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DIAGNOSING AND IDENTIFICATION OF FUNGAL MYCOFLORA DISEASES ON WHEAT SEED VARIETIES AT AMBO AGRICULTURAL RESEARCH CENTER, AMBO, ETHIOPIA

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Abstract

A plant is said to be diseased when its normal functions are disturbed and harmed. Wheat (Triticum spp) is the world's leading cereal grain which is used as staple food worldwide and its production is constrained by several biotic and abiotic factors. Fungi mycoflora which is predominantly incited among the biotic stresses causes a significant reduction in wheat grain yield and its quality. Without proper identification of the disease and the disease-causing agent, disease control measures can be a waste of time and money and can lead to further losses. Proper disease diagnosis is therefore vital. This study done during the 2021 at Ambo Agricultural research center with the objective of to differentiate the different wheat fungal mycoflora diseases in different varieties by observing mycelia symptom and diagnosing their sign using compound microscope with different magnification power. During the study Seed health test works were conducted up on the seed of three wheat Varieties namely Bollo, Tsehai and Menze to determine whether the seed is health or diseased using Agar Plate method. Planting after five, seven- and nine-days intervals, the mycelia growth for ten seeds per Petridis were recorded in germination percentages and the mean of the mycoflora were calculated. Hence Agar Plate Method wheat seed test in this study showed that the presence of the fungal pathogens on the tested three varieties during 5th, 7th and 9th days ranges from 40 to 100 infection percentage.

Keywords: Disease diagnosis, wheat varieties, fungi mycoflora, agar plate method, seed health, etc.

INTRODUCTION

Wheat (*Triticum aestivum L.*) is grown globally and is the world's most important cereal crop (Pant *et al.,* 2020). Wheat is one of the most important crops for global food security; it is widely cultivated on more than 200 million ha of land and produces more than 781 million tons annually to feed the world (Wheat Initiative, 2019). Bread wheat (*Triticum aestivum*) covers about 90% of the world wheat acreage while only 9% is covered by durum wheat (*Triticum durum*); also called Marconi wheat (Singh *et al.*

2002. Wheat is a optimum growing temperature is about 25°C with minimum and maximum growth temperatures of 3-4°C and 30- 32°C, respectively (Curtis, 2002). Plant disease epidemics impact agriculture and forestry by reducing the quantity and quality of the product, and pose a threat to food security and food safety (Strange and Scott 2005; Oerke 2006; Madden *et al.* 2007; Savary *et al*. 2012, 2017). Knowledge of the quantity of disease is fundamental to a) determine crop losses; b) conduct disease surveys; c) establish thresholds for decision making; d) improve knowledge of disease epidemiology and e) evaluate the effect of treatments (e.g. cultivar, fungicides, etc.). Plant disease intensity (a generic term) can be expressed by incidence or severity at the field/plot scale and below. Incidence is the proportion of the plant units that are diseased in a defined population or sample (Madden *et al*. 2007) while severity is the proportion of the plant unit exhibiting visible disease symptoms, usually expressed as a percentage (Bock *et al.,*2020).

Symptoms of disease on a plant may change in size, shape and color. Disease severity is often the variable that is of most importance or interest in a particular experimental situation (Paul *et al.* 2005).

Plant disease diagnosis is identification of the cause/causal agent of the disease. The causal agents may be sourced from biotic/living agent (Pathogens/parasitic microorganisms, Pests/insects or mammals feeding on or damaging plants) and abiotic agents which are the nonliving agents (damage from chemicals, weather, mechanical and nutritional problem). The diagnosis of plant diseases is a scientific art that is improved with experience and constant study. Disease Diagnosis and pathogen detection is required to detect and identify the new pathogen, determine the presence and quantity of pathogen, evaluate effectiveness of pathogen, solve complex disease incited by two or more pathogens and determine the exact disease incidence and consequently yield loss.

To diagnose a plant disease successfully, you need to be familiar with the basic classification of plants and at least how the healthy plant looks like and functions to be able to recognize when it is diseased (understanding at least basic anatomy and physiology), the characteristics of the organisms/factors that cause disease (fungi, bacteria, etc.) and the symptoms and signs associated with the major types of diseases. Signs are the visible physical presence of either the pathogen itself or the structures formed by the pathogen or signs defined as actual pathogen, parts or by-products seen on a diseased host plant Common examples of easily detected signs are those such as the fungal mycelia and spore masses of downy mildews observed on infected leaves and the bacterial ooze of Xanthomonas leaf streak disease on rice.

Symptoms - plant reactions or alterations of a plant's appearance due to a disease or disorder, i.e. the visible changes that occur in the host plant in response to infection by pathogens.Microscopes and stereoscopes are essential for the identification and

quantification of plant pathogens as well as for observing symptoms and lesions and evaluating with the objective of Diagnosing and identification of fungal mycoflora diseases on wheat seed varieties using compound microscope with different magnification power at Ambo Agricultural research center, west Shewa, Ethiopia.

METHODOLOGY/PROTOCOLS FOLLOWED

There are different methods for detecting Seed borne pathogens, which may include Visual examination, Agar Plate method, Standard blotter method, Examination of seed soaked in water, Examination of seed washing, Embryo count method, paper towel method, of dry seeds, Growing on test, Phage plaque method, PCR ELISA, Electron microscopy.

During the experiment among all I used Agar Plate method to test the availability seed borne pathogens. In the study area we were used materials are wheat seed Microscopes Simple microscope, Compound microscope, Autoclave, Pressure cooker, Hot air oven, Incubator, Inoculation chamber (Laminar flow chamber), Ultraviolet lamps, pH meter, Sensitive Balance, Haemo cytometer, Filter peppers, Para film, Refrigerator, Bunsen Burner.

Description of the Study Area

Evaluation of both in-vitro activities will be conducted at Ambo Agricultural Research Center, which is located at latitude of 8° 58' 36.5", longitude of 37° 50' 40.6", 115Kms west of from Addis Ababa and 2185 m above sea level. It is a center of excellence of Plant protection Research of the Ethiopian Institute of Agricultural Research (EIAR) with annual temperature is 110C-26oC with temperate agro-ecology. Major soil types consist of 67% clay, 18% silt, 15% sand and 1.5% organic matter. The mean annual rainfall is 1100 mm.

Media Preparation

41 g of PDA was added to distilled water (1 L) in a conical flask and magnetically stirred to dissolve while heating (30 min), autoclaved at 121oC (1 atm) for 15 minutes and cooled. The isolation room was first irradiated with UV for 30 min followed by sterilizationof medium observe the medium in conical flask and pour it on Petri dish forsolidification. After incubation period of 24-48 hrs for nutrient agar medium and 7 days for PDA.

RESULTS AND DISCUSSION

After Five days number of infected seeds were counted and expressed in percentage as mentioned in the following table.

Table 1. Wheat seeds infected by different fungal species in percentage

5th Days after Plates				
No.	Variety	Infected seed	Total Seed Plated	Infection Percentage
1	Bollo	5	10	50%
2	Tsehai	4	10	40%
3	Menze	8	10	80%

Table 2. Wheat seeds infected by different fungal species in percentage

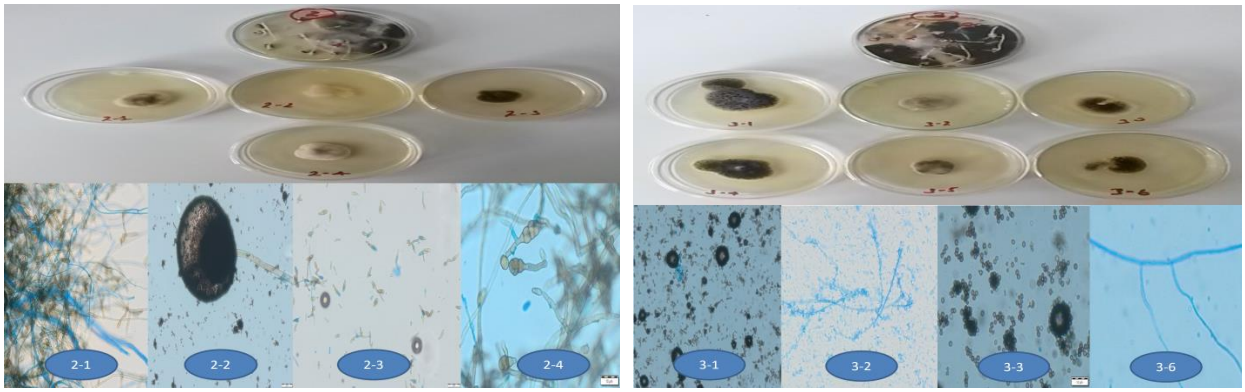
7th Days after Plates				
No.	Variety	Infected seed	Total Seed Plated	Infection Percentage
1	Bollo	7	10	70%
2	Tsehai	6	10	40%
3	Menze	9	10	80%

Table 3. Wheat seeds infected by different fungal species in percentage

9th Days after Plates				
No.	Variety	Infected seed	Total Seed Plated	Infection Percentage
1	Bollo	8	10	80%
2	Tsehai	9	10	90%
3	Menze	10	10	100%

The seed borne pathogens may cause Loss in germination, Discoloration and shriveling, Development of plant diseases of pathogen to new areas, Introduction of new strains or physiologic races of the pathogen along with new germplasm from other countries and toxin production in infected seed. The infestation/contamination of the seed may occur during harvesting, threshing and processing.

Pictures 1-1 of infected seeds were captured and illustrated as above Penicillium spices Pictures 1-5 of infected seeds were captured and illustrated as above unknown pathogen because can't have micro and macro spore. Pictures 1-3 of infected seeds were captured and illustrated as above unknown pathogen because can't have micro and macro spore.



Pictures 2-1 of infected seeds were captured and illustrated as above Wheat blast spices
Pictures 2-2 of infected seeds were captured and illustrated as above Wheat Aspergillus spices
Pictures 2-3 of infected seeds were captured and illustrated as above Fusarium Head Blight spices
Pictures 2-4 of infected seeds were captured and illustrated as above Alternaria leaf blight (*A. triticina spps*).
Pictures 3-1 of infected seeds were captured and illustrated as above Aspergillus spices
Pictures 3-2 of infected seeds were captured and illustrated as above Penicillium spices
Pictures 3-3 of infected seeds were captured and illustrated as above Aspergillus spices
Pictures 3-6 of infected seeds were captured and illustrated as above unknown fungal spices

CONCLUSION AND RECOMMENDATION

Seed health test work was conducted to determine presence or absence of seed born fungal pathogens. Seed health testing includes visual examination of seeds externally or internally, Seed washing and plating test, Blotter method, Agar plate method, Indicator test, Pathogenicity test. Among them Agar Plate method is the Simplest and inexpensive way of detecting seed borne pathogens and we were familiarized with the seed health test methods. The fungi found on the seeds were recorded and percentage of seeds infected and germination of seeds with different fungi was calculated for each pulse crops.

Citation: ShumiR and GuddissaH (2025); Diagnosing and identification of fungal mycoflora diseases on wheat seed varieties at Ambo Agricultural research center, Ambo, Ethiopia.

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