

Physicochemical, antioxidant and microstructural properties of malted finger millet (*Eleusine coracana*) flour(s) and their use in gluten-free biscuits.

By

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**Research Dissertation for the
Master of Science Degree in the Field of Study
in the Department of Food Science and Technology,
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ABSTRACT

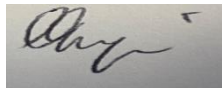
Finger millet is a gluten-free crop with outstanding malting qualities, and a low glycaemic index. The study aimed to determine the physicochemical, antioxidant, and microstructural properties of malted finger millet flours (light and dark brown) and their use in gluten-free biscuits. The two varieties of finger millet grains were germinated for 0, 24, 48, and 72 hours (h) and kilned for 8 h at 30°C. The lightness (L^*) values of malted finger millet flours significantly increased ($p \leq 0.05$), with light brown having the highest L^* value of 76.62. The moisture, ash, fibre, protein, mineral, total phenolic and total flavonoid contents and DPPH activity of both malted finger millet flours and biscuits significantly increased ($p \leq 0.05$). Both malted finger millet flours' solubility index, water and oil absorption capacity increased significantly, while the packed and loose bulk density decreased. A significant decrease in the viscosity of the cooked paste in both finger millet flours was observed. Malting resulted in an increase in the onset, peak, and conclusion temperatures. FTIR showed that malting slightly changed the peaks of both finger millet flours. Scanning electron microscopy showed that malting altered the microstructural characteristics of finger millet flours. The yellowness and whiteness index of FM biscuits significantly increased, ranging from 41.47 to 42.12 and 42.21 to 45.47 (light brown) and from 67.78 to 70.97 and 71.92 to 75.41 for dark brown, respectively. The weight, diameter, thickness, and spread ratio significantly decreased with an increase in malting in both FM biscuits. The hardness of the biscuits significantly increased from 762.53 to 1720.28 g (light brown) and 747.50 to 1918.71 g (dark brown). The LB biscuits had more desirable sensory attributes as compared to the DB biscuits. It was concluded that malted FM flour can be used to prepare nutritious and healthier gluten-free bakery products.

Keywords: Germination, functional, millet, morphological characteristics, polyphenolic compounds, and sensory properties.

DECLARATION

I, Kundai Thelma Murungweni, student number 17001077, hereby declare that the dissertation I submitted for the completion of the Master of Science degree in Food Science and Technology (MSc 6399) at the University of Venda, has not been submitted previously for a degree at this institution or any other Institution, it is my individual and original work in design and accomplishment, and all the references therein have been properly acknowledged.

Signature:



Date: 16 April 2024

Murungweni K.T

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PARTICIPATION AND CONVENTION ATTENDED

Participated in Faculty Research open day at the University of Venda. Presented a poster entitled Malting of finger millet flours and their utilization in formulation of gluten-free biscuits 1st of March 2022.

I attended the BIO Africa Convention held at the Durban International Convention Centre from the 3rd to the 6th of September 2023.

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LIST OF ACRONYMS

| | |
|---|-----------------------------------|
| a* | Redness |
| AlCl ₃ | Aluminium chloride |
| ANOVA | Analysis of variance |
| b* | Yellowness |
| Ca | Calcium |
| CW | Cell wall |
| DB | Dark brown |
| DPPH | 2,2-diphenyl-1-picrylhydrazyl |
| DSC | Differential Scanning Calorimeter |
| FM | Finger millet |
| Fe | Iron |
| FM | Finger millet |
| FRAP | Ferric reducing antioxidant power |
| FTIR | Fourier transform-infrared |
| GAE | Gallic acid equivalent |
| H° | Hue angle |
| K | Potassium |
| K ₃ [Fe (C N) ₆] | Potassium ferricyanide |
| LB | Light brown |
| L* | Lightness |
| LBD | Loose bulk density |
| M | Malted |
| Mg | Magnesium |
| Mn | Manganese |
| Na | Sodium |
| NaOH | Sodium hydroxide |
| NaNO ₂ | Sodium nitrite |
| Na ₂ CO ₃ | Sodium carbonate |
| OAC | Oil absorption capacity |
| P | Phosphorus |
| PB | Protein bodies |
| PBD | Packed bulk density |
| QE | Quercetin equivalent |
| SC | Swelling capacity |

| | |
|----------------|------------------------------|
| SEM | Scanning electron microscopy |
| SR | Spread ratio |
| SG | Starch granules |
| TFC | Total flavonoid contents |
| T _c | Conclusion temperature |
| T _o | Onset temperature |
| T _p | Peak temperature |
| TPC | Total phenolic contents |
| T _r | Temperature range |
| WI | Whiteness index |
| YI | Yellowness index |
| Zn | Zinc |
| ΔE | Total colour difference |
| ΔH | gelatinisation enthalpy |

LIST OF SI UNITS

| | |
|------|----------------|
| °C | Degree Celsius |
| G | Gram |
| H | Hour(s) |
| J/g | Joule per gram |
| Kcal | kilocalories |
| µm | Micrometre |
| M | Molar |
| Mg | Milligram |
| mL | Millilitre |
| Min | Minute(s) |
| Nm | Nanometre |
| % | Percentage |

DISSERTATION OUTLINE

This research comprises five (5) chapters, each chapter addresses a specific objective in the dissertation. Chapter One introduce the study, states the problem statement being addressed, the justification, aim, specific objectives and the hypothesis of the research study. Chapter Two presents a comprehensive outline of the literature review on finger millet including details about its nutritional components, malting of finger millet and uses to form a variety of food products and health benefits. Chapter Three focuses on the materials and methods used to analyse finger millet flours and biscuits. Chapter Four focuses on the results and discussion obtained from analysing the finger millet flour and biscuits. This section has been published in the Food Science and Nutrition Journal. Chapter Five presents the conclusions and recommendations emanating from the research findings for each specific objective of the study.

CHAPTER 1: INTRODUCTION

1.1 Background

Finger millet (*Eleusine coracana* L. Gaertn.) is a cereal crop with significant cultural, nutritional, and historical importance mainly cultivated in Africa and Asia (Mirza & Marla, 2019; Mueller et al., 2020). The plant's name derives from the crop's panicle shape, which forms several finger-like structures (Sood et al., 2019). Finger millet is a tiny, seeded cereal crop belonging to the *Poaceae* grass family. The crop has many other names, such as *mufhoho* and *uphoko* in South Africa, *poho* and *rapoko* in Zimbabwe, *madua* or *ragi* in India, and *tokuso* or *dagussa* in Ethiopia (Gull et al., 2014; Ramashia et al., 2019). Finger millet is ranked fourth in South Africa, sixth in production in Ethiopia and India, amongst other cereals such as sorghum, rice, wheat, bajra, and maize. It is a drought-resistant crop with high antioxidant like tannins and nutraceutical properties such as iron and fibre (Kandel et al., 2019). It is also a gluten-free cereal crop high in fibre, calcium, and iron with outstanding malting qualities and a low glycaemic index (Singh, 2016; Udeh et al., 2018; Anagha, 2023). Thus, finger millet is a healthy food choice for diabetic and gluten-intolerant patients (Wafula et al., 2018; Patil et al., 2023).

Finger millet is usually consumed using different processing methods such as fermentation, soaking, malting. Malting is a food processing technique that has been employed for years to transform and increase the nutritional qualities of millets (Adebisi et al., 2016; Hejazi & Orsat, 2016; Gowda et al., 2022). Malting practices differ between countries and vary among communities within a country (Alowo et al., 2018), and malted cereals are a vital part of the everyday diet of people (Swami et al., 2013; Syeunda et al., 2020). Adetokunboh et al. (2022) stated that malting caused an increase in the activities of hydrolytic enzymes, improved total sugars, amino acids, B-group vitamins, and a decrease in starch and dry matter. Malting of finger millet grains for brewing and child feeding has been traditionally practised in many parts of Africa, namely South Africa, Zimbabwe, Nigeria, Kenya, and Rwanda (Kubo, 2016; Adebisi et al., 2018; Okolo and Adejumo, 2021). In addition to its nutritional advantages, malting provides an easy, less expensive method of lowering paste viscosity and raising the calorie content of cereal slurries (Baranwal, 2017). The traditional finger millet malting process is similar to that of sorghum (Embashu & Nantanga, 2019). It includes steeping for 24 hours (h) in continuous immersion. Germination is conducted for two to three days, resulting in sprout formation. It is then followed by solar energy dehydration for one to two days at 25–30 °C (Adetokunboh et al., 2022).

Functional qualities are a food item's physical and chemical characteristics that influence its behaviour during preparation, storage, and consumption (Sachdev et al., 2021).

Flour is a flexible ingredient that may be used in various food items to enhance their nutritional content, sensory and texture quality due to its functional qualities (Awuchi et al., 2019). The cereal-based finger millet flour ingredient has several valuable qualities, making it a desirable addition to food compositions (Budhwar et al., 2020). These characteristics include the capacity to absorb water and oil, the stability of foam and emulsions, rheological characteristics, gelatinisation, flavour, and scent (Abah et al., 2020). Water absorption capacity refers to the functional property of a food ingredient or item to take in water when mixed or immersed (Adanse et al., 2021). This property is essential for determining food products' texture, stability, and overall quality properties.

Oil absorption capacity is the physical trapping of oil, which retains flavour and enhances food taste for consumers (Abah et al., 2020). Fat is used for various reasons by the human body, thus making it a vital part of the diet (Hiremath & Geetha, 2019). The bulk density of the food material can have an impact on the mouthfeel of a food item and the choice of packaging material (Awuchi et al., 2019).

The lower bulk density of the flour improves the digestion of food products, especially in children with undeveloped digestive systems (Ikujenlola & Ogunba, 2018). Finger millet (FM) flour is rich in fibre, a vital biological and nutritional element of the human body, as it increases the bulk of a diet (Hiremath & Geetha, 2019). Soluble fibre decreases cholesterol and controls blood sugar levels. The insoluble fibre helps in promoting regular bowel motions and preventing constipation (Laxmi et al., 2015; Khalid et al., 2022).

The growing awareness and diagnosis of celiac disease and other digestive disorders has increased the demand for gluten-free foods in recent years (Niland & Cash, 2018). According to Kumar et al. (2017) and Sharma et al. (2020), gluten, a protein present in wheat, barley, and rye, causes the autoimmune condition known as celiac disease.

People with celiac disease are advised to strictly stick to a gluten-free diet to control their condition and prevent health complications (See et al., 2015; Pinto-Sanchez et al., 2019). Moreover, individuals without celiac gluten sensitivity or other digestive disorders may also benefit from preventing the consumption of gluten-containing diets. Nevertheless, the notable concern with gluten-free products is the lack of nutrients because most of these products are produced using refined flour. The flour lacks essential nutrients such as total dietary fibre, protein, minerals, and vitamins (Di Nardo et al., 2019; Kulai & Rashid, 2014). On the other hand, FM flour is rich in micronutrient content, which accounts for its capacity to be a valuable ingredient in the production of gluten-free products (Gebre, 2019). Inclusion of FM flour in the formulated gluten-free biscuits might address the nutritional challenges associated with gluten-free diets and provide a healthier and more nutrient-dense alternative.

However, there is inadequate research done on the thermal and functional properties of malted finger millet and other millet flours. Nefale & Mashau (2018) observed modifications

in the functional characteristics of germinated finger millet flours. Olamiti et al. (2020), and Mudau et al. (2022a) studied modifications in functional and thermal characteristics of malted and fermented finger millet and pearl millet flours. Finger millet flour has good thermal stability and can withstand elevated temperatures without losing its nutritional value or functional properties (Mudau et al., 2022a). It is important to research malting because it might improve the nutritional value of finger millet flours, improving their worth as a food source in areas where malnutrition is a problem. Malting might contribute to finger millet's grain palatability and digestibility, which might boost its demand. Moreover, it might give farmers and the food industry opportunities to market new value-added products, boosting revenue and supporting sustainable agricultural methods. Therefore, the study determined the physicochemical, antioxidant, and microstructural properties of malted finger millet flours.

This research aims to improve the physicochemical, antioxidant and microstructural properties of malted finger millet (*Eleusine coracana*) flour(s) and their use in gluten-free biscuits create opportunities for the advancement of gluten-free food industrialisation. The findings and insights obtained from this study may contribute to the development of gluten-free biscuits that can cater to the specific dietary needs of individuals with celiac disease, heart diseases, digestive issues, diabetes, and hypertension. Exploring the impact of malting on FM and its potential application in gluten-free foods expands the range of available gluten-free options and improves the overall quality and variety of these food items.

1.2 Research problem statement and justification of study

There is an increasing public health concern about gluten food allergies and food intolerances, resulting in increased consumer demand for food products modified to meet the required dietary needs. Usually, biscuits produced from wheat flour have a high glycaemic index and a lower amount of phenolics, micronutrients, and dietary fibre. Consuming any wheat product can negatively affect celiac patients, resulting in some digestive problems. Celiac disease is an inflammation of the small intestines triggered by the gluten found in wheat, barley, and rye products. Abdominal pain, diarrhoea, or bloating are popular signs and symptoms of celiac disease. Usually, celiac disease is underdiagnosed or misdiagnosed. After diagnosis, a patient resorts to a gluten-free diet to avoid the recurrence of the disease. Finger millet does not contain gluten, thus making it a suitable option for individuals with celiac disease. It is underutilised as a food ingredient for creating value-added products in many developed countries, including South Africa. This is due to the lack of knowledge about the possibilities of processing FM grains into value-added foods and beverages. Generally, the commercial problem is that there is a consumer demand for more quality and variety of biscuit products, and bakery industries might be losing prospective revenue by failing to meet these market demands. This might be due to the lack of information on consumer satisfaction with a

commercially feasible formula to produce gluten-free dough from FM. Therefore, this study focuses on developing gluten-free biscuits from FM flour. The success of this study may encourage the cultivation and usage of the underutilised FM crop in the local communities, thus improving food security. As FM is an underutilised crop, the malting of FM may enhance and improve its nutritional composition. The study may state the most suitable flour grains and composition to use when making biscuits from FM. The findings of this study may reduce the symptoms of celiac disease by introducing a gluten-free product to the market and a variety of nutritious malted FM biscuits.

1.4 Aim of the Study

This research evaluates the physicochemical, antioxidant and microstructural properties of malted finger millet flour(s) and its utilisation in gluten-free biscuits.

1.5 Specific objectives

- To evaluate the physical properties (colour and textural characteristics) of malted finger millet flours and biscuits.
- To ascertain the thermal properties of malted finger millet flours.
- To quantify the proximate composition of malted finger millet flours and biscuits.
- To establish the antioxidant characteristics (total flavonoid and phenolic content, DPPH, and FRAP) of malted finger millet flours and biscuits.
- To estimate the mineral composition (calcium, magnesium, zinc, iron, phosphorus, and manganese) of malted finger millet biscuits.
- To examine the microstructural characteristics (Fourier transmission-infrared spectroscopy and Scanning electron microscopy) of malted finger millet flours.
- To determine the sensory properties of malted finger millet biscuits.

1.6 Hypothesis

1.6.1 Alternative hypothesis

Malting will influence the physicochemical, antioxidant and microstructural properties of malted finger millet flours and its utilisation in gluten-free biscuits.

1.6.2 Null hypothesis

Malting will not influence the physicochemical, antioxidant and microstructural properties of malted finger millet flours and its utilisation in gluten-free biscuits.

CHAPTER 2: LITERATURE REVIEW

2.1. Finger millet origin

Finger millet (*Eleusine coracana* L.) is also known as mufhoho, Uphoko, osgras, mpogo, majoloth (South Africa), zviyo, rapoko, poho, mazhovole, njera (Zimbabwe), ragi, mandua (India), bulo (Uganda), mawele, lupoko, kambale, amale, bule (Zambia), wimbi, mugimbi (Kenya), tokuso, dagussa, barankiya (Ethiopia), kaddo (Nepal), and *eleusine cultivee*, *coracan*, *koracan* (France) (Singh & Raghuvanshi, 2012; Yenasew & Urga, 2022). The word millet is constructed from the French word “mille”, which indicates a thousand millets in hand consisting of up to one thousand grains (Ramashia et al., 2021a). The cereal grain is a member of the subfamily *Chloridodeae* of the *Poaceae* family, which has its origins in Ethiopia (Ramashia et al., 2018).

Finger millet is cultivated in more than 25 countries in (Gebreyohannes et al., 2021). These countries are South Africa, Nigeria, Ethiopia, Uganda, Ghana, Zimbabwe, Kenya, Malawi, Tanzania, and Zambia (Mathur, 2012; Wambi et al., 2021). The total amount of millet produced annually worldwide is nearly 4.5 to 5 million tons (Harish et al., 2024). India alone produces 2.5 million tons with some African countries producing around two million tons of grain (Serna-Saldivar, 2016). It is a prime staple food in certain areas of east African countries such as Kenya, Uganda, and Rwanda and central African countries such as Angola and Republic of Congo. The grain is found in different colours which include light brown, brown and white grains (Ramashia et al., 2019).

2.2. Structure of finger millet grain

Finger millet grains are round with a diameter that ranges from 1.0 to 1.5 mm (Siwela, 2009). The colour of FM grains is used for differentiation, whereby white grains (Figure 2.1A) are mainly used in the baking industries, light brown grain types are used for making porridge, and dark brown (Figure 2.1B) is used in fermenting traditional beer in Southern Africa (Sood et al., 2017). Figure 2.1C illustrates the two (2) varieties of white and light brown FM grains.

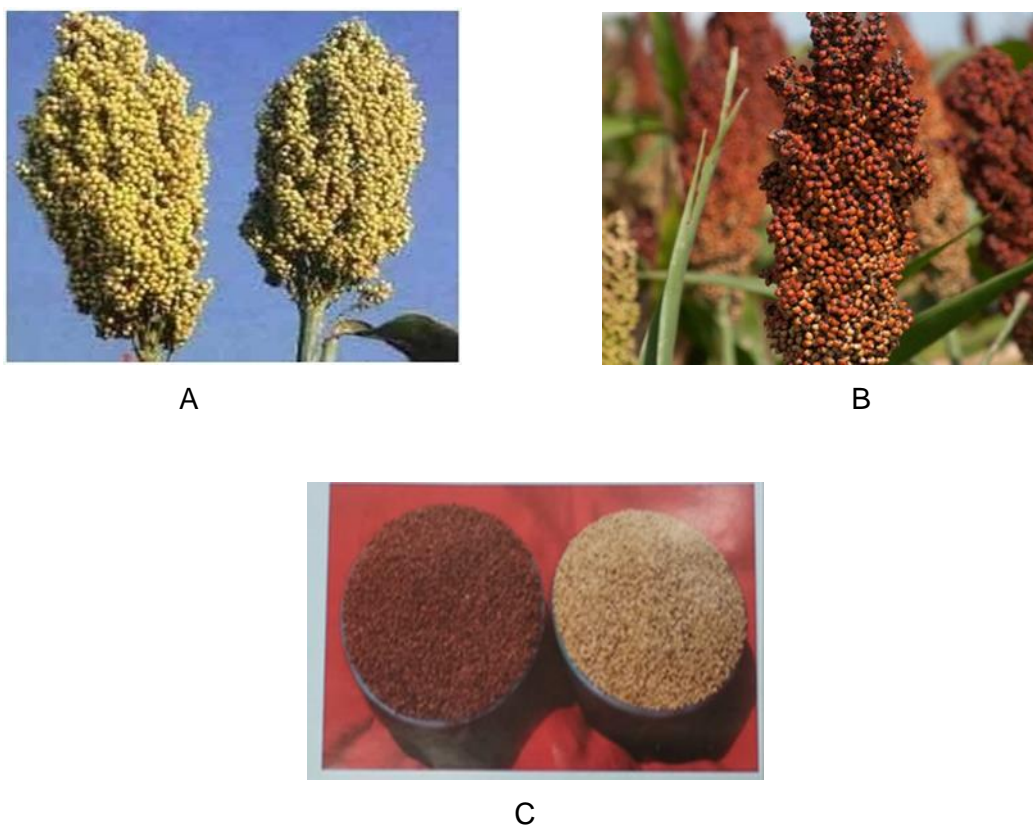


Figure 2.1: **A-** White Finger millet; **B-** Brown Finger millet; **C-** Brown and white finger millet (<https://www.bing.com/images/search?q=types+of+finger+millet&form>, Accessed 20 September 2021).

Sood et al. (2017) found that, unlike other millets such as teff, pearl, foxtail, and fonio millet, FM grains exhibit a distinct characteristic known as an utricle instead of a true caryopsis. In this unique grain structure, the pericarp of the FM grain does not completely fuse with the testa (Ramashia et al., 2019). In contrast, caryopses, which are single-seeded fruits, are observed in other millets. Caryopses are characterised by the tight adhesion of the pericarp or fruit coat to the grain, as described by Korolyuk and Polosmak (2010), and typically have a seed coat that appears brick red in colour, as mentioned by Patel et al. (2014). The unique characteristic of FM grains allows for easy removal of the pericarp when grinding the grains using a pestle and mortar. Another notable structural feature of FM grains is the presence of a five-layered testa, which is believed to contribute to the superior fibre content of the grain (Shobana et al., 2013). The major physiological components of FM grains include the germ, endosperm, and pericarp (Figure 2.2). The endosperm, which is the largest part of the grain, is closely attached to the seed coat and is used in flour production (Chandrashekar, 2010). The pericarp, also known as the outer glume, acts as a protective layer for the grain. It comprises three layers of varying thickness, namely the inner endocarp, the middle mesocarp, and the outermost epicarp (Taylor & Duodu, 2017). During further treatment, the non-edible pericarp is separated from the kernel (Ramashia et al., 2019).

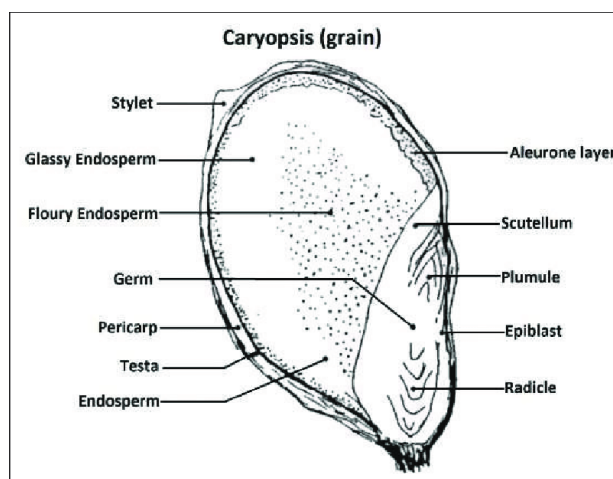


Figure 2.2: Structure of finger millet

<https://www.bing.com/images/search?view=detailV2&ccid=ASG5F3SX&id=D7E12DC41C38C5301B5D6BE641D62BFF629C9FC8&thid=OIP.ASG5F3SXjiXYJEXv2yliEwH>, Accessed 15 April 2024)

2.3. Nutritional compositions of finger millet

Ensuring nutritional quality is crucial for maintaining overall health and well-being. Hence, addressing the issues of food insecurity and malnutrition is necessary when considering the nutritional content (Singh & Raghuvanshi, 2012). Finger millet is a satisfactory source of various nutrients (Table 2.1), including protein (5.9%–10.3%), carbohydrates (72.6%–82.6%), dietary fibre (2.0%–3.6%), (Chandra et al., 2016; Ramashia et al., 2019). The carbohydrate content of FM ranges from 72% to 79.5% (Singh et al., 2018). It serves as a primary staple food for individuals with metabolic disorders such as celiac disease, diabetes, and obesity, as well as those belonging to low socio-economic groups (Mathanghi & Sudha, 2012).

Table 2. 1: Chemical composition of finger millet (g/100g).

| Carbohydrate | Protein | Fibre | Minerals | Fat | References |
|--------------|---------|-------|----------|-----|--|
| 83.3 | 8.2 | 3.5 | 2.7 | 1.8 | Jaybhaye et al., 2014; Yenasew and Urga (2022) |
| 72.6 | 7.7 | 3.6 | 2.6 | 1.5 | Amadou et al., 2013; Yenasew and Urga (2022) |
| 76.4 | 10.3 | 3.1 | 2.4 | 0.8 | David et al., 2014 |
| 72.6 | 7.3 | 3.6 | 3.0 | 1.3 | Chandra et al., 2016 |
| 85.6 | 6.5 | 3.4 | 2.0 | 1.3 | Lansakara et al., 2016 |
| 75.3 | 5.9 | 2.0 | 2.5 | 1.8 | Ramashia et al., 2019 |
| 79.0 | 7.7 | - | 2.7 | 1.8 | Katake et al., 2016 |

2.3.1. Protein composition

The average protein composition of FM ranges from 5.9 to 10.3 g/100g, (Table 2.1). Admassu et al. (2013) investigated FM varieties, with had protein content 7.7 g/100g. The most significant storage protein in FM is the prolamin fraction, which constitutes 25-36% of the total protein (Serna-Saldivar, 2016). Protein digestibility is a crucial quality parameter because it reflects the amount of protein absorbed from a meal. The FM protein exhibit a digestibility in the range of 50 to 88% (Singh & Raghuvanshi, 2012). It is worth mentioning that FM has a more balanced essential amino acid profile than other types of millet because it has more lysine, threonine, and valine (Hassan et al., 2021). Essential amino acids are particularly important from a nutritional perspective, as they are carbon skeletons that humans cannot synthesise and should be obtained via dietary sources (Tomar et al., 2022). The grain consists of nine (9) important amino acids, which include isoleucine, valine, phenylalanine, methionine, threonine, lysine, histidine, and tryptophan. These amino acids can be ranked based on their content levels, with leucine, valine, isoleucine, phenylalanine, threonine, lysine, methionine, histidine, and tryptophan appearing in descending order (Amadou et al., 2013; Ramashia et al., 2019). In contrast to other millet grains, FM has a high content of lysine, methionine, threonine, and valine (Ramashia et al., 2019). This superior balance of crucial amino acids in FM, particularly due to its high lysine, threonine, and valine content, makes it

advantageous for achieving a well-rounded amino acid profile in food products (Gull et al., 2014).

Table 2.2: Essential amino acids found in finger millet.

| Amino acid | Amadou et al., 2013 (mg/100g) | Ramashia et al., 2019 (g/100g protein) | Chandra et al., 2016 (mg/100g) |
|---------------|----------------------------------|---|-----------------------------------|
| Histidine | 130 | 230 | 2.2 |
| Methionine | 210 | 290 | 2.5-3.1 |
| Lysine | 220 | 220 | 2.2 |
| Tryptophan | 100 | - | 1.1-1.5 |
| Threonine | 240 | 430 | 3.4-4.2 |
| Phenylalanine | 310 | 600 | 4.1-5.2 |
| Isoleucine | 400 | 430 | 4.3 |
| Leucine | 690 | 1080 | 6.6-9.5 |
| Valine | 480 | 630 | 4.9-6.6 |

2.3.2. Carbohydrates

Finger millet contains a significant amount of carbohydrates, ranging from 72.6 to 85.6 g/100g of grain (Gebre, 2019). Starch, the primary carbohydrate present in grains, is a significant source of physical energy in the diet of humans. Yenasew and Urga (2022) reported a carbohydrate content of 72.6 to 83.3 g/100g which was like what Murungweni et al. (2023) observed. In FM, the starch composition varies from 59.4 to 70.2% of the dry matter, with approximately 80 to 85% amylopectin and the residual of 15 to 20% being amylose (Dayakar et al., 2017). It also contains approximately 1.5% of reducing sugars and 0.03% of non-reducing sugars (Pragya & Rita, 2012). Table 2.1 illustrates the range of carbohydrate amounts in FM.

2.3.3. Lipids

In contrast to other millets, which typically contain a fat content ranging from 1.0 to 1.8% (Jaybhaye et al., 2014), FM has a fat content of less than 2% (Table 2.1). The low-fat level and high fibre content of FM contribute to its functional properties. Fat is a concentrated source of energy and is an essential part of the structure of cell membranes, enabling normal bodily functions. The quality of fat is important, and FM stands out because of its elevated

levels of unsaturated fatty acids, including linoleic acid, oleic acid, and palmitic acid, which make up approximately 74% of the total fatty acids existing (Najdi Hejazi & Orsat, 2017).

2.3.4. Dietary fibres

In the small intestines, dietary fibre resists enzymatic breakdown and absorption, while in the large intestines, it undertakes complete or incomplete fermentation (Graf et al., 2014). Finger millet is a rich source of crude fibre, comprising 2.0 to 3.6% of the overall mass of the FM grain (Table 2.1). Amadou et al. (2013) state that FM exhibits a higher total dietary fibre (TDF) content (22.0%) compared to grains like maize (12.8%), wheat (12.6%), and rice (4.8%). Research has associated ingesting whole grains rich in fibre with a reduced chance of having cardiovascular disease and type 2 diabetes (Archundia et al., 2017). Dietary fibre improves digestive health, lowers cholesterol and lipid absorption, promotes satiety, reduces the danger of gastrointestinal infection, and regulates postprandial insulin response, among other benefits (Graf et al., 2014).

2.3.5. Minerals

Minerals play a vital role in maintaining glucose homeostasis, nerve impulse transmission, and electrolyte balance and serve as enzyme cofactors within the human body (Gordillo-Bastidas et al., 2016). Finger millet stands out with its higher overall mineral (ash) content, as shown in Table 2.3. In comparison, Ethiopian staple grains such as tef, wheat, maize, sorghum, rice, and barley have total mineral amounts of 1.4, 1.6, 2.8, 1.7, 3.1, and 1.7%, respectively (Gebre, 2019). Table 2.3 displays the calcium, magnesium, phosphorus, zinc, iron, potassium, and manganese content found in FM grains. Notably, FM is high in calcium and iron, both essential elements for optimal health (Chandra et al., 2016). Anaemia, which is prevalent among Ethiopian reproductive-age women, can be addressed by including FM in their regular diet, as it helps combat iron deficiency while also preventing bone disorders caused by insufficient calcium intake (Ambati & Sucharitha, 2019). Chandra et al. (2016) reported a magnesium content of 137 which was similar to the findings of this study.

Table 2. 3: Minerals found in finger millet.

| Minerals | Admassu et al., 2009 | Chandra et al., 2016 | Hiremath et al., 2018 | Ramashia et al., 2019 |
|-------------|----------------------|----------------------|-----------------------|-----------------------|
| Phosphorous | 122 | 283 | 228 | 190 (130-250) |
| Calcium | 257 | 344 | 282 | 398 |
| Magnesium | 168 | 137 | 166 | 140 (78 - 201) |
| Zinc | 1.8 | 2.3 | 2.79 | 2.3 |
| Phosphorus | - | 150 - 250 | - | - |
| Iron | 27 | 3.6 | 3 | 9.1 (3.3 -14.9) |
| Potassium | - | 408 | - | 460 (430 - 490) |
| Copper | | 0.18 - 0.79 | | 0.18-0.79 |
| Manganese | 28.35 | 5.49 | 25 | 33 (17.6 - 48.4) |

2.3.6. Vitamins

Vitamins are essential for various aspects of human health, including promoting vision, regulating cell growth and development, acting as antioxidants, serving as enzyme cofactors in metabolic processes, and supporting other physiological functions (Del Mondo et al., 2020). Finger millet constitutes a range of vitamins, both fat-soluble and water-soluble, with a particularly high concentration of vitamins A and B complex (Dayakar et al., 2017; Ramashia et al., 2019). Due to its high vitamin and mineral contents, FM has the potential to be a valuable crop in South Africa to address hunger, especially among vulnerable groups such as children, pregnant and lactating women. Devi et al. (2014) and Samtiya et al. (2020) reported high levels of water-soluble vitamins and fat-soluble vitamins A and B complex in FM grains (Table 2.4). However, dried FM grains lack vitamin C (Siwela, 2009).

Table 2.4: Major vitamins in finger millet

| Vitamins | mg/100 g | References |
|---------------------------|-------------|--------------------------------------|
| Vitamin A (Retinol) | 6.00 | Siwela, 2009, Ramashia, 2018 |
| Vitamin B1 (Thiamine) | 0.20 - 0.48 | Saleh et al., 2013 |
| Vitamin B2 (Riboflavin) | 0.12 | Devi et al., 2014), Ramashia, 2018. |
| Vitamin C (Ascorbic acid) | 1.00 | Siwela, 2009, Shobana et al. (2013). |
| Niacin | 1.00 - 1.30 | Saleh et al., (2013). |

2.3.7. Antioxidant properties of finger millet

Antioxidant properties were identified in several compounds present in different millets (Table 2.5). Millet constituents such as phenolics, carotenoids, dietary fibre, and tocopherols, which are rich in micronutrients, revealed notable antioxidant capabilities, primarily concentrated in the bran layers. Fermentation and germination processes can improve millets with additional antioxidants, such as peptides (Fernandez-Orozco et al., 2008; Amadou et al., 2013). Cereals mostly contain phenolic acids and tannins as their main phenolic compounds, with flavonoids available in smaller amounts (Van Hung, 2016). These compounds act as scavengers of free radicals, metal chelators, reducing agents, and quenchers of singlet oxygen molecules (Devi et al., 2014).

2.4. Impact of processing methods on the nutritional value of finger millet grains

Malting or germination of cereals can lead to biochemical transformations, producing a malt with an improved nutritional composition that may be used in various traditional recipes (Nkhata et al., 2018). Saleh et al. (2013) indicate that the germination of proso millet leads to a rise in total sugars and free amino acids, accompanied by a decline in dry weight and starch content. Furthermore, there is a notable rise in tryptophan, lysine, and nonprotein nitrogen. In the case of pearl millet, germination significantly improves the *in vitro* digestibility of protein from 14% to 26% and starch from 86% to 112%, surpassing the benefits achieved through blanching alone (Sarita & Singh, 2016). Antinutrients like phytic acid, polyphenols, and tannins tend to form complexes with proteins, but they are reduced by soaking, germination, dry heating, and debranning (Annor et al., 2017).

Furthermore, germination leads to a reduction of crude fat and protein in foxtail millet (Gowda et al., 2022). This reduction is caused by the loss of nitrogenous substances of low molecular mass during the rinsing and soaking of millet grains, as well as lipid hydrolysis and oxidation of fatty acids during germination (Choudhury et al., 2011; Saleh et al., 2013).

The process of germination or malting has a significant impact on the extractability and accessibility of minerals like iron, calcium, and zinc in FM and pearl millet while simultaneously reducing antinutritional elements like phytic acid (Tharifkhan et al., 2021). Studies have shown that germination and probiotic fermentation can lead to notable improvements in niacin, thiamine, protein fractions, total lysine, soluble fibre, carbohydrates, and the *in vitro* availability of iron, calcium, and zinc in food combinations (Arora et al., 2011). The increased activity of phytase during germination may contribute to a decline in phytate content, resulting in improved mineral availability. Antinutrients such as polyphenols and saponins, which can hinder mineral absorption, are also degraded during germination, further enhancing mineral availability (Grewal & Jood, 2006).

Subsequently, the process of germination in millet grains can be utilised alone or in a mixture with other processing techniques to create a nutrient-rich malt that can be incorporated into different healthy and nutritious food products, including supplemental foods, composite flours, infant formula, and food combinations (Pragya & Rita, 2012; Jagati et al., 2021). However, to facilitate the widespread adoption of millet utilisation, industrial-scale malting with specialised germination and precise control systems for germination conditions is necessary to ensure the production of high-quality malt products that are convenient to handle and eaten by greater populations (Kilfoil, 2020).

The total content of phenolics and tannins in FM grain genotypes can vary (Hassan et al., 2021). Pale-coloured grains have significantly lower levels of total phenolics, and tannins compared to brick-red-coloured grains. Red-coloured varieties with pigmented testa, located in the outer tissue of the grain, have a high tannin content (Siwela et al., 2007). Certain compounds found in FM grains, such as phytate, phytic acid, tannins, trypsin inhibitors, and flavonoids, reduce mineral bioavailability, thereby affecting the overall nutritional quality of FM grains (Chauhan, 2018). However, the primary polyphenols in FM are phenolic acids and tannins, with flavonoids present in lower amounts (Mitharwa et al., 2021). Polyphenols, including those in FM grains, play a vital role as antioxidants and support the immune system of the body (Nithiyantham et al., 2019). Tannins available in the outside layer of the grain serve as a physical blockade against fungal invasion (Smýkal et al., 2014) and play crucial roles in both plant and human biological functions. However, their antinutritional effects can hinder nutrient digestibility and mineral absorption to a certain extent (Ramashia et al., 2019). Studies have demonstrated that certain processing techniques, like soaking, decortication, fermentation, malting, and steaming, can enhance the availability of these nutrients (Gowda

et al., 2022). Tannins, on the other hand, can inhibit growth by affecting the function of the pancreas and thyroid gland, as well as causing liver abnormalities. The presence of tannin compounds can impact the colour, flavour, and nutritional quality of cereals and FM (Ramashia et al., 2019). Additionally, tannins contribute to the antioxidant capacity of FM foods, which plays an important role in promoting healthy ageing and preventing metabolic diseases (Bhat et al., 2018).

The xylo-oligosaccharides (XOs) mentioned in Table 2.5 consist of D-xylose units connected by -1,4 glycoside linkages, and they can exist as xylotriose, xylobiose, or xylotetrose with a low degree of polymerisation (DP 2-10) (Jamaldheen et al., 2019; Liang & Liang, 2019). Most oligosaccharides contain phenolic acids with ester bonds, such as coumaric, ferulic, and caffeic acids (Samanta et al., 2012). The hydrolysis of arabinoxylans from grain and millet bran yields XOs. The notable rise in ferulic and syringic acid content in FM could explain its remarkable antioxidant activity (Veenashri & Muralikrishna, 2011). Additionally, phenolic acids, polysaccharides with sugars containing the $> C = O$ (uronyl/acetyl) moiety, and substantial polymerisation levels contribute to the antioxidant characteristics of these polysaccharides (Liang & Liang, 2019).

Table 2.5: Antioxidants found in finger millet.

| Name of compound | Major types | Mechanisms of action | References |
|-----------------------|---|---|---|
| Xylo-oligosaccharides | Finger millet | Most oligosaccharides are composed of ester-bound aromatic acids. Besides aromatic acids, polysaccharides exhibit significant antioxidative properties owing to the inclusion of sugars containing > C = O (acetyl/urony) units and the type/extent of polymerisation. | Devi et al., 2014 |
| Tannins | Finger millet | By means of different coupling methods, procyanidin o-quinone has the capacity to generate oligomeric compounds that preserve the amount of hydroxyl groups. These molecules can act as prooxidants, inducing the creation of reactive oxygen species through futile redox cycling. | Siwela et al., 2007; Suma and Urooj, 2012 |
| Phenolic acids | Finger millet, proso, kodo, foxtail, pearl millet | These compounds possess the ability to give hydrogen atoms to electron-deficient free radicals utilising hydroxyl groups found on benzene rings. This process leads to the creation of a resonance-stabilized phenoxyl radical, which exhibits reduced reactivity. | Udeh et al., 2017; Devi et al., 2014; Chandrasekara and Shahidi, 2011 |
| Flavonoid | Proso, pearl, foxtail, kodo, finger millet | The presence of multiple hydroxyl groups gives out notable antioxidant properties to the molecules. These properties are improved by the incorporation of a double bond and carbonyl function in the heterocycle, as well as via the polymerisation of the nuclear structure. These changes result in a more stable flavonoid radical through conjugation and electron delocalisation, thereby increasing its overall activity. | Suma and Urooj, 2012; Bangoura et al., 2013 |
| Carotenoids | Foxtail, pearl, kodo, proso, finger millets | Carotenoids, working as antioxidants, effectively neutralise single oxygen and free radicals. | Viswanath et al., 2009 |
| Vitamin E | Foxtail, proso, little, foxtail millets | The biological action of tools is due to their antioxidant effect in biological membranes, and it prevents lipid peroxidation. | Asharani et al., 2010 |

2.5. Microstructural analysis

The techniques of small-angle neutron and X-ray scattering are referred to as small-angle scattering. Both methods can examine structures ranging from a few hundred nanometres to billions of nanometres in size (Eyssautier et al., 2011). In these techniques, X-ray photons or neutron radiation are elastically scattered by a sample, and the resulting scattered radiation distribution is analysed to gather information about the size of the sample, shape, and orientation (Jeffries et al., 2021). Molecular technologies such as differential scanning calorimetry (DSC), X-ray diffractometry, high-performance anion-exchange chromatography (HPAEC), gel-permeation chromatography (GPC), nuclear magnetic resonance (NMR), and attenuated total reflectance Fourier transform infrared (FTIR) can be employed for analysis. X-ray diffractometry (Table 2.6) is used to determine the crystalline structures and degree of crystallinity of starch (Blazek & Gilbert, 2011; Tian et al., 2019).

Kumar et al., (2022) indicates that scanning electron microscopy (SEM) is a valuable tool for studying the microstructures of grains from cereals and pseudocereals, as well as their by-products. Gull et al. (2016) states that microstructure studies have revealed that starch granules in FM and pearl millet flour exhibit polygonal shapes with visible edges and no surface pores. Joardder et al. (2017) notes that the term food microstructure refers to the spatial arrangement of cells and intercellular spaces in food materials. Various food products, such as meat, fruits, vegetables, and bread, all possess unique microstructures. During food preparation, existing food structures are disrupted, and new structures are formed. Sharma and Bhardwaj (2019) point out that changes in food structure during preparation can affect food quality and lead to the breaking down of the food compound.

Microstructural analysis provides understandings into the physical arrangement and distribution of components within food. Food texture is influenced by microstructural patterns such as particle size, shape, and arrangement. By analysing the microstructure, food developers can shape formulations and processing methods to get desired texture characteristics.

Table 2. 6: Current technology on microstructure study.

| Microstructure Structure level | Technology | Function | Technology | Reference |
|--------------------------------|------------|--------------------------------|--|---|
| Molecular level | XRD | Crystal type and degree | Gelatinisation by limit temperature or water | Han et al., 2011; Purohit et al., 2019 |
| | DSC | Thermal Properties | Retrogradation by temperature rotations | Lin and Lin, 2015; Mohamad et al., 2015 |
| | GPC | Molecular weight distributions | Starch-lipid | Lange et al., 2016; Ji et al., 2018 |
| | FTIR | Functional groups absorbance | Debranching by enzymes | Hu et al., 2016; Su et al., 2018 |
| Cell and tissues | SEM | Surfaces changes | Alter granules size | Sharma and Bhardwaj, 2019 |
| Food level | RVA | Rheological characteristics | Minimal process | Balet et al., 2019 |

Note: XRD- X-ray diffraction analysis, DSC- Differential scanning calorimetry analysis, GPC- Gel permeation chromatography analysis, FTIR- Fourier Transform Infrared Spectroscopy analysis, SEM- Scanning electron microscope analysis, RVA- Rapid visco analyser.

2.6. Utilisation and application of finger millet

Rathore et al. (2016) state that finger millet grains are a key ingredient in the production of composite flour, extruded products, baked goods, and other cereal-based foods that are gluten-free. Ramashia et al. (2019) point out that the specific types of FM-based foods can vary greatly between countries and regions, and many food items made in underdeveloped nations remain uncommercialised. Gebre (2019) observes that in affluent countries, commercially available FM-based products include macaroni, spaghetti, pasta, vermicelli, noodles, and flakes, which are widely consumed. Pathak and Kochhar (2018) note that in the production of extruded snacks, FM is often combined with buckwheat and amaranth as an alternative to wheat and maize. In Ethiopia, Zewdu et al. (2018) report that FM is used to create a variety of food items such as injera, a fermented pancake-like bread, porridge, local beverages, chechebsa, a traditional breakfast, bread, and soup. The straw of FM is also used as animal feed. A staple food in Ethiopia called injera may be produced with or without tef (*Eragrostis tef*), which is the preferred grain for injera preparation. Hotessa and Robe (2020) indicate that traditional Ethiopian beverages like *areki* and *tella* are produced from various cereals, including FM. Hailu and Gebreyohans (2017) recommend the use of mango-flavoured FM juice to enjoy the nutritional benefits of the crop.

2.7. Health benefits of finger millet

Finger millet possesses various health benefits and is a functional and nutraceutical food due to its nutritional value and positive effect on health. It is rich in minerals, dietary fibre, and phytochemicals, and has a low glycaemic index, making it beneficial for overall well-being (Shobana et al., 2013; Kumar et al., 2016; Tripathi et al., 2022). The consumption of FM has been linked to various health advantages, including its effectiveness against celiac disease, type 2 diabetes, gastrointestinal diseases, cancer, heart attacks (strokes), allergies, constipation, and anaemia (Ambati & Sucharitha, 2019; Tripathi et al., 2022). Kandel et al. (2019) note that pregnant women can benefit from including FM in their diet and report that FM has also been used to treat animal diarrhoea. In Southern India, Manikam et al. (2018) says that healthcare professionals recommend FM-based diets for infants above 6 months of age. Onipe and Ramashia (2022) report that the seed coat of FM is particularly rich in phenolic compounds, dietary fibre, minerals, and vitamins. The antioxidant activity of FM can vary depending on the colour of the grain. Chethan and Malleshi (2007) indicate that brown varieties have been found to contain higher levels of polyphenols (0.08-3.47%) in contrast to white varieties (0.04-0.09%). Similarly, Xiang et al. (2019) observed that coloured red or

reddish and brown FM grains exhibit higher concentrations of phenolic compounds compared to white varieties.

Duodu and Awika (2019) state that the phytochemicals and antioxidants present in FM act as scavengers of free radicals, helping to regulate extreme cellular oxidation and providing protection against heart attacks and certain cancer types in humans. Nagaraju et al. (2020) reported that FM's low glycaemic index, which lets glucose slowly enter the bloodstream after digestion, has been linked to a lower risk of diabetes. Studies have suggested that the capacity of FM to manage type 2 diabetes may be attributed to its richness in calcium and magnesium contents, which help in lowering blood glucose levels (Chandra et al., 2016; Kanorwala et al., 2022). Furthermore, Kumar et al. (2016) notes that the insoluble dietary fibres found in FM resist digestion, add bulk to the stool, and act as a natural laxative. He et al. (2022) indicates that this promotes bowel motility, retains water, and prevents constipation, colon malignancies, gastrointestinal disorders, and heart diseases. Woomer and Adedeji (2021) assert that another notable characteristic of FM is its gluten-free nature, making it a suitable substitute to wheat-based cereals. Ambati and Sucharitha (2019) observe that gluten is a major trigger for celiac disease; therefore, the consumption of gluten-free grains like FM by individuals with the condition is recommended.

2.8. Biscuits and biscuit production

Biscuits are typically prepared using a blend of flour, sugar, oil, and various other ingredients. In the case of gluten-free biscuits, alternative flours such as rice, starches, and maize flour are commonly used (Di Cairano et al., 2018). Researchers are actively exploring the development of alternative flours that can improve the functional, nutritional, and sensory properties of gluten-free biscuits (Deora et al., 2014; Rai et al., 2018). However, unlike in other baked goods, the formation of a gluten network is not necessary to produce biscuits (Mert & Demirkesen, 2016). Kaur et al. (2020) notes that biscuits incorporating FM have higher levels of crude fibre, iron, calcium, and zinc compared to control biscuits. Thapliyal and Singh (2015) observe that this suggests that FM biscuits could potentially offer consumers the inherent health benefits of FM, addressing calcium deficiencies and contributing to the prevention of other lifestyle diseases.

2.9 Concluding remark

Malting of finger millet has been shown to improve its nutritional profile by increasing the bioavailability of nutrients and reducing anti-nutritional factors. Moreover, the application of microstructural technologies allows for a deeper understanding of the changes occurring during malting, providing insights into how these processes affect the overall quality and

functionality of finger millet-based products. These technologies can help in the development of innovative food products that retain the nutritional benefits of finger millet while offering improved sensory characteristics and enhanced functionality.

CHAPTER 3: MATERIALS AND METHODS

3.1. Sample Collection

Ten kilograms of light brown (LB) and dark brown (DB) finger millet (FM) grains were obtained from street vendors and local markets in Thohoyandou, Limpopo, South Africa. Analytical grade reagents and chemicals were purchased from Merck Chemicals (PTY) Ltd., Germiston, South Africa.

3.2. Malting of Finger Millet Grains and Flour production

Undesirable materials such as grain chaff and other impurities were eliminated from the light brown (LB) and dark brown (DB) FM grains through a process of winnowing, sorting, and screening. After the visual sorting process, the LB and DB grains were cleaned using cold water to eliminate foreign substances such as dirt and stones. They were then placed in a 5 L bucket of cold water and allowed to soak for 10 h at a temperature of 30 °C. After the soaking period, the grains were drained, weighed, and divided into four equal portions. Individual grains of FM were placed on clean cheese fabrics and permitted to germinate for 0, 24, 48, and 72 h. Water was periodically sprayed on the FM grains every 4 h to facilitate germination. The germinated grains were then kilned for 8 h in an oven dryer 30 °C, resulting in a distinctive malt fragrance. Afterwards, the kilned grains were milled into fine flours using a miller (Retsch ZM 200 miller, Germany). The two malted FM flours were sifted using a 250 µm sieve to obtain fine and uniform flour, packaged in polyethylene bag, and refrigerated at 4 °C until use. Two separate batches of malted FM flours were prepared, and analyses were duplicated to ensure reliability.

3.3 Biscuits Production

The production of the LB and DB finger millet variety biscuits was carried out using a slightly modified technique as described by Punia and Dhull (2019). The formulated biscuits consisted of 112.5 % of FM flour, 28 % of sugar, 0.75 % of baking powder, 33 % of baking margarine, 6.75 % of vanilla essence, and 60 % of water. These ingredients were manually blended with a spatula in a container to form a dough that was rolled and flattened to attain a desired thickness of 0.7 cm. Circular-shaped biscuit cutters with a diameter of 5.3 cm were used to cut the dough into individual biscuits. The shaped dough was baked in a preheated Defy oven (Model DSS700, Midrand, South Africa) for 18 min at 180 °C. After baking, the biscuits were

allowed to cool at room temperature for 20 min before being packaged in plastic and stored in a refrigerator for further investigation. The whole process of biscuit production and subsequent investigations were replicated and done in triplicate to ensure the validity of the outcomes, and nine biscuits were investigated for each formulation. Biscuits produced from unmalted flours were used as a control.

3.4. Proximate Composition Analysis

3.4.1. Moisture Content

The moisture content of the finger millet flours and biscuits were assessed in triplicate using the AACC (2000) International approved method 44-15. The finger millet flours and biscuits were dried in a forced-air oven set at 105 °C until a constant mass is achieved. Subsequently, the dried samples were cooled in a desiccator. The percentage of moisture content will be calculated using the Equation 1.

$$\text{Moisture content (\%)} = \frac{\text{moisture loss}}{\text{sample weight}} \times 100 \quad (1)$$

3.4.2. Fat Content

The oil content was assessed in triplicate following the AACC (2000) International approved method 30-25.01. After drying, the flour and biscuit samples were crushed into powder using a grinder, and the fat content was determined through a 4 h solvent extraction process utilising petroleum ether (40 – 60 °C) in an automated Soxhlet extractor. The calculation of the fat content will be carried out using Equation 2.

$$\text{Fat content (\%)} = \frac{\text{weight of extract}}{\text{sample weight}} \times 100 \quad (2)$$

3.4.3. Ash Content

The ash content was analysed in triplicate following the AACC (2000) International approved method 08-01.01. Heating of the silica crucibles was done and were cooled in a desiccator and were weighed at room temperature. Dried samples weighing 5 g were measured into the crucibles, covered, and were left in the muffle furnace at 550 °C overnight. After cooling in a desiccator, the crucibles were re-weighed, and the percentage of ash was calculated using Equation 3

$$\text{Ash content (\%)} = \frac{\text{weight of residue}}{\text{sample weight}} \times 100 \quad (3)$$

3.4.4. Fibre Content

Flour and biscuit powdered samples weighing 1 g were combined with a solution composed of H₂SO₄ (1.25 g) and potassium hydroxide at a concentration of 100 mL per 200 mL in a 400 mL beaker. The mixture was boiled under reflux for 30 min. Subsequently, the solution underwent filtration through a lined, fluted funnel and be washed with water. The resulting residue, along with 100 mL of solution, was boiled in a beaker for 30 min and then dried in an oven dryer at 105 °C for 1 h. The dried residue was weighed and heated in a muffle furnace at 540 °C for 3 h and re-weighed after cooling in a desiccator for 1 h. The calculation of crude fibre was performed using Equation 4 from the AACC (2000) procedures.

$$\text{Crude fibre (\%)} = \frac{W_2 - W_3}{W_1} \times 100 \quad (4)$$

Where: W₁ = weight of sample

W₂ = weight of crucible + sample

W₃ = weight of flour + crucible + ash

3.4.5. Protein Content

Protein analysis was conducted using the AOAC Kjeldahl method (955.04) with certain adjustments. The nitrogen content in the protein was determined, and a multiplication factor of 6.25 was applied to obtain the protein content. The flatbread, once dried, will be ground into a uniform powder capable of passing through a 20-mesh screen. A 1 g sample was mixed with 10 mL of concentrated H₂SO₄ in a Kjeldahl flask for digestion. A selenium catalyst tablet was added, and the mixture was heated under a fume cupboard until a clear solution is achieved. The resulting digest was diluted to a volume of 100 mL in a volumetric flask for subsequent analysis. For the distillation step, 10 mL of the digest was combined with 10 mL of a 45% NaOH solution in a Kjeldahl distillation apparatus. The mixture was distilled into 10 mL of a 4% boric acid solution containing indicators such as methyl red. In the titration stage, 50 mL of the distillate was collected and titrated against 0.02N EDTA until it reached a deep red endpoint. A reagent blank went through digestion, distillation, and titration. The nitrogen content was calculated using Equation 5, while the protein content was determined using Equation 6.

$$\%N = \frac{(\text{Titration in mL} - \text{Blank in mL}) \times 1.4 \times 0.1}{\text{Mass of Sample (g)}} \quad (5)$$

$$\% \text{ Protein} = \% N \times 6.25 \quad (6)$$

3.4.6. Carbohydrate Content

The carbohydrate content of malted finger millet flours and biscuits was determined based on difference method as described by Farzana et al. (2017). The sum of the moisture, protein, ash, fat, and fibre content was subtracted from one hundred to obtain the carbohydrate content percentage (Equation 7).

$$\text{Carbohydrate}(\%) = 100 - (\% \text{ Moisture} + \% \text{ Protein} + \% \text{ Ash} + \% \text{ Fat} + \% \text{ Fibre}) \quad (7)$$

3.4.7. Energy Content

The energy value of malted finger millet flours and biscuits was determined as stated by Farzana and Mohajan (2022) using Equation 8.

$$\text{Energy value (kcal)} = \text{g}/100 \text{ g carbohydrate} \times 4 + \text{g}/100 \text{ g fat} \times 9 + \text{g}/100 \text{ g protein} \times 4 + \text{g}/100 \text{ g fibre} \times 2$$

3.5. Determination of Colour Properties

The colour properties of LB and DB finger millet flours and biscuits were measured using a ColourFlex spectrophotometer (Hunter Associates Laboratory in Reston, Virginia, USA) after calibration with white and black tiles. The colour was interpreted by Hunter values such as L^* , a^* , and b^* . The a^* designated hue on the green (-) to red (+) axis, whereas b^* denoted hue on the blue (-) to yellow (+) axis, yellowness index (YI) denoted yellowness, and whiteness index (WI) denoted whiteness. L^* denotes lightness, which runs from black to white (0-100) (Murungweni, et al., 2023). The chroma (C), hue angle (H°), total colour difference (E), YI, and WI were calculated using Equations 8, 9, 10, 11 and 12 (Murungweni et al., 2023).

$$\Delta E = \sqrt{(L^* - L_c)^2 + (a^* - a_c)^2 + (b^* - b_c)^2} \quad (8)$$

$$\text{Hue } (H^\circ) = \tan^{-1} \left\{ \frac{b^*}{a^*} \right\} \quad (9)$$

$$Chroma = \sqrt{(a^*)^2 + (b^*)^2} \quad (10)$$

$$YI = \frac{142.86b^*}{L^*} \quad (11)$$

$$WI = \sqrt{(100 - L^*)^2 + (a^*)^2 + (b^*)^2} \quad (12)$$

3.6. Determination of Functional Characteristics

3.6.1 Packed and Loose Bulk Density

The density of finger millet flours was calculated using the procedure suggested by Amandikwa et al. (2015), which involves measuring the packed bulk density and loose bulk density. Ten grams of finger millet flour were weighed and placed in a measuring cylinder of 25 mL. The packed bulk density was established by slightly beating the cylinder at the bottom repeatedly until the flour volume became constant. The finger millet flour mass per unit volume was utilised to calculate the packed and loose bulk densities (g/cm³) using Equation 13.

$$\text{Bulk density} = \frac{\text{Flour weight (g)}}{\text{Volume cm}^3} \quad (13)$$

3.6.2 Water and Oil Absorption Capacity

Water and oil absorption capacities of finger millet flours were measured using the method described by Mudau et al. (2022a). One gram of finger millet flour was weighed into a 50 mL centrifuge tube, and 10 mL of sunflower oil was added. The mixture was mixed thoroughly using a vortex stirrer for 30 min at room temperature (25 °C) and centrifuged for 25 min at 3000 rpm (Rotina 380 R- Labotech Ecotherm centrifuge, Midrand, South Africa). The volume of water or oil absorbed was measured by subtracting the original volume from the volume after centrifugation.

3.6.3 Swelling Power

The swelling power of malted finger millet flours was assessed using a method stated by Adebiji et al. (2016). Finger millet flours were added to a 100 mL measuring cylinder until it reached the 10 mL level and filled with distilled water to the 50 mL mark. The measuring cylinder was firmly closed and inverted for 2 min. After that, the measuring cylinder was flipped and left to stand for 30 min. The volume of the flour samples was then measured.

3.6.4. Solubility Index

The solubility index of finger millet flours was measured following a method described by Chandrasekar et al. (2022). About 0.1 gram of finger millet flour was mixed with 10 mL of distilled water in a centrifuge tube and shaken at 70 °C for 30 min, followed by cooling in cold water for 5 min. Afterwards, the tubes were centrifuged at 1700 rpm for 5 min. The liquid portion was evaporated using a water bath at 100 °C until it reached a constant weight. The weight of the flour liquid was then used to calculate the solubility.

3.6.5. Viscosity (cold & hot) Pastes

The viscosity of the malted finger millet flours was determined using a Brookfield viscometer (RV model, Brookfield Engineering, Inc., Stoughton, USA) following a procedure outlined by Ramashia et al. (2018). A beaker containing 10 g of each finger millet flours was filled with 90 mL of distilled water, and the mixture was left to hydrate for 30 min. The blend was intermittently stirred until it forms a slurry, and the viscosity of the slurry was quantified while the viscosity of the cold paste was observed. A water bath was used to heat the slurry until it reached its boiling point at 95 °C. Viscosity was observed after boiling and cooling of the paste at 30 °C.

3.7. Polyphenolic Compounds and Antioxidant Activity Measurement

Fifty grams of finger millet flours and biscuits was mixed with 500 mL of methanol. The mixture was then centrifuged (Rotina 380 R- Labotec Ecotherm (Pty) Ltd, Midrand, SA) for 10 min at 3000 rpm (Mudau et al., 2022a). Whatman no.1 filter paper was used to filter the extract and evaporated; different centrifuge tubes were stored in the refrigerator at 4 °C until used.

3.7.1. Total Phenolic Content (TPC)

The TPC extracted from finger millet flours and biscuits was evaluated using a method by Dimov et al. (2018) with minor adjustments. About 0.2 millilitre of the flour sample was incorporated in test tubes with 2.5 mL of Folin-Ciocalteu that had been diluted 5 times in 5 mL of distilled water. Seven and half millilitres of 15% sodium carbonate was added to the tubes after 5 min, and the mixture was vortexed (Model 36110740, Separation Scientific, South Africa) and afterwards, it was kept in the dark for 30 min. A spectrophotometer (UV-1600, Shimadzu Tokyo, Japan) was used to measure the absorbance values at 760 nm. The

standard curve was produced using gallic acid, and the outcomes were expressed as mg of gallic acid per gram of the sample flour.

3.7.2. Total Flavonoid Content (TFC)

A slightly altered version of Mahloko et al. (2019) approach was used to determine the TFC of finger millet flours and biscuits extract. The extract of the flour samples was mixed with 5% NaNO_2 (0.3 mL) in a tube, and the mixture was left to react for 5 min before 10% AlCl_3 (0.6 mL) was added. Distilled water and 2 mL of 1 M NaOH were added after 6 min and vortexed. The absorbance values at 510 nm were measured with a spectrophotometer. The quercetin standard ($R^2 = 0.9992$) was used to produce the standard curve, and results were presented in milligrams of quercetin per gram of flour samples (mgCE/g).

3.7.3. DPPH (2,2-Diphenyl-1-picryl-hydrazyl) Free Radical Scavenging Activity

Following the procedure outlined by Ramashia et al. (2021), DPPH assay of finger millet flours and biscuits samples (LB & DB) was measured. Two millilitres of each sample and 2 mL of 0.1 mM DPPH were mixed in 95% ethanol. After being stirred up ferociously, the combination was left to sit for 30 min at 25°C under low light. A UV spectrophotometer was used to measure the absorbance of the mixture at 517 nm. The standard curve was produced using a gallic acid solution, and results were expressed as a percentage of the inhibition of DPPH radical activity. The calibration curve equation was $y = 3.6574x + 0.0363$; R^2 was 0.9986.

3.7.4. Ferric Reducing Antioxidant Power (FRAP)

The FRAP assay of finger millet flours and biscuits was measured following a method described by Lou et al. (2017). A test tube containing 100 mL of the extracted sample from malted FM flours combined with 1 mL of methanol was used. The mixture was thoroughly blended with 2.5 mL of 1% $\text{K}_3[\text{Fe}(\text{C}_2\text{O}_4)_3]$ and 0.2 M phosphate buffer before being centrifuged for 20 min at 5000 rpm (Rotina 380 R-Labotech Ecotherm, Midrand, South Africa). One millilitre of distilled water and 0.1 mM FeCl_3 solution were added to the resultant supernatant. Utilising a spectrophotometer, the transmission density of the mixture was then measured at 700 nm. A bigger absorbance combination suggested a larger reducing power.

3.8. Determination of Thermal Characteristics

The thermal characteristics of finger millet flours were measured using differential scanning calorimetry (DSC 4000, Perkin-Elmer, Shelton, CT, USA). An empty DSC pan was used as the control, and then 25 mg of finger millet flours was placed in a sealed pan on a weighing balance. Heating of the pan was done from 20 to 130 °C at a rating of 10 °C per min. Pyris thermal system software linked to DSC was used to record the onset, peak and conclusion temperatures as the gelatinisation temperature and enthalpy range of malted and control finger millet flours (Mudau et al., 2022a).

3.9. Fourier-Transform Infrared Spectra

The Nicolet 8700 FTIR spectrometer (Thermo Scientific, Inc., located in Santa Clara, CA, USA) was used to analyse the functional groups of finger millet flours following a method by Adebiji et al. (2016). The spectral analysis included wavelengths spanning from 400 to 4000 cm^{-1} . About 0.5 grams of flour was prepared and placed on the instrument for analysis, and the spectra of the flour samples was obtained. The instrument ran 32 scans for each collected spectrum.

3.10. Scanning Electron Microscopy (SEM)

The method described by Gull et al. (2014) was used to determine the microstructure of finger millet flours with slight modifications. Using a gold palladium layer, a coater was employed for coating finger millet flour samples. The samples were placed on a sample holder throughout the coating process. To examine the microstructure of finger millet flours, scanning electron microscopy (Model: JSM 6610-LV, Chicago, IL, USA) was used. The analysis was performed at a magnification of 1000 \times and a scale of 20 μm .

3.11. Mineral analysis

Mineral content of the LB and DB finger millet biscuits was quantified using a slightly modified method of da Silva et al. (2021). A Perkin-Elmer Titan MPS absorption spectrometer was utilised for the analysis. Approximately 400 mg of biscuit sample was weighed into a digestion vessel. About 5.0 mL of HNO_3 and 2.0 mL of H_2O_2 was added. The mixture was carefully stirred with a clean glass bar. The vessel was then left to settle for 10 min before closure. The digestion procedure was done using a microwave with a specific program set. The temperature conditions for the microwave digester were 25 – 170 °C for 10 min, 170 – 240 °C for another 10 min, at 1000 W. After digestion, ventilation was done for 20 min at room

temperature. The mixtures were then allowed to cool down in 50 mL volumetric flasks before subsequent analysis.

3.12. Physical Properties of Malted Finger Millet Biscuits

The weight, diameter, thickness, and spread ratio of the LB and DB finger millet biscuits were analysed. A digital weighing balance was used to measure the weight of the biscuits (Ramashia et al., 2021b) while the diameter and thickness were determined by Vernier calliper. The spread ratio was calculated by dividing the diameter by the thickness, using the formula (W/T), where W represented the diameter and T represented the thickness of the biscuit. All analyses were done in triplicate to ensure accuracy and consistency.

3.13. Texture Characteristics

A Texture Analyzer (TA-XT2i, Stable Micro Systems, Surrey, UK) was employed to measure the hardness of the LB and DB finger millet biscuits. The equipment comprised five-kilogram cell loads, a three-point bend ring, and a heavyweight platform. The test speed was set at 3.0 mm/s, and the trigger force was set at 50 g, using automatic settings. The peak force recorded during the analysis was considered as the hardness value of the FM biscuits (Ksüz & Karaka, 2016).

3.14. Sensory Evaluation

An ethical clearance (FSEA/22/FST/06/0902) was obtained from the internal Ethics Committee at the University of Venda. The sensory evaluation of both FM biscuits was carried out in a well-ventilated sensory evaluation room at 25 °C using semi-trained panellists (including staff members and students, $n = 70$). Selected panellists were regular consumers of biscuits, and this was done to avoid bias. Prior to the evaluation, the panellists were briefed on the product and instructed to assess biscuits for appearance, colour, aroma, taste, mouthfeel/texture, and overall acceptability, rating them using a nine-point hedonic scale. The scale varied from 1 (dislike extremely) to 5 (neither like nor dislike) to 9 (like very much). The biscuits, consisting of eight pieces of each sample, were put on white plastic plates. The panellists were instructed to taste the biscuits in the order they were presented on the plates and were requested to rinse their mouth with the water provided before and after each tasting (Mudau et al., 2022b).

3.15. Statistical analysis

The analyses were performed in triplicate, and the results are presented as mean \pm standard deviation (SD). Data analysis was conducted using SPSS software version 26.0 (SPSS, Chicago, Illinois, USA) through one-way analysis of variance (ANOVA). Mean values were compared using the Duncan multiple range test, with a significance level set at $p < 0.05$.

CHAPTER 4: RESULTS AND DISCUSSION

4.1 Impact of malting periods on the colour properties of light and dark brown finger millet flours and biscuits.

The influence of malting periods on the colour properties of finger millet (FM) flours is displayed in Table 4.1a. The lightness (L^*) of the light brown (LB) and dark brown (DB) flour samples ranged from 73.92 to 76.62 and 70.56 to 74.71, respectively. The L^* values significantly increased in both FM flours. The colour of malted FM flour is often lighter than that of control FM flour (Agrahar-Murugkar et al., 2015). This is because when FM grains were immersed in water during malting, enzymatic activities took place and broke down complex molecules, including the colour pigments of the grain (Udeh et al., 2018; Adetokunboh et al., 2022). Nefale and Mashau (2018) observed an increased in L^* values of germinated FM flours.

Table 4.1b displays the colour profile of LB and DB finger millet biscuits. Figure 4.1 displays the colour images of the two FM biscuits. The L^* values of the FM biscuits ranged from 31.52 to 33.39 (LB) and 25.26 to 28.80 (DB). The L^* values significantly ($p < 0.05$) decreased with an increase in malting in both FM biscuits. Sharma et al. (2016) mentioned that low L^* values in the biscuits might be attributed to high protein content (Table 4.2b) and the Maillard reaction during the baking process. Additionally, the baking process might have intensified the darker colour of the biscuits (Nguyen et al., 2022). Sharma et al. (2016) observed a decrease in L^* values in germinated millet biscuits.

The redness (a^*) values of the malted and control FM flours ranged from 3.23 to 3.81 (LB) and 2.53 to 3.65 (DB), and the yellowness (b^*) values ranged from 7.62 to 9.49 (LB) and 6.84 to 7.09 (DB), respectively. A significant decrease in a^* values was observed in both FM flours. Malted FM flour production involves heating and drying, affecting some pigments. The presence of phenolic compounds like tannin in the testa and pericarp of the grain, which were reduced by leaching, could be responsible for the colour shift during malting (Devi et al., 2014), particularly in a^* values. The a^* values ranged from 7.88 to 8.06 (LB) and 4.92 to 5.42 (DB), and no significant difference ($p < 0.05$) was observed in both FM biscuits. The b^* values ranged from 9.15 to 9.72 (LB) and 6.96 to 7.15 (DB). The b^* values significantly decreased in LB biscuits with an increase in malting time; on the other hand, there was no significant difference ($p > 0.05$) in DB biscuits. Therefore, the reduction in b^* colour parameters of LB biscuits might be associated with the low pigment concentration on the surface of the biscuits caused by the absorption of water.

Tannins undergo polymerisation reactions when exposed to oxygen and enzymes (Siwela, 2009). These polymers might have influenced the colour changes of the malted FM flours. The b^* values significantly increased in LB flours, with a decrease in DB flours. The

decrease in b^* colour parameter of malted DB flour might be associated with decreased pigment concentration, such as tannins on the surface of FM flours by water absorption (Table 4.1a). Degradation of tannins during soaking for 10 h, probably due to the diffusion of soluble tannins into the water, might have contributed to colour loss (Boon et al., 2010). Moreover, some oxidative enzymes such as polyphenol oxidase and peroxidase were activated during malting, which resulted in browning, thus increasing the b^* (yellowness) of LB flour. Yenasew and Urga (2023) observed increased b^* values in germinated FM varieties.

The chroma of the FM flours ranged from 8.52 to 10.02 (LB) and 7.29 to 8.38 (DB). A significant increase in the chroma of the LB flours was noted, along with a decrease in the DB flours. Korus et al. (2017) and Olamiti et al. (2020) observed that the higher chroma values with the increase in malting resulted in the liberation of glycones from conjugated glycosides triggered by the stimulation of enzymes or by the synthesis of flavonoids. The high concentration of a^* and b^* colour values in LB flours could have increased chroma, and as the concentration decreased, the colour got darker (Alotaibi et al., 2021). The leaching of polyphenols during malting might have contributed to the variations in chroma of the malted FM flours (Radonjić et al., 2021). The Chroma of the FM biscuit samples ranged from 11.75 to 12.66 (LB) and 9.92 to 10.19 (DB). There was no significant difference ($p < 0.05$) between LB and DB malted finger millet biscuits.

Hue angle (H°) is the qualitative aspect of colour, typically based on greenish, reddish, and other hues (Olamiti et al., 2020). Pathare et al. (2013) and Emery et al. (2021) mentioned that hue values between 0° and 90° correspond to the red hue, whereas values beyond 90° correspond to the yellow hue. The hue angle values for the FM flour samples ranged from 63.43 to 71.20° (LB) and 64.23 to 70.36° (DB), respectively. At 72 h of malting, a higher H° was observed in LB flours in contrast to the lower DB flours. However, the hue angles for both malted FM flours were less than 90° , indicating reddish-yellow (Kortei & Akonor, 2015). Variations in the values of H° of FM flours could be due to differences in malting periods that produced different protein content (Table 4.1b) and soluble sugars. Proteins can partake in Maillard browning reactions with reducing sugars, leading to the formation of brown pigments thus affecting the H° . Azeez et al. (2022) observed an increase in hue angle in germinated brown FM flours.

Hue values ranging from 0 to 90° , correspond to the red hue, while values at 90° indicate yellow. The hue angle of the FM biscuits ranged from 49.40 to 51.12° (LB) and 51.80 to 55.75° (DB). The hue angles for both FM biscuits were between 0° to 90° thus falling under the red hue. The ΔE ranged from 0.00 to 3.26 (LB) and 0.00 to 3.62 (DB), and a significant difference was observed in both FM biscuits. The variations might be attributed to the increased availability of reducing sugars in both malted FM biscuits, which contributed to a more intense Maillard reaction because of the extended baking time (Adedara et al., 2021).

Table 4.1a: Impact of malting on colour profile of malted finger millet flours.

| Malting time (h) | L* | a* | b* | Chroma | Hue angle (H°) | Total colour different (ΔE) | YI | WI |
|-------------------------------|---------------------------|--------------------------|--------------------------|---|---|-----------------------------|----------------------------|----------------------------|
| LB finger millet flour | | | | | | | | |
| Control | 73.92 ± 0.54 ^a | 3.81 ± 0.13 ^b | 7.62 ± 0.26 ^a | 8.52 ± 0.28 ^a | 63.43 ± 0.41 ^a | - | 14.73 ± 0.21 ^a | 102.41 ± 6.21 ^a |
| M24 | 75.85 ± 0.50 ^b | 3.25 ± 0.16 ^a | 8.29 ± 0.85 ^b | 8.91 ^{bc} ± 0.75 ^{ab} | 68.59 ± 2.93 ^b | 2.12 ± 0.84 ^{ab} | 15.78 ± 0.18 ^{ab} | 108.91 ± 6.18 ^b |
| M48 | 76.42 ± 0.80 ^c | 3.20 ± 0.13 ^a | 8.82 ± 0.14 ^c | 9.38 ± 0.18 ^b | 70.05 ^{bc} ± 0.46 ^b | 2.84 ± 0.78 ^b | 16.49 ± 0.15 ^b | 117.33 ± 5.3 ^c |
| M72 | 76.62 ± 0.81 ^d | 3.23 ± 0.05 ^a | 9.49 ± 0.18 ^d | 10.02 ± 0.18 ^c | 71.20 ± 0.08 ^b | 3.34 ± 0.47 ^b | 16.45 ± 0.14 ^b | 129.48 ± 7.23 ^d |
| DB finger millet flour | | | | | | | | |
| Control | 70.56 ± 0.99 ^a | 3.65 ± 0.03 ^d | 7.56 ± 0.06 ^d | 8.38 ± 0.03 ^c | 64.23 ± 0.77 ^a | - | 15.31 ± 0.19 ^c | 99.59 ± 3.51 ^c |
| M24 | 74.16 ± 1.34 ^b | 2.53 ± 0.01 ^b | 7.09 ± 0.27 ^c | 7.53 ± 0.26 ^b | 70.36 ± 0.71 ^b | 3.80 ± 1.68 ^b | 13.66 ± 0.11 ^b | 85.94 ± 2.98 ^b |
| M48 | 74.57 ± 0.23 ^c | 2.49 ± 0.04 ^a | 6.84 ± 0.15 ^a | 7.29 ± 0.13 ^a | 70.00 ± 0.62 ^b | 4.24 ± 0.83 ^b | 13.10 ± 0.12 ^a | 82.08 ± 2.75 ^a |
| M72 | 74.71 ± 0.71 ^d | 2.65 ± 0.11 ^c | 6.91 ± 0.11 ^b | 7.40 ± 0.14 ^{ab} | 69.99 ± 0.50 ^b | 4.32 ± 1.28 ^b | 13.21 ± 0.13 ^a | 82.65 ± 2.80 ^a |

Values are illustrated by average ± standard difference. Different characters in the same line are notably different at $p < 0.05$. LB = light brown, DB = dark brown, M = malted (24, 48, 72 h), L* = lightness, a* = redness, b* = yellowness, chroma, H° = hue angle and ΔE = total colour difference, YI = yellowness index, WI = whiteness index.

Table 4. 1b: Colour profile of malted finger millet biscuits.

| Malting (h) | L* | a* | b* | Chroma | Hue | ΔE | YI | WI |
|--------------------------|---------------------------|--------------------------|--------------------------|---------------------------|---------------------------|--------------------------|---------------------------|----------------------------|
| LB finger millet biscuit | | | | | | | | |
| Control | 33.39 ± 1.94 ^b | 8.06 ± 0.63 ^a | 9.72 ± 0.47 ^b | 12.66 ± 0.73 ^b | 50.72 ± 1.20 ^b | - | 41.59 ± 0.63 ^a | 67.78 ± 0.84 ^a |
| M24 | 31.52 ± 1.52 ^a | 7.88 ± 0.24 ^a | 9.15 ± 0.71 ^a | 11.75 ± 0.41 ^a | 51.05 ± 1.09 ^b | 2.12 ± 0.94 ^a | 41.47 ± 0.61 ^a | 69.56 ± 0.92 ^b |
| M48 | 31.54 ± 1.07 ^a | 7.97 ± 0.59 ^a | 9.30 ± 0.47 ^a | 12.26 ± 0.18 ^a | 49.40 ± 1.44 ^a | 2.20 ± 0.90 ^a | 42.12 ± 0.75 ^b | 70.97 ± 1.60 ^c |
| M72 | 31.67 ± 1.50 ^a | 7.92 ± 0.56 ^a | 9.33 ± 0.49 ^a | 12.00 ± 0.22 ^a | 51.12 ± 1.39 ^b | 3.26 ± 0.93 ^b | 42.09 ± 0.72 ^b | 69.87 ± 0.98 ^{bc} |
| DB finger miller biscuit | | | | | | | | |
| Control | 28.80 ± 0.81 ^c | 5.42 ± 0.45 ^a | 6.96 ± 0.31 ^a | 10.19 ± 0.46 ^a | 51.80 ± 1.66 ^a | - | 42.68 ± 0.78 ^a | 71.92 ± 0.97 ^a |
| M24 | 25.94 ± 0.58 ^b | 5.28 ± 0.58 ^a | 7.04 ± 0.43 ^a | 9.92 ± 0.56 ^a | 55.75 ± 2.59 ^c | 3.01 ± 0.40 ^a | 44.66 ± 0.90 ^b | 74.71 ± 1.76 ^{bc} |
| M48 | 25.26 ± 1.28 ^a | 4.92 ± 0.32 ^a | 7.10 ± 0.37 ^a | 10.03 ± 0.48 ^a | 53.23 ± 0.58 ^b | 3.62 ± 1.31 ^b | 45.47 ± 1.02 ^b | 75.41 ± 1.81 ^c |
| M72 | 26.07 ± 1.40 ^b | 5.31 ± 0.66 ^a | 7.15 ± 0.65 ^a | 10.12 ± 0.91 ^a | 53.81 ± 0.95 ^b | 3.24 ± 0.60 ^a | 42.21 ± 0.71 ^a | 74.52 ± 1.52 ^b |

Values are illustrated by average ± standard deviation, n = 3. Different characters in the same column are notably different (p < 0.05). M = malted (0, 24, 48, 72 hours). YI = yellowness index, WI = whiteness index, LB = Light brown, DB = Dark brow

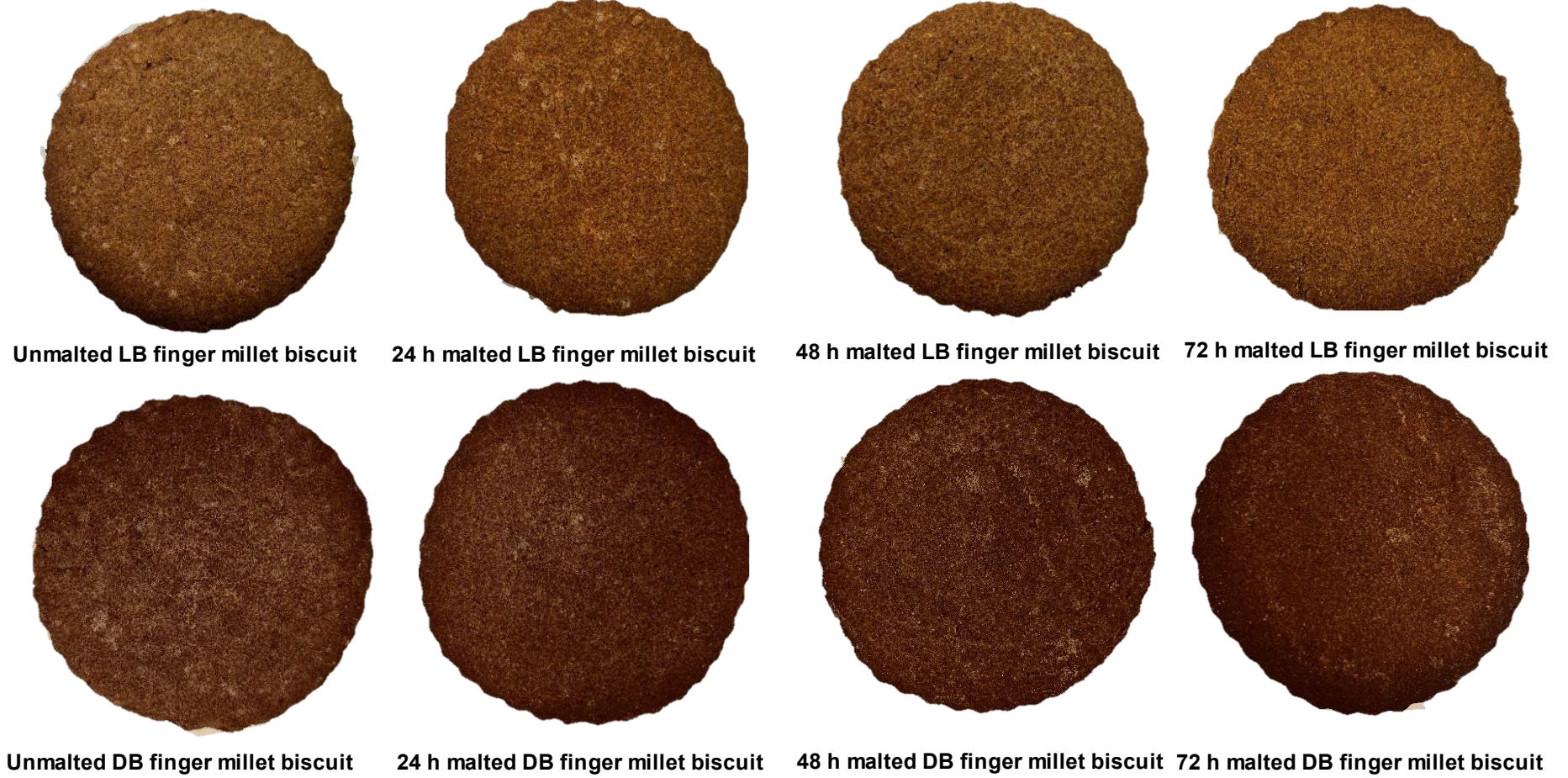


Figure 4. 1: Finger millet biscuit images. LB = light brown, DB = dark brown, h = hours

The extent of the colour shift between the malted and control FM samples is indicated by the total colour difference (ΔE) (Wirkijowska et al., 2020). The change of colour ranged from 0.00 to 3.34 (LB) and 0.00 to 4.34 (DB). Both FM flour samples showed no significant difference. Amadou and Moussa (2018) observed similar results in germinated millets. ΔE were ranging from 0.00 to 3.26 (LB) and 0.00 to 3.24 (DB).

The whiteness index (WI) and the yellowness index (YI) of FM flours are presented in Table 4.1a. The WI values of LB flours significantly increased ($p < 0.05$), with values ranging from 102.41 to 129.48. Nevertheless, the WI values of DB flour significantly decreased with an increase in malting time. The decrease in WI of malted DB flour might be due to the natural darker colour. These observations are in tandem with what Nguyen et al. (2022) observed with a decrease in WI in germinated millet flours. The YI values significantly increased in LB flours, with an increase in malting ranging from 14.73 to 16.45, whereas the YI in DB flours significantly decreased. The decreasing trends in YI of malted DB flours could be attributed to the lower b^* values observed. Table 4.1b shows the YI and WI of malted FM biscuits, with values ranging from 41.47 to 42.12 and 67.78 to 70.97 (LB), 42.21 to 45.47, and 71.92 to 75.41 (DB), respectively. There was a significant difference observed in YI and WI in both malted FM biscuits. The baking ingredients, Maillard reaction, presence of reducing sugars, and heat might have contributed to the increase observed in YI and WI values (Dimov et al., 2018).

4.2. Proximate composition of malted light and dark brown finger millet flours and biscuits

The effect of malting periods on the proximate composition of FM flours is displayed in Table 4.2a. A significant increase in the moisture content with the increased malting time was observed in both FM flours, with values ranging from 10.60 to 11.09% (LB) and 10.51 to 11.09% (DB), respectively. During the malting process, the FM grains absorbed water, which triggered the liberation of enzymes that broke down complex starches and proteins into simpler components that the growing plant could use for energy. Thus, the moisture content of the grain increased. Abioye et al. (2018) mentioned that the increased in the moisture content of malted FM flour was linked to the fact that whole grains absorbed moisture from the soaking water during germination, and more cells within the grains were moistened as the soaking duration increased. The influence of malting on the proximate composition of LB and DB finger millet biscuits is depicted in Table 4.2b. The moisture content of FM biscuits varied from 6.64 to 7.41% (LB) and 6.98 to 7.22% (DB), respectively. There was no significant difference between the malted FM biscuits and the control samples. The moisture content of

all biscuit samples was under 10%, and this is the permitted moisture content for a longer shelf life of the biscuits since most undesirable bacteria do not survive in this condition (Ramashia et al., 2021b). As similar to this study Adebisi et al. (2016) observed low moisture content in malted pearl millet biscuits.

The ash content of the malted FM flours ranged from 1.57 to 2.27% (LB) and 1.46 to 2.27% (DB). There was a significant increase ($p < 0.05$) in the ash content of malted LB and DB flours from 24 to 72 h. This was a positive effect as it resulted in more nutrient-rich flour. Guzmán-Ortiz et al. (2019) also stated that enzymes break down complex proteins and carbohydrates into simpler parts for the growing plant to use as energy during germination. Thus, the longer germination period activated enzymes, and more minerals were released from the grain. This study's ash content can be linked to Lande et al. (2017) observations on a high ash content in malted FM flours. Yenasew and Urga (2022) found that the ash content of finger millet flour decreased with longer germination periods. The decrease in ash level of germinated FM flour may be due to the elimination of shoots, roots, and bran layers. The ash content of control and malted FM biscuits ranged from 1.22 to 1.63% (LB) and 1.19 to 1.70% (DB). There was a significant increase ($p < 0.05$) in ash content in both FM biscuits, along with an increase in malting. The increase in ash content could be attributed to the high ash content observed in both FM flours (Murungweni et al., 2023). Poudel (2021) and Poornakala (2022) also observed an increase in the ash content of malted sorghum biscuits and germinated FM and pomace biscuits.

Malting increased the total dietary fibre (TDF) of FM flours with values ranging from 1.90 to 3.01% (LB) and 1.81 to 3.01% (DB), respectively. As anticipated, a significant increase ($p < 0.05$) in the TDF of both FM flours as malting increased was observed. As the malting time increased, the concentration of both soluble and insoluble fibre in the grain increased, leading to a subsequent increment in the fibre content of the resulting flours. Obadina et al. (2017) observed an increase in the TDF of pearl millet flour samples with the length of the malting time. The total dietary fibre (TDF) of the LB and DB biscuits ranged from 3.17 to 3.58% and 3.23 to 3.59%. Malting significantly increased the TDF of both FM biscuit samples. The gradual increase noted in both malted FM biscuits might be due to the high fibre seen in the malted FM flours (Murungweni et al., 2023). Caprez et al. (1986) and Sharma et al. (2018b) stated that the increase of TDF during the processes of baking and germination may arise from the generation of fibre-protein complexes that endure heat and are computed as dietary fibres. Sharma et al. (2016) observed high TDF in biscuits incorporated with germinated minor millet.

The fat content of finger millet flours ranged from 1.36 to 1.72% (LB) and 1.35 to 1.70% (DB), respectively. A significant decrease in the fat content of both FM flours was observed as the malting period increased. The decrease in fat content in both malted FM flours was

attributed to enzymes hydrolysing triacylglycerol to release free fatty acids during germination (Murungweni et al., 2023). Moreover, β -oxidation of free fatty acids took place to produce essential energy to support the growth of seeds (Jan et al., 2017; Saithalavi et al., 2021). As a result, low fat content was expected throughout germination (Cornejo et al., 2015). Kumar et al. (2021) also observed a decrease in the fat content of germinated finger millet, these observations are similar to the findings of this study. The fat content of the FM biscuit samples varied from 18.11 to 22.85% (LB) and 18.13 to 23.15% (DB) which was significantly higher as compared to the two malted flours. The increase in malting significantly reduced the fat content of both malted FM biscuits. This could be because of the fatty acid oxidation and lipid hydrolysis that transpired throughout seed germination and the activity of lipolytic enzymes that utilised fat as a source of energy (Hiremath & Geetha, 2019; Kaushik et al., 2021). The hydrolysed glycerol joins the carbohydrate pool, and fatty acids are oxidised, which causes the fat content of the malted grain to decrease (Maldonado-Celis et al., 2019). The low-fat content has the potential to extend the shelf life of malted FM biscuits by reducing the occurrence of rancidity (Yenasew & Urga, 2022). Rana et al. (2021) observed comparable results in sprouted *ragi* biscuits. Some authors have reported an increase in the fat content of biscuits (Adebiyi et al., 2017), and this was attributed to the addition of fat during dough preparation.

Table 4.2a: Impact of malting time (h) on the proximate composition of finger millet flours.

| Malting time (h) | Moisture (%) | Ash (%) | Fibre (%) | Fat (%) | Protein (%) | Carbohydrate (%) | Energy (kcal/100g) |
|-------------------------------|----------------------------|--------------------------|---------------------------|---------------------------|---------------------------|---------------------------|-----------------------------|
| LB finger millet flour | | | | | | | |
| Control | 10.60 ± 0.80 ^a | 1.57 ± 0.37 ^a | 1.90 ± 0.10 ^a | 1.72 ± 0.09 ^c | 10.80 ± 0.50 ^a | 72.03 ± 0.69 ^a | 353.00 ± 0.75 ^a |
| M24 | 10.75 ± 0.81 ^{ab} | 1.68 ± 0.08 ^a | 2.36 ± 0.09 ^b | 1.59 ± 0.08 ^{bc} | 11.20 ± 0.87 ^b | 71.96 ± 0.54 ^a | 352.66 ± 2.530 ^a |
| M48 | 10.93 ± 0.54 ^{bc} | 1.80 ± 0.03 ^b | 2.85 ± 0.11 ^c | 1.42 ± 0.04 ^{ab} | 11.89 ± 0.18 ^c | 71.56 ± 0.04 ^a | 351.30 ± 0.99 ^a |
| M72 | 11.09 ± 0.50 ^c | 2.27 ± 0.04 ^c | 3.01 ± 0.02 ^c | 1.36 ± 0.03 ^a | 12.20 ± 0.36 ^c | 71.44 ± 0.29 ^a | 350.50 ± 0.72 ^a |
| DB finger millet flour | | | | | | | |
| Control | 10.51 ± 0.99 ^a | 1.46 ± 0.40 ^a | 1.81 ± 0.11 ^a | 1.70 ± 0.02 ^c | 10.75 ± 0.55 ^a | 72.57 ± 0.47 ^d | 352.53 ± 1.32 ^d |
| M24 | 10.92 ± 1.34 ^b | 1.65 ± 0.04 ^b | 2.38 ± 0.13 ^b | 1.58 ± 0.10 ^{bc} | 11.48 ± 0.07 ^b | 71.56 ± 0.31 ^c | 351.97 ± 0.86 ^c |
| M48 | 11.01 ± 0.23 ^c | 1.77 ± 0.06 ^c | 2.82 ± 0.11 ^c | 1.40 ± 0.05 ^{ab} | 11.85 ± 0.35 ^c | 71.34 ± 0.59 ^b | 351.17 ± 0.62 ^b |
| M72 | 11.09 ± 0.71 ^d | 2.21 ± 0.10 ^d | 3.01 ± 0.13 ^{cd} | 1.35 ± 0.03 ^a | 12.30 ± 0.36 ^d | 71.24 ± 0.87 ^a | 349.25 ± 3.39 ^a |

Values are illustrated by average ± standard difference. Different characters in the same column are notably different ($p < 0.05$). LB = light brown, DB = dark brown, M = malted (24, 48, 72 h).

Table 4.2b: Impact of malting time (h) on the proximate constitution of finger millet biscuits.

| Malting time (h) | Moisture (%) | Ash (%) | Fibre (%) | Fat (%) | Protein (%) | Carbohydrate (%) | Energy Content (kcal/100g) |
|----------------------------------|--------------------------|---------------------------|---------------------------|----------------------------|--------------------------|----------------------------|-----------------------------|
| LB finger millet biscuits | | | | | | | |
| Control | 7.41 ± 1.04 ^a | 1.22 ± 0.10 ^a | 3.17 ± 0.06 ^a | 22.85 ± 0.65 ^d | 8.54 ± 0.12 ^a | 60.18 ± 0.37 ^d | 476.13 ± 6.30 ^d |
| M24 | 6.85 ± 0.56 ^a | 1.45 ± 0.03 ^b | 3.19 ± 0.05 ^b | 21.72 ± 0.29 ^c | 8.74 ± 0.08 ^b | 59.43 ± 0.25 ^c | 469.27 ± 0.44 ^c |
| M48 | 6.71 ± 0.30 ^a | 1.57 ± 0.06 ^b | 3.34 ± 0.07 ^{bc} | 19.78 ± 0.38 ^b | 9.19 ± 0.05 ^c | 58.11 ± 0.91 ^{bc} | 459.15 ± 2.52 ^b |
| M72 | 6.64 ± 0.55 ^a | 1.63 ± 0.08 ^c | 3.58 ± 0.03 ^d | 18.11 ± 0.70 ^a | 9.50 ± 0.03 ^d | 57.49 ± 0.86 ^a | 448.86 ± 4.97 ^a |
| DB finger millet biscuits | | | | | | | |
| Control | 7.22 ± 0.48 ^a | 1.19 ± 0.07 ^a | 3.23 ± 0.09 ^a | 23.15 ± 1.08 ^d | 8.60 ± 0.34 ^a | 60.15 ± 0.91 ^d | 473.68 ± 4.00 ^d |
| M24 | 7.13 ± 0.60 ^a | 1.32 ± 0.08 ^{ab} | 3.24 ± 0.09 ^{ab} | 20.66 ± 0.45 ^{bc} | 8.76 ± 0.09 ^b | 59.27 ± 1.17 ^c | 462.75 ± 2.60 ^{bc} |
| M48 | 7.07 ± 0.57 ^a | 1.44 ± 0.11 ^{bc} | 3.38 ± 0.08 ^c | 19.75 ± 0.12 ^b | 9.22 ± 0.19 ^c | 58.82 ± 0.51 ^{bc} | 458.42 ± 2.42 ^b |
| M72 | 6.98 ± 0.32 ^a | 1.70 ± 0.03 ^d | 3.59 ± 0.07 ^d | 18.13 ± 0.78 ^a | 9.51 ± 0.08 ^d | 56.75 ± 1.35 ^a | 449.02 ± 4.43 ^a |

Values are illustrated by average ± standard deviation, n = 3. Different letters in the same column are notably different (p < 0.05). M = malted (0, 24, 48, 72 hours), LB = light brown, and DB = dark brown

The protein content of FM flours ranged from 10.80 to 12.20% (LB) and 10.75 to 12.30% (DB). There was a significant increase in the protein content of both FM flours with the increased in the malting time. The increased in protein content of the malted FM flours might be due the formation of enzymes or an encompassing change following degradation of other constituents (Ijarotimi, 2012; Owhero et al., 2019). Moreover, the increased in protein content could be the effect of dry weight loss because of the utilisation of some fats and carbohydrates during respiration and the synthesis of some amino acids during germination (Devi et al., 2015; Saithalavi et al., 2021). The protein content of FM flours increased with a longer malting time. This was in line with earlier research findings that crop protein content increased during germination (Abioye et al., 2018). Hejazi and Orsat (2017); Chauhan et al. (2018) observed a high protein content in germinated FM flour. The protein content of the FM biscuits varied from 8.54 to 9.50% (LB) and 8.60 to 9.51% (DB). A significant ($p < 0.05$) increase in the protein content with the increase in malting time in both FM biscuits was observed. According to Sharma and Gujral (2020), the increase may be the result of enzyme production after the breakdown of other malt components like carbohydrates and lipids. In addition, the high protein content observed in the FM flours also contributed to the increment in the protein content both FM biscuits. Adebisi et al. (2017) observed a high protein content in malted pearl millet biscuits.

The carbohydrate content of FM flours ranged from 71.44 to 72.03% (LB) and 71.24 to 72.57% (DB). No significant difference was observed in the LB flour samples, but a significant decrease was noted in DB flour samples. Malting enhanced the enzymatic degradation of carbohydrates in DB flour into simple sugars by stimulating endogenous enzymes like α -amylase, improving digestibility because starch was broken down to give energy for the seed growth (Oghbaei & Prakash, 2016; Samtiya et al., 2020). Malting reduces the carbohydrate content of grains, which may benefit individuals with certain health conditions, such as diabetes or obesity (Ojedokun et al., 2020). Gowda et al. (2022) observed low carbohydrate contents in malted pearl and kodo millet. The carbohydrate content of the LB and DB finger millet biscuits ranged from 57.49 to 60.18% and 56.75 to 60.15%, respectively. There was a significant ($p > 0.05$) decrease in the carbohydrate content with an increase in malting time. The decrease in both FM biscuits could be attributed to the breakdown of carbohydrate content into simple sugars (Nkhata et al., 2018).

The energy content of FM flours ranged from 350.50 to 353.00 kcal/100g (LB) and 349.25 to 352.53 kcal/100g (DB), respectively. A significant decrease was observed in the energy value of both malted FM flours compared to both control FM flours. The germination process caused the breakdown of complex carbohydrates, which reduced carbohydrates in the flour (Saithalavi et al., 2021). Kulla et al. (2021) observed a decreased in the energy content of malted pearl millet flour. The energy values of FM biscuit samples ranged from

448.86 to 476.13 kcal/100 g (LB) and 449.02 to 473.68 kcal/100 g (DB). The energy content significantly ($p > 0.05$) decreased with an increase in the malting period in both FM biscuits. Bolarinwa et al. (2016) and Himabindu and Devanna (2017) reported low carbohydrate content in malted sorghum-soy biscuits and biscuits incorporated with malted kodo and little millet. Therefore, the incorporation of malting decreased the carbohydrate content.

4.3. Impact of malting on functional characteristics of light and dark brown finger millet flours

Table 4.3 shows the impact of malting on the functional properties of FM flours. The solubility index of FM flours significantly increased with an increase in malting time from 24 to 72 h compared to the control samples. The values ranged from 1.78 to 3.80% (LB) and 1.79 to 3.90 (DB), respectively. The increase in solubility index might be attributed to the elevated changes that starch molecules undergo that cause them to break down into simpler sugars, resulting in these sugars having a high solubility index for both malted FM flours (Nefale & Mashau, 2018). The malting process decreased the size of the particles in millet flour, resulting in an increase in the surface area of the flour, which enhanced its ability to absorb water and, consequently, increased its solubility index (Panda et al., 2020). Kumar et al. (2021) found that as the germination time of FM increased, the water solubility index showed a significant increase ($p \leq 0.05$).

The packed bulk density of malted and control FM flour samples ranged from 0.74 to 0.81 g/mL (LB) and 0.73 to 0.82 g/mL (DB), respectively. The packed bulk density of both FM flour samples significantly decreased with an increase in malting periods. The same trend was also observed for loose bulk density. The decrease in bulk density (packed and loose) might be attributed to the degradation of complex composites, such as proteins and starch, caused by alterations during germination (Ocheme et al., 2015). The breakdown of starch during malting decreased starch content, resulting in low bulk density (Oti & Akobundu, 2008; Ojha et al., 2018). The changes that took place at the molecular level during malting could have resulted in a reduction of inter-particle attraction, thereby reducing the bulk density. Low bulk density flour is suitable for infant formulation and might be useful in various bakery products (Okoye et al., 2010). Adebisi et al. (2016) and Obadina et al. (2017) observed a decrease in bulk density of malted finger and pearl millet flours.

Table 4.3: Functional characteristics of malted light brown and dark brown finger millet flours.

| Malting time (h) | SOLB (%) | PBD (g/g) | LBD (g/g) | WAC (g/g) | OAC (g/g) | SP (mL) | Visc Cold paste (cP) | Visc Cooked paste (cP) |
|-------------------------------|--------------------------|--------------------------|--------------------------|---------------------------|--------------------------|---------------------------|---------------------------|----------------------------|
| LB finger millet flour | | | | | | | | |
| Control | 1.78 ± 0.01 ^a | 0.81 ± 0.01 ^b | 0.68 ± 0.01 ^c | 1.55 ± 0.01 ^a | 1.31 ± 0.01 ^a | 10.38 ± 0.04 ^a | 19.59 ± 0.66 ^a | 424.00 ± 1.00 ^d |
| M24 | 2.05 ± 0.06 ^b | 0.79 ± 0.01 ^b | 0.64 ± 0.03 ^c | 1.68 ± 0.01 ^{ab} | 1.36 ± 0.01 ^b | 10.00 ± 0.05 ^a | 19.52 ± 0.67 ^a | 376.00 ± 1.00 ^c |
| M48 | 2.93 ± 0.15 ^c | 0.75 ± 0.01 ^a | 0.59 ± 0.01 ^b | 1.70 ± 0.02 ^d | 1.42 ± 0.01 ^c | 9.89 ± 0.01 ^a | 20.00 ± 0.51 ^b | 335.67 ± 1.15 ^b |
| M72 | 3.80 ± 0.10 ^d | 0.74 ± 0.01 ^a | 0.54 ± 0.01 ^a | 1.72 ± 0.02 ^c | 1.49 ± 0.01 ^d | 9.88 ± 0.01 ^a | 20.62 ± 0.77 ^c | 285.00 ± 5.00 ^a |
| DB finger millet flour | | | | | | | | |
| Control | 1.79 ± 0.02 ^a | 0.82 ± 0.02 ^c | 0.68 ± 0.01 ^d | 1.54 ± 0.02 ^a | 1.32 ± 0.01 ^a | 10.15 ± 0.06 ^a | 19.59 ± 0.95 ^a | 418.00 ± 1.00 ^d |
| M24 | 2.09 ± 0.11 ^b | 0.79 ± 0.01 ^b | 0.63 ± 0.02 ^c | 1.69 ± 0.02 ^b | 1.36 ± 0.01 ^b | 10.00 ± 0.03 ^a | 20.33 ± 0.79 ^c | 386.00 ± 1.00 ^c |
| M48 | 3.03 ± 0.15 ^c | 0.74 ± 0.01 ^a | 0.58 ± 0.01 ^b | 1.73 ± 0.01 ^c | 1.43 ± 0.02 ^c | 9.92 ± 0.03 ^a | 20.08 ± 0.99 ^b | 321.00 ± 1.00 ^b |
| M72 | 3.90 ± 0.10 ^d | 0.73 ± 0.10 ^a | 0.53 ± 0.01 ^a | 1.78 ± 0.02 ^d | 1.48 ± 0.01 ^d | 9.89 ± 0.01 ^a | 20.46 ± 0.76 ^d | 271.33 ± 1.53 ^a |

Values are illustrated by average ± standard difference. Different characters in the same column are notably different ($p < 0.05$). LB = light brown, DB = dark brown, M = malted (24, 48, 72 h). SOLB = solubility, PBD = packed bulk density, LBD = loose bulk density, WAC = water absorption capacity, OAC = oil absorption capacity, SP = swelling power, Visc = viscosity (cold and cooked paste), cP = specific heat capacity.

The water absorption capacity (WAC) of malted and control FM flours ranged from 1.55 to 1.72 g/g (LB) and 1.54 to 1.78 g/g (DB). A significant increase in the WAC of both FM flours with an increase in malting periods was observed. The formation of molecules such as soluble sugars, which have a high water-holding capacity, may have contributed to the improved WAC in both malted FM flours (Nefale & Mashau, 2018; Oskaybaş-EmLek et al., 2021). Abah et al. (2020) stated that flour that can absorb water may contain a higher proportion of hydrophilic components, such as polysaccharides. Moreover, the low WAC observed in control FM flours suggests low water-binding hydrophilic groups (Adebowale et al., 2005; Mudau et al., 2022a). Yenasew and Urga (2023) observed similar increases in the WAC of germinated FM flours.

The oil absorption capacity (OAC) of FM flour samples ranged from 1.31 to 1.49 g/g (LB) and 1.32 to 1.48 g/g (DB), respectively. The OAC values of both FM flour samples showed a significant increase with malting time. The increase in OAC observed during malting could be attributed to the breakdown of starch during germination, as hydrolysed starch has a higher capacity to absorb both water and oil (Horstmann et al., 2017). Ojha et al. (2018), observed an increase in the OAC of malted sorghum flour and suggested that the increased ability of sorghum flour to bind with oil indicated its potential usefulness in food formulations where retaining oil was a significant factor to consider. Thus, the increased OAC in both malted FM flour samples suggested an increased potential in food formulations. Nazni and Shobana (2016) observed an increase in OAC during the germination of barnyard and foxtail millet samples.

The swelling capacity of FM flour samples ranged from 9.88 to 10.38 g/g (LB) and 9.89 to 10.15 g/g (DB), respectively. The swelling power of both malted FM flours significantly decreased with an increase in malting time. Low swelling power in both malted FM flours could be attributed to alterations in protein content or quality, as some proteins may form complexes with starch molecules and reduce their ability to swell (Wilson et al., 2022). Nefale and Mashau (2018) and Yenasew and Urga (2023) noted a decrease in swelling power in germinated FM flours.

The viscosity of FM flours is presented in Table 4.3. Finger millet flours' cold paste viscosity values ranged from 19.52 to 20.62 cP (LB) and 19.59 to 20.46 cP (DB), respectively. The hot paste values of FM flours ranged from 285.00 to 424.00 cP (LB) and 271.33 to 418.00 cP (DB), respectively. A significant difference in the cold paste viscosity was observed in both FM flours. Malting increased the cold paste viscosity of flour by altering its composition, specifically by increasing the level of soluble fibre and decreasing the amount of starch (Claver et al., 2010; Aswalekar et al., 2021). This increase in soluble fibre was significant ($p < 0.05$) since it formed gels that contributed to the cold paste viscosity of the flour and could hold water effectively (Shand, 2000; Ahmed et al., 2019). A significant reduction ($p > 0.05$) in the viscosity

of the cooked paste with each malting stage in both FM flours was observed. The reduction was mainly caused by alterations in the composition of flour and enzymatic activity, which decreased its starch content (Atuna et al., 2022). The decrease in viscosity in both malted FM flours showed that the flour was suitable for producing infant foodstuffs (Kaushik et al., 2021). Sharma et al. (2021) observed similar results in germinated kodo millet flours.

4.4. Impact of malting period on the polyphenols and antioxidant capacity of light brown and dark brown finger millet flours and biscuits

Table 4.4a illustrates the polyphenols and antioxidant activity of malted FM flours. Comparing the malted and control FM flours, TPC increased significantly ($p < 0.05$) from 67.74 to 94.22 mg (GAE)/100 g (LB) and 68.57 to 95.40 mg (GAE)/100 g (DB). Enzymes that break down the cell wall became functional during malting and altered the structure of the cell wall of the grain, increasing the content of phenolic chemicals in FM flours (Arya, 2022). During the malting process, the effectiveness of anti-nutrients such as tannins and phytic acid decreases (Ram et al., 2020). These anti-nutrients have an affinity for phenolic compounds. Therefore, removing these anti-nutrients increases the overall phenolic content of malted finger millet flours (Yousaf et al., 2021). Azeez et al. (2022) recorded an increased TPC in germinated brown FM flour. The polyphenols and antioxidant capacity of the LB and DB finger millet biscuits are presented in Table 4.4b. The total phenolic content significantly increased ($p < 0.05$) from 57.03 to 77.18 mg (GAE)/100 g (LB) and 53.77 to 75.76 mg (GAE)/100 g (DB) with an increase in malting time. A significant ($p < 0.05$) increase was also observed in total flavonoid content, ranging from 3.14 to 3.31 mg QE/g (LB) and 3.18 to 3.28 mg QE/g (DB). Taylor and Duodo, (2015) stated that the increase in the total phenolic and total flavonoid contents likely resulted from the liberation of phenolic compounds that were previously bound within the cell walls of FM grains. The release of bound phenolic compounds occurs when cell wall-breaking enzymes, primarily esterases, act on these bonds, triggering the liberation of phenolic compounds (Perales-Sánchez et al., 2014). Adebisi et al. (2017) and Bello et al. (2017) observed an increase in the total phenolic and total flavonoid contents of malted pearl millet biscuits.

The TFC of light and dark brown FM flours ranged from 7.22 to 10.18 mg QE/g (LB) and 7.28 to 10.21 mg QE/g (DB). A significant ($p < 0.05$) increase in TFC in both FM flour samples at each malting period was observed. The increase in TFC at each malting period might be because of the metabolic alterations that took place in the grain and produced secondary metabolites or flavonoids (Kaur & Gill, 2021). Furthermore, malting resulted in the liberation of glycones from conjugated glycosides triggered by the stimulation of enzymes or

by the alteration or synthesis of flavonoids (Sharma et al., 2022). Sharma et al. (2018) observed that malting at 48 h substantially increased the TFC of foxtail millet flour.

Table 4.4a: Polyphenols and antioxidant activity of malted light brown and dark brown FM flours.

| Malting time (h) | TPC (mg GAE/g) | TFC (mg QE/g) | DPPH (%) | FRAP (mg GAE/g) |
|-------------------------------|---------------------------|---------------------------|---------------------------|--------------------------|
| LB finger millet flour | | | | |
| Control | 67.74 ± 1.01 ^a | 7.22 ± 1.05 ^a | 67.31 ± 1.83 ^a | 0.93 ± 0.00 ^a |
| M24 | 71.42 ± 1.26 ^b | 8.48 ± 0.74 ^{ab} | 73.32 ± 1.69 ^b | 0.87 ± 0.02 ^b |
| M48 | 81.55 ± 1.64 ^c | 9.30 ± 0.97 ^b | 82.52 ± 0.86 ^c | 1.33 ± 0.10 ^c |
| M72 | 94.22 ± 3.71 ^d | 10.18 ± 0.95 ^b | 90.70 ± 2.29 ^d | 1.88 ± 0.12 ^d |
| DB finger millet flour | | | | |
| Control | 68.57 ± 0.77 ^a | 7.28 ± 0.94 ^a | 76.61 ± 0.87 ^a | 0.96 ± 0.00 ^a |
| M24 | 72.53 ± 1.26 ^b | 8.41 ± 1.05 ^b | 78.02 ± 4.91 ^b | 0.90 ± 0.02 ^a |
| M48 | 83.62 ± 1.40 ^c | 9.41 ± 0.78 ^c | 87.43 ± 1.38 ^c | 1.41 ± 0.17 ^b |
| M72 | 95.40 ± 4.59 ^d | 10.21 ± 0.41 ^d | 95.14 ± 1.59 ^d | 1.87 ± 0.12 ^c |

Values are illustrated by average ± standard difference. Different symbols in the same column are notably different at $p < 0.05$. LB = light brown, DB = dark brown, M = malted (24, 48, 72 h), TPC= total phenolic content, TFC = total flavonoids content, DPPH = 2,2-Diphenyl- 1-picrylhydrazyl, FRAP = ferric reducing antioxidant power.

Table 4. 4b: Antioxidant properties of malted finger millet biscuits.

| Malting (h) | TPC (mg GAE/g) | TFC (mg QE/g) | DPPH (%) | FRAP (mg GAE/g) |
|--------------------------|---------------------------|--------------------------|---------------------------|---------------------------|
| LB finger millet biscuit | | | | |
| Control | 57.03 ± 6.64 ^a | 3.14 ± 0.34 ^a | 54.24 ± 1.23 ^a | 0.82 ± 0.03 ^a |
| M24 | 66.06 ± 5.26 ^b | 3.17 ± 0.22 ^a | 60.02 ± 0.53 ^b | 0.89 ± 0.05 ^c |
| M48 | 73.21 ± 7.14 ^c | 3.29 ± 0.05 ^b | 67.31 ± 4.72 ^c | 0.93 ± 0.01 ^c |
| M72 | 77.18 ± 6.98 ^d | 3.31 ± 0.08 ^b | 73.32 ± 1.34 ^d | 0.87 ± 0.02 ^b |
| DB finger millet biscuit | | | | |
| Control | 53.77 ± 4.63 ^a | 3.18 ± 0.49 ^a | 58.58 ± 0.86 ^a | 0.88 ± 0.04 ^a |
| M24 | 61.17 ± 5.12 ^b | 3.26 ± 0.07 ^b | 69.11 ± 2.15 ^b | 0.92 ± 0.06 ^b |
| M48 | 68.74 ± 5.28 ^c | 3.27 ± 0.15 ^b | 76.61 ± 1.22 ^c | 0.96 ± 0.15 ^c |
| M72 | 75.76 ± 8.64 ^d | 3.28 ± 0.18 ^b | 84.68 ± 1.78 ^d | 0.90 ± 0.07 ^{ab} |

Values are illustrated by average ± standard deviation, n = 3. Different characters in the same column are notably different (p < 0.05). M = malted (0, 24, 48, 72 hours). TPC= total phenolic content, TFC = total flavonoids content, LB = Light brown, DB = Dark brown.

The DPPH values significantly (p < 0.05) increased in both FM flours, ranging from 67.31 to 90.70% (LB) and 76.61 to 95.14% (DB), respectively. The activity of enzymes that released bound antioxidants and the reduction of anti-nutritional factors during malting contributed to the high values of DPPH in both FM flours. The increase in DPPH values suggested that the antioxidant capacity of both FM flours increased, which positively influences consumers' health (Nkhata et al., 2018). Sharma et al. (2021) observed an increment in DPPH radical scavenging activity with malting time in kodo millet. The FRAP values of FM flour samples ranged from 0.87 to 1.88 mg/g GAE (LB) and 0.90 to 1.87 mg/g GAE (DB), respectively. A

significant ($p < 0.05$) increase in FRAP with the malting periods in both FM flours from 48 and 72 h was observed. The consumption or transformation of antioxidants by the enzymes generated during the malting process could have contributed to the low FRAP assay at 24 h in both malted FM flours (Yang et al., 2021). DPPH values of FM biscuits significantly ($p < 0.05$) increased from 54.24 to 73.32% (LB) and 58.58 to 84.68% (DB) with an increase in malting time. The scavenging activity is directly proportional to the DPPH value of the biscuits; thus, a higher DPPH value indicates a stronger ability of both FM biscuit samples to scavenge free radicals (Mudau et al., 2022b). Therefore, the high DPPH observed at 72 h in both malted FM biscuits suggested higher scavenging activity. The high DPPH values in malted FM flours could be due to an increase in the accessibility of certain polyphenolic compounds that were previously bound to polysaccharides and proteins. This implied that the ability to scavenge DPPH free radicals was positively linked to the total phenolic contents (Sharma et al., 2016). Chauhan et al. (2015) stated that the antioxidants of biscuits might be further enhanced during the baking process. The generation of brown colour during the baking process increased the antioxidant properties of the malted FM biscuits. Chauhan et al. (2015), Sharma et al. (2016), and Oskaybaş-EmLek et al. (2021) observed DPPH increments in germinated amaranth, minor millet, and lentil biscuits.

In contrast, the increased FRAP assay at 48 to 72 h in both malted FM flours was likely due to the production of new antioxidants and further generation of metabolites produced during malting, as well as the release of phenolic compounds (Yang et al., 2021). Sharma et al. (2018) observed an increase in FRAP in malted foxtail millet flours. Foods that are high in antioxidants may reduce oxidative stress and decrease the risk of conditions like cardiovascular disease, cancer, and neurodegenerative disorders (Pham-Huy et al., 2008). FRAP values of FM biscuit samples ranged from 0.82 to 0.93 mg/g GAE (LB) and 0.88 to 0.96 mg/g GAE (DB). A significant ($p < 0.05$) increase from 24 to 48 h of malting was observed, with a significant decrease at 72 h in both FM biscuits. The baking process might have led to the formation of new antioxidant compounds or the release of bound phenolics, further contributing to the increased FRAP activity (Reis & Abu-Ghannam, 2014). The application of heat could have resulted in the disruption of the cell wall, facilitating the liberation of more antioxidant compounds. Consequently, this might have led to an increase in antioxidant activity (Balasubramaniam et al., 2019). Omoba et al. (2015) stated that heat applied during baking increases the antioxidant capacity. Baking involves high temperatures, which can initiate the Maillard reaction between reducing sugars and amino acids in food ingredients. This reaction leads to the formation of various compounds with antioxidant properties. Antioxidants are stabilisers of lipids, preventing excessive oxidation associated with ageing and cancer (SK and Sudha, 2012; Pisoschi and Pop, 2015). In contrast, the significant decrease in FRAP

values at 72 h could be due to the depletion or complete utilisation of the antioxidants during germination (Yang et al., 2021).

4.5. Thermal characteristics of malted light and dark brown finger millet flours

The thermal properties of the malted and control FM flours are presented in Table 4.5. The more extended malting period in both the LB and DB flours increased the onset temperature (T_o), peak temperature (T_p), and conclusion temperature (T_c). A significant difference in the malted and control FM flours in relation to the onset, peak, and conclusion temperatures was observed. The FM flour samples malted for 72 h exhibited the elevated temperatures required for gelatinisation. The peak temperature increase after malting could be due to the buildup of proteolytic enzymes produced by indigenous microorganisms, which degraded the walls of the grain cell, leading to a greater liberation of starch and larger dimensions of crystalline structures in the sample (Mudau et al., 2022a). Malting modified the structure of the macromolecular or configuration of amylose and amylopectin present in flour granules and caused variations in gelatinisation temperatures (Su et al., 2020). Similarly, as observed in this study, Sharma et al. (2018) and Li et al. (2020) observed an increase in (T_o) and (T_p) of germinated millet, sorghum, and foxtail millet.

The differences in gelatinisation temperatures, including T_o , T_p , and T_c values, observed in both malted FM flours might be attributed to inherent alterations in granule size, morphology, distribution of starch, and organisation of internal starch fractions within the granules (Nagaprabha et al., 2017). The observation of increased gelatinisation temperatures in both FM flours at 72 h of malting indicated that greater force was necessary to initiate the starch gelatinisation. An increase in gelatinisation temperatures observed in malted LB and DB flours might be attributed to the generation of amino acids, which resulted from modifying proteins during germination (Gebremariam et al., 2014; Azeez et al., 2022). Starches that exhibit lower gelatinisation temperatures, like those in both control and 24 h malted flours, have superior culinary quality (Waters et al., 2006; Tomar et al., 2022). No significant ($p > 0.05$) differences were observed in both malted FM flours in relation to the range of gelatinisation temperatures. These temperatures affect the texture, viscosity, stability, cooking time and energy consumption of the products that can be produced from the flours.

Table 4. 5: Thermal characteristics of malted light brown and dark brown finger millet flours

| Malting time (h) | T _o (°C) | T _p (°C) | T _c (°C) | T _r (°C) | ΔH (J/g) |
|-------------------------------|---------------------------|---------------------------|---------------------------|--------------------------|--------------------------|
| LB finger millet flour | | | | | |
| Control | 76.03 ± 1.15 ^a | 80.41 ± 0.93 ^a | 86.04 ± 0.45 ^a | 3.33 ± 1.56 ^a | 5.38 ± 0.76 ^d |
| M24 | 78.22 ± 1.09 ^b | 82.64 ± 0.81 ^b | 87.21 ± 0.32 ^b | 3.08 ± 0.63 ^a | 5.04 ± 0.68 ^c |
| M48 | 79.62 ± 1.02 ^c | 83.26 ± 0.72 ^c | 89.15 ± 0.28 ^c | 3.65 ± 1.20 ^a | 4.80 ± 1.12 ^b |
| M72 | 82.18 ± 0.95 ^d | 85.21 ± 0.65 ^d | 90.26 ± 0.15 ^d | 3.82 ± 1.42 ^a | 4.32 ± 0.97 ^a |
| DB finger millet flour | | | | | |
| Control | 74.16 ± 1.35 ^a | 75.91 ± 1.26 ^a | 80.12 ± 0.95 ^a | 8.78 ± 1.84 ^a | 4.65 ± 1.72 ^c |
| M24 | 75.20 ± 1.24 ^b | 76.18 ± 1.18 ^b | 80.83 ± 0.92 ^b | 8.54 ± 1.22 ^a | 4.52 ± 1.55 ^c |
| M48 | 76.07 ± 1.14 ^c | 76.01 ± 1.13 ^a | 81.13 ± 0.83 ^c | 8.88 ± 0.56 ^a | 4.30 ± 0.99 ^b |
| M72 | 77.52 ± 1.08 ^d | 77.70 ± 1.09 ^c | 82.15 ± 0.78 ^d | 9.38 ± 0.67 ^a | 4.15 ± 1.09 ^a |

Values are illustrated by average ± standard difference. Different letters in the same column show notable differences ($p < 0.05$). LB = light brown, DB = dark brown, M = malted (24, 48, 72 h). T_o= onset temperature, T_p = peak temperature, T_c= conclusion temperature, ΔH= gelatinisation enthalpy.

4.6. Fourier-Transform Infrared Spectra (FTIR) of malted light and dark brown finger millet flours

Figure 4.2 illustrates the FTIR analysis of the functional groups in malted and control FM flours. There were no significant changes in the spectra of malted and control FM flours, but slight variations in the intensity of the peaks were observed. The LB peaks ranged from 3000 to 3600 cm⁻¹, and the DB peaks ranged from 3000 to 3650 cm⁻¹. A broad stretch was observed, indicating the presence of the O-H group. This was caused by water, alcohol, carboxylic acids, and the interaction between protein and starch (Kaur & Prasad, 2022).

The peaks of the FM flours were in the range of 3253 to 3275 cm⁻¹ (LB) and 3252 to 3277 cm⁻¹ (DB) in the O-H region, respectively. The peaks detected in the spectra could be attributed to the vibrational stretching of the O-H. Variations in the intensity of the peaks might be associated with variations in the moisture content of the flours resulting from the malting process, as shown in Table 4.2a. As the malting period increased, the width of the C-H band

in both malted FM flours widened, with peaks absorption falling between 2914 to 2920 cm^{-1} (LB) and 2925 to 2927 cm^{-1} (DB), respectively. The widening was most likely due to vibrations stretching of both aliphatic and aromatic C-H bonds (Olamiti et al., 2020). The peak variations could be related to variations in fat content in both malted FM flours (Table 4.2a).

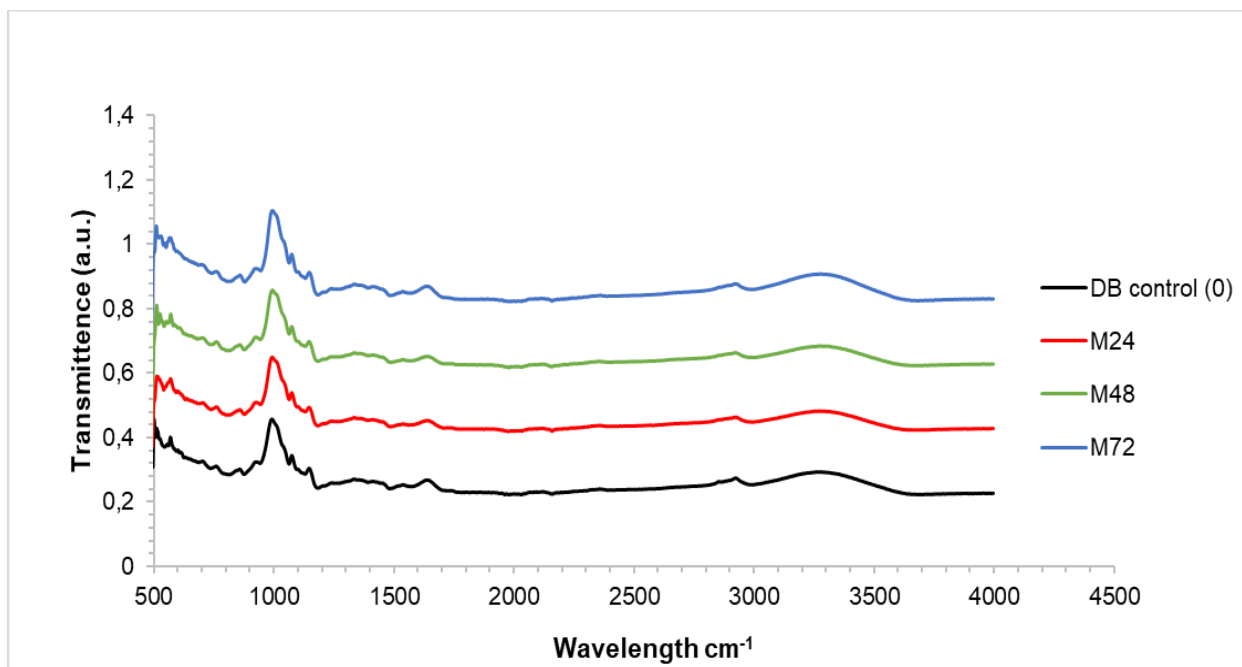
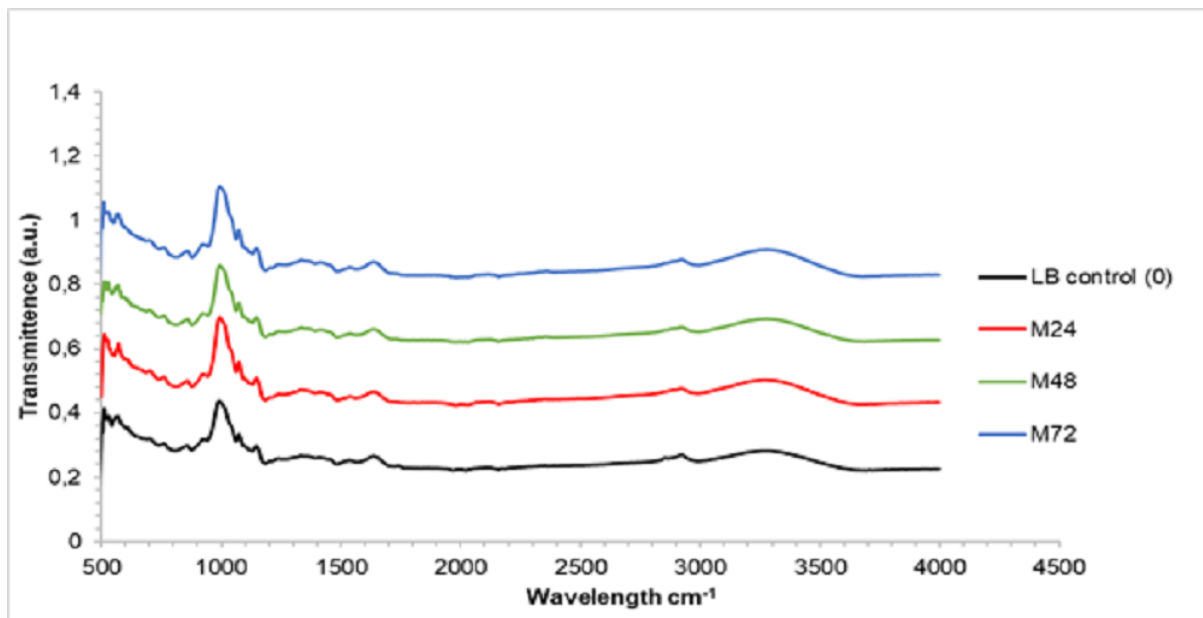


Figure 4. 2: FTIR of malted FM flours. FM = finger millet, LB control (0) = light brown, DB control (0) = dark brown, M= malted (24,48,72 h).

The more pronounced intensities of amide I peak in both malted FM flours might be due to high protein content (Table 4.2a) resulting from the malting of FM flours (Olamiti et al., 2020). Additionally, this study observed many bands in the fingerprint area ($800 - 1600 \text{ cm}^{-1}$). Fingerprint region refers to a specific range of infrared wavelengths where complex molecular vibrations occur. FTIR analysis provided important information on the composition of the malted and control FM flours which could be used to analyse quality and nutritional value (Kaur & Prasad, 2022). The peaks shown on the FTIR graph corresponded to various functional groups present in both malted FM flours, and the variations in these peaks provided insights into changes in the flour composition. Adebisi et al. (2016) observed similar changes in malted pearl millet flour.

4.6. Scanning electron morphology of malted light and dark brown finger millet flours

Figure 4.3 illustrates the scanning electron microscopy of malted and control FM flours. The control LB (light brown) FM flour had a tightly packed arrangement of protein bodies (PB), but malting caused the breakdown and disintegration of this structure. Both the control LB and DB (dark brown) flours displayed a variety of starch granules (SG) in terms of size, with some being small and others large. Furthermore, the SG was linked together within the PB. On the other hand, the malted FM flours had smaller PB and a greater number of liberated SG than the control FM flours. This could be caused by the fact that the process of malting dismantled large compounds of starch into simpler molecules, resulting in the liberation of SG. The liberated SG in both malted FM flours had variations in size and form, ranging from small to large, featuring polygonal, spherical, and oval shapes.

Moreover, scanning electron microscopy revealed that FM malted flours had more holes between the SG than control FM flours. In linkage to this study, Li et al. (2017) also observed pits and holes in germinated starches. Further liberation of SG was observed at 72 h in both malted FM flours, which might have been due to the breakdown of the cell wall. Faltermaier et al. (2015) and Tian et al. (2018) indicated that cell wall breakdown influenced the SG. There were differences in the thermal properties, WAC, solubility, and swelling power. This might be because the starch composition changed, making the flours less dense (Khoza et al., 2021).

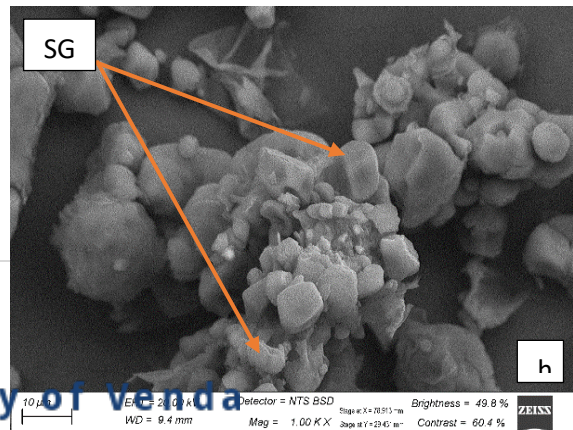
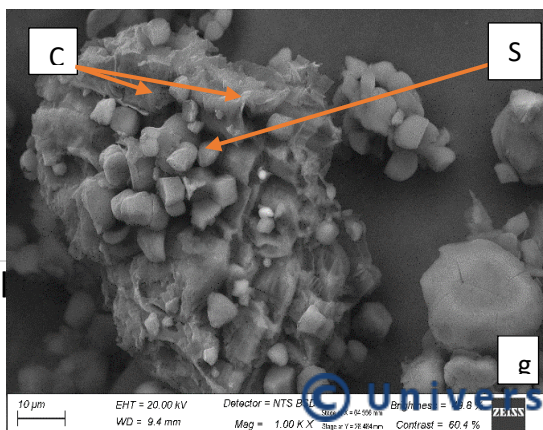
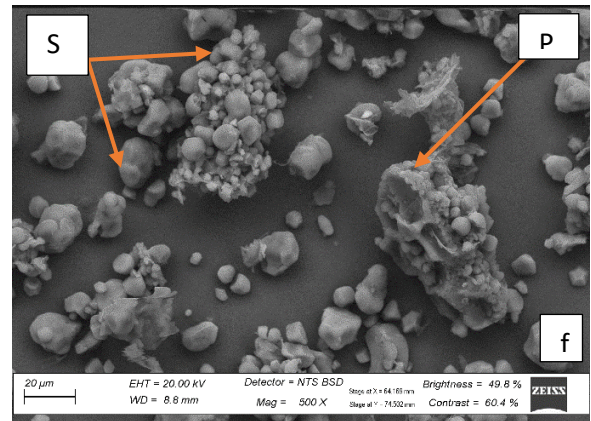
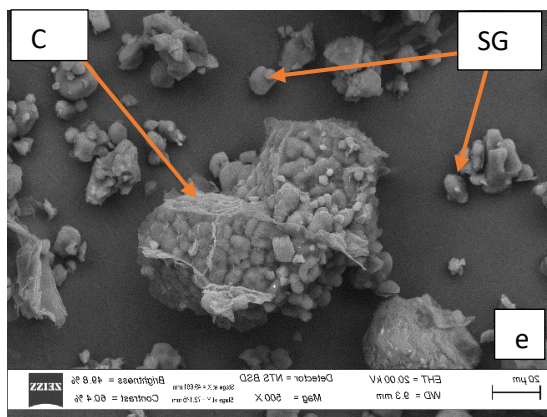
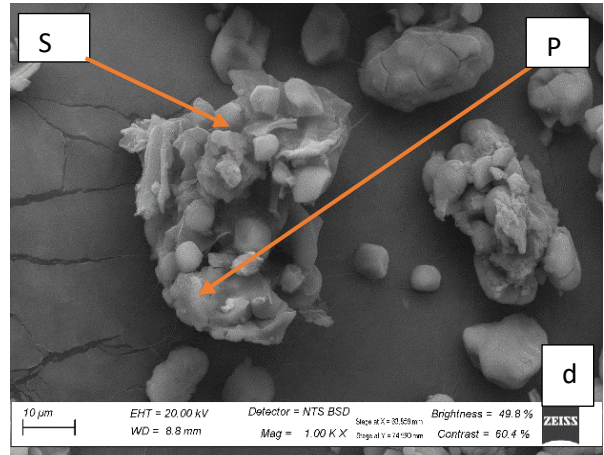
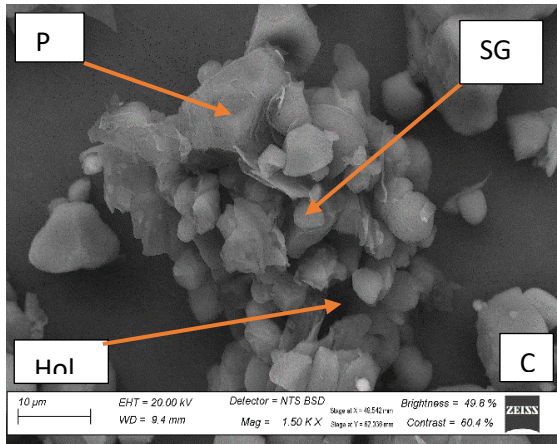
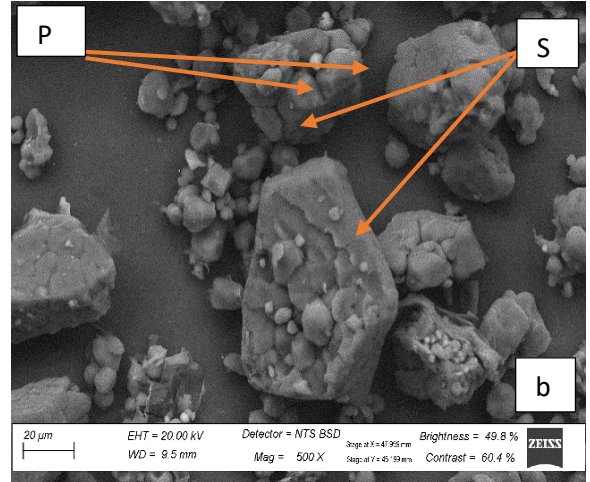
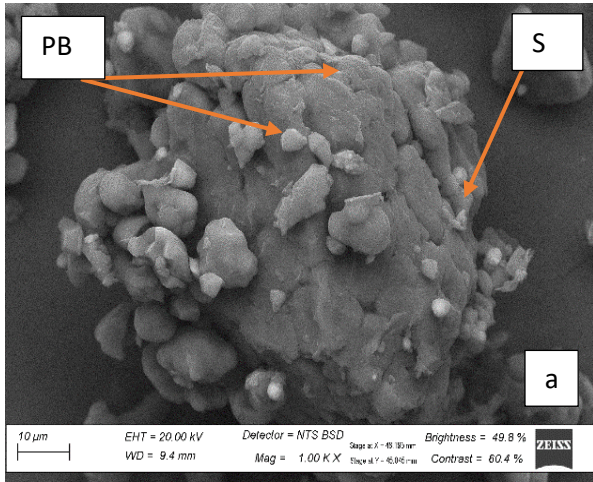


Figure 4. 3: Scanning electron morphology of malted LB and DB finger millet (FM) flours, (a) unmalted LB flours, (b) unmalted DB flours, (c) 24 h malted LB flours, (d) 24 h malted DB flours, (e) 48 h malted LB flours, (f) 48 h malted DB flours, (g) 72 h malted LB flours, (h) 72 h malted DB flours, FM = finger millet; DB = dark brown, LB = light brown; PB = protein bodies, CW= cell walls, SG = starch granules.

4.8. Mineral content of biscuits

Finger millet (FM) is an excellent mineral-rich food with substantial amounts of calcium (Ca), iron (Fe), phosphorus (P), potassium (K), magnesium (Mg), manganese (Mn), and zinc (Zn) (Gowda et al., 2022). The mineral compositions of the control and malted FM biscuits are presented in Table 4.6. In both FM biscuits, the mineral content increased with an increase in malting time. Biscuits produced from the flours malted for 72 h significantly ($p < 0.05$) displayed higher levels of both major and minor minerals in contrast to the mineral content of control FM biscuit samples.

Krishnan et al. (2012) and Adebisi et al. (2017) stated that malting improves the accessibility of minerals by promoting their synthesis and facilitating the solubilisation of cell walls. Hence, the increase of minerals like Ca, Mg, P, Fe, manganese (Mn), and Zn was most likely due to the malting process. Such observations may also be attributed to Maillard reactions that occurred during the baking process. Delgado-Andrade et al. (2011) stated that the Maillard reaction might lead to the generation of complexes or the breakdown of compounds, possibly influencing the accessibility and solubility of minerals. The Maillard reaction during the baking process might potentially lead to the liberation of bound mineral elements, as a result, the biscuit samples exhibited an improved mineral content (Adebisi et al., 2017). Chinma et al. (2022) observed an increase in Ca, Mg, Fe, P, and Zn in biscuits made from germinated FM and Bambara groundnut flour. Although germination may break down antinutrients, the baking process may also impact on the mineral content, these may have a negative effect depending on the food.

Table 4. 6: Impact of malting on macro-and microminerals of finger millet biscuits (mg/100 g dry basis).

| Parameter | Light brown finger millet biscuit | | | | Dark brown finger millet biscuit | | | |
|-----------------|-----------------------------------|----------------------------|----------------------------|----------------------------|----------------------------------|----------------------------|----------------------------|----------------------------|
| | Control | M 24 h | M 48 h | M 72 h | Control | M 24 h | M 48 h | M 72 h |
| Minerals | | | | | | | | |
| Ca | 347.33 ± 1.21 ^a | 372.15 ± 1.48 ^b | 398.34 ± 0.58 ^c | 445.22 ± 1.36 ^d | 329.19 ± 1.18 ^a | 363.51 ± 1.23 ^b | 392.32 ± 1.35 ^c | 438.17 ± 1.15 ^d |
| Mg | 131.21 ± 1.66 ^a | 144.36 ± 1.24 ^b | 152.18 ± 1.06 ^c | 166.24 ± 0.44 ^d | 128.13 ± 1.29 ^a | 140.72 ± 0.97 ^b | 153.44 ± 1.42 ^c | 164.57 ± 1.17 ^d |
| P | 8.91 ± 1.12 ^a | 10.12 ± 0.94 ^b | 12.40 ± 1.09 ^c | 15.33 ± 1.18 ^d | 8.64 ± 1.10 ^a | 9.89 ± 0.73 ^b | 12.11 ± 1.09 ^c | 14.36 ± 1.14 ^d |
| Fe | 9.62 ± 0.18 ^a | 10.78 ± 0.34 ^b | 11.85 ± 0.16 ^c | 13.11 ± 0.22 ^d | 9.70 ± 0.12 ^a | 10.53 ± 0.52 ^b | 11.76 ± 0.28 ^c | 12.98 ± 0.60 ^d |
| Mn | 3.58 ± 0.72 ^a | 4.12 ± 0.83 ^b | 5.29 ± 0.79 ^c | 6.34 ± 0.91 ^d | 3.44 ± 0.63 ^a | 4.06 ± 0.59 ^b | 5.14 ± 0.61 ^c | 6.03 ± 0.85 ^d |
| Zn | 3.27 ± 0.76 ^a | 5.54 ± 0.80 ^b | 7.31 ± 0.88 ^c | 8.29 ± 0.93 ^d | 3.24 ± 0.73 ^a | 4.87 ± 0.55 ^b | 5.73 ± 0.71 ^c | 7.95 ± 0.77 ^d |

Values are illustrated by average ± standard deviation, n = 3. Different characters in the same column are notably different. M = malted (0, 24, 48, 72 hours).

Ca = Calcium, Mg = Magnesium, P = Phosphorus, Fe = Iron, Mn = Manganese, Zn = Zinc.

The increased mineral content of malted FM biscuits can be viewed as an approach to tackling mineral deficiencies in children and adults. Minerals have essential roles in the human body, which include the regulation of the immune system and heartbeat, the synthesis of bone tissue and hormones, as well as the transmission of nerve impulses (Gharibzahedi & Jafari, 2017). Insufficient consumption of minerals such as Ca, Fe, and Zn has negative impacts on human health. Calcium deficiency is linked to conditions like osteoporosis and rickets, particularly predominantly in Africa and Asia. In addition, inadequate consumption of Fe can result in anaemia in individuals (Al-Naseem et al., 2021). The improvement of mineral content through the process of malting holds great promise for positively impacting the health and well-being of human beings.

4.9. Physical characteristics of biscuits

Table 4.7 displays the physical characteristics of the control and malted LB and DB finger millet biscuits. The weight of the malted FM biscuits varied from 12.83 to 13.23 g (LB) and 12.25 to 12.98 g (DB). Both the control FM biscuits had significantly ($p < 0.05$) higher weights compared to the malted FM biscuits. Wang et al. (2023) indicated that high dietary fibre in biscuits, as noted in Table 4.2b, specifically β -glucan, leads to improved moisture retention after baking, thus resulting in high weight. Mudau et al. (2022b) observed that an elevated amount of dietary fibre results in an increased weight of the biscuits. Agrahar-Murugkar et al. (2015) recorded low weight in malted FM biscuits.

Diameter ranged from 4.51 to 4.67 cm (LB) and 4.21 to 4.50 cm (DB), and thickness ranged from 0.67 to 0.72 cm (LB) to 0.67 to 0.74 cm (DB). The decrease in diameter and thickness in both malted FM biscuits could be attributed to the high TDF values. Dietary fibre has water-absorbing properties, and it absorbs moisture during baking (Kobets et al., 2016). The high TDF in both malted FM biscuits resulted in the dough becoming denser and less spreadable and expandable, leading to a low diameter and thickness of the FM biscuits. The alterations in diameter and thickness were reflected in the spread ratio of both FM biscuits.

The spread ratio of FM biscuits ranged from 6.21 to 7.23 (LB) and 6.09 to 6.95 (DB). A significant ($p < 0.05$) decrease in spread ratio with an increase in malting time was observed. The decrease in spread ratio noted in both malted FM biscuits could be attributed to the higher presence of water-absorbing components like protein and fibre (Agrahar-Murugkar et al., 2015; Di Cairano et al., 2021). In addition, the low spread ratio of malted FM biscuits might be attributed to the enzymatic degradation of the protein and starch into smaller sugars and peptides, which resulted in the high hydrophilic nature of the biscuits (Sharma et al., 2016). Adebiji et al. (2017) and Chinma et al. (2022) observed a low spread ratio on malted pearl millet and Bambara groundnut FM biscuits.

Table 4. 7: Physical characteristics of malted finger millet biscuits.

| Malting (h) | Weight (g) | Diameter (cm) | Thickness (cm) | Spread ratio | Hardness (g) |
|--------------------------|---------------------------|---------------------------|---------------------------|--------------------------|-----------------------------|
| LB finger millet biscuit | | | | | |
| Control | 13.23 ± 0.66 ^c | 4.67 ± 0.49 ^c | 0.72 ± 0.07 ^{ab} | 7.23 ± 0.56 ^c | 762.53 ± 2.12 ^a |
| M24 | 13.13 ± 0.94 ^b | 4.51 ± 0.29 ^a | 0.71 ± 0.04 ^{ab} | 7.21 ± 0.50 ^c | 1065.61 ± 3.25 ^b |
| M48 | 12.87 ± 0.35 ^a | 4.60 ± 0.46 ^b | 0.67 ± 0.05 ^a | 6.64 ± 0.58 ^b | 1334.52 ± 1.78 ^c |
| M72 | 12.83 ± 0.94 ^a | 4.58 ± 0.56 ^{ab} | 0.67 ± 0.04 ^a | 6.21 ± 0.93 ^a | 1720.28 ± 3.56 ^d |
| DB finger millet biscuit | | | | | |
| Control | 12.98 ± 0.85 ^c | 4.50 ± 0.43 ^c | 0.72 ± 0.05 ^{ab} | 6.95 ± 0.90 ^d | 747.50 ± 2.38 ^a |
| M24 | 12.43 ± 0.13 ^b | 4.40 ± 0.26 ^b | 0.74 ± 0.07 ^{ab} | 6.44 ± 0.84 ^c | 1048.10 ± 3.43 ^b |
| M48 | 12.24 ± 0.21 ^a | 4.47 ± 0.28 ^{bc} | 0.67 ± 0.06 ^a | 6.32 ± 0.82 ^b | 1502.29 ± 3.32 ^c |
| M72 | 12.25 ± 0.29 ^a | 4.21 ± 0.03 ^a | 0.72 ± 0.04 ^{ab} | 6.09 ± 0.29 ^a | 1918.71 ± 3.70 ^d |

Values are illustrated by average ± standard deviation, n = 3. Different characters in the same column are notably different ($p < 0.05$). M = malted (0, 24, 48, 72 hours), LB = Light brown, DB = Dark brown.

The hardness of FM biscuits ranged from 762.53 to 1720.28 g (LB) and 747.50 to 1918.71 g (DB). A significant ($p < 0.05$) increase in hardness in both FM biscuit samples was observed with the increase in malting. The high hardness values of both malted biscuits could be due to starch conversion and protein modification during malting, which resulted in denser, firmer, and harder biscuits (Sahin et al., 2019). Moreover, the presence of additional free sugars that became available during the germination process might have also contributed to the high hardness. Zucco et al. (2011) state that the sugar present in flour undergoes crystallisation when cooling, resulting in a hardening effect on the biscuit. Nevertheless, the hardness of the biscuits was influenced by the amount of moisture present, the protein content, and the extent of gelatinisation of the starch (Di Cairano et al., 2021). Agrahar-Murugkar et al. (2015) stated that biscuits made from sprouted and malted composite flours were harder. Adebiji et al. (2017) observed an increase in the hardness of malted pearl millet biscuits.

4.11. Sensory evaluation of biscuits

Table 4.8 shows the sensory attributes of the LB and DB malted FM biscuits. The panellists did not perceive a significant difference ($p < 0.05$) in the appearance of both malted FM biscuits. The average colour scores at 48 and 72 h of malting in both FM biscuits were significantly lower than those of the control biscuits. The colour of the DB biscuits malted for 48 and 72 h was not preferred by the panellists. The observed darker colour of the DB biscuits (Figure 4.1) may have created the impression among the panellists that they were overbaked, which potentially affected how they perceived them. The aroma of a product plays a vital role in determining its acceptance or rejection, thus making it the most crucial factor in the decision-making process (Vivek et al., 2020). The panellists preferred the aroma of both the control and 24 h malted biscuits than the 48 and 72 h malted FM biscuits. Regarding taste, the control FM biscuits received higher scores compared to the malted FM biscuits. Additionally, the light brown FM biscuits received better scores in all sensory attributes than the dark brown FM biscuits.

At 72 h of malting, low average scores on taste were observed. The prolonged malting process could be a result of the excessive enzymatic activity, leading to a stronger and less desirable flavour profile (Filipowska et al., 2021). Regarding texture, the panellists indicated that the LB biscuits had a more appealing mouthfeel than the DB biscuits. The extended malting duration could have caused changes in texture, resulting in a less appealing mouthfeel. The panellists also described the texture of the FM biscuits to be overall hard and less preferable. Adebisi et al. (2017) and Adebisi et al. (2016) observed high texture values in malted pearl millet biscuits. Overall, the LB biscuits were more acceptable than the DB biscuits.

Table 4. 8: Sensory liking scores of malted finger millet biscuits.

| Malting (h) | Appearance | Colour | Aroma | Taste | Texture | Overall acceptability |
|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| LB finger millet biscuit | | | | | | |
| Control | 7.88 ± 1.46 ^a | 7.92 ± 1.22 ^b | 7.26 ± 1.62 ^a | 6.17 ± 2.62 ^a | 6.12 ± 2.45 ^a | 6.61 ± 1.90 ^a |
| M24 | 7.72 ± 1.32 ^a | 7.82 ± 1.44 ^b | 7.20 ± 1.58 ^a | 5.94 ± 1.93 ^a | 5.88 ± 1.74 ^a | 5.40 ± 1.43 ^b |

| | | | | | | |
|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| M48 | 7.64 ± 1.58 ^a | 7.50 ± 1.48 ^a | 7.01 ± 1.70 ^a | 5.72 ± 1.69 ^a | 5.75 ± 1.80 ^a | 5.10 ± 1.52 ^b |
| M72 | 7.61 ± 1.64 ^a | 6.92 ± 1.53 ^a | 6.92 ± 1.53 ^a | 5.62 ± 1.77 ^a | 5.64 ± 1.67 ^a | 4.92 ± 2.02 ^b |
| DB finger millet biscuit | | | | | | |
| Control | 6.94 ± 1.48 ^a | 6.92 ± 1.53 ^a | 6.80 ± 2.20 ^a | 5.40 ± 1.41 ^a | 5.26 ± 1.46 ^a | 5.88 ± 1.12 ^a |
| M24 | 6.86 ± 1.33 ^a | 6.81 ± 1.47 ^a | 6.72 ± 1.38 ^a | 4.96 ± 1.75 ^a | 4.95 ± 1.66 ^a | 5.56 ± 1.49 ^b |
| M48 | 6.82 ± 1.41 ^a | 6.76 ± 1.35 ^a | 6.63 ± 1.15 ^b | 4.55 ± 1.57 ^b | 4.76 ± 1.88 ^a | 5.22 ± 1.24 ^c |
| M72 | 6.77 ± 1.20 ^a | 6.72 ± 1.41 ^a | 6.42 ± 1.49 ^b | 4.48 ± 1.22 ^b | 4.62 ± 1.68 ^b | 4.95 ± 1.78 ^c |

Values are expressed by mean ± standard deviation, n = 3. Different superscripts in the same column are significantly different (p < 0.05). M = malted (0, 24, 48, 72 hours). LB = Light brown, DB = Dark brown.

CHAPTER 5: GENERAL CONCLUSION AND RECOMMENDATION

Variations in nutritional composition in finger millet (FM) flours were observed throughout a 72-h malting. Nutritional composition dictates the physicochemical characteristics as well as food applications. Malting led to a higher mineral content, functional and thermal properties of finger millet flours. Both malted finger millet flours exhibit improved thermal stability, making them suitable for various processing and cooking techniques. Malting also modified the microstructural characteristics and functional groups of finger millet flours. Taking into consideration that malting improves the antioxidant properties of finger millet extracts, these malted finger millet flours may be used as functional ingredients in preparing healthy grain-based products such as weaning foods and gluten-free bakery products for people suffering from celiac disease. This study demonstrates that malting significantly ($p < 0.05$) improved the nutritional value of both LB = (Light brown) and DB (Dark brown) FM biscuits. The malting process enhanced antioxidants, fibre, protein, and energy content, thus making these biscuits a healthier choice. Malting increased the hardness of both FM biscuits. L^* values decreased with an increase in malting in both FM biscuits. The panellists preferred the 24 h malted FM biscuits compared to the other malted biscuit samples. LB biscuits had more desirable and favourable attributes compared to DB biscuits. Malted FM biscuits offer consumers a convenient and tasty snack that contributes to a balanced diet.

These findings highlight the potential of malting to create functional food products with enhanced nutrition, promoting overall health and well-being. More studies about the influence of malting on the antinutritional factors, and amino acids of malted FM biscuits should be done. Performing microstructural analyses using X-ray diffraction on malted FM biscuits can provide more insightful information. The food industry should explore the commercialisation and utilisation of malted FM but also contribute to the economic well-being of local rural farmers in Africa to produce gluten-free biscuits or alternative food products derived from malted FM flours.

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APPENDICES

APPENDIX A: ARTICLE PUBLISHED

ARTICLE PUBLISHED

Murungweni, K. T., Ramashia, S. E., & Mashau, M. E. (2023). Effect of malting on physicochemical, antioxidant, and microstructural properties of finger millet (*Eleusine coracana*) flours. *Food Science & Nutrition*, 00,1–17. <https://doi.org/10.1002/fns3.3790> v

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DOI: 10.1002/fns3.3790

ORIGINAL ARTICLE

Food Science & Nutrition WILEY

Effect of malting on physicochemical, antioxidant, and microstructural properties of finger millet (*Eleusine coracana*) flours

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Funding Information
Agricultural Research Council, Grant/Award Number: E601

Abstract

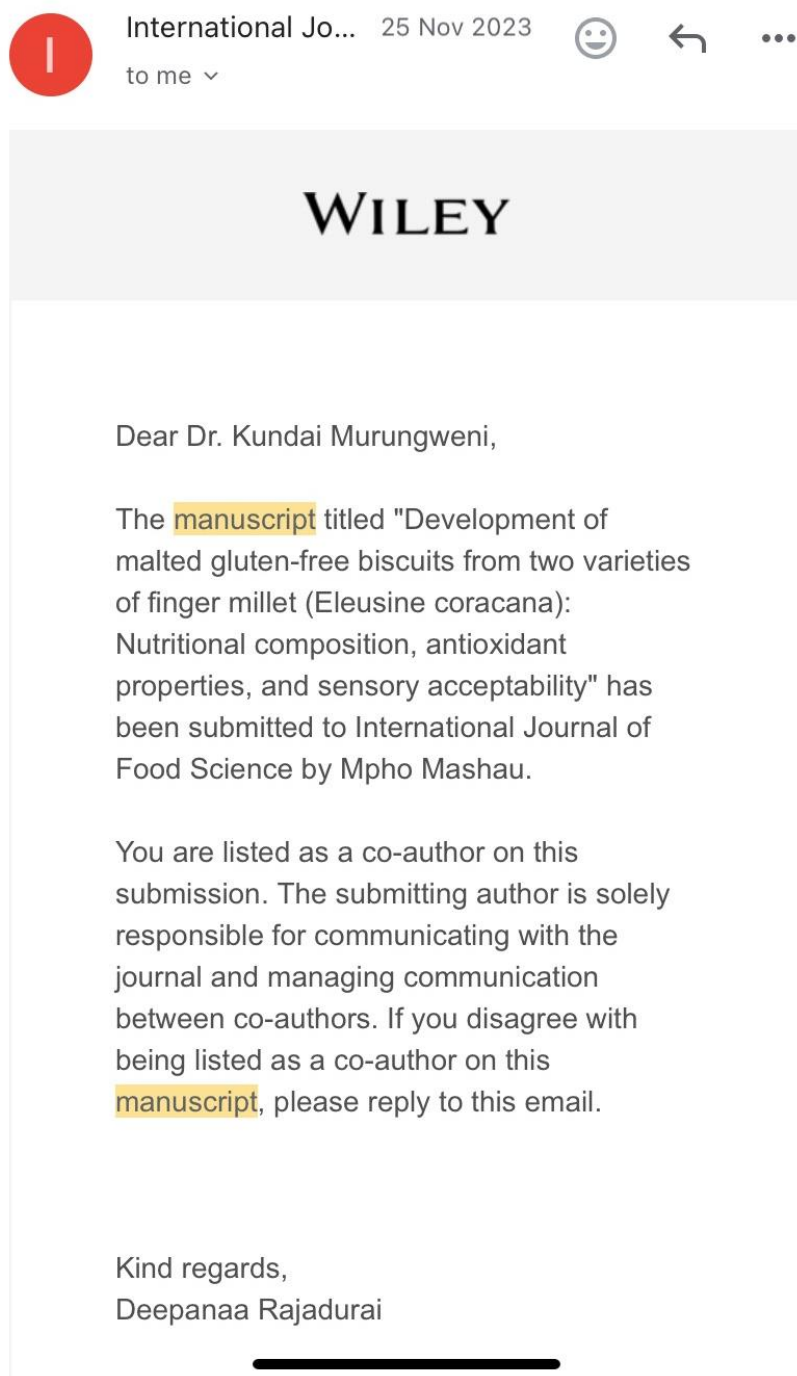
Finger millet (*Eleusine coracana* L. Gaertn.) is a gluten-free crop with a high amount of fiber, calcium and iron, outstanding malting qualities and a low glycemic index. The study aimed to determine the physicochemical, functional, antioxidant and microstructural properties of malted finger millet (light and dark brown) flours. The two varieties of finger millet grains were germinated for 0, 24, 48 and 72 h and kilned for 8 h. The lightness (L*) values of malted finger millet flours significantly increased, with light brown having the highest L* value of 76.62. The hue angle and total color differences (ΔE) of the malted finger millet flours increased significantly ($p \leq .05$), and values ranged from 63.43° to 71.20° (light brown) and 2.12° to 4.32° (dark brown), respectively. The moisture, ash, fiber, protein, total phenolic, total flavonoids contents and DPPH activity of both malted finger millet flours significantly increased. On the contrary, the fat, carbohydrate, energy contents and FRAP activity significantly decreased with each malting period of both finger millet flours. Both malted finger millet flours' solubility index, water and oil absorption capacity increased significantly while the packed and loose bulk density decreased. Malting had no significant effect on the viscosity of the cold paste; however, a significant decrease in the viscosity of the cooked paste in both finger millet flours was observed, with values ranging from 285 to 424.00 cP (light brown) and 271.33 to 418.00 cP (dark brown), respectively. Malting resulted in changes in the thermal properties of finger millet flours with an increase in the onset, peak and conclusion temperatures. Fourier-Transform Infrared Spectra showed that malting slightly changed the peaks of both finger millet flours. Scanning electron microscopy showed that malting altered the microstructural characteristics of finger millet flours. The results showed that malted finger millet flours are promising raw materials for gluten-free bakery products.

KEYWORDS

functional, germination, Millet, polyphenolic compounds, thermal and morphological characteristics

APPENDIX B: MANUSCRIPT UNDER REVIEW

Murungweni, K.T., Ramashia, S.E., & Mashau, M.E. (2023). Development of malted gluten-free biscuits from two varieties of finger millet (*Eleusine coracana*): Nutritional composition, antioxidant properties and sensory acceptability. International Journal of Food Science.



APPENDIX C: ETHICAL CLEARANCE

ETHICS APPROVAL CERTIFICATE

RESEARCH AND INNOVATION
OFFICE OF THE DIRECTOR

NAME OF RESEARCHER/INVESTIGATOR:
Ms KT Murungweni

STUDENT NO:
17001077

PROJECT TITLE: Physicochemical, antioxidant and microstructural properties of malted finger millet (*Eleusine coracana*) flour(s) and their utilisation in gluten-free biscuits.

ETHICAL CLEARANCE NO: FSEA/22/FST/06/0902

SUPERVISORS/ CO-RESEARCHERS/ CO-INVESTIGATORS

| NAME | INSTITUTION & DEPARTMENT | ROLE |
|------------------|-------------------------------------|------------------------|
| Dr SE Ramashia | UNIVEN, Food Science and Technology | Supervisor |
| Mr M Mashau | UNIVEN, Food Science and Technology | Co - Supervisor |
| Ms KT Murungweni | UNIVEN, Food Science and Technology | Investigator – Student |

Type: **Masters Research**

Risk: **Minimal risk to humans, animals, or environment (Category 2)**

Approval Period: **February 2023 - February 2024**

The Human and Clinical Trials Research Ethics Committee (HCTREC) hereby approves your project as indicated above.

General Conditions

While this ethics approval is subject to all declarations, undertakings and agreements incorporated and signed in the application form, please note the following:

- The project leader (principal investigator) must report in the prescribed format to the REC:
 - Annually (or as otherwise requested) on the progress of the project, and upon completion of the project.
 - Within 48hrs in case of any adverse event (or any matter that interrupts sound ethical principles) during the course of the project.
 - Annually a number of projects may be randomly selected for an external audit.
- The approval applies strictly to the protocol as stipulated in the application form. Would any changes to the protocol be deemed necessary during the course of the project, the project leader must apply for approval of these changes at the REC. Would there be deviation from the project protocol without the necessary approval of such changes, the ethics approval is immediately and automatically forfeited.
- The date of approval indicates the first date that the project may be started. Would the project have to continue after the expiry date; a new application must be made to the REC and new approval received before or on the expiry date.
- In the interest of ethical responsibility, the REC retains the right to:
 - Request access to any information or data at any time during the course or after completion of the project,
 - To ask further questions; Seek additional information; Require further modification or monitor the conduct of your research or the informed consent process.
 - withdraw or postpone approval if:
 - Any unethical principles or practices of the project are revealed or suspected,
 - It becomes apparent that any relevant information was withheld from the REC or that information has been false or misrepresented.
 - The required annual report and reporting of adverse events was not done timely and accurately,
 - New institutional rules, national legislation or international conventions deem it necessary.

ISSUED BY:

UNIVERSITY OF VENDA, RESEARCH ETHICS COMMITTEE

Date Considered: February 2023

Name of the HCTREC Chairperson of the Committee: Prof MS Maputle

Signature

Ms Maputle



APPENDIX D: PROOFREADING REPORTS

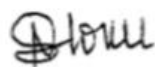
Editing and Proofreading Report

26 June 2023

This letter serves to confirm that I, Dr I. Ndlovu of the Department of English, Media Studies and Linguistics at the University of Venda, have proofread and edited an article titled “Insights into the malting effect on the physicochemical, antioxidant and microstructural properties of finger millet (*Eleusine coracana* L) flours” by Kundai Thelma Murungweni, Shonisani Eugenia Ramashia and Mpho Edward Mashau.

I carefully read through the document, focusing on proofreading and editorial issues. The recommended suggestions are clearly highlighted and can either be accepted or rejected using the Microsoft Track Changes Function.

Yours Sincerely



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Editing and Proofreading Report

15 August 2023

This letter serves to confirm that I, Prof. I. Ndlovu of the Department of English, Media Studies and Linguistics at the University of Venda, have proofread and edited an article titled “Development of malted gluten-free biscuits from two varieties of finger millet (*Eleusine coracana*): Nutritional composition, antioxidant properties, and sensory acceptability” by Kundai Thelma Murungweni, Mpho Edward Mashau and Shonisani Eugenia Ramashia.

I carefully read through the document, focusing on proofreading and editorial issues. The recommended suggestions are clearly highlighted and can either be accepted or rejected using the Microsoft Track Changes Function.

Yours Sincerely



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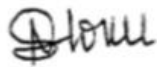
Editing and Proofreading Report

19 November 2023

This letter serves to confirm that I, Prof. I. Ndlovu of the Department of English, Media Studies and Linguistics at the University of Venda, have proofread and edited a research chapter by Thelma Murungweni.

I carefully read through the document, focusing on proofreading and editorial issues. The recommended suggestions are clearly highlighted and can either be accepted or rejected using the Microsoft Track Changes Function.

Yours Sincerely



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