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Maize Common Smut (*Ustilago maydis*) Infection Mechanism and Its Management Options

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Abstract: Maize indeed plays a crucial role in Ethiopia's food security, but its productivity faces significant challenges due to maize smut (Ustilago maydis (DC.) Corda). U. maydis poses a serious threat as it can infect different parts of the maize plant, leading to substantial yield losses ranging from 40 to 100%. Understanding the lifecycle of U. maydis is essential for devising effective control strategies. This pathogen exhibits a dimorphic lifestyle, with haploid sporidia that are not infectious but can grow as saprophytes. However, infection occurs when compatible diploid sporidia mate, initiating filamentous growth and ultimately leading to host infection. Teliospores facilitate the pathogen's survival and spread. Implementing good agronomic practices includes removing infected plants before the pathogen bursts, deep plowing, crop rotation, disease-free seed, recommended nitrogen fertilization, and plant density. Using advanced methods like bi-1 gene silencing and UmRrm75 gene deletion could help make maize more resistant to U. maydis and weaken its ability to infect other plants. Additionally, seed dressing with Carboxin 15% and Thiram 13% has proven effective against seed-borne U. maydis. By combining cultural practices, genetic resistance, and chemical control measures, farmers can mitigate the impact of maize smut and improve maize productivity in Ethiopia.

Keywords: teliospores, maize, smut, karyogamy, gene silencing, gene deletion.

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INTRODUCTION

Maize holds significant importance globally as a staple food, feed, and industrial crop, contributing substantially to the economy and trade. In Ethiopia, maize ranks second in terms of cultivation area, but it leads in productivity and total production (1), underscoring its vital role in the country's agricultural landscape. However, despite its importance, maize productivity in Ethiopia lags behind the global average (1, 2) due to various biotic factors, with fungal diseases being a major concern (3).

Maize smut, among the fungal diseases affecting maize in Ethiopia, stands out as a significant threat. Reports indicate that this disease is 100% prevalent across west Wollega, Oromia, and Ethiopia (4). It was also reported from Sudan for the first time, with a 5% incidence in Yei and Morobo counties (5). This disease primarily affects warm and moderately dry regions, leading to substantial damage to maize stalks and ears through the formation of large galls (6). Studies have shown that these galls can cause yield losses ranging from 40% to 100% (6, 7), highlighting the economic impact of maize smut on farmers.

Effective management of maize diseases like smut depends on understanding the causal agent's infection mechanisms, inoculum sources, and transmission methods. *U. maydis, a biotrophic basidiomycete, parasitizes maize and induces tumor formation on all aerial parts of the plant (8), ultimately resulting in stunted growth and yield reduction (9).*

To combat maize smut effectively, it's crucial to explore various management options. These may include cultural practices such as crop rotation, sanitation measures like the removal of infected plant debris, and the use of disease-resistant maize varieties. Additionally, chemical control methods like seed treatment with fungicides can also help in managing the disease.

BIOLOGY OF U. MAYDIS

Taxonomy of *U. maydis*

Ustilago maydis, the causative agent of maize smut, belongs to the Kingdom Fungi, Phylum *Basidiomycota*, Class Ustilaginomycetes, Order Ustilaginales, Family Ustilaginaceae, Genus *Ustilago*, and Species *U. maydis* (10). This taxonomic classification provides insight into its evolutionary relationships and biological characteristics.

Lifecycle of *U. maydis*

The fungus exhibits three distinct phases in its lifecycle: diploid, haploid, and dikaryon (11). In laboratory conditions, both diploid and haploid mycelia can grow rapidly (John, 1968). However, dikaryon formation and subsequent development are restricted to the host tissue, particularly maize. During infection, cells with compatible mating types fuse to form a heterokaryon, which is essential for completing the fungal lifecycle (12, 13, 14). It's worth noting that successful infection and completion of the lifecycle require that the alleles at that mating *locus* are different, ensuring genetic diversity and successful reproduction.

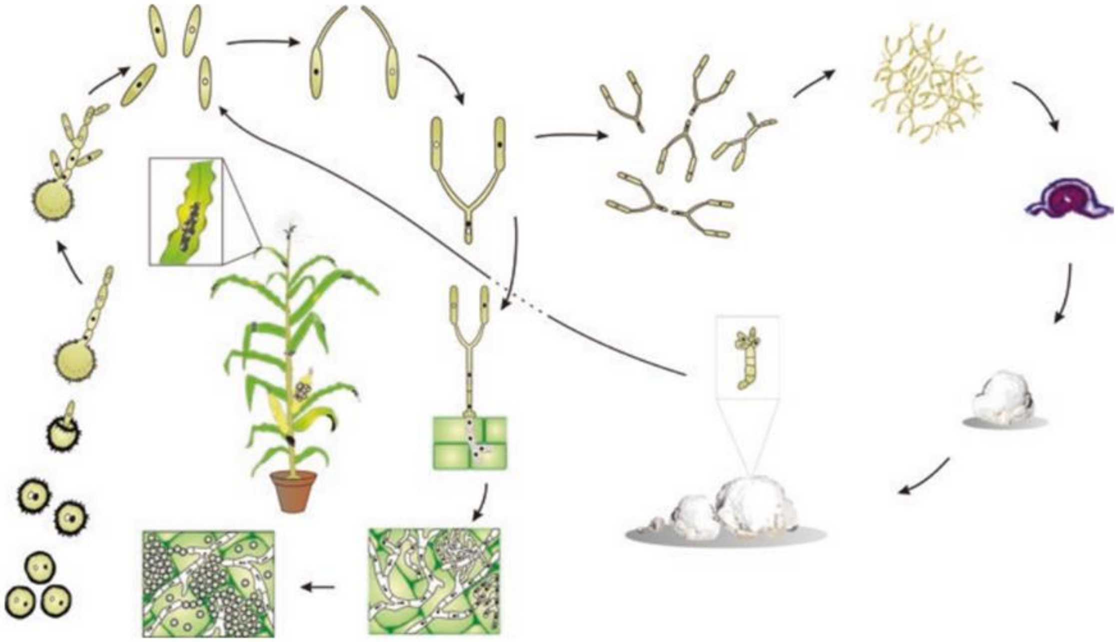

Figure 1; The Life Cycle of *U. maydis* (13).

Teliospores serve as the sexual, thick-walled resting spores of smut fungi, enabling them to survive adverse conditions for several years. Karyogamy, the fusion of two haploid eukaryotic cells, occurs during the formation of teliospores, ensuring genetic recombination and diversity within the fungal population (Figure 1).

In the lifecycle of *U. maydis*, haploid cells formed after basidium development mature into basidiospores, each containing a single complete set of chromosomes. This dimorphic fungus primarily infects the host plant during the dikaryotic stage, characterised by the presence of two genetically distinct nuclei within the same hyphal cell (15).

1. *maydis* host infection mechanism

The infection mechanism of *U. maydis* involves a series of steps aimed at penetrating various organs of the maize plant, including leaves, stems, ears, and tassels. Initially, the fungus grows along the epidermis of these organs before penetrating between epidermal cells to gain access and subsequently spreading throughout the sub-epidermal cells. During this process, U. maydis forms specialized infection structures called appressorium, aided by cell wall-degrading enzymes, to facilitate penetration into the host cells (16).

Within 3 to 5 days of infection, there is a lot of growth (hypertrophy) and too much cell division (hyperplasia). This is the start of the tumour formation caused by the fungus (17). The interaction between Ustilago maydis and the host plant is tissue-specific, with different responses seen in cell types that are on the epidermis and those that are inside the plant.

As the fungal hyphae move through plant cells, they grow between cells, making close connections that weaken the host's defences by blocking the supply of nutrients (18). This intricate interplay between the pathogen and the host highlights the sophisticated strategies employed by U. maydis in maize to establish infection and promote tumor formation.

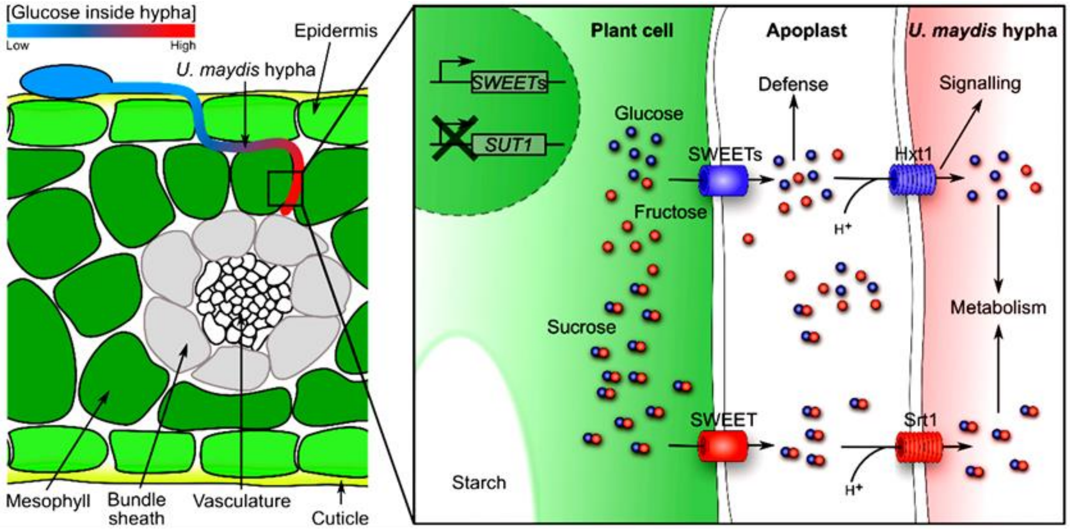

Figure 2: A complex overview of sugar flux in *U. maydis*-infected maize leaf tissue (18).

As depicted in Figure 2, U. maydis employs sophisticated strategies to manipulate the host plant's sugar metabolism for its own benefit, ultimately aiding in its growth and proliferation within the maize tissue. Here's a breakdown of the key points:

1. ***Maydis hyphal growth:*** *The fungus grows between the epidermis and mesophyll cells, reaching the leaf bundle sheath and vasculature.*

2. **Glucose accumulation**: Glucose accumulates primarily towards the tip region of the hyphae, likely serving as a nutrient source for the growing fungus.

3. **Expression of sweets**: Upon infection, U. maydis induces the expression of sweets. These are proteins that facilitate the leakage of hexoses (like glucose) and sucrose into the apoplast, the extracellular space between plant cells.

4. **Uptake of sugars by Maydis**: The fungus takes up the leaked hexoses and sucrose via specific transporters called Srt1 and Hxt1. This uptake mechanism allows U. maydis to access the sugars present in the plant tissue.

5. **SUT1 expression blockage**: Maydis block SUT1, a protein responsible for loading sugars into the phloem, which is the vascular tissue responsible for transporting nutrients throughout the plant. By inhibiting SUT1, U. maydis prevents the normal phloem loading process.

6. **Starch accumulation**: By blocking SUT1 expression and disrupting normal sugar transport, U. maydis induces the accumulation of starch within the infected leaf tissue. Starch serves as a storage form of glucose in plants.

Depending on the specific organs of the maize plant targeted, distinct patterns emerge in the progression of U. maydis infection and tumour formation. Around four days after infection (dpi), a large biotrophic interface forms on seedling leaves, with hyphae often colonising meristematic tissue at the base of the leaf blade. After that, cells in the bundle sheath and mesophyll turn into tumour cells, which causes tumours to grow in the fully differentiated distal blade tissue (19). By 13 dpi, both epidermal and mesophyll cells undergo significant enlargement, with mesophyll cells often tripling in size (19).

In the anthers, U. maydis reaches sub-epidermal cells around three days post-infection, which causes extra-periclinal divisions in the somatic tissues. Even though middle-layer cells usually die early on during anther development, infected anthers show hyperplasia and hypertrophy, turning them into noticeable areas of large growth (20). By 7–10 dpi, the surface of infected tassel organs, including anthers, becomes distorted and enlarged (21). By 15 dpi, mature tumours begin to split open, releasing diploid teliospores (22).

Transmission and over-wintering of *U. maydis*

The life cycle of U. maydis includes transmission and overwintering mechanisms that are critical for its survival and proliferation. Wind and rain can spread common smut spores, which can overwinter in the soil and remain viable for several years. Smut galls can form at wound sites caused by a variety of factors, such as insect feeding, hail, or wind damage. Warmth and moisture are favorable conditions for gall development, with susceptibility increasing during drought or wet conditions, especially during or after pollination. It's important to note that each point of infection represents a separate event, as the fungus is not systemic within the maize plant (23).

Understanding the intricacies of *U. maydis*'s life cycle, infection dynamics, and transmission mechanisms is crucial for implementing effective management strategies to control maize smut disease and minimise its impact on maize production.

1. *Maydisimpact* on humans’s health

It has a minimal direct health effect on humans, causing respiratory tract diseases such as allergies and asthma (24). However, its culinary use (25) and potential bioactive compounds that have positive effects on human health make it an interesting subject for further research (26). It is also used as a food (25).

Management Options

Cultural, genetic, and chemical strategies are available for managing U. maydis-caused maize smut.

Cultural practices:

- It is important to remove common smut-infected maize plants before smut galls rupture (27, 28).
- Adjusting plant densities and nitrogenous fertilisation to reduce *maydis* infection. Higher plant densities and nitrogen fertilisation can favour maize smut infection (29, 30, 31), so optimal levels should be determined.
- Deep plowing was used to bury survival spores.
- Rotate your crops using non-cereal plants to break the cycle of disease.
- Use disease-free maize seeds to stop the initial infection.

Genetic approaches:

- Virus-induced gene silencing (VIGS) targets specific maize genes involved in the interaction with*maydis* (32, 33), such as Terpene synthase 6/11 (*tps6/11*) (34, 35), Endochitinase B (ecb), Bowmann-Birk type trypsin inhibitor (bti), and Bax inhibitor 1 (*bi-1*) (36). Silencing *bi-1* has shown promising results in reducing *U. maydis* colonisation and disease development (36).
- Targeted transcriptional induction of tps6/11 to control*maydis* disease development in maize (36).

Genetic Modification:

- Targeting RNA-binding proteins in maydis, such as the UmRrm75 gene, are involved in filamentation and pathogenicity (37, 38, 39). Deletion of this gene results in reduced mating and post-mating filamentous growth of *U. maydis*, as well as decreased virulence in maize (38).

Chemical control

- Maize seed dressing with Carboxin 15% and Thiram 13% has been identified as an effective option to control seed-borne *maydis*, providing chemical protection against initial infection.

CONCLUSION

It's evident that maize plays a crucial role in Ethiopia's agriculture. However, the disparity between the national average yield and the global average underscores the need to address factors contributing to this productivity gap, including biotic factors such as maize smut caused by *U. maydis*. The prevalence of maize smut in Ethiopia highlights the importance of effective management strategies to mitigate its impact on maize production. Understanding the biology and lifecycle of the causative agent, Ustilago maydis, is crucial for devising management options. According to the information provided, here are some key points and management strategies:

- **Understanding *Ustilago maydis*:** *This fungus has a dimorphic lifestyle, transitioning between haploid sporidia and filamentous growth when mating on the plant surface.* It has three phases: diploid, haploid, and dikaryon. Teliospores are the sexual, thick-walled resting spores of smut fungi.

- **Spread and survival:** Common smut spores overwinter in the soil and remain viable for several years. Wind and rain spread them.

- **Management options:**

- **Cultural practices:**
- Infected plants can be removed before smut bursts, which can help reduce the spread.
- Adjusting plant density and using the recommended nitrogen fertilizer can also help with disease management.
- Deep plowing to bury survival spores, as well as crop rotation with non-cereals, can disrupt the disease cycle.

Genetic approaches:

- Silencing the bi-1 (Bax inhibitor 1) gene in maize can improve resistance to U. maydis.
- In maize, deleting genes like UmRrm75 in U. maydis can decrease virulence.

Chemical control

- Maize seed dressing with Carboxin 15% and Thiram 13% has shown effectiveness in controlling seed-borne *maydis*.
- **Integrated Pest Management:** Combining multiple management strategies, including cultural, genetic, and chemical approaches, can provide a holistic and sustainable approach to effectively reduce the incidence and severity of maize smut, thereby minimising yield losses and ensuring sustainable maize production.

Implementing these management options, combined with proper monitoring and early detection, can help mitigate the impact of maize smut on productivity in Ethiopia and other affected regions.

DISCLOSURE STATEMENT

The authors are not aware of any affiliations, memberships, or financial holdings that might be perceived as affecting the objectivity of this review.

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