CN (59.77 mg/kg), AV+CN (49.77 mg/kg) and MO (19.36 mg/kg) whereas MO (48.67 mg/kg) was significantly ( $P < 0.05$ ) higher in zinc content compared to AV+CN (47.08 mg/kg), AV (46.02 mg/kg) and MO+CN (42.50 mg/kg). K, Mn, and Cu also reported a higher content for *Aloe vera* treated fruits compared to other coatings. The mineral content value for Ca, Mg, P was lower in MO banana treated fruits while AV coated fruits recorded the highest value. According to this findings, AV coated fruit has a significant ( $P < 0.05$ ) higher value for Ca, Mg, K, Cu, Mn, P and Al mineral content followed by MO+CN, AV+CN and MO at the end of storage. Overall addition of chitosan nanoparticles to plant extract of *Aloe vera* and moringa did not show a significant effect in mineral content except for MO+CN coated fruits which showed better results than using MO alone. According to a statistical investigation of the mineral content in coated bananas, AV coated bananas appeared to be the healthiest. Its bananas are rich in Na, Al, Zn, Mn, K, Cu, and Mg, Ca, and P, and moderate in Mg, Ca, and P. Variances in the levels of minerals contained in banana fruits can be linked to a range of factors, including the banana variety used, geographical location differences, soil qualities, banana ripening stage, pre-treatment method employed, and drying process time (Wall, 2006).

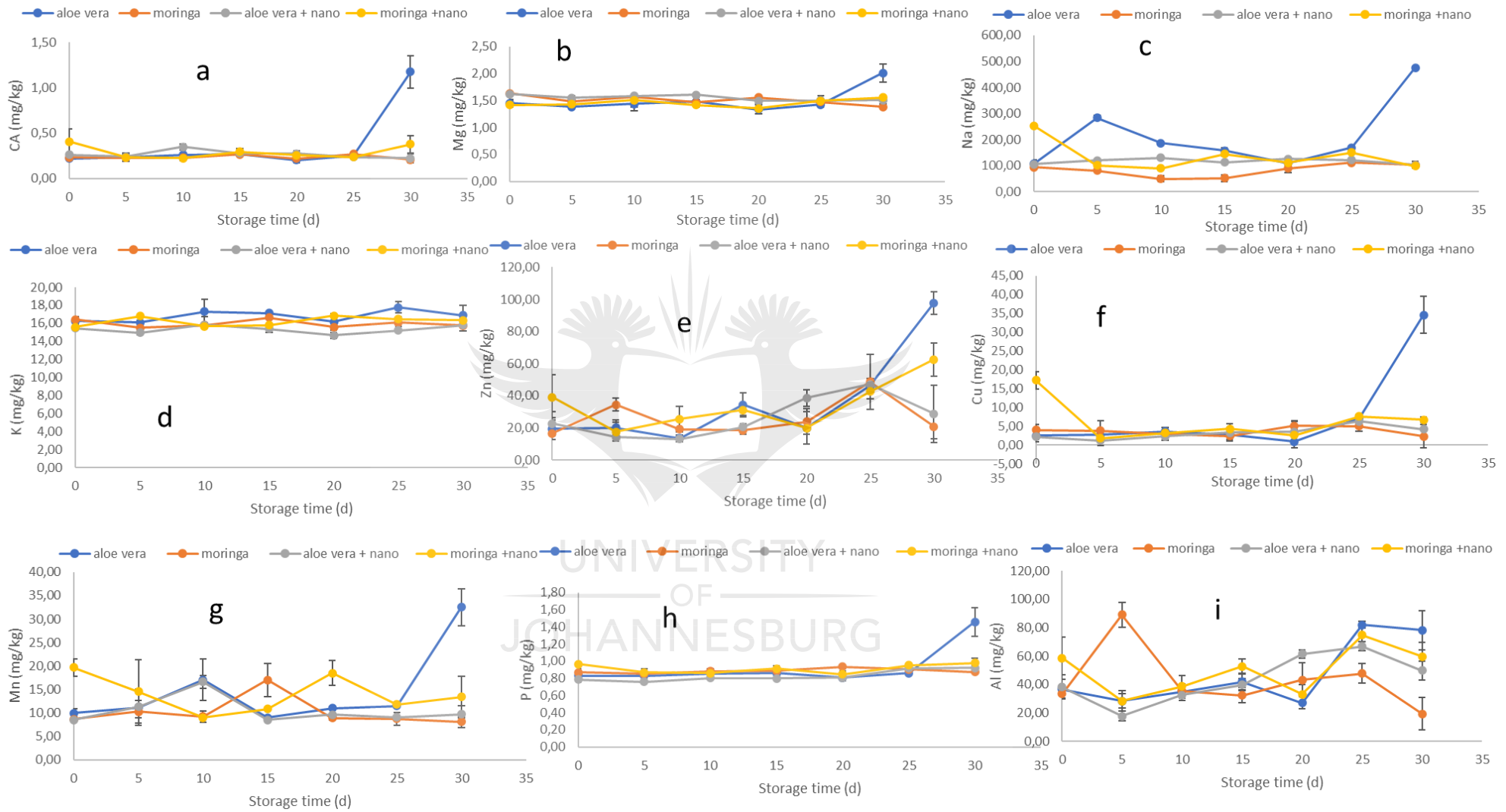




Figure 4.10: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the mineral content of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 d. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.10 Protein content

The protein content of bananas coated with four different treatments are shown in Fig 4.11. On days 15, 25, and 30, there were significant ( $p < 0.05$ ) differences in all the coated fruit. AV protein value was 4.88% and increased to 5.54 %, followed by AV+CN from 4.78 to 5.01 % respectively while MO was reduced it from 4.99 % to 4.83 % followed by MO+CN from 4.68% to 4.55 % respectively from day 0 to 30. After day 30 of storage, AV had a significant ( $P < 0.05$ ) higher value for protein compared to MO, AV+CN, and MO+CN. According to our findings, the AV coated banana had the highest protein content, whereas the MO+CN coated banana had the lowest. This could have occurred because of the fruit's semi-permeable coating, which altered the internal atmosphere (Islam et al., 2018). Our result is like that of Islam et al. (2018) who used chitosan and guava leaf extract to extend the shelf life of fruits such as banana, carambola, and tomato. Protein is necessary for the human body to provide an appropriate supply of essential amino acids (Ashokkumar et al., 2018).

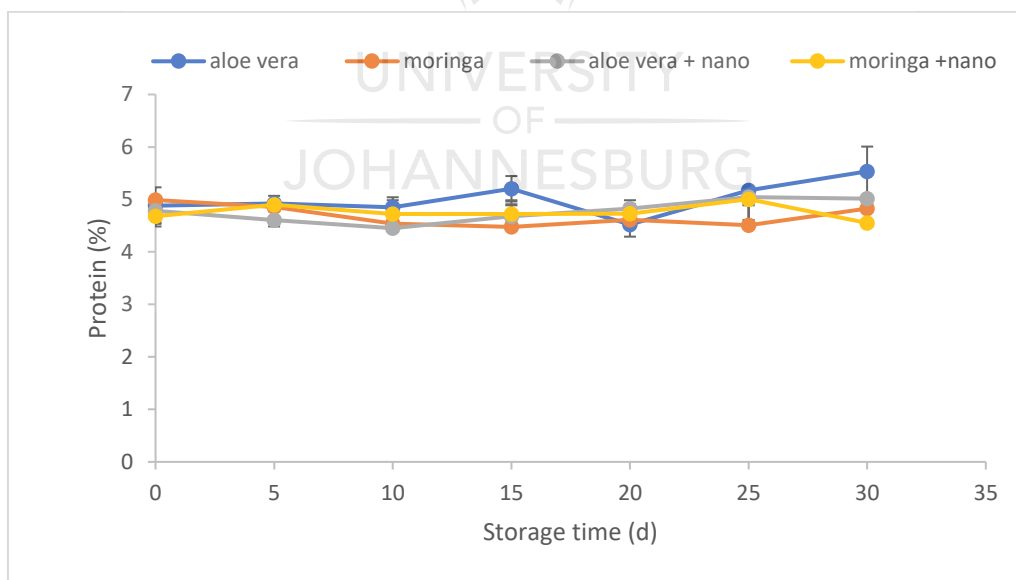


Figure 4.11: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the protein content of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 d. Error bars represent standard error (SE) at  $n = 3$

#### 4.5.11 Consumer evaluation

Consumer acceptance of fruit coated with an edible surface coating is still a big challenge, as the organoleptic qualities of coated produce may be altered by the coating (Guerreiro et al., 2015). As a result, sensory evaluation plays a crucial role in the creation of edible coatings. On day 26 of the storage period, sensory analysis of coated banana fruits revealed changes in colour, texture, odour, and overall acceptability (Fig 4.12). The panellists gave the highest ratings to the fruits treated with MO+CN in all the parameters assessed. However, when compared to all other treatments, fruit coated with M+CN received the greatest marks in colour, texture, and odour, as well as the highest overall acceptability. After 26 days, the MO+CN treatment had greater colour (6.0 scores), texture (7.0 score), odour (7.0 score), and overall acceptability (6.0 score) ratings followed by the AV+CN treatment, which had colour (5.0 score), texture (6.0 score), odour (6.0 score), and overall acceptability (5.0 score) ratings (4.0 score), AV had colour (5.0 score), texture (6.0 score), odour (6.0 score), and overall acceptability (4.0 score) ratings and MO had colour (4.0 score), texture (5.0 score), odour (6.0 score), and overall acceptability (4.0 score) ratings. These sensory results are in line with parameters such as texture where MO+CN retained better firmness of bananas compared to other treatments and disease incidence rates were reduced which might have contributed to the acceptability scores of bananas by the panellists. In addition, a frequency test was carried out to ascertain the percentage of participants that liked the coated bananas. MO+CN was accepted by 67% of the panellists and AV+CN by 61% compared to AV (33%) and MO (30%). So, these further buttresses the claim that MO+CN and AV+CN edible coatings can be employed to extend the shelf life of banana fruits and improve their quality during storage because they were accepted by more than half of the panellists. Similar results were seen in a prior study by Maqbool et al. (2011), who found that banana fruits coated with 10% GA with 1.0% CH had an overall superiority after 33 days of storage. Senescence and a drop in TSS cause sensory quality to deteriorate (Ali et al., 2016; Shah et al., 2017). It has been found that combining plant extracts with edible coatings considerably

delays senescence as well as the loss of TSS, resulting in sensory qualities that are preserved for a longer duration of storage (Khaliq et al., 2019).

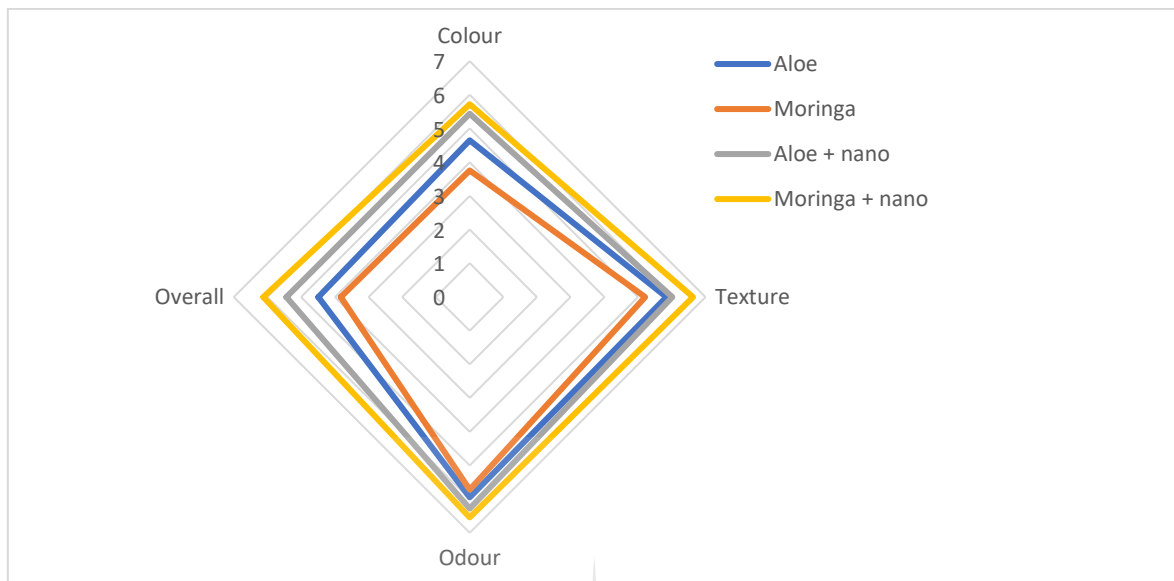


Figure 4.12: Sensory evaluation scores of colours, texture, odour, and overall acceptability of Cavendish bananas treated with *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano and stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH after 25 days. Mean of 57 panellists

#### 4.5.12 Shelf life

The shelf life of a product is an important indicator of its quality (Ali et al., 2010). For the selling and preservation of fresh fruits, longer shelf life is recommended (Chen et al., 2019; Mendy et al., 2019). Fig 4.13 depicts the impact of edible coating of AV, MO, AV+CN, MO+CN coatings on banana shelf-life stored for 30 days at room temperature. The longest banana shelf life was found to be 30 days in MO+CN coated bananas followed by AV+CN for 28 days, whereas the shortest shelf life was found to be 26 days for MO and AV was 25 days. Slower physiological processes (such as respiration) and weight loss linked with less microbiological activity may contribute to the banana's longer shelf life. These findings are in line with those of Mondal et al. (2011). From the result in Fig. 4.13, it was obvious that adding chitosan nanoparticles to plant extracts of *Aloe vera* and moringa coatings improved the banana and extended the shelf life further than using edible coating alone. Furthermore, these

findings are in line with those of Khatri et al. (2020), who found that the combination of *Aloe vera* gel and chitosan has superior effects to the individual coatings throughout the storage period, extending the shelf-life of tomato fruits by up to 42 days compared to 35 days for *Aloe vera* or chitosan alone.

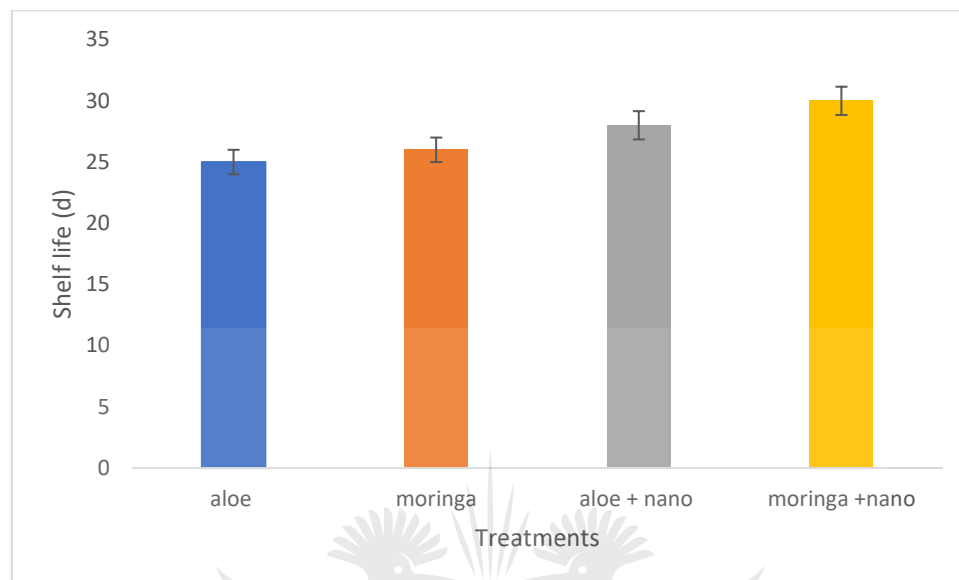


Figure 4.13: The effect of *Aloe vera*, moringa, *Aloe vera* + nano, and moringa + nano on the shelf life of bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH from 0 to 30 days. Error bars represent standard error (SE) at  $n = 3$

#### 4.6 Conclusion

The banana is a climacteric fruit that is often harvested at its ripe green stage. As the fruit ripens, chlorophyll decomposes, carotenoids emerge in the skin, and respiration and ethylene production increase. Overall, the study found that banana fruit treated with a combination of edible coatings and chitosan nanoparticles had lower rates of respiration, TSS, decay rate, and higher firmness values than banana fruit coated with edible coating alone, and this was consistent throughout the storage period. Reduced rate of respiration, moisture, and textural changes resulted in better fruit quality and shelf life, according to these findings. By delaying ethylene production and respiration rate, the addition of CN to AV and MO plant extracts demonstrated remarkable success in reducing banana metabolism, resulting in a net gain in banana storability. Although edible coatings protected fruit quality and shelf life in banana fruit

in a previous study, the addition of chitosan nanoparticles further increased the shelf life to 30 days compared to 24 days in the previous study for bananas. The results revealed that MO+CN was the most effective treatment for minimizing weight loss, firmness loss, and delaying colour changes. In addition, MO+CN preserved the sensory quality of banana fruit after 30 days of storage. This research suggests that edible coatings containing chitosan nanoparticles could be beneficial in the banana sector for preserving postharvest quality and extending shelf life.

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## CHAPTER 5: EFFECT OF NANOPARTICLE ENRICHED PLANT EXTRACT-BASED COATINGS ON THE SHELF LIFE OF ETHYLENE TREATED CAVENDISH BANANAS

### 5.1 Abstract

Banana is a climacteric and very perishable tropical fruit. Among the various preservation procedures, the edible coating is an excellent strategy to extend the shelf life of fruits. As a result, the current study looked at the effects of *Aloe vera* 50% + chitosan nanoparticles 2% (AV+CN) and *Moringa oleifera* 10%+chitosan nanoparticles 2% (MO+CN) on the postharvest quality of ethylene treated and non-ethylene treated Cavendish bananas. After coating, the fruit were stored at room temperature ( $18 \pm 1^\circ\text{C}$ ) and 35-45% RH. Weight loss, firmness, colour, disease incidence, total soluble solids (TSS), respiration and ethylene rate, sensory evaluation, total phenolic content and shelf life were all measured at five-day intervals. The results showed that MO+CN significantly ( $p < 0.05$ ) reduced firmness loss, disease incidence, respiration rate and ethylene production for ethylene treated bananas after 25 days storage compared to AV+CN coating. However, these changes were similar to those observed in non-ethylene treated bananas coated with MO+CN. The MO+CN treatment had a greater colour, texture, odour, and overall acceptability than AV +CN for both ethylene treated, and non-ethylene treated bananas when assessed by consumers. Therefore, it is concluded that the MO+CN combination can be utilized to extend the storage life of both ethylene-treated and non-treated bananas.

**Keywords:** *Chitosan, consumer evaluation, ethylene rate, respiration rate, postharvest quality*

### 5.2 Introduction

Bananas are an important source of health-promoting phytochemicals, according to several studies (Someya et al., 2002; Davey et al., 2007). Most banana cultivars, such as 'Cavendish,' are harvested at the pre-climacteric fully-mature-hard green stage, and ripening is induced by exposing them to a certain dose and duration of ethylene in commercial airtight ground warehouses. The use of ethylene gas in ripening rooms is the most common approach used in developed countries. Ethylene, a plant-produced

ripening hormone, plays an important physiological function in the ripening process and can thus be utilized as a controlled treatment for fruit to reach the desired aesthetic standards and fruit quality (Barry & Giovannoni, 2007; Kulkarni et al., 2011). Following ethylene treatment, bananas go through a series of physical and biochemical changes during ripening, including weight loss, peel chlorophyll degradation and browning, and pulp sugar content, while others, such as pulp hardness and total acidity decrease (Waliszewski et al., 2007; Baez-Sanudo et al., 2009; Maqbool et al., 2011; Fernando et al., 2014; Wang et al., 2014). The climacteric character of bananas, on the other hand, causes a quicker ripening process and a shorter shelf life at ambient temperatures after ethylene treatment (Zhang et al., 2010). Once ripening begins, the peel turns a uniform and quick yellow with brown flecks in 2-4 days at 20°C (de Martino et al., 2007; Ahmed & Palta, 2016). As a result, good and proper postharvest handling procedures are essential for ensuring the quality of banana fruits and extending their shelf life. Edible coatings have been increasingly popular in recent years as a means of preserving the postharvest quality of banana fruits. Edible coatings, which are made of natural and biodegradable materials, are seen to be one of the most promising ways to extend the post-harvest storage life of fruits (Campos et al. 2011). These coatings are thin layers of materials that offer a barrier against moisture and gas transmission between the skin and the external environment while also being edible (Nawab et al., 2017). Plant extracts, on the other hand, are readily available and inexpensive all around the world and are also commonly used as a postharvest treatment (Anjum et al., 2016). Plant edible coating alone or in combination with other coatings has been studied and are beneficial in retaining the postharvest quality of banana and extending its shelf life at ambient temperature. Take for example, edible coating formulations based on guava leaf and lemon extract (Tabassum et al., 2018), gum ghatti and clove oil (Joshi et al., 2018), starch edible coating blended with sucrose esters (Thakur et al., 2019), kurdi gum and farsi gum coating (Shahbazi & Shavisi, 2020) and lemon peel extract (Jodhani & Nataraj, 2021) all reduce quality loss and improve the commercial value of banana fruits. Another plant-based natural covering with antibacterial properties is *Aloe vera* gel (Mendy et al., 2019). In recent years, the use of *Aloe vera* in postharvest fruits and vegetables has expanded dramatically (Ali et al., 2019; Ali et al., 2020). Moringa leaf extract is also abundant in antioxidants and has

antibacterial properties against a variety of microbes or microorganisms (Marrufo et al., 2013). Oranges were shown to have a longer shelf life and better quality when moringa was combined with carboxymethyl cellulose (Adetunji et al. 2013). Moringa leaf extract was added to chitosan and carboxymethyl cellulose to improve the postharvest quality of avocado, and the result showed that the combination of moringa and carboxymethyl cellulose was the most effective treatment in reducing respiration rate, had higher values of firmness, suppress postharvest diseases thereby maintaining overall quality compared with the moringa and chitosan and uncoated fruits (Tesfay & Magwaza, 2017). This indicates that moringa plant components have a large potential for application as an ingredient in the creation of various functional food items, such as edible coatings. Moringa and other natural ingredients were tested on the storability and fruit quality features of the 'Fuerte' avocado by Yousef et al. (2015). Certain additives, such as antibrowning and antimicrobial agents, could be added to edible coatings to improve the performance of the edible coating material (Dhall, 2013). Chitosan is a biodegradable and non-toxic polysaccharide with outstanding biocompatibility making it a great antibacterial substance that has been approved for use in food (Lei et al., 2014). Chitosan-based coatings have shown to be useful in extending the shelf life and increasing the quality of banana fruits (Seseno et al., 2014; Sikder & Islam 2019). Chitosan, as an antibacterial agent, can be used in plant extract edible coatings to improve their efficacy in terms of preserving fruits from microbial deterioration and therefore extending their shelf life (Deb Majumder & Ganguly, 2020), but the antibacterial and barrier characteristics of chitosan nanoparticles are thought to be superior. Nanoparticles, when included in a polymer matrix, can improve mechanical and barrier qualities, as well as operate as an antibacterial agent, as in the case of chitosan, with benefits for food quality and safety (Belbekhouche et al., 2011; Martelli et al., 2013). Chitosan nanoparticles have been shown to have high selective permeability, limiting weight loss, and retaining firmness in fruits for longer (Eshghi et al., 2014; CorreaPacheco et al., 2017; Nguyen et al., 2020). Various research on the application of chitosan nanoparticles as edible coatings on a variety of fruits have been published, including bananas (Lustriane et al., 2018; Esyanti et al., 2019), strawberries (Eshghi et al., 2014), and apples (Gardeshs et al., 2016). Several earlier studies have employed polymers such as

chitosan and *Aloe vera* to extend the shelf life of fruits (Khatri et al., 2020; Shah & Hashmi, 2020; Amin et al., 2021; Seyed et al., 2021). According to Arroyo et al. (2020), Kasim et al. (2021), and La et al. (2021), nanoparticles can be combined with edible coatings to extend fruit shelf life but there is still little research on the use of edible coatings mixed with nanoparticles as fruit coating. According to the literature study, there were no studies on *Aloe vera* and moringa with chitosan nanoparticles as an edible coating to extend the shelf life and maintain the quality of banana fruit. Our study in Chapter 4 showed that enriching edible coating with chitosan nanoparticle improves the postharvest quality of non-ethylene treated bananas. This study aims to assess the usage of *Aloe vera* and moringa coatings mixed with chitosan nanoparticles as an edible coating on ethylene treated banana and assess if the efficacy of the coatings is consistent for preserving the post-harvest quality attributes of both ethylene treated and non-ethylene treated banana during room temperature storage.

### 5.3 Materials and Methods

#### 5.3.1 Materials

Two sets of Cavendish bananas were purchased from Novasun Limitada, Komatipoort, Mpumalanga, South Africa. The first set were at maturity stage A1 according to Fig 3.1 and they were stored at  $19 \pm 2^{\circ}\text{C}$  for seven days until they reached ripening stage A2 (Fig 3.1) before the edible coating was applied. The second set of mature green banana at ripening stage A1 according to the Fig 3.1 were directly pre-treated with ethylene gas for ripening induction for 48 hours in commercial airtight ground warehouses at the Joburg Market. Then, at the warehouse, bananas with uniform hands at ripening stage B1 (Fig 3.1) were randomly picked and transported to the Botany laboratory of the University of Johannesburg. The selected bananas were medium to large with an average weight ranging from 82 to 192 g. Bruised or diseased fruit were discarded, and the bananas were stored at  $19 \pm 2^{\circ}\text{C}$  before coatings were applied. Fresh leaves of *Aloe vera* were harvested from the Johannesburg Botanical Garden (26°08'60.00"S 28°00'0.00"E). *Moringa oleifera* leaves were harvested from the Agricultural Research Council in Mbombela (25°27'04.06"S 30°58'09.01"E). All chemicals used in this study were purchased from Sigma-Aldrich (South Africa).

## 5.3.2 Methods

### 5.3.2.1 Experimental design

Each treatment comprised three replicates, each with 60 fruits. The fruit was dipped in treatment solutions for 1 min before air drying for 10-15 min on a laboratory bench at  $19 \pm 2^\circ\text{C}$ . *Aloe vera* 50% + chitosan nanoparticle 2% (AV+CN) and *Moringa oleifera* 10% + chitosan nanoparticle 2% (MO+CN) were assigned to both ethylene treated and non-ethylene treated bananas.

### 5.3.3 Preparation of coatings

#### 5.3.3.1 *Aloe vera* enriched with chitosan nanoparticle (AV+CN)

A mechanical procedure was used to obtain the pulp (gel) and liquid fractions according to Khaliq et al. (2019) with some modifications by adding ascorbic and citric acids. Firstly, the leaves of *Aloe vera* were washed with distilled water to remove dirt. Then, aloin (a yellow-coloured liquid) was extracted by cutting the base of the leaves. The skin was carefully separated from the colourless parenchyma gel using a knife, and the gel was then separated from the epidermis using a spoon. The gel was homogenized with a blender for 6 min. The mixture obtained was filtered using a muslin cloth to remove fibrous fractions. Four percent (v/v) ascorbic acid was added as an antioxidant. *Aloe vera* gel was diluted with distilled water to make 50:50 portion (v/v) and 2% (v/v) glycerol was added as a plasticizer. Based on earlier work with modifications, this study created chitosan nanoparticles chemically (ionic gelation) (Esyanti et al., 2019). At room temperature, chitosan (0.2% w/v) was dissolved in acetic acid (0.5% v/v) and homogenized with a magnetic stirrer. To increase the wettability of the solution, Tween 80 (0.1% v/v) was added. The sodium tripolyphosphate (TPP) solution was produced by dissolving it in deionized water (1 mg/mL). Nanoparticle solutions were made by swirling chitosan solutions containing Tween surfactant and TPP droplets at a volume ratio of 5:1 in a beaker glass at room temperature for 30-60 min. After adding TPP to chitosan solution, nanoparticles were developed spontaneously. To minimize particle size, the mixture was homogenized in a high-speed blender. Coating solutions (AV+CN) were prepared by adding chitosan nanoparticle (2%) to *Aloe vera* 50% on a magnetic stirrer. The remaining volume was

made up by adding distilled water to obtain a homogeneous and smooth mixture of coating solutions. Each batch required around 8 L of coating solution. The mixtures were stirred at room temperature for 2 h to form emulsions for coating the bananas (Hadian et al. 2017; Shahbazi 2018).

### **5.3.3.2 *Moringa oleifera* enriched with chitosan nanoparticle (MO+CN)**

A mechanical procedure was used to obtain the liquid extract according to Hafez et al. (2019) with some modifications by adding glycerol. The leaves of *Moringa oleifera* were obtained from the stalk and stem. Firstly, the leaves were washed with distilled water to remove dirt and afterward spread on clean tissue paper. The yellow and black coloured leaves were discarded, and green leaves were air-dried on the laboratory bench. After drying they were placed in a zip bag and stored in a freezer. The leaves were dried on an open bench for 72 h and later ground to a powder using a coffee grinder. Fifty grams of leaf powder was diluted with 500 mL of distilled water in a beaker. The mixture was heated to about 80°C in a water bath for 10 min. The solution was then cooled at room temperature. A sieve (100 µm) was used to filter the extract. The moringa was stored at 4°C. The extract was diluted with distilled water to get 10:90 (v/v) to produce an edible coating for the banana. The moringa coating concentration (10% v/v) was selected according to previous work done in the laboratory and glycerol (2% v/v) was added as a plasticizer. Based on earlier work with modifications, we created chitosan nanoparticles chemically (ionic gelation) (Esyanti et al., 2019). At room temperature, chitosan (0.2% w/v) was dissolved in acetic acid (0.5% v/v) and homogenized with a magnetic stirrer. To increase the wettability of the solution, Tween 80 (0.1% v/v) was added. The sodium tripolyphosphate (TPP) solution was produced by dissolving it in deionized water (1 mg/mL). Nanoparticle solutions were made by swirling chitosan solutions containing Tween surfactant and TPP droplets at a volume ratio of 5:1 in a glass beaker at room temperature for 30-60 min. After adding TPP to chitosan solutions, nanoparticles were developed spontaneously. To minimize particle size, the mixture was homogenized in a high-speed blender. Coating solutions (MO+CN) were prepared by adding chitosan nanoparticle (2%) to moringa 10% on a magnetic stirrer. The remaining volume was made up by adding distilled water to obtain a homogeneous and smooth mixture of coating solutions. Each batch required around 8 L of coating solution. The mixtures

were stirred at room temperature for 2 h to form emulsions for coating the bananas (Hadian et al. 2017; Shahbazi, 2018).

### **5.3.3.3 Application of coatings and storage**

Sorted bananas were assigned into three groups at random (each with 60 fruits and three replications). Ethylene treated bananas in each group were immersed for 1 min in one of the following treatments: *Aloe vera* 50% + chitosan nanoparticle 2% (AV+CN) and *Moringa oleifera* 10% + chitosan nanoparticle 2% (MO+CN). The same process was repeated for bananas without ethylene treatment. The fruit was left to air dry for 15 to 20 min at room temperature to allow a thin layer to be formed on the fruit. The fruits were relocated to a dark storage room with temperature and relative humidity ranging from 17 to 19°C and 35 to 45% respectively using a Hobo data logger, throughout the experiment. On day 0, and after 5, 10, 15, 20, 25 and 30 days of storage, observations on several postharvest qualities were evaluated. A range of quality parameters such as colour, weight loss percentage, firmness, total soluble solid, decay rate, ethylene, and respiration rate, total phenolics, and the sensory test was assessed at regular intervals to monitor the postharvest changes of fruit during storage.

## **5.4 Postharvest quality parameters**

### **5.4.1 Colour**

A colour reader CR-10 plus (Konica Minolta, Japan) was used to determine the colour of the banana fruit's skin which included L\*, a\*, and b\*. Four fruits were randomly picked in each replicate for each phase of the measurements, and readings were taken from three separate spots on the fruit. The colour index: L\* (white-black), a\* (red-green), and b\* (yellow-blue), were then measured and reported (Murmu and Mishra, 2017) from the CIELAB scale.

### **5.4.2 Weight loss**

At the start of the experiment, four fruits from each replicate were marked and kept separate for periodic weighing using a digital balance (Delta range, Switzerland). Following treatment (day 0), and at various intervals (days 5, 10, 15, 20, 25, and 30) during storage, the weight of individual lots was recorded. The percentage loss of initial

weight was used to calculate cumulative weight losses. Total weight loss was calculated as the difference between the initial and final fruit weights after 24 days of storage (Sharmin et al., 2015). Weight loss was determined by taking their initial and final weights differences and expressed as a percentage:

$$\text{Percentage weight loss} = \left( \frac{W_i - W_f}{W_i} \times 100 \right)$$

Where  $W_i$  = initial fruit weight

$W_f$  = final weight loss

#### **5.4.3 Firmness**

Fruit flesh firmness was measured with an 8 mm round stainless-steel probe, attached to a fruit pressure tester (Model FT 327, Effegi, Italy). With the aid of a cutting blade, a thin section of the epicarp was removed (the thickness did not exceed 1 mm). The measurements were taken on the pulp of the bananas at two different locations in their equatorial region. The mean of the measurements from each fruit sample was used to calculate the values in kilograms (kg), which were then converted to Newtons (N).

#### **5.4.4 Total soluble solids (TSS)**

The refractive index of fruit juice was measured at 20°C with a hand refractometer (Atago, Japan) to determine the soluble solids. The refractometer prism was washed and calibrated with distilled water to give a 0% reading before each sample was measured for the estimation of sucrose expressed in TSS. The banana pulp (50 g) with 150 mL of distilled water was homogenized using a blender to get a filtrate. To obtain the % TSS reading directly from the instrument, a drop of banana juice squeezed from the fruit pulp of each fruit of the three replications of each treatment was mounted on the prism glass of the refractometer. Brix was used to represent the findings (AOAC, 1995; Lopez-Palestina, 2018).

#### **5.4.5 Respiration and ethylene rate**

The banana samples (n=2) were put in a 1000 mL airtight plastic container with a rubber septum on the lid at room temperature according to Nasrin et al. (2017). An F-950 three gas analyzer (Felix Instrument, America) with a syringe was placed through



the rubber septum into the container to measure the amount of CO<sub>2</sub> produced in percentage and ethylene rate in ppm after covering for 2 h. The respiration rate and ethylene rate were converted to mg CO<sub>2</sub> emitted per kg per h and µL C<sub>2</sub>H<sub>4</sub> emitted per kg per h, respectively. The respiration rate and ethylene were then determined using the following equation:

$$\text{Respiration rate} = \frac{\text{Amount of respiration}}{\text{Weight of fruit}} \times \text{incubation time}$$

$$\text{Ethylene rate} = \frac{\text{Amount of ethylene}}{\text{Weight of fruit}} \times \text{incubation time}$$

#### 5.4.6 Disease incidence

Disease incidence means the percentage of banana infected with diseases. In this study, the apparent black patches and visible symptoms were regarded as disease, The disease incidence of bananas was calculated by using the following formula (Hossain & Iqbal, 2016):

$$\text{Disease incidence (\%)} = \frac{\text{Number of bananas infected}}{\text{Total number of bananas}} \times 100$$

#### 5.4.7 Determination of total phenolic content

Total phenolics (TP) were analysed according to Murmu and Mishra (2018) using the Folin Ciocalteu reagent (FCR) method with slight modification. Pulp tissue (3 g) was homogenized in a glass tube with 30 mL of methanol using a hand blender. They were centrifuged at 25°C for 30 min at 2500 g. The supernatant was collected and used for the analysis of total phenols. The methanol extract (0.5 mL) was mixed with 5 mL of deionized water and 0.5 mL of the Folin-Ciocalteu reagent (FCR) (Sigma Aldrich, USA) was added. After 10 min, 1.5 mL of saturated 20%(w/v) aqueous sodium carbonate solution was added to the mixture. The mixture was mixed vigorously and allowed to stand for 30 min at room temperature. The absorbance was measured at 760 nm of the reaction mixtures against the blank (methanol) using a spectrophotometer (WPA, Biochrom England). The content of phenolics in the extracts was expressed as mg of gallic acid equivalent per 100 g of the fresh weight (mg GAE/g FW).

#### 5.4.8 Consumer evaluation

On days 25 and 26 of storage, a sensory evaluation was conducted by 57 and 59 untrained panellists ranging in age from 20 to 40 years old. They were made up of University of Johannesburg students and staff. On a standard 9-point hedonic scale (dislike extremely = 1; dislike very much = 2; dislike moderately = 3; dislike slightly = 4; neither like nor dislike = 5; like slightly = 6; like moderately = 7; like very much = 8; like extremely = 9), and participants were asked to score the bananas from each treatment for colour, texture, odour, and overall acceptability. The panellists were served all the samples together, which were coded with random three-digit numbers (Shirani & Ganesharane, 2009). A consent form was given out at the start of the experiment, and the procedure was explained. Three samples corresponding to the treatments were examined by each panellist. Ethical approval and Institutional permission were obtained from the Faculty of Sciences Ethics Committee with reference number (2020-07-13/Ngobese\_odetayo) before panellists from the university were recruited for participation.

#### **5.4.9 Shelf life**

Shelf life was assessed by conducting a visual observation. In this present study, the shelf life of bananas was graded on a scale of 0 to 100% for visual yellowing of peel to brown colour daily. When bananas showed more than 80% browning, they were deemed to have reached the end of their shelf life (Hassan & Mahfouz, 2010; Taduri et al., 2017)

#### **5.4.10 Statistical analysis**

To establish the significant difference between the treated and control fruits on the same day of storage, a one-way analysis of variance (ANOVA) was done using IBM SPSS statistical software (SPSS v 27). At  $p < 0.05$ , multiple comparisons were done using the Turkey Post hoc test to separate the means.

### **5.5 Result sand Discussion**

#### **5.5.1 Colour**

Colour is a criterion used to anticipate the ripening and aging processes of bananas, which are represented by changes in the skin colour of the banana from green to yellow at the early stages of development and yellow to brown as the fruits mature

(Soradech et al., 2017). Fruit colour probably adds more to the consumer's evaluation of quality than any other aspect. As a result, the colour of the peel is a crucial post-harvest selection criterion and may also indicate its level of decay, disease infestation, maturity, and/or contamination (Mikulic-Petkovsek et al., 2015). Fig 5.1 A and B show the colour changes  $L^*$  values of ethylene treated and non-ethylene treated bananas that have been stored for 25 and 30 days respectively. As seen in Fig 5.1A, all samples had a decrease in  $L^*$  value as the storage time progressed. On day 25 of storage, the  $L^*$  value of AV+CN was 42, and the  $L^*$  value of MO+CN was 41 for ethylene treated banana. Non ethylene treated banana coated with AV+CN and MO+CN had lower  $L^*$  values (37.51 and 39.19, respectively) at the end of 30 days storage. The ethylene treated bananas had completely turned brown at day 25 so they were not further investigated (Fig 5.4 A). Based on colour readings, negative  $a^*$  values reflect the green colour of fruits, while positive  $a^*$  values portray the red colour which indicates the initiation of fruit ripening (Basulto et al., 2009). Ethylene treated fruit had negative  $a^*$  values from 0 to 5 days and turned positive from 10 to 25 days (Fig 5.2 A) while non-ethylene treated bananas had negative  $a^*$  values from day 0 to 15 and turned positive only at day 20 to day 30 (Fig 5.2 B). Non-ethylene treated bananas coated with AV+CN (6.71) had the lowest  $a^*$  value compared to MO+CN (6.73) at the end of the storage period. In comparison to AV+CN (11.08), MO+CN had a lower  $a^*$  value of 10.02 for ethylene treated bananas after storage. The progressive increase in  $a^*$  value over time for ethylene treated banana could be attributed to chlorophyll breakdown, which resulted in a reduction in greenness and an increase in redness (Chen & Ramaswamy, 2002; Gomes et al., 2013; Ding & Ling, 2014). As a result of chlorophyll breakdown, the  $b$  values of ethylene treated bananas increased from day 0 to 15 while non-ethylene treated bananas almost had the same value until a slight increase at day 20. Banana skin colour changes from green to yellow during ripening, making the skin carotenoids more visible (Gol & Rao, 2011). The results indicated that the degradation of chlorophyll pigments in fruit peel in ethylene treated fruits was more rapid than in non-ethylene treated fruits. This could be due to accelerated rate of diffusion of commercial exogenous ethylene in the peel of ethylene treated bananas which triggered the degradation of chlorophyll pigments during the peak and post climacteric stages (Tanuja et al., 2021). In non-ethylene treated fruits, there was less chlorophyll

degradation in the peel tissues in the absence of exogenous ethylene application which probably depended on ethylene produced by the pulp (Kulkarni et al., 2011). From day 15 to 25,  $b^*$  values of MO+CN dropped from 41 to 25 and AV+CN from 45 to 27 for ethylene treated bananas (Fig 5.3 A), and it had completely turned brown and lost its marketability, thus it was not examined further. In Fig 5.3 B, there was a decrease in  $b^*$  values from day 25 to 30 for both AV+CN and MO+CN coatings which showed that bananas were turning from yellow to brown for non-ethylene treated bananas (Fig 5.4 B). The statistical analysis for this study showed significant difference ( $P < 0.05$ ) for L,  $a^*$  and  $b^*$  value for ethylene treated banana but no significant difference for non-ethylene treated banana coated with MO+CN and AV+CN during storage. According to this study, AV+CN was able to preserve the colour of ethylene treated banana fruits during storage thereby delaying ripening and fruit senescence which generally results in lower fruit firmness (Rehman et al., 2020). The good effect of *Aloe vera* gel and chitosan nanoparticle could be explained by its hydrophobic feature, which acted as a gas exchange barrier between the fruit and the surrounding environment, preventing external interference (Olivas et al., 2008). For non-ethylene treated, we cannot conclude because both coating has almost same values and there was no significant difference among the coatings. According to Parven et al. (2020) and Rehman et al. (2020), *Aloe vera* gel coating delayed the colour change of papaya and guava fruits during storage when compared to control fruits. Our findings concur those of Sarduni et al. (2020), who coated bananas with pectin concentration and discovered that the colour was sustained for the 5 days of storage at room temperature.

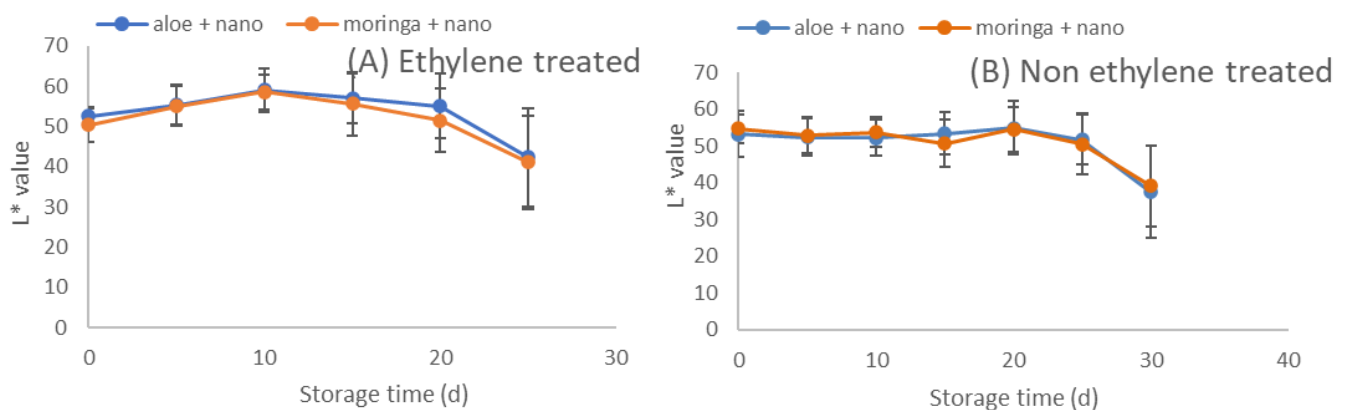


Figure 5.1: Changes in L\* values of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

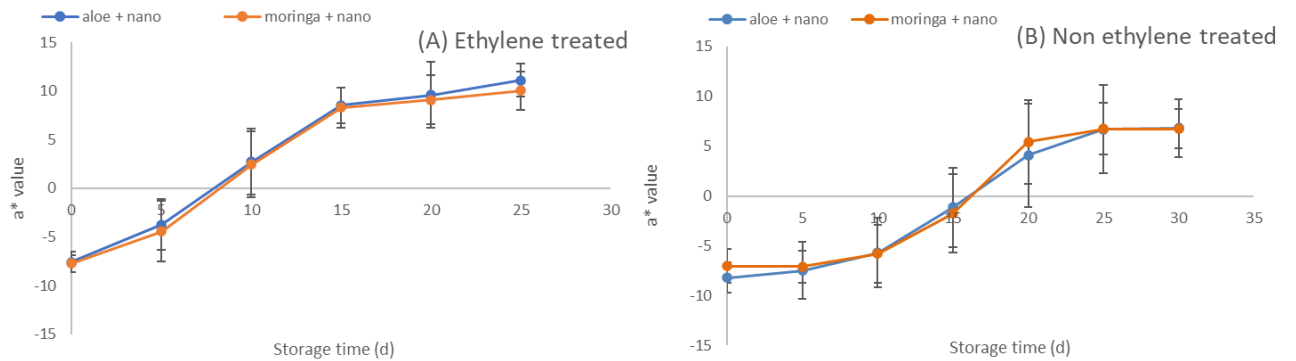


Figure 5.2: Changes in a\* values of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

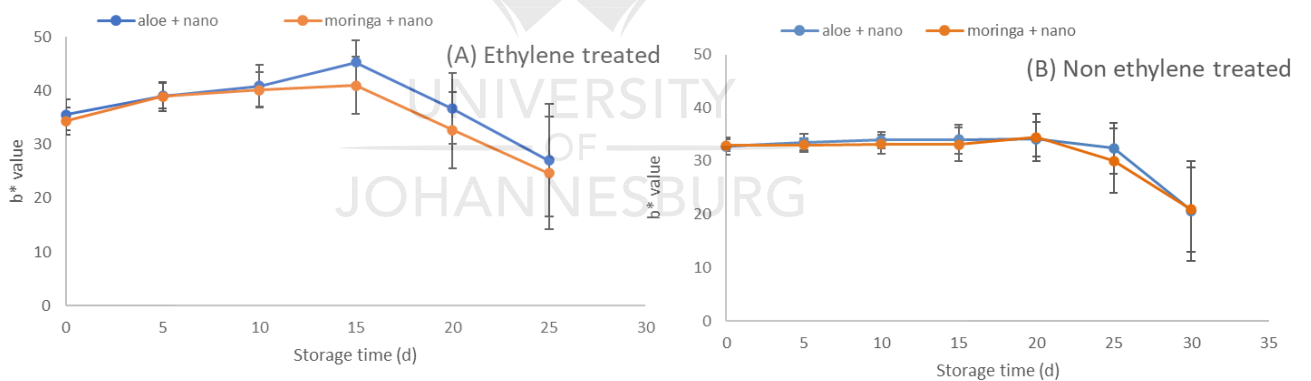
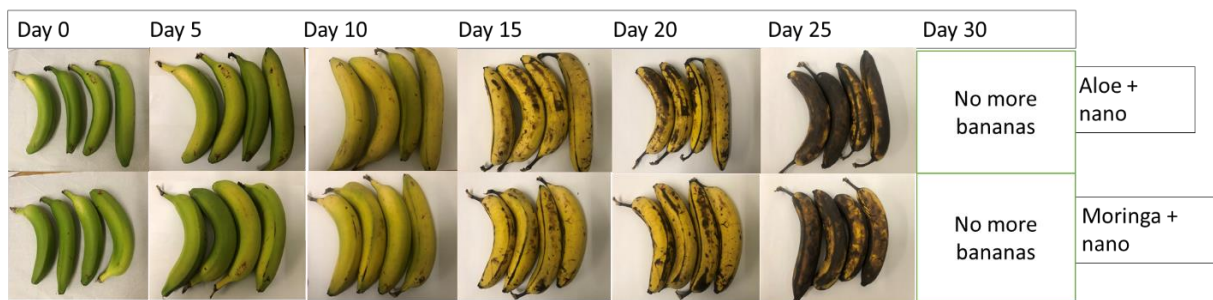


Figure 5.3: Changes in b\* values of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

(A) Ethylene treated bananas



(B) Non ethylene treated bananas



Figure 5.4: Changes in ripening stages of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH

**5.5.2 Weight loss (WL)**

Stomatal transpiration and direct evaporation are the predominant causes of water loss in fruits (Moggia et al., 2017). Reducing weight loss not only ensures good financial returns but also helps to extend the shelf life of fresh produce. As a result of the coating process, the creation of a protective coating on the surface of the bananas reduced water loss, and the inclusion of the chitosan nanoparticles increased moisture retention during storage (Kittur et al., 2001). The weight loss of ethylene-treated and non-ethylene-treated bananas coated with MO+CN and AV+CN over time is shown in Fig 5.5 A and B. Weight loss began after storage and continued throughout the storage period, regardless of the banana or coatings employed in this experiment. At day 25 of storage, MO+CN showed a lesser weight loss of 20% compared to AV+CN which had 24% weight loss for ethylene treated bananas (Fig 5.5A) while non-ethylene treated had weight loss of 21% and 22% for MO+CN and AV+CN respectively (Fig 5.5B). The ethylene treated bananas had reached the end of their shelf life at day 25 and no more measurements were taken. Exogenous ethylene may have hastened early ripening of bananas, resulting in greater weight loss. This increase in weight loss

during storage of ethylene-treated banana fruits could be owing to an increase in respiration rate, resulting in faster ripening according to Mahajan et al. (2008). At 30 days of storage, the non-ethylene treated bananas coated with MO+CN exhibited a smaller weight loss of 23%) than the AV+CN (26%) coated banana. At 30 days of storage, the non-ethylene treated bananas coated with MO+CN exhibited a smaller weight loss of (23%) than the AV+CN (26%) banana. According to this study, MO+CN coating helped minimize weight loss in both ethylene treated and non-ethylene treated bananas during storage, even though it was not statistically significant. The semipermeable barrier generated by these coatings resulted in a modified atmosphere between the fruit and the environment, resulting in a reduction in weight loss observed in coated fruit (Park, 1999). Similarly, Shahbazi and Shavisi (2020) discovered substantial variations in the WL of bananas coated with kurdi gum versus farsi gum-based coatings due to the formation of a coating on the top of the skin acting as an additional barrier. The findings were similarly consistent with those of Dong and Wang (2018), who found that incorporating ginseng extract into guar gum edible coatings reduced sweet cherry weight loss by lowering the respiration rate and water loss during storage at room temperature. According to Seyed et al. (2021), adding chitosan and calcium chloride to *Aloe vera* coatings improves their efficiency and considerably minimizes mango fruit weight loss. The antibacterial capabilities of the chitosan nanoparticles, which minimize the loss of carbon atoms generated by the respiration process, may have contributed to the weight loss reduction when chitosan nanoparticle was added to moringa coating (Belbekhouche et al., 2011; Martelli et al., 2013). This finding in this study agreed with that of La et al. (2021) who employed ZnO nanoparticles with chitosan/gum arabic coating and found that when nanoparticles were introduced, the weight loss was low in banana fruits. Previous research using alginate or chitosan supplemented with olive leaf extract on sweet cherries found similar findings, concluding that edible coatings established a physical barrier against moisture loss and lowered transpiration rates (Zam, 2019). The combination coatings of moringa extracts, CMC, and chitosan were utilized to prevent weight loss in papaya fruit, and the combined coating reduced weight loss when compared to control fruits (Langa, 2018). Pea starch and guar gum supplemented with oleic acid and shellac

were also shown to considerably reduce the weight loss of oranges during storage (Saber et al., 2018).

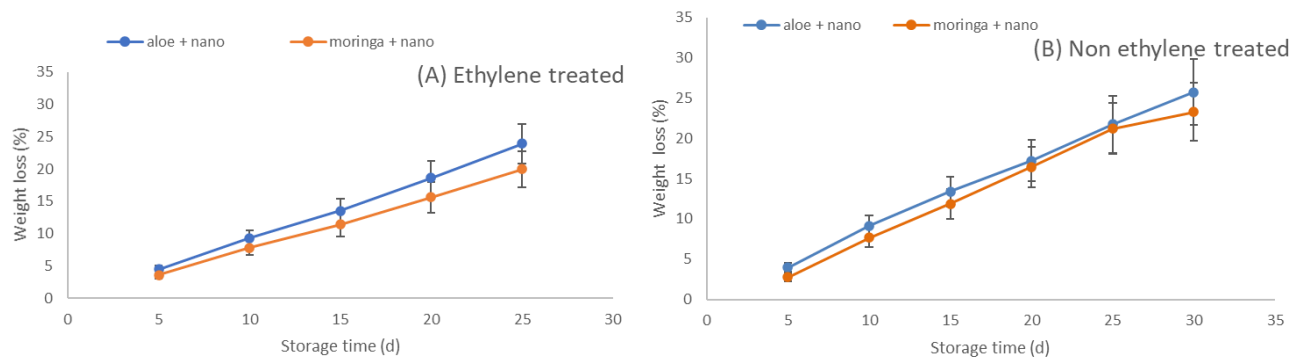


Figure 5.5: Changes in weight loss of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.3 Firmness

Fruit firmness is a key factor that determines the postharvest life and quality of fruit, according to Ali et al. (2011). Both ethylene-treated and non-ethylene-treated bananas lost firmness during storage (Fig 5.6 A and B). After being picked, bananas go through a post-ripening phase in which the texture softens as the fruit matures, eventually making the fruit perishable (Maduwanthi et al., 2017). Fruit softening is primarily caused by enzyme activity on carbohydrate polymers, which leads to degradation of the middle lamella of the cell wall of cortical parenchyma cells (Al Eryani-Raqeeb et al., 2009; Azene et al., 2014). On day 25 of storage, MO+CN edible coatings had a significantly ( $P < 0.05$ ) higher firmness of 37 N than AV+CN (34 N) for ethylene treated bananas (Fig. 5.6A). On day 30, Fig. 5.6B shows that non-ethylene treated bananas coated with MO+CN had a higher firmness of 44 N than AV+CN (40 N) coatings, but the difference was not statistically significant between the two coatings. The action of cell wall degrading enzymes, which hydrolyse starch to soluble sugars and protopectin to water-soluble pectin, is linked to the softening of fruit flesh (Ali et al., 2011). Firmness was higher in non-ethylene treated bananas compared to ethylene treated bananas with MO+CN coating having the greatest firmness value according to the



results of this investigation. This finding is consistent with that of Saltveit (1999) and Ahmad et al. (2001), who found that exogenous ethylene treatment lowered the firmness of banana fruits by increasing the softness of mature bananas due to starch hydrolysis and weight loss during ripening. Although ethylene gas application accelerated banana ripening leading to softening and breakdown of fruits, MO+CN coating considerably delayed ripening of bananas exposed to ethylene prior to coating during storage, as well as non-ethylene treated bananas compared to using AV+CN coating according to this study. The presence of bioactive substances in the edible coatings may also be responsible for the preservation of the firmness and stability of the coated fruit cell wall (Dong and Wang, 2018). Bananas coated with a rice starch edible coating blended with sucrose esters and treated with ethylene gas and stored at  $20 \pm 2^\circ\text{C}$  yielded similar results (Thakur et al., 2019). Fresh fruits treated with edible biopolymer coatings, such as bananas (Maqbool et al. 2011; Lo'ay & Dawood 2017), mangos (Kittur et al. 2001), and strawberries (Azarakhsh et al. 2014; Shahbazi 2018), exhibited superior firmness quality than untreated ones.

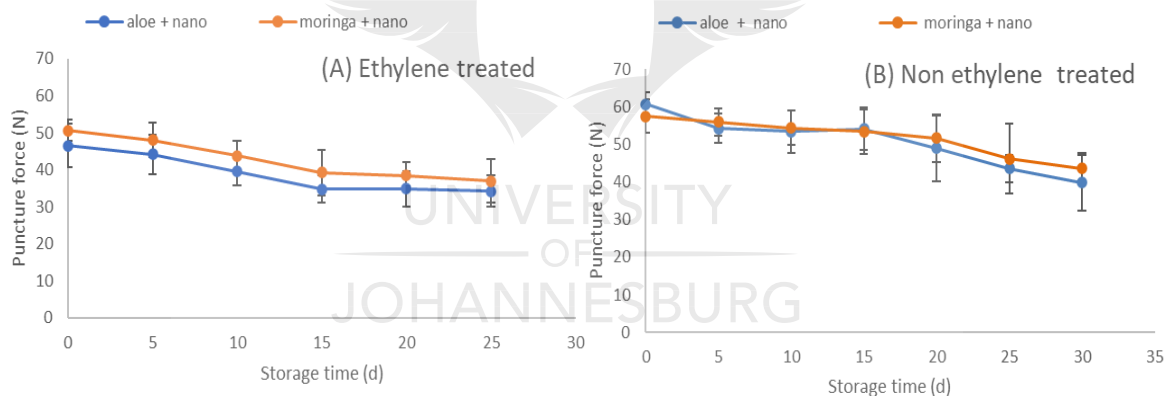


Figure 5.6: Changes in firmness of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

#### 5.5.4 Total soluble solids (TSS)

The amount of TSS in the fruit increases as it matures and ripens, thus the soluble solids content of the fruit can be used as a valuable indicator of maturity or ripeness stage (Ngnambala, 2013). TSS ( $^\circ\text{Brix}$ ) increased consistently from the first to the last day of storage for both ethylene and non-ethylene treated bananas at room

temperature, as shown in (Fig 5.7 A and B). Total soluble solids are an essential component in fruit consumer acceptance. The steady increase in total soluble solids during fruit storage could be attributed to the breakdown of cell wall polysaccharides (Sun et al., 2013) or an increase in sugar concentration as a result of the higher respiration rate of the fruit (Burdon et al., 2016). At days 25 and 30, the AV+CN coating had the highest TSS value (39 °Brix, 33 °Brix), while the MO+CN coating had the lowest TSS value (37 °Brix, 32 °Brix) for ethylene treated and non-ethylene treated bananas, respectively. The fruits coated with MO+CN treatment, which provided an excellent semipermeable layer that modified the internal atmosphere by inhibiting ethylene generation, had the lowest TSS at the end of the storage period (Ali et al., 2010). The higher increase in the TSS of the ethylene treated bananas may be associated with the faster degradation of starch and dehydration of fruit during storage. Moreover, a similar explanation for the increase in TSS during storage has been provided by Thakur et al. (2019) who studied the effect of rice starch edible coating blended with sucrose esters on controlling the postharvest physiological activity of ethylene treated Cavendish bananas at  $20 \pm 2^{\circ}\text{C}$ . The moringa enriched chitosan nanoparticles successfully slowed the increase in TSS for both ethylene treated and non-ethylene treated banana according to this finding. Chitosan nanoparticles may cause a decrease in the soluble solids content of bananas by decreasing respiration and metabolic activity in the fruit, causing the ripening process to be delayed (Lustriane et al., 2018). When chitosan is given as nanoparticles, this feature is improved or strengthened, as evidenced by the reduced concentration of soluble solids in bananas coated with edible coatings and nanoparticles. In papaya (Ali et al., 2011) and banana (Suseno et al., 2014; Zahoorellah et al., 2017), chitosan treatment has been shown to reduce fruit TSS during storage. A similar result was reported when Shah & Hashmi 2020, treated mango with chitosan- *Aloe vera* coating, where there was a reduction in TSS value for chitosan + *Aloe vera* treated fruits compared to chitosan alone and the control.

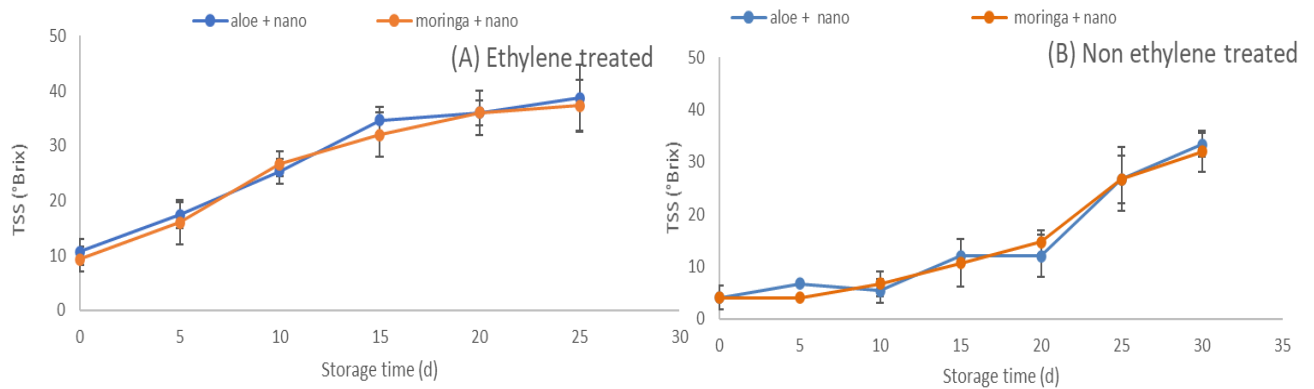


Figure 5.7: Changes in total soluble solids (TSS) of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.5 Respiration rate (RR)

Ripening in climacteric fruits like bananas is marked by a considerable and quick increase in respiration rate, as well as extensive metabolic change (Wills & Golding, 2016). Ethylene-treated bananas coated with AV+CN had a lower respiration rate (41.43 mg/kg/h) on day 0 and increased until day 15, before falling to 38 mg/kg/h on day 20 to 25, whereas ethylene-treated bananas coated with MO+CN had a higher value (49 mg/kg/h) on day 0 and reduced to 28 mg/kg/h on day 25 of storage. MO+CN treatment exhibited a significantly reduced respiration rate ( $p < 0.05$ ) than AV+CN treatment for ethylene treated banana fruits after 25 days (Fig. 5.8A). Non-ethylene treated bananas coated with AV+CN had a lower respiration rate (16 mg/kg/h) at day 0, increasing to 45 mg/kg/h at day 20, then falling to 30 mg/kg/h at day 30, whereas non-ethylene treated bananas coated with MO+CN had a lower value (18 mg/kg/h) at day 0, increasing to (55 mg/kg/h) at day 20, then falling to 18 mg/kg/h at day 30 of storage (Fig. 5.8B). The lower respiration rate in non-ethylene treated banana fruits is attributed to the less gas interchange and therefore the lower oxygen availability to the fruit tissues for respiration (Barman et al., 2011). By producing a modified atmosphere surrounding the fruit, edible coatings limit the exchange of gases and water vapor, lowering the fruit's respiration rate and protecting fruit quality (Raghav et al., 2016). Application of ethylene gas to banana may have accelerated fruit senescence which may be linked to the increase in respiration rate in ethylene treated fruit. The

exogenous ethylene treatment can induce ripening of bananas with increased rate of respiration and increased level of endogenous ethylene (Dominguez & Vendrell, 1994; Maduwanthi & Marapana, 2019). According to this study, both ethylene-treated and non-ethylene-treated bananas were reported to have lower respiration rates after 25 and 30 days of storage when coated with MO+CN respectively. Mthembu et al. (2018) investigated the influence of moringa-based edible coatings on papaya quality and shelf life. Their findings revealed that treated papaya fruit with moringa-based edible coatings had a significantly lower respiration rate. The results of this investigation were comparable to those of other studies that used rosehip essential oil and *Aloe vera* gel as edible coatings on plum (Martinez-Romero et al., 2017) and *Aloe vera* gel and chitosan on strawberry (Nasrin et al., 2017). Furthermore, moringa-based edible coatings inhibited fungus growth. Finally, a moringa-based edible coating may be employed by the fruit industry, and it would be preferable to synthetic chemicals because it poses no health risks.

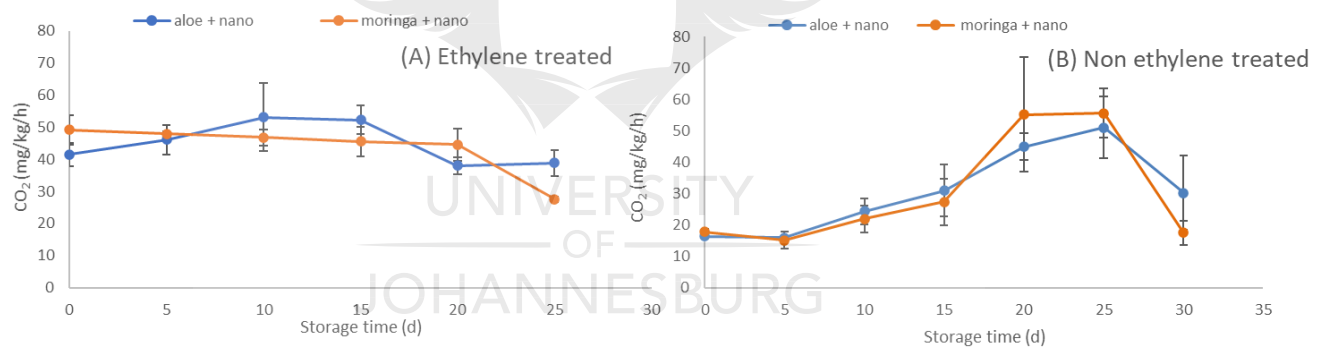


Figure 5.8: Changes in respiration rate of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.6 Ethylene

Ethylene is a physiologically active compound that occurs naturally in plants and is thought to be a hormone. It is biologically active and is responsible for the ripening of fruit. When climacteric fruits, such as bananas, begin to ripen, ethylene production

skyrockets, coinciding with a sharp increase in respiration (Kader, 2002). One of the most important physiological indicators of ripening and senescence during storage is fruit ethylene production (Ahmed & Palta, 2016). Fig 5.9 A and B shows the differences in ethylene production between ethylene-treated and non-ethylene-treated bananas coated with MO+CN and AV+CN. For ethylene treated banana, MO+CN had a significantly ( $P < 0.05$ ) lower ethylene rate ( $306 \mu\text{L/kg/h}$ ) than AV+CN ( $449 \mu\text{L/kg/h}$ ) at day 25 (Fig. 5.9A). The ethylene rate of MO+CN ( $144 \mu\text{L/kg/h}$ ) was significantly ( $p < 0.05$ ) lower than of AV+CN ( $227 \mu\text{L/kg/h}$ ) for non-ethylene treated bananas at day 30 (Fig. 5.9B). The commercial exogenous ethylene treatment applied on bananas can induce ripening with increased rate of respiration and increased level of endogenous ethylene rate (Dominguez & Vendrell, 1994; Maduwanthi & Marapana, 2019). This might be responsible for why ethylene rate was higher in ethylene treated bananas compared to non-ethylene treated ones at the end of storage. MO+CN outperformed AV+CN in terms of ethylene production rates after storage for both ethylene treated, and non-ethylene treated bananas. These findings confirm that the MO+CN coating reduced banana ethylene production, thus delaying ripening and senescence. This reduction could be achieved by constructing a semipermeable membrane over the fruits, which alters the inside environment, delaying metabolic activity and potentially reducing ethylene production (Khaliq et al., 2016). These findings agree with earlier research by Naeem et al. (2018), Shahbazi. (2018), and Shahbazi & Shavisi. (2020) demonstrating that edible coatings can help slow down the ethylene rate. MO+CN, on the other hand, reduced the amount of ethylene produced. Similarly, Shah and Hashmi (2020) found that when a composite coating based on chitosan and *Aloe vera* was applied to bananas, the production of ethylene was reduced. Furthermore, the combination of *Aloe vera* gel and rosehip oil in plum, nectarine, and peach dramatically reduced ethylene production (Paladines et al. 2014). Therefore, the combination uses of moringa, and chitosan nanoparticles suppressed ethylene production in banana fruit, according to this study.

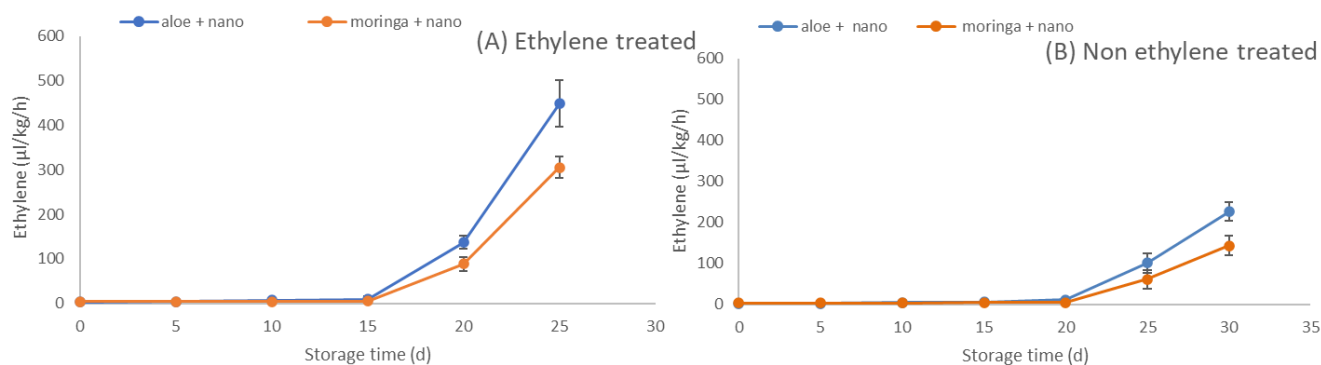


Figure 5.9: Changes in ethylene production of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.7 Disease incidence

One of the major issues affecting the quality of fresh horticulture products during storage is postharvest diseases (Sapper et al., 2019). During the ripening phase, the presence of diseases in fruit becomes increasingly apparent. Several diseases commonly reveal themselves in ripe fruits. The ethylene-treated banana coated with the MO+CN showed a significantly reduced disease incidence ( $p < 0.05$ ) than the AV+CN after 25 days of storage, as shown in Fig 5.10A. On day 15, disease incidence in ethylene-treated bananas coated with MO+CN was 33%, climbed to 58% on day 20, and reached 75% on day 25, while AV+CN had 42% on day 15, 75% on day 20, and rose to 100% on day 25. The non-ethylene treated bananas coated with the MO+CN had a lower disease incidence than the AV+CN after 30 days of storage, but there were no statistically significant differences. Disease incidence in non-ethylene-treated bananas covered with MO+CN was 8% on day 20, grew to 33% on day 25, and peaked at 58% on day 30, while AV+CN had 33% on day 20, 33% on day 25, and climbed to 67% on day 30 (Fig. 5.10B). Ethylene treated bananas developed disease incidence only at day 15, while non-ethylene treated bananas developed disease incidence only at day 20, with MO+CN coatings having the lowest rate in both bananas. This is because edible coatings can not only slow down the rate of respiration and ripening, but they can also greatly slow down the growth of bacteria, fungi, and molds, all of which are known to cause disease incidence in banana fruits.

In the current study, the barrier created by using composite coatings inhibited pathogen growth and retarded the ripening and senescence process of banana fruit, resulting in lower disease incidence according to Murmu and Mishra. (2018). Our findings agreed with Tabassum et al. (2018) and Rahman et al. (2020) who found a reduced rate of disease incidence in banana samples, with clear evidence of treated samples being more acceptable than control samples. Edible coatings enhanced with plant extracts such as *Ficus hirta* successfully reduced disease incidence in coated Naveloranges according to investigations of Chen et al. (2018).

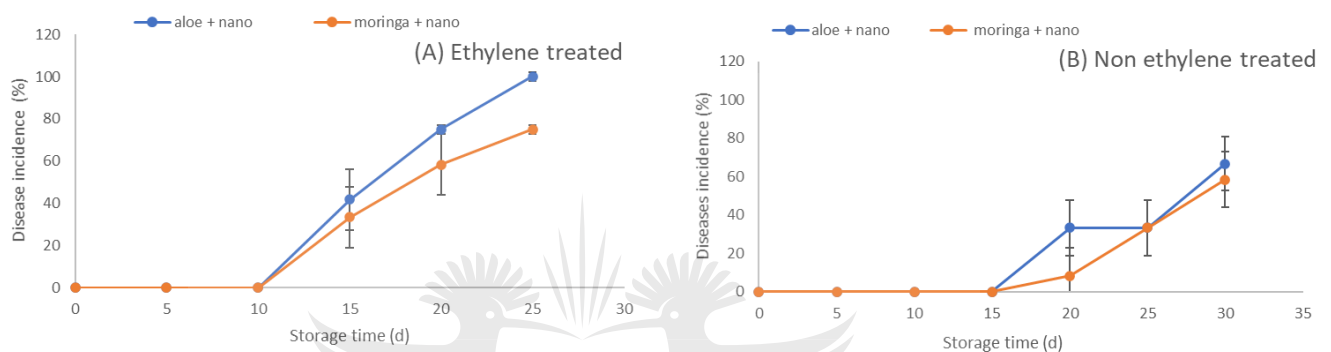


Figure 5.10: Changes in disease incidence of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.8 Total phenolic content

The total phenolic content of ethylene-treated and non-ethylene-treated bananas coated with MO+CN and AV+CN during storage is shown in Fig. 5.11 A and B. Fruit phenol content gradually decreased throughout the first 10 days of storage for ethylene treated bananas but increased in non-ethylene treated bananas. The activity of peroxidase and polyphenol oxidase enzymes is thought to be responsible for the decrease in phenolic compound concentrations in all fruit samples till the end of storage (Ali et al., 2011; Chiabrando & Giacalone, 2015). The total phenolic content of ethylene-treated bananas differed significantly ( $p < 0.05$ ) on days 1, 10, and 25 of storage. For ethylene treated bananas at day 25, *Aloe vera* supplemented with chitosan nanoparticles (27 mg GAE/g FW) exhibited significantly greater phenolic compound retention, followed by moringa enriched with chitosan nanoparticles (20 mg

GAE/g FW). On day 30, the phenolic content of non-ethylene treated bananas coated with MO+CN was higher (22 mg GAE/g FW) than that of AV+CN (19 mg GAE/g FW). Such results could be attributable to the edible coating's general preservation effects, which acted as a barrier to gaseous exchange, reducing oxidation processes and delaying fruit senescence (Maqbool et al., 2011; Alali et al., 2018). Hassan et al. (2018) found that edible coatings reduced gas exchange and respiration, which suppresses phenolic component oxidation. The current experiment's findings were in line with those of Tzortzakis et al., (2019), who found that coating tomato fruit with *A. vera*-sage essential oil improved the preservation of phenolic components during storage.

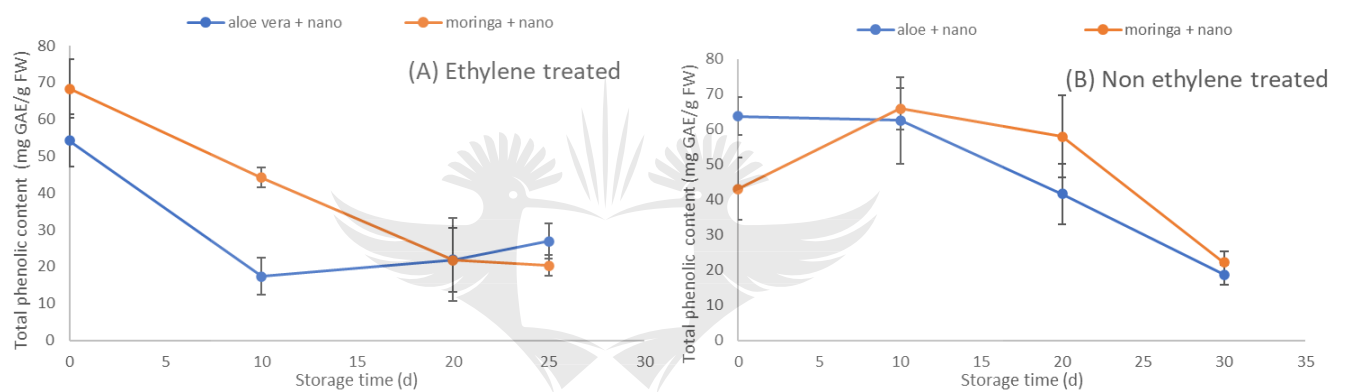


Figure 5.11: Changes in total phenolic content of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

### 5.5.9 Consumer evaluation

Consumer acceptance of fruit treated with edible coatings is a major concern, as these coatings can alter the fruit's sensory properties. Because treatments and storage time can change the quality of fruit, and edible coatings are usually consumed with the food product, the sensory aspect is very important (Rojas-Graü et al., 2009). Sensory investigation of ethylene-treated and non-ethylene treated banana storage showed changes in colour, texture, odour, and overall acceptability (Fig. 5.12 A and B). After 25 days, the MO+CN treatment had considerably greater colour (5.0 score), texture



(6.0 score), odour (6.0 score), and overall acceptability (6.0 score) ratings than AV+CN which had colour (5.0 score), texture (6.0 score), odour (6.0 score), and general acceptability (5.0 score) for ethylene treated bananas (Fig. 5.12A). The MO+CN treatment had higher colour (6 score), texture (7.0 score), odour (7.0 score), and overall acceptability (6.0 score) compared to the AV+CN treatment, which had lower colour (5 score), texture (6.0 score), odour (6.0 score), and overall acceptability (5.0 score) for non-ethylene treated banana ratings after 26 days of storage (Fig. 5.12B). MO+CN received the top ratings in terms of colour, texture, and odour, as well as overall acceptance for both ethylene treated and non-ethylene treated bananas. These sensory results are in line with parameters such as texture where MO+CN retain firmness of bananas compared to AV+CN and disease incidence rates were reduced which might have contributed to the acceptability scores of bananas by the panellists. Non-ethylene treated bananas coated with MO+CN were accepted by 67% and AV+CN by 61% of the panellists while non-ethylene treated bananas coated with MO+CN was accepted by 64% and AV+CN by 56% of the panellists from the result of the frequency test of this study. So, these further buttresses the claim that MO+CN and AV+CN treatments can be employed as an edible coating to extend the shelf life of banana fruits and improve their quality during storage because they were accepted by more than half of the panellists. According to Shahbazi & Shavisi, (2020), integrating ethanolic *Prosopis farcta* extracts into Kurdi gum (KG) and Farsi gum (FG) based coatings on banana fruits resulted in more acceptable sensory scores than the untreated samples. Similar results were seen in a prior study by Kubheka et al. (2020), who found that avocado fruits coated with CMC and Moringa had the highest overall acceptability compared to other treatments.

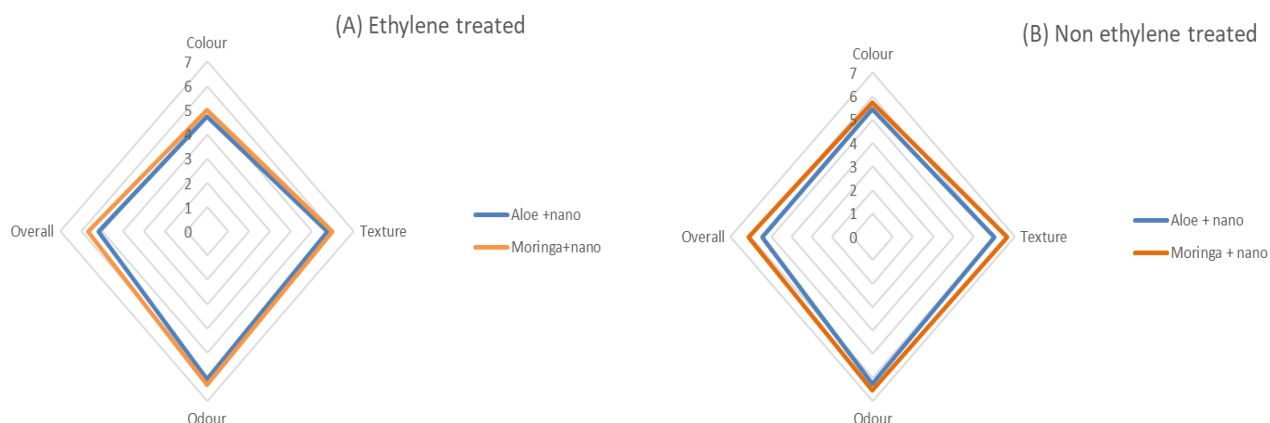


Figure 5.12: Sensory evaluation scores of colour, texture, odour, and overall acceptability of ethylene treated bananas (A) and non-ethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Mean of 57 and 59 panellists for each sample using the following hedonic scale: 1 = dislike 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

### 5.5.10 Shelf life

Banana is a typical climacteric fruit, once ripening has been initiated with ethylene, the fruit exhibits a short shelf life because of rapid quality deterioration (Xu et al., 2007). Fig. 5.13 A and B depicts the impact of AV+CN and MO+CN edible coatings on the shelf life non-ethylene treated and ethylene treated bananas stored for 25 and 30 days respectively. The longest banana shelf life was found to be 30 days in non-ethylene treated bananas coated with MO+CN followed by AV+CN for 28 days (Fig. 5.13B), whereas the shortest shelf life was found to be 25 days in ethylene treated bananas coated with MO+CN and AV+CN (Fig. 5.13A). Slower physiological processes such as respiration and weight loss linked with less microbiological activity may contribute to the banana's longer shelf life. According to this study non-ethylene treated bananas coated with MO+CN had the longest shelf life of 30 days while ethylene treated bananas has shortest shelf life of 25 days. Exogenous ethylene treatment induces a sequence of physiological and biochemical changes in banana fruit leading to the development of characteristic soft flesh tissue and short shelf life that impacts on consumer acceptability (Valérie Passo Tsamo et al., 2014). This might

be the reason why non-ethylene treatment had longer shelf life than ethylene treatment according to this study. Coating fruits with chitosan nanoparticles and chitosan coatings to extend their shelf life has also been documented in banana (Lustriane et al., 2018), strawberry (Eshghi et al., 2014), litchi (Dong et al., 2004), longan (Jiang & Li 2001), and mango (Chien et al., 2007). Oranges were shown to have a longer shelf life and better quality when moringa was combined with CMC (Adetunji et al. 2013). As a result of the usage of edible coatings and chitosan nanoparticles, the shelf life of bananas was extended in the current study.

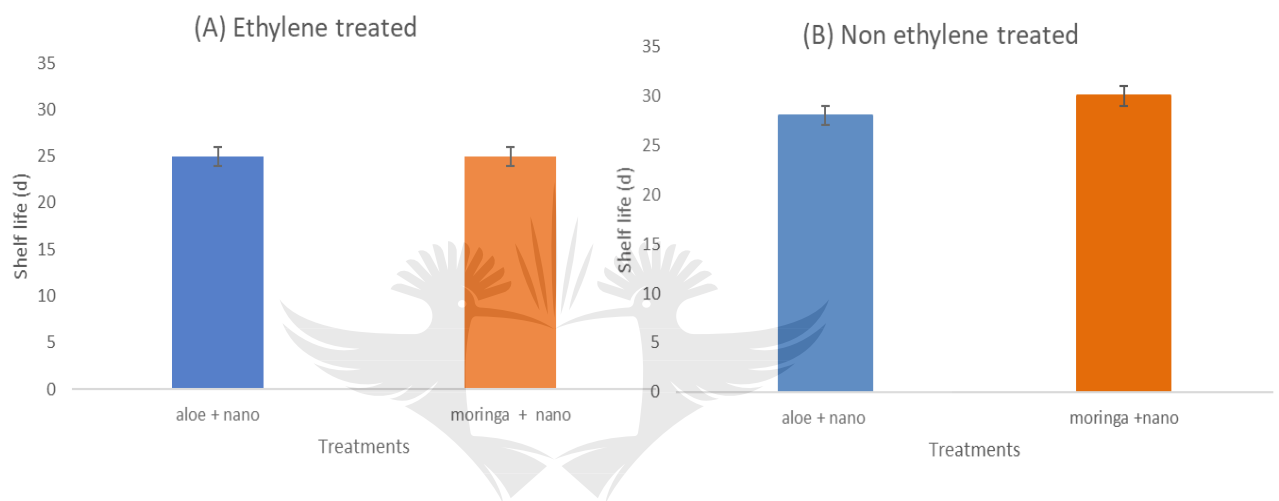


Figure 5.13: Shelf life of ethylene treated bananas (A) and nonethylene treated bananas (B) stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Error bars represent standard error (SE) at  $n = 3$

## 5.6 Conclusion

In addition to ethylene treatment and cold storage, both of which are employed in the fresh produce sector, edible coating of moringa with inclusion of chitosan nanoparticle is a creative and unique technique to maintaining the quality of banana for extended periods of time. The delay in ripening was observed in both ethylene-coated and non-ethylene-coated bananas treated with MO+CN, according to this study. By reducing weight loss, firmness, and respiration rate during storage, the moringa plus chitosan nanoparticle coatings on the fruit surface resulted in a delay in senescence. Furthermore, the sensory study revealed that the MO+CN coating had a higher overall

acceptance score for both ethylene treated, and non-ethylene treated bananas than using AV+CN coating. Fruits have long been treated with edible coatings containing natural compounds to enhance their shelf life. For the first time, this study discovered that moringa plant extracts in combination with chitosan nanoparticles improved post-harvest quality of both ethylene treated and non-ethylene bananas. Overall, the shelf life of non-ethylene treated bananas coated with MO+CN was extended by 4 more days which is a good advantage for the farmers who don't have access to ethylene treatment and cold storage. To expand the study on other fruits and vegetables, more research should be done to establish additional qualities of coatings created from diverse plant extracts supplemented with chitosan nanoparticles.

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## CHAPTER 6: CONCLUSIONS AND RECOMMENDATIONS FOR FURTHER RESEARCH

### 6.1 Introduction

Cavendish banana are among the most important sub-tropical fruits grown in South Africa and are planted for sale in local markets or self-consumption and only a fraction of all bananas is sold in the world market. Bananas are characterized as climacteric fruits because of their respiration behaviour and ethylene production. This means that after harvesting, metabolic activity and ripening continue, making banana fruits perishable commodities. Also, banana fruit ripens quickly, resulting in considerable physicochemical and biochemical changes such as starch to sucrose conversion, mineral reduction, and flavour development once picked, making green banana fruit highly perishable (Mohapatra et al., 2010; Alkarkhi et al., 2011). The quality features of banana fruit, such as colour, texture, and flavour, decline quickly after harvest resulting in a limited shelf life of about 6–8 days under ambient storage (Jiang et al., 1999; Boudhrioua et al., 2003). Thus, extending the shelf life of bananas is a pressing need that has drawn widespread attention; to preserve the freshness of fruits, a variety of technologies, including but not limited to cold storage, climate-controlled storage, edible coatings, and hypobaric storage, have been extensively used. While cold, modified atmosphere, and controlled atmosphere storage technologies are regarded expensive means of preserving bananas, edible coatings are dependable and cost-effective techniques of extending the shelf life of bananas by slowing down ripening processes for an acceptable period. One of the promising factors in the preservation of climacteric fruits like bananas is the edible coating. Because of its human health benefits and antibacterial capabilities, *Aloe vera* gel and moringa have received attention from the scientific community as one of the promising bio-preservatives. Edible coatings formulated from plant extracts are biodegradable compounds with antifungal properties that can improve the shelf life of fresh produce. Therefore, the aim of this study was to evaluate the effect of edible coating of *Aloe vera* and moringa coatings and addition of chitosan nanoparticle on postharvest quality of Cavendish banana fruits at room temperature.

## **6.2 To evaluate the effect of selected edible plant extract-based coatings on shelf life of Cavendish bananas**

The effects of *Aloe vera* and moringa plant extract coatings on the shelf life of bananas was explored in Chapter 3. The use of edible coatings based on *Aloe vera* and moringa on banana fruits has been demonstrated to slow down the ripening process. Banana ripening is the consequence of a complex set of internal and external changes that are linked to the internal atmosphere and metabolism of the banana as it ripens. In comparison to the control, the results showed that both coatings reduced the ripening process and maintained banana quality. This may be due to the coating's capacity to delay weight loss, fruit firmness decline, delay in TSS increase, retain protein and mineral content and overall phenolic loss reduction. Furthermore, coated bananas had a shelf life of 24 days, but control bananas had a shelf life of 20 days, implying that coating with *Aloe vera* and moringa only extended shelf life of banana by 4 days. As a result of this finding, chitosan nanoparticles were added to the aloe and moringa plant extract edible coatings in Chapter 4 to improve the coating properties.

## **6.3 To evaluate the effect of nanoparticle enriched plant extract based coatings on shelf life of Cavendish bananas**

The shelf life and sensory quality of Cavendish bananas were studied when *Aloe vera* and moringa were combined with chitosan nanoparticles. Quality parameters such as weight loss, firmness, colour, respiration, and ethylene rate, disease incidence, TSS, total phenolic content, mineral analysis, and consumers test were all analysed. When compared to AV+CN, AV, and MO at room temperature, MO+CN significantly ( $p < 0.05$ ) slowed down firmness loss, ethylene and respiration rate. It also reduced disease incidence and TSS rise but there were no statistically significant differences reported. It has been proven that adding chitosan nanoparticles to edible coatings not only improves the quality characteristics listed above, but also extends the shelf life by 10 days compared to edible coating alone which extended shelf by 4 days from Chapter 3 findings. However, regardless of the treatment applied to the banana fruits, the overall acceptability of the fruits determines the treatment's effectiveness or recommendation. Our research revealed that edible coatings had considerable impacts on overall acceptability of 'Cavendish' banana fruits, with MO+CN and AN+CN

coated fruits having the highest acceptability and AV and MO coated banana fruits having the lowest acceptability. Overall, MO+CN's superior capacity to maintain fruit texture and overall quality during storage may have aided in the retention of greater sensory qualities than other coatings. Also, the frequency test showed it was accepted by more than 50% of participants that took the consumer test. We may conclude from this research that MO+CN coatings were effective in improving the shelf life of banana fruits at room temperature.

#### **6.4 To evaluate the effect of nanoparticle enriched plant extract –based coatings on shelf life of ethylene treated Cavendish bananas**

The samples utilized in this chapter were ethylene treated and non-ethylene treated bananas. For the evaluation of chosen quality criteria at room temperature, edible coating of *Aloe vera* and moringa enriched with chitosan nanoparticle were applied to external surfaces of bananas. In comparison to the non-ethylene treated bananas coated with MO+CN and AV+CN which had a shelf life of 28 and 30 days respectively, ethylene treated bananas had shelf life of 25 days for both coatings. In conclusion, our findings show that moringa coating combined with chitosan nanoparticles considerably slowed ripening and can extend postharvest life by delaying fruit quality parameters such as fruit firmness, TSS, colour, weight loss, respiration, and ethylene rate, total phenolics, and disease incidence for both ethylene treated and non-ethylene treated bananas stored at  $18 \pm 1^\circ\text{C}$  and 35-45% RH. Although exogenous ethylene application promoted the ripening of the bananas, the use of MO+CN coating significantly delayed the ripening of the ethylene treated bananas. According to this study, MO+CN coating was more effective for prolonging the storage life of bananas without ethylene application for up to 30 days compared with ethylene treated ones which had shelf life of 25 days. Finally, the sensory study revealed that the MO+CN coated fruit had a higher visual acceptance from consumers test than AV+CN coating. These findings show that MO+CN could be a valuable biochemical tool for preserving banana fruit quality and extending postharvest life. However, more research is needed to completely comprehend the process by which MO+CN coatings affect ethylene mechanism in banana and the effect of coatings on phytochemicals and antioxidant activities of bananas.

## 6.5 Future possibilities

In South Africa, the use of edible coatings of *Aloe vera* gel, moringa leaf extract and chitosan nanoparticles seems to be a novel technique that has not been explored before in the banana industry. Therefore, this study could be a starting point for more research into the impact of nanoparticle coating on biochemical and molecular characteristics throughout the ripening process of fruits. Although the results of this study appear encouraging, we only looked at one concentration level for edible coating and chitosan nanoparticle in this investigation. In the future, different concentrations of edible coatings and chitosan nanoparticles should be investigated. Furthermore, while the treatments were evaluated on a small scale in a controlled laboratory environment, it still needs to be tested on a commercial large scale to see how it responds before it can be used confidently in the banana fruit sector. Additionally, we also need to test the antifungal effect of these coatings against a major disease in banana which is Anthracnose caused by the fungus *Colletotrichum musae* which results in major economic loss during storage. Doing all the above will further strengthen our research findings and see the possibility of adopting these coatings in the future for the banana fruit industry. It is hoped that the data from this study will be of great importance and provide understanding about how best to extend the shelf life of not only banana fruits, but also other climacteric fruits such as mango, avocado, papaya, and other similar fruits.

## 6.6 Final comments and summary conclusions

The goal of this study was to examine how employing edible coatings alone or in combination with chitosan nanoparticles in edible coatings might help preserve the postharvest quality of a popular banana cultivar (Cavendish) and how it can complement current fresh produce technology. In the fresh produce market, the process involves inducing bananas with ethylene gas for a few days and then storing them in cold storage to achieve the appropriate aesthetic standards and fruit quality before being sold by market agents. The picture showing changes in ripening stages of banana in Fig 5.4 A and B depicts even yellow colour for bananas treated with ethylene and non-treated ones at the end of storage meaning the coatings work quite well with ethylene treated banana and non-ethylene treated ones. The use of chitosan

nanoparticles with edible coatings could be a novel way to keep banana fruit fresh after harvest. Disease incidence, weight loss, and delayed changes in banana fruit associated to ripening were all reduced by adding chitosan nanoparticles in edible coatings. Total phenolic loss was successfully decreased using the same technique. Incidence of postharvest disease in fresh produce is a well-known supply-chain constraint that has a negative impact on their marketability. Antifungal and antibacterial activities of *Aloe vera*, moringa and chitosan nanoparticles have been reported. These findings imply that using an edible coating enhanced with chitosan nanoparticles to delay physicochemical changes and maintain nutritional characteristics in banana fruit could be an environmentally acceptable strategy to delay physicochemical changes during room temperature storage. It is also worth noting that a composite coating of moringa and chitosan nanoparticles improved the post-harvest characteristics of both ethylene and non-ethylene treated bananas when stored at room temperature. As a result, these coatings can assist farmers in extending the shelf life of bananas as well as improving the quality of bananas after harvest in the fresh produce chain.

## 6.7 References

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