BINDURA UNIVERSITY OF SCIENCE EDUCATION
FACULTY OF AGRICULTURE AND ENVIRONMENTAL SCIENCE
DEPARTMENT OF CROP SCIENCE

ASSESSMENT OF FUNGAL LOAD AND MOISTURE CONTENT OF MAIZE IMPORTED FROM SOUTH AFRICA INTO ZIMBABWE

BY

MARUSETA NELSON
B231516B

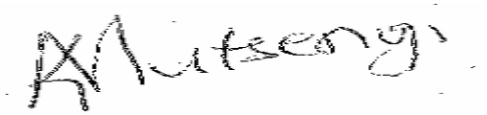
A dissertation submitted in partial fulfilment of the requirements for the degree of Bachelor of Agricultural Science Honours Degree in Crop Science

DEPARTMENT OF CROP SCIENCE
JUNE 2026

DECLARATION

I, Nelson Maruseta, hereby declare that this research project is from my own work and has not been submitted to any University or other Tertiary Institution for the award of a degree.

Signed:  **Date:** 26/06/2026

Signed: …  **Date:** ……28/08/2026

Supervisor: Mr Mutsengi

APPROVAL FORM

The undersigned certify that they have read and recommend to Bindura University of Science Education for acceptance, a dissertation entitled "Assessment of Microbes and Physicochemical Characterization of Maize Imported from South Africa into Zimbabwe," submitted by Maruseta Nelson **(B231516B)** in partial fulfilment of the requirements for the Bachelor of Agricultural Science Honours Degree in Crop Science.

DEDICATION

I dedicate this project to my dearest family and friends, whose massive encouragement, patience, and unwavering belief in me made it possible for me to complete this work successfully. Their constant support of mutual understanding during difficult moments, and motivation throughout this journey gave me the strength and confidence to persevere. I am sincerely grateful for their love, guidance and sacrifices, which have been a foundation for my achievements.

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ABSTRACT

Maize is one of the major staple foods consumed in Zimbabwe; however, fungal contamination of imported maize poses significant food safety and public health risks. This study aimed to identify fungi associated with imported maize from South Africa, determine yeast and mould counts, and assess the relationship between moisture content and fungal contamination.

A cross-sectional laboratory-based study was conducted using imported dried maize samples collected from selected storage and distribution points in Zimbabwe between January and March. Standard microbiological methods were used for fungal isolation and identification. Potato Dextrose Agar was used for culturing fungi, while fungal identification was based on macroscopic and microscopic characteristics. Yeast and mould counts were determined using serial dilution and plate count methods. Physicochemical analysis focused mainly on moisture content determination using the oven drying method. Data were analysed using descriptive statistics and inferential statistical tests. Completely randomized observational design with sub-sampling was implemented.

The study identified four major fungal genera associated with imported maize, namely *Aspergillus spp., Candida spp., Fusarium spp., and Penicillium spp*. The results further showed that there was no statistically significant difference ($p<0.05$) in fungal occurrence and counts among maize imported during January, February, and March. In addition, a slight positive relationship was observed between moisture content and fungal counts, suggesting that increased moisture may promote fungal proliferation.

The study concludes that imported maize acts as a source of fungal contamination and potential mycotoxin exposure in Zimbabwe. The findings highlight the importance of routine microbiological surveillance, moisture control, and improved storage conditions to ensure the safety and quality of imported maize.

Keywords: maize, fungal contamination, Aspergillus spp., moisture content, imported maize, Zimbabwe, yeast and mould count

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LIST OF ACRONYMS

Acronym	Meaning
ANOVA	Analysis of Variance
CFU/g	Colony Forming Units per gram
GM	Genetically Modified
GMB	Grain Marketing Board
GIS	Geographic Information System
IPPC	International Plant Protection Convention
MLND	Maize Lethal Necrosis Disease
NA	Nutrient Agar
NGS	Next Generation Sequencing
PCR	Polymerase Chain Reaction
PDA	Potato Dextrose Agar
PHL	Post-Harvest Losses
PRDC	Plumtree Rural District Council
RSA	Republic of South Africa
RT-PCR	Real Time Polymerase Chain Reaction
SPSS	Statistical Package for the Social Sciences

CHAPTER 1: INTRODUCTION

1.0 Introduction

Maize (*Zea mays* L.) constitutes a large part of the daily diet in both rural and urban populations of Zimbabwe. Out of the three major food crops (maize, wheat and sorghum) grown in Zimbabwe, maize is by far the largest cultivated crop, on average by 70–80% of the total population (Odunze & Uwizeyimana, 2019). However, maize from local farmers is not sufficient to feed the people and livestock in Zimbabwe, so stock feed companies strongly rely on grain imports from South Africa and Zambia. In Zimbabwe, maize is controlled by the government, which means that the government controls its purchase and sale, and the terms of such regulations are defined by the Grain Marketing Board (GMB) Act. In terms of the GMB Act (section 33 (1)), "no person shall sell or otherwise dispose of any controlled product within the prescribed area except to the GMB". White maize, which is preferred by the people, accounts for most of the maize produced and consumed by Zimbabwean citizens, while yellow maize, although grown in significant quantities, is primarily used as livestock feed (Odunze & Uwizeyimana, 2019).

Food supply and price stability crucially depend on global trade, because the import of agricultural products is necessary for countries to import goods during local failures and shortages of crops to avoid the occurrence of food scarcity and food price volatility (Matsikidze, 2025). The system of trade stimulates economic growth by offering opportunities for formation in the supply chain for farmers, traders and industries. It improves economies of global trade as it makes available a variety of fruits and vegetables which enhance the diets in the locality and provide it with necessary vitamins and minerals. Technological advancement in agriculture is enabled through the exchange of agricultural technologies that enhance productivity, sustainability and efficiency in agriculture (Ayala-Zavala *et al*., 2014).

Africa's global grain trade is vital but it has its own challenges in that it can introduce new pests and diseases. These risks have devastating effects on agriculture and food supply, including the possibility of bringing in foreign microbes, pests and diseases. The microorganisms are not visible to the naked eye, but PCR can detect pathogens' genetic material in small quantities. A study by Victor *et al*., (2013) highlighted that imported grains are prone to contamination during

the transportation, storage and handling processes. These pests and diseases are typically introduced through contaminated packaging materials, shared transport vehicles, inadequate phytosanitary inspection at ports of entry, and the co-mingling of consignments from multiple sources during bulk handling and storage.

1.1 Objectives

Main Objective

To find out the phytosanitary hazards associated with imported maize from the Republic of South Africa (RSA).

Specific Objectives

1. To identify types of fungi introduced through maize importation in Zimbabwe.
2. To compare the total number of fungal moulds introduced in Zimbabwe through maize imports from South Africa across three different months.
3. To characterise the physicochemical properties of maize imported from RSA and find the relationship between moisture content and fungal contamination.

1.2 Hypothesis

- H0: Maize imported into Zimbabwe is not primarily contaminated by Aspergillus, Fusarium, and Penicillium species.
- H0: Maize imported from South Africa has a total viable yeast and mould count $\leq 10^4$ CFU/g.
- H0: Maize imported from South Africa has a total viable yeast and mould count $\leq 10^4$ CFU/g.

1.5 Statement of the Problem

There is little documented evidence of the particular types and numbers of microorganisms introduced into Zimbabwe via imported maize. Fungal contaminants such as *Aspergillus* and *Fusarium* species are particularly problematic because they create mycotoxins that jeopardize food safety and result in post-harvest losses. Similarly, high yeast and mould burdens affect grain quality and shelf life, although the entire microbial burden of Republic of South Africa (RSA) maize consignments remains unknown.

In addition, physicochemical properties such as moisture content, protein, and aflatoxin levels directly influence microbial growth and grain marketability. If imported maize fails to meet national quality standards, it may increase spoilage, economic losses, and health risks for consumers. Despite these concerns, there is no comprehensive study characterizing both the microbial and physicochemical profile of maize imported from South Africa into Zimbabwe.

Reported hazards associated with such contamination include aflatoxin-related hepatotoxicity and carcinogenicity in humans, immune suppression, and growth stunting in children, as well as reduced weight gain, organ damage, and increased mortality in livestock fed contaminated grain.

1.6 Justification

Imported grains like maize are prone to introducing new crop diseases. Early identification of such microbes allows the Plant Quarantine Services to update phytosanitary protocols and prevent outbreaks that would reduce future harvests (Orcid, 2021). No study using laboratory and molecular techniques has been carried out to authenticate the visual inspection results at the port of entry. Hence, the study seeks to check the accuracy of visual inspections and advise stakeholders. This study will aid in preventing the introduction of new field pathogens and crop diseases. Moreover, the results of this study will enhance millers and stock feed companies' ability to reduce post-harvest losses and contract disputes. Furthermore, risks to both animal and human health and safety will be reduced due to early detection of mycotoxins.

CHAPTER 2: LITERATURE REVIEW

2.0 Literature Review

2.1 Drivers of Low Maize Production in Zimbabwe

In Zimbabwe, over 70 percent of the population depends on agriculture for their livelihoods according to Muoni *et al*.,(2025) . However, climate change is threatening agricultural productivity and exacerbating some of Zimbabwe's key agricultural challenges: low soil fertility, reliance on rain-fed systems, poorly functioning markets, and farmers' limited access to credit, knowledge and best practices. Smallholder farmers in Zimbabwe are vulnerable to the impacts of climate change due to naturally low rainfall, yet they heavily rely on rain-fed systems. However, regardless of their vulnerability, farmers continue to grow maize (*Zea mays* L.) and chances of crop failures are very high, and their yields remain extremely low (Manyanga et al., 2025).

Still on the drivers of low maize production in Zimbabwe, a vast area of agricultural land is owned by smallholder farmers. Smallholder farmers continuously grow crops without external fertiliser inputs, mainly due to lack of purchasing power and scarcity of mineral fertilisers. Over time, nutrient mining results in poor soil fertility degradation and soil organic matter depletion (Chipomho *et al.*, 2020). Poor soil fertility and weed infestation are among the major constraints to maize production in Southern Africa. Nutrient and weed management strategies are needed to improve efficiencies on farms.

2.2 Risks of Importing Grains

Grain trade has expanded dramatically in Southern Africa (Lesotho importing wheat and maize from South Africa, and Zimbabwe importing maize from Zambia) during the past few decades to serve as a source of greater food availability and diversity, especially in many regions (Aguiar *et al.*, 2020). Nevertheless, international trade is associated with risks to agriculture such as the spread of invasive pests and diseases, chemical contamination, and disruption of domestic agricultural markets (Victor *et al*., 2013). These risks have negative economic, food security and environmental sustainability implications for local economies (Ummah, 2019). The high demand for grains has resulted in an increase in the volume of imports, which the regulatory bodies are unable to inspect and control thoroughly on every shipment. As a result, many foreign pests and diseases have been inadvertently introduced, which can completely destroy native agricultural

ecosystems. Documented examples in the region include the introduction of the Fall Armyworm (*Spodoptera frugiperda*) into sub-Saharan Africa via international trade routes, and outbreaks of Maize Lethal Necrosis Disease linked to the movement of contaminated seed and grain across borders.

2.3 Grain Pathogens and Diseases

Imported grains are one of the most important risks for introducing invasive pests and plant diseases. Many of the pests and pathogens introduced into an importing country cause substantial agricultural damage (Tshikhudo *et al*., 2025) A study by Kholik & Zaman, (2025) states that the Mediterranean fruit fly (*Ceratitis capitata*), a very damaging pest, has accidentally been brought into several areas through imported fruits. Now, in the United States (US), it is quarantined and stringent quarantine measures have been implemented to prevent its establishment. The outbreak of fruit fly is capital intensive to control. Similarly, *Bactrocera dorsalis*, the oriental fruit fly, has been introduced to China through fruit imports and has caused extensive damage to domestic fruit production (Coulibaly *et al*., 2014; Wan *et al*., 2011)

Based on the studies mentioned above, maize production can be hindered by the introduced pests and diseases. These invasive pests have a chain of effects on the whole agricultural sector, as they increase the demand for chemical control measures, leading to negative impacts on the environment and human health. In addition, an invasive species, once established, is almost impossible to eradicate, and usually requires long-term management strategies that place a financial burden on affected countries. Stricter phytosanitary measures have to be instigated by governments, but the implementation comes with challenges including high costs and trade restrictions by partner countries which fear for their own agricultural security (Syahri et al., 2019). The occurrence of mycotoxins in maize is largely dependent on environmental and agronomic factors. As such, maize is typically contaminated by both field fungi (those occurring at any stage throughout the growth cycle of the crop prior to harvest) and storage fungi (those occurring at any stage throughout the post-harvest phase of the crop). These fungi may produce mycotoxins in the maize given an enabling environment. Pest infestation also plays a significant role in contamination of raw maize by mycotoxins, either in the field or during storage, for example fumonisins (Hove et al., 2016).

2.4 International Regulations on Importation of Grains

Agricultural regulations are fundamental to global food security, addressing a complex array of issues that ensure the availability, accessibility, stability, and utilization of food worldwide (Kholik & Zaman, 2025). These regulations form the backbone of agricultural policy frameworks, guiding safe and sustainable food production, distribution, and consumption. Governments and international organizations have established strict regulations to protect agricultural systems (Islam & Mcgowan, 2024). The International Plant Protection Convention (IPPC) has developed principles of phytosanitary measures to prevent the introduction and spread of pests in trade in agricultural commodities. Many countries have adopted stringent quarantine inspections and traceability systems to make sure they meet the safety standards. These regulatory frameworks are only effective if enforcement agencies have strict monitoring mechanisms in order to strictly enforce rules and penalties on violators. Corruption and the lack of coordination between trade and agricultural ministries may result in the emergence of gaps which allow non-conforming shipments to enter the domestic market (Tshikhudo *et al*., 2025). To strengthen these regulatory frameworks, greater international collaboration is required, along with greater technological capability and investment in research to develop innovative solutions.

2.5 Morphological and Biochemical Characterization of Plant Pathogens

Macroscopic and microscopic observations of fungal and bacterial isolates are followed by biochemical and molecular characterization in order to confirm species identity (Demeni *et al*., 2025). Characterization of fungal isolates is usually based on colony morphology, pigmentation, spore formation and hyphal structure. A study by Demeni *et al.*, (2025) further characterized *Fusarium oxysporum* from imported maize based on cottony white colonies, reddish pigmentation on PDA, and curved macroconidia and septate hyphae under light microscopy. Differentiation of closely related fungal species requires such morphological characterization.

2.6 Impact of pH, Moisture % and Protein % on Grain Quality

A study by (Angelovič *et al.*, 2015) emphasized that drying of maize is vital for safe storage. Drying of maize either elongates the shelf life of maize or improves quality. However, unsuitable drying could decrease its feeding and processing quality. During the post-harvest treatment, grain

is cleaned from a substantial part of impurities and grain is dried to a moisture of 14 to 14.5% of its weight, if necessary. The purpose of the storage is to preserve and improve the grain quality. As grain ages, lipid and protein degradation leads to an increase in free fatty acids, causing the pH to drop. A pH below 6.2 is a strong indicator of storage deterioration, indicating rancidity and reduced nutritional quality.

2.7 Post-Harvest Microbial Infections and Their Impacts

A study by Adeyeye *et al*., (2022) states that mycotoxins are low molecular weight metabolic secretions from fungi which, when ingested, could be dangerous and produce serious side effects or undermine the health of humans or animals. Consumption of foods contaminated with mycotoxins could result in vomiting, weight loss, tumour growth, and death depending on the level of toxicity. There are many mycotoxins secreted by fungi and they are found in legumes, dried root and tuber crops as well as in cereal grain crops. Most mycotoxins are toxic to human organs like blood, kidneys, skin, or the central nervous system. Across Africa, staple crops such as maize, groundnuts, sorghum and millet are mostly contaminated with several mycotoxins produced by diverse fungi (Bhunjun *et al.*, 2021)

Mycotoxin contamination of agricultural produce may occur either in the field before harvesting or after harvesting when there are favourable post-harvest storage conditions. Apart from mycotoxin contaminations that occur in crops which are improperly stored, mycotoxins are also produced in the field when crops are infected with certain diseases. Deterioration of grains after harvesting is a global issue and it commonly occurs during storage and transportation (Abdullahi & Dandago, 2021). Pest infestation and microbial and rodent attacks are the causes of post-harvest losses in grains. Among all, fungal attack is the most disastrous, and it causes losses and health-threatening problems in improperly dried grains. Attack by fungi and pests depreciates grain qualities during medium and long-term storage.

The grain industry's primary challenge in developing countries is inadequate storage facilities and inadequate knowledge and experience on post-harvest management of grains (Brahmi & Tyagi, 2019). Storage management of harvested grains and measures to prevent insects from thriving in stored grains is of great concern to food processing and storage. Storage structures and facilities extend shelf life and reduce post-harvest losses by protecting grains against environmental factors, rodents and insects (Chowdhury *et al*., 2022).

There is a long history of the search for control measures to combat post-harvest microbial infection, many of which focus on chemical and biological control (Jongman *et al*., 2022). Commonly used fungicides to control *Penicillium* spp. in apples and pears in Europe and North America include thiabendazole and fludioxonil, but resistance to fungicides has raised concerns about fungicide use and human health, and biological options are being sought. Post-harvest pathogens have also spread to different regions with the global trade of fresh produce. A study by Rigby *et al.*, (2021) stressed that latent infections on imported fruits may not show up until after distribution and storage. The results of that study support this study, where imported maize is not thoroughly assessed at the entry point and stakeholders strongly rely on results from the source country. Studies on the identification of isolated latent infections in imported grains were improved by molecular techniques such as Polymerase Chain Reaction (PCR) and Next Generation Sequencing (NGS) in China and Australia (Monteiro *et al*., 2018)).

2.8 Cross-Border Plant Pathogen Transmission

Plant pathogen transmission across borders is a serious threat to global agriculture, food security and biodiversity (Hulme, 2021). The risk of introducing invasive pathogens to new environments has increased as more plant materials, fruits, vegetables and seeds move through international trade. Fungi, bacteria, viruses, and even nematodes are pathogens that can pass through contaminated agricultural products, soil, water and even insect vectors to cause devastating outbreaks in areas that were never affected before (Gai & Wang, 2024). Plant pathogens have been introduced from one border to another, leading to dramatic economic losses, lower crop yields and increased use of chemical control methods, which adds to environmental unsustainability (Davy et al., 2025).

A historically notorious example of cross-border pathogen transmission is the outbreaks of new pathogen races of various crop species including cereals, fresh fruits and vegetables, exacerbated by cross-border traffic aided by anthropogenic activities. The attack of maize and tomato crops by the equally damaging *Spodoptera frugiperda* and *Tuta absoluta* respectively in Southern African countries in 2016 to 2017 is a good example of how anthropogenic behaviour is linked to plant biosecurity challenges (Mudada *et al*., 2022)

Alegbeleye *et al*., (2022) noted that cross-border plant disease outbreaks have also been attributed to bacterial pathogens. The bacterium *Xylella fastidiosa* causes such diseases as

Pierce's disease of grapes, citrus variegated chlorosis, and olive quick decline syndrome, and has been spreading fast across continents. *X. fastidiosa* was first detected in Italy in 2013, where it is devastating olive orchards, resulting in economic losses in excess of €1 billion ((Telesca *et al*., 2024). Since then, the pathogen has been reported in France, Spain and Portugal, most probably due to the importation of contaminated ornamental plants. Among other important bacterial pathogens, *Ralstonia solanacearum*, the causal agent of bacterial wilt in tomatoes, potatoes, and bananas, has been found in various locations caused by contaminated water sources and international trade of infected plant materials. The introduction of *Ralstonia solanacearum* Race 3 Biovar 2 into Africa has had a devastating effect on the production of potato, with reported outbreaks in Kenya, Ethiopia, and Rwanda (Aguiar *et al*., 2020).

Dash *et al*., (2021) added that viruses have also exhibited high potential for cross-boundary movement as a result of globalization and climate change, which have expanded the range of insect vectors. A highly transmissible plant virus that has rapidly spread across many continents after initial detection in the Middle East in 2014 is the Tomato Brown Rugose Fruit Virus (ToBRFV). The virus has been reported in Europe, North America and Asia, presumably because tomato seeds were contaminated in international trade (Caruso *et al.*, 2022). Likewise, Banana Bunchy Top Virus (BBTV), which causes severe stunting of banana plants, has been spreading across Asia, the Pacific Islands and Africa through infected planting materials and aphid vectors (Lopes Geber & Fraga da Silva, 2025).

2.9 Control of Cross-Border Pathogen Transmission

Dubey *et al.*, (2021) highlighted that the introduction of plant pathogens across borders is intensified by insufficient biosecurity measures, weak quarantine enforcement, or climate variability. The study further noted that imported plant materials are often used as a reservoir of pathogens otherwise absent in pest-free regions that lack phytosanitary controls. Preventing entry of foreign pathogens is very stringent for the importation of agricultural products into the European Union and the United States, which requires pre-export inspections, quarantine treatments, and pathogen-free certification. Nevertheless, despite these measures, there are still cases of undetected introduction of pathogens, such as *Monilinia fructicola*, a fungal pathogen causing brown rot in stone fruits, which emerged in Europe through imports of North and South American origin (Salih et al., 2021; Zaman et al., 2025).

Another major factor influencing the movement of plant pathogens across borders is climate change (Noack *et al*., 2024). In many cases, and particularly in new regions, new conditions for successful survival and establishment of invasive pathogens were created by rising temperatures, altered precipitation patterns and extreme events (Brooks *et al*., 2022). Diseases have been linked to climate variation, which in Africa has seen an increase in the spread of Maize Lethal Necrosis Disease (MLND), a viral disease caused by the co-infection of Maize Chlorotic Mottle Virus (MCMV) and Sugarcane Mosaic Virus (SCMV). MLND was first identified in Kenya in 2011 and has since spread to Uganda, Tanzania, and Rwanda, where there have been outbreaks associated with the cross-border trade of infected maize seed (Hill *et al.*, 2020).

In order to reduce the extension of plant pathogens across borders, there is a need for surveillance, early detection, and strict quarantine protocols. Real Time Polymerase Chain Reaction (RT-PCR) and NGS have been utilized as molecular diagnostic tools for diagnosing latent infections in imported plants (Olubukola O. Babalola, 2012). Moreover, there is the use of remote sensing technologies and machine learning models to identify early the progression of disease outbreaks over large agricultural landscapes. Indeed, cross-border pathogen transmission should also be addressed through international collaboration. The IPPC and other regional plant health organizations are important for the establishment of global phytosanitary standards and the coordination of response strategies to new plant disease threats (Dubey *et al*., 2021).

CHAPTER 3: MATERIALS AND METHODS

3.0 Materials and Methods

3.1 Site Description

The study was conducted at Plumtree Border Post, which is situated in Matabeleland South Province (MSP) of Zimbabwe, in Mangwe District. Bulawayo is 110 km from Plumtree Border Post. The Botswana border is an essential point of entry for goods and people to Zimbabwe and neighbouring countries. It is a cross-border trade hub, importing grains and vegetables from South Africa. The study site coordinates are: S 20°32′26.00988″ E 27°44′15.09612″. The border post is accessible from Bulawayo via the A7 road (Bulawayo–Plumtree Road), travelling south-west through Esigodini and Marula towards the border with Botswana.

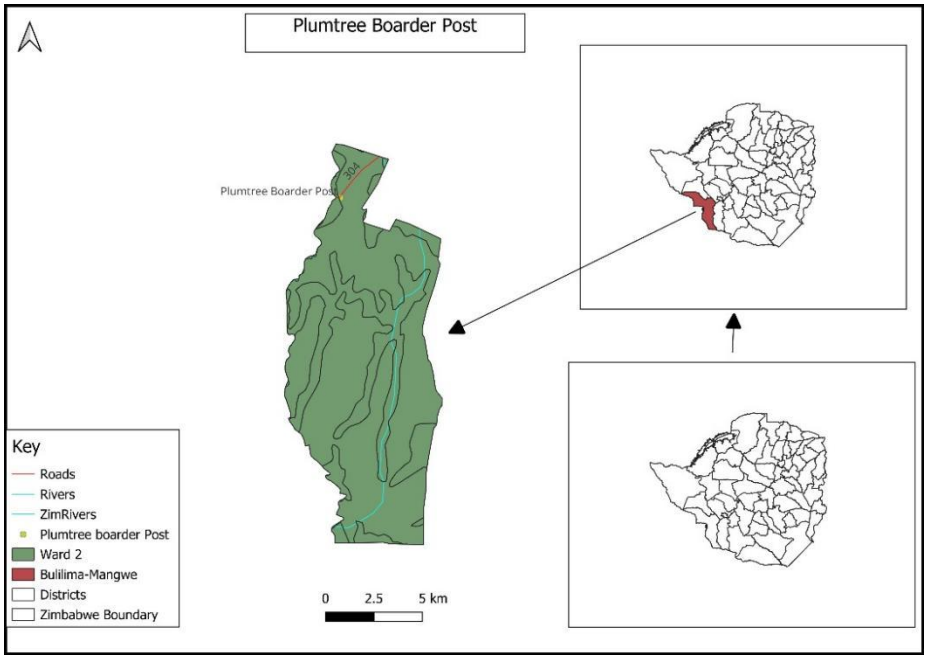


Figure 3.1: Plumtree Border Post

Source: Geographic Information System (GIS) shape files from Plumtree Rural District Council (PRDC)

3.2 Sampling Procedure

3.2.1 Site Selection

The Plumtree Border Post plays a pivotal role in evaluating agricultural risks tied to pathogenic microorganisms due to its position as a primary entry point for goods and travellers between Zimbabwe and neighbouring countries, particularly South Africa. This site is a hub for the importation of grains, making it essential for addressing cross-border biosecurity concerns. Its strategic importance lies in its capacity to regulate and inspect large volumes of imported grains, which have the potential to introduce harmful pathogens that can impact local agriculture and food security. The proximity to South Africa, a major agricultural producer, amplifies the risks associated with pathogens due to high trade activity. Thus, Plumtree Border Post represents a critical node for mitigating risks and protecting Zimbabwe's agricultural integrity.

3.2.2 Study design

Completely Randomized Observational Design with Sub-sampling was used where maize trucks are the experimental units. The 2 sampling points per truck and 3 lab objectives act as replicates. The study measured naturally occurring fungal load and moisture.

3.2.3 Data collection

The grains were collected using a systematic stratified random sampling method. This approach ensured an unbiased and representative selection of samples (where every third 30-tonne truck was selected from the front of the queue). Stratification allowed only the representation of yellow maize trucks being sampled (Sabillon, 2014). Plumtree Border Post was selected due to its high volume of imported grains from South Africa, which provided an ideal environment for investigating the prevalence of pathogenic microorganisms.

The study examined 18 kg of yellow maize. Each 30-tonne truck consisted of two sample points. From each sample point, 600 g of yellow maize was sampled and stored in a khaki envelope before being sent to the lab. From the 600 g sampled, 200 g was used to achieve objective 1, 200 g for objective 2, and the last 200 g to obtain objective 3. A total of 5 trucks were selected per month. A total of 15 trucks were sampled from January 2026 to March 2026.

3.2.4 General management and Laboratory Tests

Experimental research was conducted to find out the hazards associated with imported maize from RSA using a cross-sectional laboratory-based analytical study. Collected samples were ground before being sent to the lab. The experiment utilized essential laboratory reagents, including sodium hypochlorite for sterilization, sterile distilled water for washing, and peptone water as the diluent. Chloramphenicol and gentamicin were used as selective agents to inhibit undesired microbial growth. Lactophenol Cotton Blue stain was employed for the microscopic identification of fungal isolates. Vadillo-Dieguez *et al*., (2024) underscored the role of sodium hypochlorite in effective microbial surface sterilization, while pointing to peptone water as a critical diluent that maintains microbial viability during sample preparation. (Luciano-Rosario *et al*., 2020) reiterated the selective utility of chloramphenicol and gentamicin in minimizing non-target microbial growth.

Potato Dextrose Agar (PDA) was used for fungal isolation. Nutrient Agar (NA) was used to promote bacterial growth. The media were prepared according to the manufacturer's instructions. A Biobase incubator was used to sterilize the agar. (Luciano-Rosario *et al*., 2020) highlighted that Potato Dextrose Agar (PDA) is a widely used medium for fungal isolation due to its nutrient-rich composition.

The plates were inoculated under sterile conditions. The samples were surface sterilized using 0.1% sodium hypochlorite. The plates were incubated under optimal growth conditions specific to the target microorganisms. Fungal cultures on Potato Dextrose Agar were incubated at 28°C for 72 hours, while bacterial cultures on NA were incubated at 37°C for 48 hours. This allowed for effective colony development and differentiation. Duniya *et al*., (2018) emphasized the importance of appropriate incubation conditions, stating that temperature and time settings tailored to the microbial target can significantly influence colony development. Meena *et al*., (2021) described the pour plate and spread plate techniques as indispensable for achieving uniform microbial distribution on culture media.

For fungal colonies, macroscopic characteristics, such as colour, texture, and growth patterns, were documented. Additionally, Lactophenol Cotton Blue staining was used for microscopic observations to identify fungal species accurately. Lactophenol Cotton Blue staining is regarded

as a gold standard for fungal identification, particularly in visualizing hyphal structures and reproductive elements.

Collected samples were sent to National Foods for physicochemical tests (moisture %, protein %, and pH). Oven drying method was used to measure moisture content from the grinded maize samples.

3.4 Data Analysis

Descriptive statistics using IBM SPSS version 27.0.1 were used to identify types of fungi introduced through maize importation in Zimbabwe. An ANOVA test was used to compare the total number of fungal moulds introduced in Zimbabwe through maize imports from South Africa across three different months. To assess the relationship between moisture content and fungal mould count, Pearson correlation was performed using SPSS version 27.0.1.

CHAPTER 4:

RESULTS

4.1 Types and Frequency of Fungi

A total of four types of fungi were identified from the research (*Aspergillus spp.*, *Candida spp.*, *Fusarium spp.* and *Penicillium spp.*). *Aspergillus spp.* contributed 10%, 13.3% was from *Candida spp.*, *Fusarium spp.* was most prevalent (43.3%), followed by *Penicillium spp.* with 33.3%. Table 4.1 shows the type and frequency of fungi.

Table 4.1: Types of fungi and frequency

Type of fungi (spp)	Frequency	Percent
Aspergillus spp	3	10.0
Candida spp	4	13.3
Fusarium spp	13	43.3
Penicillium spp	10	33.3
Total	**30**	**100.0**

4.2 Total Number of Moulds Across Three Months

The Kolmogorov-Smirnov test indicated that the total number of fungi from January to March followed a normal distribution curve, $D = 0.143$, $p = 0.118 > 0.05$. The ANOVA results indicated no statistically significant differences in fungal counts among the three months sampled ($p = 0.500$). This indicates that the month of sampling did not significantly influence fungal contamination levels in imported maize. Table 4.2 shows the ANOVA table indicating the results of fungal mould counts across three different months (January, February and March).

Figure 4.1 shows the types of fungi mould counts across three different months. As indicated by Figure 4.1, February had the highest fungal diversity, followed by March, then January.

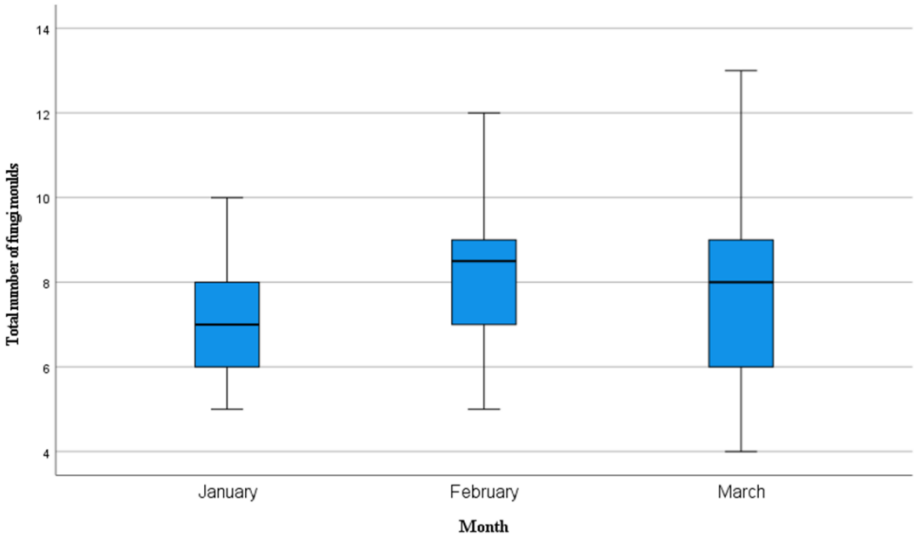


Figure 4.1: Fungi mould counts across three different months

4.3 Relationship Between Moisture Content and Fungal Contamination

The Kolmogorov-Smirnov test showed that moisture content data were normally distributed, D = 0.123, p = 0.200 > 0.05. Pearson correlation analysis was conducted to determine the relationship between moisture content and total fungal count in imported maize samples. The results showed a non-significant weak positive correlation between moisture content and fungal count (r = 0.163, n = 30 p = 0.389). Therefore, moisture content did not significantly influence fungal contamination levels in the maize samples analysed. Although fungal counts tended to increase slightly with increasing moisture content, the relationship was weak and statistically insignificant. This suggests that factors other than moisture content also contribute to fungal contamination in the imported maize samples.

Figure 4.2 shows the relationship between moisture content (%) and mould count in imported maize from South Africa. A slight positive correlation ($R^2 = 0.027$) between moisture content and mould count is indicated by the regression line ($y = 2.61 + 0.4x$).

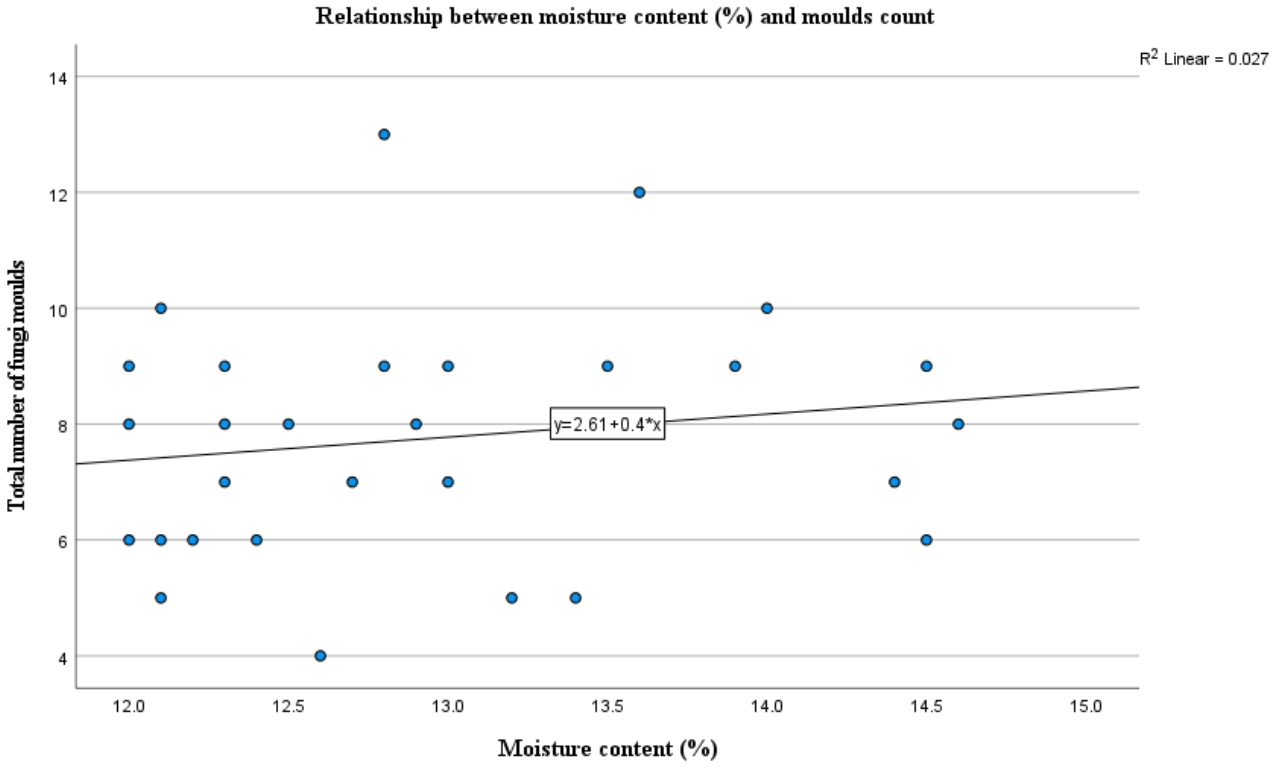


Figure 4.2: Relationship between moisture content (%) and mould count

CHAPTER 5: DISCUSSION

5.0 Results Discussion

5.1 Types and Frequency of Fungi

The predominance of *Aspergillus spp.*, *Fusarium spp.*, and *Penicillium spp.* in imported maize from South Africa supports research by (Patrick Ije *et al.*, 2024) which identified these fungi as the most common contaminants of stored cereal grains, particularly maize. These fungi are commonly associated with poor storage conditions, high humidity, and prolonged transportation periods (Poroșnicu *et al.*, 2026). *Fusarium spp.* produce fumonisins and mycotoxins harmful to humans and livestock, as indicated by (Poroșnicu *et al*., 2026)

Overall, the findings suggest that imported maize acts as a vehicle for introducing potentially toxigenic fungi into Zimbabwe, posing risks to food safety, grain quality, and public health.

5.2 Total Number of Moulds Across Three Months

The findings of the research also highlighted the absence of significant differences in fungal occurrence across three different months (January, February, and March). The results indicate either constant environmental factors like temperature and humidity, or constant storage and transportation conditions. Such observations are in line with a study by (D. C. Meena et al., 2020) who stated that stored maize fungal populations remained relatively unchanged under constant storage conditions, for example January to March being in the same season.

5.3 Relationship Between Moisture Content and Fungal Contamination

The study showed a positive relationship between moisture content and fungal counts. Such results agree with a study by Palacios *et al*., (2022) which states that increased moisture promotes fungal growth in stored maize. Moisture is one of the most important factors influencing fungal proliferation, because fungi require available water for metabolic activity and colonization.

The slight positive correlation observed indicates that although moisture contributed to fungal growth, other factors such as temperature, duration of storage, grain damage, aeration, and transportation conditions may also have influenced fungal contamination.

CHAPTER 6: CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion and Recommendations

The study identified the major fungal contaminants associated with imported dried maize in Zimbabwe. Four major fungal genera, namely *Aspergillus spp., Candida spp., Fusarium spp.*, and *Penicillium spp.*, were isolated from the maize samples. The predominance of these fungi indicates that imported maize may serve as a vehicle for the introduction of potentially toxigenic and spoilage microorganisms into the country.

The research found that there was no significant ($p > 0.05$) difference in fungal occurrence and counts among maize imported during January, February, and March. This suggests that fungal contamination remained relatively consistent throughout the study period and may largely originate from pre-harvest, storage, transportation, or handling conditions before importation.

In addition, the study demonstrated a slight positive relationship between moisture content and fungal counts. This finding confirms that moisture contributes to fungal proliferation in maize, although other environmental and storage factors may also influence fungal growth.

Based on the findings of the study, the following recommendations are proposed: relevant authorities such as the Ministry of Agriculture, and grain inspection agencies should implement routine fungal screening of imported maize at border entry points and storage facilities like the GMB and other milling companies.

Furthermore, future monitoring programs should include testing for mycotoxins such as aflatoxins and fumonisins, due to the presence of *Aspergillus spp.* and *Fusarium spp.*, at border entry points and storage facilities like the GMB and other milling companies.

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APPENDIX

Appendix A: Raw Data – Fungal Counts and Moisture Content

Sample Code	Type of Fungi	Total Number of Fungi	Border Post	Date	Moisture Content %
J	*Penicillium spp*	8	Plumtree	January	14.6
J	*Fusarium spp*	9	Plumtree	January	13.5
J	*Fusarium spp*	10	Plumtree	January	14.0
J	*Fusarium spp*	9	Plumtree	January	13.9
J	*Fusarium spp*	12	Plumtree	January	13.6
J	*Fusarium spp*	8	Plumtree	January	12.9
J	*Fusarium spp*	5	Plumtree	January	13.2
J	*Fusarium spp*	9	Plumtree	January	14.5
J	*Penicillium spp*	7	Plumtree	January	14.4
J	*Penicillium spp*	6	Plumtree	January	14.5
F	*Candida spp*	10	Plumtree	February	12.1
F	*Penicillium spp*	8	Plumtree	February	12.3
F	*Penicillium spp*	7	Plumtree	February	12.7
F	*Penicillium spp*	8	Plumtree	February	12.0
F	*Candida spp*	6	Plumtree	February	12.1
F	*Candida spp*	9	Plumtree	February	12.0

Sample Code	Type of Fungi	Total Number of Fungi	Border Post	Date	Moisture Content %
F	*Penicillium spp*	6	Plumtree	February	12.0
F	*Penicillium spp*	5	Plumtree	February	12.1
F	*Penicillium spp*	7	Plumtree	February	12.3
F	*Candida spp*	6	Plumtree	February	12.2
M	*Fusarium spp*	4	Plumtree	March	12.6
M	*Fusarium spp*	6	Plumtree	March	12.4
M	*Aspergillus spp*	5	Plumtree	March	13.4
M	*Aspergillus spp*	8	Plumtree	March	13.6
M	*Penicillium spp*	9	Plumtree	March	12.8
M	*Fusarium spp*	7	Plumtree	March	13.0
M	*Fusarium spp*	8	Plumtree	March	12.5
M	*Fusarium spp*	13	Plumtree	March	12.8
M	*Fusarium spp*	9	Plumtree	March	12.3
M	*Aspergillus spp*	9	Plumtree	March	13.0

Table 4.2: ANOVA table showing fungal counts across three different months transfer the ANOVA to the appendix, the table below will surfice

Source	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	6.067	2	3.033	0.710	0.500
Within Groups	115.300	27	4.270		
Total	**121.367**	**29**			

Table 4.3: Relationship between moisture content and fungal count

Correlations	Statistic	Total number of fungi	Moisture content %
Total number of fungi	Pearson Correlation	1	0.163
	Sig. (2-tailed)		0.389
	N	30	30
Moisture content %	Pearson Correlation	0.163	1
	Sig. (2-tailed)	0.389	
	N	30	30

AUTHOR ACTION ITEMS

The items below are supervisor comments that require your own study data, findings, or judgement to resolve. General/editorial corrections have already been applied directly in the chapters above (shown highlighted in yellow). One comment (Comment 28, botanical name of maize) was found to already be addressed in the text and needed no change. Please supply the missing information for each item below, then remove this section.

Comment 5 — Methodology (Data collection)

Clarify whether physicochemical assessment was based on moisture content alone, or whether other physical properties were also measured. State this explicitly.

Comment 13 — Introduction

Briefly describe the most common fungal contaminants of imported maize and their negative effects, and use this section to clearly state the research gap.

Comment 17 — Research Objectives

Make the stated objective more specific (currently reads as broad/general).

Comment 19 — Hypotheses

Explain why Aspergillus, Fusarium and Penicillium specifically were selected as the target genera for this hypothesis.

Comment 38 — Literature Review

Clearly articulate the specific research gap that this study addresses, supported by the literature reviewed.

Comment 43 — Methodology (Study site coordinates)

This marker had no accompanying comment text in the reviewed file — please check with your supervisor whether an action is still required here.

Comment 50 — Methodology (Moisture content determination)

State the oven-drying temperature (°C) and duration (hours) used to determine moisture content.

Comment 52 — Methodology / Data Analysis

State whether the data met the assumptions required for ANOVA (e.g. normality, homogeneity of variance) before the test was run, and where this was tested.

Comment 58 — Results

Move/specify the normality and assumption-testing information (e.g. Kolmogorov–Smirnov results) under the Data Analysis subsection of the Methodology chapter, rather than only in the Results.

Comment 59 — Results (Figure 4.1)

Explain how differences in fungal diversity across the three months were determined — state the statistical test or criteria used.

Comment 61 — Results (Moisture content)

Provide the actual moisture content range recorded across the study samples.

Comment 65 — Discussion

Discuss any other fungal species/genera identified in your samples beyond Aspergillus, Fusarium and Penicillium, if applicable.

Comment 66 — Discussion

Compare your results with studies on local/Zimbabwean fungal contaminants, and note whether any species identified is uncommon or of particular concern.

Comment 68 — Discussion

State what the actual weather conditions (temperature, humidity) were during January–March to support the claim of 'constant environmental factors'.

Comment 70 — Discussion

Correct this discussion: the correlation being discussed was NOT statistically significant. Rewrite so the interpretation accurately reflects a non-significant result, rather than implying agreement/disagreement as if it were significant.

Comment 73 — Discussion

Confirm and clearly state that the relationship was not statistically significant, including the exact p-value, rather than leaving this ambiguous.