

A Review on Seed Borne Mycoflora Associated with Different Oilseed Crops and their Management

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ABSTRACT

The mycoflora associated with seeds at the stage of storage bring about several undesirable changes making them unfit for consumption and sowing. Further, association of mycoflora adversely affects quality and health of seeds. Many fungal and bacterial species have been detected in seed samples of oil seed crops. Fungi including Alternaria sp., Curvularia sp. Fusarium sp., Helminthosporium sp., Penicilium sp., Mommoniella sp., Aspergillus sp., Mucor sp. and Rhizopus sp. have been found associated with the oilseeds and among these, Alternaria sp. as well as Aspergillus sp. are the most destructive pathogen of oilseeds. Ninety percent of food crops are propagated by seed. A most widely grown crops in world agriculture (groundnut, sesame, mustard etc) are all affected by seed borne disease. The aim of this study is to identify those mycoflora associated with different oil seed crops and their management. There is a significant economic impact of seed borne diseases particularly in underdeveloped countries where routine chemical seed treatment of seed is prohibitively expensive, and individual farmers can suffer huge yield reduction.

Key words: Mycoflora, Seed treatment, Penicilium sp., Mommoniella sp., Aspergillus sp.

INTRODUCTION

Oilseeds are raised in almost all the parts of the country. Interestingly, in some regions of the country, they are considered as important oil-seeds. Oilseeds are the source of oil-cake as well as vegetable oil. However, the export of oilseeds has been curtailed to meet the increasing demands of the country. The oilseed accounts for 13% of the Gross Cropped Area, 3% of the Gross National Product and 10% value of all agricultural commodities. This sector has recorded annual growth rate of area, production and yield @

2.44%, 5.47% and 2.96% respectively during last decade (1999-2009). During the last few years, the domestic consumption of edible oils has increased substantially and has touched the level of 18.90 million tonnes in 2011-12 and is likely to increase further. With per capita consumption of vegetable oils at the rate of 16 kg/year/person for a projected population of 1276 million, the total vegetable oils demand is likely to touch 20.4 million tonnes by 2017. Groundnut is one of the main oilseeds of India. In fact, it is the leading producer in the world.

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Tropical climates are considered favourable for the growth of groundnuts. This oilseed is raised as a kharif crop. It is not grown mainly in the winter season. Growth of groundnuts is extremely high if sown in well-drained sandy loams. The main months when it is sown are June and July. Gujarat is the main producer of groundnut in India. Other important groundnuts states of the India are Andhra Pradesh, Maharashtra and Tamil Nadu. Mustard is another significant oil-seed in India. It is cultivated mainly in winter season. They are mainly cultivated as Rabi crop. They are grown as mixed crop with gram and wheat. Uttar Pradesh and Rajasthan are the principal producers in the country. The oil that is extracted from the mustard seeds are used for cooking in India. Other producers of oil seeds in India are Madhya Pradesh, Haryana, West

Bengal, Assam and Punjab. Sesame is one of the main oilseeds produced in the country. India generates almost one-third of the total production^{1,4}. Even the oil that is extracted from this seed is edible. Uttar Pradesh, Rajasthan, Madhya Pradesh, Orissa are the major producers of this oil-seed. Linseed is another popular oil-seed in India. Maharashtra, Bihar and Madhya Pradesh are the chief producers of linseed. Castor seed is another major oil-seed produced in India. This country produces more than one-fifth of the total production of castor seed in the world. Castor oil that is extracted from this seed is used as a lubricant, hair oil and is also used in manufacturing soaps. This oil-seed is generally cultivated in light soils and are grown as a Rabi crop. It is grown in Gujarat and Andhra Pradesh. Cotton seed is a major oil seed produced in the country. The oil extracted from these seeds is used in the manufacture of hydrogenated oil. India is of the biggest producers of this particular oil seed. Thus, it can be said that India is a huge producer of oil seeds and most of its states produce one form of oil seed or the other. Despite the rapid spread of the crop, a disheartening trend is that the productivity is going down in recent years. The full potential of the crop is so far from

being exploited and the yield levels of the country are the lowest in the world due to several biotic and abiotic factors. Among the several biotic limiting factors for successful oilseed production, susceptibility to disease is one of the major constraints. Several diseases are known to cause yield loss in the oilseed crops and many of these diseases are caused by seed borne mycoflora viz., *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus* sp., *Fusarium* sp., *Mucor* sp., *Alternaria* sp., *Colletotrichum* sp., *Macrophomina phaseolina*, *Penicillium* sp., *Botrytis* sp. etc. Among these, *Alternaria* sp., *Aspergillus niger*, *Aspergillus flavus* and etc. has been considered as a potentially destructive on many oilseed crops in different countries and. In India, it has been reported from many parts of the country including Andhra Pradesh, West Bengal etc. and is known to cause reduction in flower size, number of seeds per head, seed yield per plant, seed weight and also oil content⁵. The loss in yield varies from 25- 60 percent depending on the stage and the extent of infection⁶. Leaf spot or leaf blight a seed borne disease has shown variable trends over the years with huge loss in total production. Thus, it realizes the importance of healthy and viable seed, which is free from any type of fungal or bacterial infection. To increase the production of oilseeds qualitatively and quantitatively, farmer requires healthy quality seeds with high percentage of germination and purity. Hence, it is imperative that the seeds must be tested before they are sown in the field. Another adverse effect of seed borne pathogen is that it will contaminate the areas which were disease free previously. So, it necessitates the eradication of seed borne inoculum through various seed treatments and through the enforcement of proper domestic and international quarantine acts and procedures. Seed treatment is the oldest practice in plant protection and now, this is an attractive delivery system for either fungal or bacterial bio-protectants. Seed treatment with bio-protectants provides economical and relatively non polluting delivery systems for protective materials compared to the other

field application systems. However the uses and expectations of seed treatments with chemicals are greater today but due to the impact of environment regulations they have either banned or restricted the use of older chemicals like organo mercurial fungicides, because of their residual toxicity. Bio-protectants applied to seeds may not only protect the seeds but also may colonize and protect roots and increase the plant growth. However, biological agents have tended to be somewhat less effective and more variable than chemical seed treatments. There are stray reports in literature on seed borne mycoflora of various oilseed crops in West Bengal particularly in Birbhum district with reference to detection and identification methods and management practices against them.

Historical background of the pathogen:

Alternaria blight is a destructive disease of rapeseed-mustard throughout the world and appears each year in crop fields. The disease is caused by different species of *Alternaria* viz. *A. brassicae* (Berk) Sacc, *A. brassicicola* (Schw.), *A. raphani* Grows and Skolo and *A. alternata* (Fr) Keisskr. Among than *A. brassicae* is much more destructive and occurs more frequently than *A. brassicicola* on *B. juncea*⁷. The literature on the disease is voluminous, however, in the present review, only those aspects which have a direct bearing on the investigation, have been included. The genus *Alternaria* was first described by Nees in 1817 with type species *A. tenuis*; which was later renamed as *A. alternata*. Berkeley, noticed fungal infection on plant belonging to the family Brassicaceae and indentified the fungus as *Macrosporium brassicae* (Berk) which was later renamed as *A. brassicae* (Berk) Sacc by Saccardo in 1886. In India, *Alternaria* blight was first observed on sarson from Tirhoot in 1901 but the fungus was thought to be new and described as *Sporodochium brassicae*. Later Mason, first observed the *Alternaria* species from a herbarium material of sarson from Pusa (Bihar) India. Leaf spot of safflower caused by *A. carthami* Chowdhury is common in all the safflower growing regions of the world. The

disease was reported from India by Chowdhury, and subsequently from erstwhile USSR, Ethiopia and Kenya, Africa, Australia, Pakistan, and Italy. More than 15 per cent yield loss of safflower was caused by *Alternaria* spp. in USA⁸. In India it is the major destructive disease of safflower and estimated to be causing 25-60 per cent yield loss every year⁹. Preliminary surveys on the intensity of *Alternaria* leaf blight in northern India revealed 27-90 per cent yield loss when the disease appeared at early stages of crop growth¹⁰, Siddaramaiah and Hedge and Mahabaleswarappa, reported the severity of leaf blight of safflower in the state of Karnataka. Severe leaf blight leading to blighting of leaves was documented in Orissa¹¹. Patil and Jadav and Indi *et al.*, The economic losses caused due to *A. carthami* in Maharashtra region¹². The occurrence of *Alternaria* leaf blight leading to considerable economic losses in Madhya Pradesh¹³. In India, linseed powdery mildew due to *Oidium lini* was first reported by Padwick. Powdery mildew of flax caused by *Oidium lini* Skorik was first recorded from Turkey¹⁴.

Mechanism of seed infection by fungi:

Fungal infection of seed borne pathogens may reach the ovule of the seed at any stage from the initiation of ovule formation to the mature seed. The fungal plant pathogens vary in their modes of multiplication and attack on the host plant^{15,16,17,18} fungal propagules/spores germinate and the growing hyphae determine the entry of the pathogens in plant tissue including the fruit and seed. The ovule and the seed develop in the pistil which is enclosed by other floral appendages in the flower and seed. The position and structure of seed, including the physical environment during seed development, determines the successful seed infection. The physiological and biochemical factors inside the fruit and seed further control the establishment of successful infection. Fungi may be biotroph (obligate parasite) or necrotroph (saprophytic). Biotroph cause minimal damage to the host seed, have a narrow host range, while necrotrophs cause apparent damage to the host cell and have a

wide host range. The fungi, depending upon the time of infection and environment condition cause superficial or deep infection. Biotrophs usually establish deeper into the tissues including embryo. Necrotroph which degrade tissues through their enzymatic activity, as they spread, are rarely transmitted to the embryo through the mother plant. The mechanism of infection of the ovule and seed is also dependent upon the nature of disease in the plant and the mechanism of transmission of infection into the seed^{19,22}.

Mechanism of seed infection by bacteria:

The important bacterial species that cause seed infection are *Xanthomonas Pseudomonas*, *Acidovorax*, *Burkholderia*, *Rathayibactor*, *Clavibacter*, *Curtobacterium* and *Pontoea*. The bacteria may cause seed infection, i.e., carried on the surface of the seed or seed infection, that occurs in the seed coat and other parts of the seed. In both the cases, there is either failure in seed germination or the appearance of disease symptoms in seedlings and plants. Unlike fungi, bacteria lack mechanism for forcing their way physically

through protective barriers, such as cuticle, epidermis and bark. The developing ovules and seeds occupy a specific position in angiosperms. In order to reach the ovule or seed of a plant, the infection must find passages for invasion of the ovary or developing fruits. Bacterial diseases of seeding may be established either from seed-borne infection or may take place through soil, air, water, insects, and nematodes. The entry of phytopathogenic bacteria may be passive and occur through natural openings (stomata, lenticels, hydathodes, or nectariferous surfaces) or through wounds including scars left by dropping of hairs, cracks²³.

Important seed borne disease of oilseed crops:

Any infectious agent associated with the seed, having the potential of causing a disease of a seedling or a plant, is termed as seed borne pathogens. In this case seed may or may not exhibit disease symptoms. This term includes all plant pathogenic microorganisms (fungi, bacteria, nematode) and the viruses which are carried in, on or with the seed²⁴.

Crop	Diseases	Pathogens
Groundnut	Collor rot Seed rot	<i>Macrophomina phaseolina</i> . <i>Aspergillus flavus</i> and <i>A.niger</i>
Sunflower	downy mildew white rot charcoal rot	<i>Plasmopara halstedii</i> <i>Sclerotinia spermophila</i> <i>Rhizoctonia bataticola</i>
Safflower	Rust	<i>P.carthami</i>
Rape seed mustard	Alternaria leaf and pod spot Downy mildew	<i>Alternaria brassicae</i> and <i>A.brassicicola</i> <i>Peronospora parasitica</i> .
Linseed	Foot rot stem canker	<i>Septoria linicola</i> <i>Colletotrichum lini</i>
Sesame	leaf blight sesame blight leaf blotch	<i>Alternaria sesamicola</i> <i>Corynespora cassiicola</i> <i>Drechslera sesame</i>
Flax	Steam canker Anthracnose	<i>Aureobasidium lini</i> <i>Colletotrichum lini</i>

MATERIAL AND METHODS

Various microorganisms have been reported as causative agent of seed borne disease. Fungi and bacteria are most important pathogens among them. Scientists are able to discover methods to detect and identified. There are various techniques to detect the presence of pathogens associated with infection of seeds. Methods are described as follows –

Collection of various oilseeds samples:

For studying mycoflora associated with oilseeds (sesame, mustard, nut) were collected from various region of oil seeds grower. The collected seed samples were shade dried and stored in paper bags at room temperature for further studies. All the seed samples were examined by visual seed inspections and

occurrence of seed mycoflora was analysed by following methods:

Standard blotter method:

Three pieces of filter paper^{25,26}, were properly soaked in sterilized water and were placed at the bottom of a 9 cm well labelled plastic Petri dishes. Twenty (20) seeds per Petri dish were placed using a pair of forceps and making sure that seeds are placed equidistantly under aseptic conditions. The lids of each Petri dish were held in place with gummy cello tape. The Petri dishes containing seeds were incubated at room temperature ($25^{\circ} \pm 2^{\circ}\text{C}$) for 7 days under alternating cycles of light and darkness of 12 hours each.

Agar plate method:

PDA was prepared and sterilized in an autoclave. In agar plate method²⁷, 20 ml of potato dextrose agar was distributed to each of the sterile Petri plates under aseptic conditions. After cooling, crop seed samples were transferred²⁸. to the plate containing PDA medium. Twenty seeds per Petri plate were kept at equidistance in a circle and incubated at room temperature ($25^{\circ} \pm 2^{\circ}\text{C}$) under 12 hours alternating cycles of light and darkness for 7 days and observed everyday for the growth of fungi²⁹. The per cent seed mycoflora and percentage frequency of various fungal species were calculated. The incidence of fungi on seed performed under these methods were calculated as follows:

$$\% \text{ incidence} = \frac{\text{Number of infected seeds}}{\text{Number of plated seeds}} \times 100$$

Rolled towel method: The method developed by Warham, was followed. Germinability of the seed was determined in the laboratory at room temperature ($30^{\circ} \pm 2^{\circ}\text{C}$). A total of 200 seeds were randomly taken from each variety and 50 seeds were placed between a pair of moist paper towels³⁰. Four replications were used for each variety. The towels were rolled and the ends were closed by robber band and covered by polyethylene paper to prevent drying. First observation was taken after 5 days and final count was taken after 14 days of incubation period pertaining to (a) % germination, (b) non germinated seed (hard seed and rotten seed), (c) shoot length and, (d) root length. For determination of seed mycoflora the fungal growth on infected seed were taken with the needle and observed under compound microscope. For determination of seedling vigor ten seedlings (normal/abnormal) was randomly selected from each paper and their individual shoot and root length was measured³¹.

Deep freeze method:

This method was developed to detect slow growing pathogens. Three hundred seeds of moderately infected were placed at the rate of 20 seeds per plate on moistened blotters in the way as described under Standard blotter method. The Petri plates were incubated at $20^{\circ} \pm 2^{\circ}\text{C}$ for 24 h under alternate cycles of 12 h NUV light and darkness, for next 24 hours the plates are incubated at -20°C in dark and then kept back under original conditions for next five days. After eight days of incubation, the seeds were examined under stereobinocular microscope³². For the surface sterilization the seed in all methods were sterilized by 0.1% mercuric chloride solution to 1 to 2 min then washed by sterilized water³³. The frequency of the fungus was calculated by the following formula:

$$\frac{\text{No. of seeds containing a particular fungus}}{\text{Total seeds used}} \times 100$$

Identification of seed mycoflora:

The identification of fungi was done based on the morphological and colony characters of the

pathogens. Ten fungi were noticed on the oilseed samples collected from the different oilseed growing areas of Birbhum district,

West Bengal. *Aspergillus niger*, *Aspergillus flavus* often appeared in many samples along with species of *Rhizopus* sp, *Fusarium* sp and *Mucor* sp *Alternaria* sp were found mostly in the seed samples of safflower and linseed. Spore morphology and colony characters are given below.

***Aspergillus niger*.**

Colony of *Aspergillus niger* on seed grows slowly, consisting of a compact to fairly loose white to faintly yellow basal mycelium, which bears abundant erect and initially crowded conidial structures. Conidial heads are typically large and black, compact at first, spherical or split into two or looser to reasonably well defined columns. Conidiophores are smooth, hyaline or faintly brownish near the apex. Two series of conidia bearing the cells (supporting cells and phialides) are produced but in some heads only phialides are present. Conidia are typically spherical at maturity. Often very rough or spiny, mostly 4-5µm diameter and very dark in colour or with conspicuous longitudinal striations.

***Aspergillus flavus*.**

Colony of *Aspergillus flavus* on seed is usually spreading and very light. Yellow to deep yellow-green, olive-brown. Conidiophores are swollen apically with numerous conidia bearing cells in long chains. Conidiophores are heavily walled, hyaline coarsely and rough end. They can be one or two series of conidia bearing cells (Phialides and supporting cells). Conidia are typically spherical to sub-spherical, conspicuously spiny, variably 3-6 µm in diameter.

***Fusarium* sp.**

The fungus produces abundant loose, aerial white mycelium on incubated seed. In this mycelium several shiny, hyaline, transparent to milky white spherical droplets are seen hanging at the tip of long thin stalks. These stalks are primary conidiophores, which arise laterally from the hyphae in the aerial mycelium. The hanging droplets are most false heads in which conidia are produced. Microconidiophores (microphilides) that bear the microconidia are very long and slender and

measures 15-40 x 2-3 µm, where as those bearing macroconidia (macro-conidiophores) are short and measure 10-25 x 3-4.5 µm. Microconidia are hyaline, 1-2 septate oval, ellipsoidal to sub- cylindrical and measure 5-20 x 2.8-7 µm. Macroconidia are hyaline, stout, measured 22-75 x 35- 7 µm, subcylindrical (or) slightly curved, with short blunt and rounded apical cells and indistinctly conidia are thick, with dorsal and ventral surfaces parallel to most their length. They are mostly 3 septate but 4-7 septate conidia are common chlamydospores are formed singly and in pairs or in clusters in sporocidia. They are glubose to subglubose, smooth (or) rough-walled and 6-11 µm in diameter.

***Rhizopus* sp.**

Colony of *Rhizopus* sp on seed grows slowly, consisting of a compact to fairly loose pink mycelium, sporangiophores grow in cluster they are stout and stiff, the characteristic features is rhizoids present at the base of the sporangium, vegetative body is highly branched coenocytic in nature. The sporangium contain either one or both type of sporangiospore.

***Trichoderma* sp.**

Colony of *Trichoderma* sp that are grown from seeds sample on media are fast growing, conidia typically form within one week in compact or loose tufts in shade of green or yellow or less frequently white. A yellow pigment may be secreted in to the agar, especially on PDA. Some species produce a character *Bacillus* sp. rustic sweet or coconut odour. Conidiophores are highly branched and the difficult to define or measure loosely or compactly tufted, often formed in distinct concentric rings or borne along the aerial hyphae. Main branches of the conidiophores produced lateral side branches that may be paired or not. Conidia typically dry but in some species they may be held in drops of clear green or yellow liquid.

***Pythium* sp.**

Pythium sp. is a genus of parasitic oomycotes. Most species are plant parasites. *Pythium* spp like some other fungi in this family are usually characterized by their production of

coenocytic hyphae without septations. They generally contain a single oospore. They contain an elongated and club-shaped antheridium.

Bacillus sp.

Rod-shaped, endospore forming bacteria. Aerobic or facultatively anaerobic. Irregular and large in shape, undulated margin, elevation umbonated, white and dull colour, dry or rough textured.

Penicillium sp.

Penicillium spp are commonly known as contaminants. The colonies of *Penicillium* sp are rapid growing, flat, filamentous and velvety, woolly or cottony in structure. The colonies are initially white and become blue green, gray green, olive gray, yellow or pinkish in time. Colonies of *Penicillium* sp are often dominated by copious clear to yellow or brown exudates at the centres. Hyphae are septate, hyaline measuring 1.5 to 5 µm in diameter with simple or branched conidiophores. Metulae are secondary branches that form on conidiophores. The metulae carries the flask shaped phialides. The organisation of the phialides and the conidiophores are very typical. They form brush like clusters, which are also referred as “Penicilli”. The conidia (2.5 – 5 µm in diameter) are round, unicellular and visualized as unbranching chains at the tip of phialides.

Macrophomina sp.

Conidia are aseptate, hyaline, ellipsoidal to obovoid. Pycnidia are larger than sclerotia, dark brown to black and scattered throughout the surface. Hyphae are thick, grey to brown to black or dull white to light brown. Produces a profuse aerial mycelium with pycnidia and sclerotia. Sclerotia are black, shiny, and irregular shaped. Pycnidia are larger than sclerotia, dark brown to black and scattered throughout the surface.

Pseudomonas sp.

Aerobic bacilli forming bacteria. Motility is by a single polar flagellum. Oval and medium in shape with wavy margin, umbonated, diffusible green colour, mucoid textured with fruity odor and usually gram positive.

Status and Economic importance of the disease :

Alternaria blight of rapeseed-mustard had been reported from many countries viz. India, Italy, USA, UK and several other European countries, Canada, Iran etc³⁴. Besides India, it is a significant disease in Australia, Africa, England, Germany, France, Sri Lanka, Spain and Sweden. Among several fungal diseases severe damage or the seed germination in crucifers due to Alternaria blight caused by *A. brassicae* (Berk). The losses caused by Alternaria blight pathogens were up to 4.8 per cent in oil content in and losses ranged from 15.0 to 71.0 per cent due to Alternaria blight in India³⁵. Kolte, reported that the disease caused an average yield loss of 16-47% in yellow sarson and 35-38% in mustard. Mc Donald and Ingram, and Bains and Tewari, reported that the seed production of Brassica had been greatly reduced to 30.0-40.0% by the attack of this disease which invaded siliqua and penetrated the seed besides damaging the assimilating tissues of the leaves and stem. The loss in oil content of seeds from heavily infected rapeseed-mustard brassicae ranged from 14.58 to 35.97 per cent depending on the cultivars particularly in *B. juncea* it ranged from 14.12 to 29.07 per cent in India³⁶. The yield losses ranged from 20.0 to 30.0 per cent in Canada. Hong and Fitt, reported that Alternaria blight resulted in yield losses up to 71.3 per cent in Brassica crops. The disease may be caused by *A. Brassicae* and *A. brassicicola* singly or concordantly³⁷. They further reported 26.5% infection by *A. brassicicola* and 22.6% by *A. brassicae* whereas rest 50.9% was accounted for concomitant infection of the two species. Kumar and Kolte, observed that Alternaria blight was one of the important disease of Brassica responsible for average yield loss of 10.0 to 70.0 per cent in different parts of the Northern India depending upon the severity. Kolte, reported that Alternaria blight disease caused by *A. brassicae* (Berk) Sacc. had been reported from all the continents of the world and was one of the important disease of Indian mustard causing up to 60 per cent yield losses.

The *Alternaria* blight occurred frequently in Nepal with average yield losses in the range of 3.2-5.7 per cent in yield and 14.6 to 36.0 per cent in oil content. Due to lack of availability of the sources of resistance against brassicae within the family Brassicaceae. *Alternaria* blight was considered the most damaging and widespread fungal disease of Brassica in India³⁸. Mondal reported yield losses upto 10.0 to 40.0 per cent every year by *Alternaria* blight caused by *A. brassicae* in old alluvial zone of northern part of West Bengal. Chattopadhyay reported that the *Alternaria* blight caused by *A. brassicae* was the most destructive disease of Indian mustard which caused up to 47.0 per cent yield loss. Sharma et al, reported that *B. juncea* along with *B. oleracea* are two important crucifer's crops of India which are facing serious yield and quality loss in production due to *A. brassicae* (Berk) Sacc. Yield losses 63% due to powdery mildew in linseed were estimated by Sharma and Khosla.

Management of Seed Borne Disease by various methods:

Seed treatment, in broad term, is the application of physical, chemical, biological, or organic means to provide protection to seeds and subsequent sprouts to improve the establishment of healthy crops.

Physical Seed Treatment:

Physical seed treatment usually refers to apply the heat for seed treatment in order to kill the seedborne pathogens with minimal damage to the seed tissues.

A) Hot water treatment:

Hot water treatment has been used since 1920's and before the advent of systemic fungicides in the 1960's, was the only treatment available to eradicate deep-seated infections of seeds. Bacterial blight of cluster bean (*X. campestris* pv. *cyamopsidis*) the seeds are kept at 56° C for 10 min; and bacterial blight of sesame (*X. campestris* pv. *sesami*) at 52° C for 10 min. by different workers. Hot vegetable oil treatment has also been explored for control of *Phomopsis longicolla* infection in soybean seeds, but causes adverse effect on seed germination³⁹.

B) Dry Heat treatment:

Treatment of seeds with dry heat is found partially effective and has been little used in practice. The dry heat treatment has been found to eliminate various seed borne mycoflora. Microwave heating also reduced transmission of soybean mosaic virus, with little reduction in germination in seeds treated at 8.5% moisture content, but germination was considerably reduced in seeds treated at 16% moisture content⁴⁰.

C) Radio-frequency heat treatment:

Exposure oil seeds to radio-frequency heat treatment at various temperatures levels resulted in the reduction of seed-borne fungus *Alternaria padwickii* to varying percentages. Other fungi such as *Fusarium* sp. *Curvularia lumata*, and *Trichoderma* sp have also been found to be partially controlled. However, this type of seed treatment decreased seed qualities, viability, moisture content and vigor by accelerated aging by increasing the temperatures⁴¹.

Biological Seed Treatment :

In the narrowest sense, biological control involves suppressing pests or organism with other organism. Biological control agents used as seed treatments are microorganisms that protect seeds and seedlings from various pathogens. Pursuit of alternatives to chemical pesticides and an increasing interest in organic crop production technologies have stimulated increased scientific development of biological control agents over the past 20 years. Advances have been achieved over this period through a greater understanding of the control mechanisms used by these agents especially in the soil. Some limited progress is noticed in the technology formulation due to certain difficulties with these agents such as, storage stability, shelf life of products, and an erratic biological efficacy after application to the seed, have slowed the growth and adoption of the technology. In addition, limited understanding of the *rhizosphere* ecology, the release of some products which did not meet performance expectations, the uncertainty of the size of the commercial opportunity, the strength of patents and the cost of registration,

have hindered the development of their market. Of the biological control agents patented by early 1999. 84 percent were bacteria and 16 percent were fungi. The bacteria included species of *Streptomyces*, *Pseudomonas*, *Bacillus* and *Enterobacter*. Species of *Pseudomonas* and *Bacillus* made up the vast majority of those products. Fungal products consisted of various species of *Phomopsis*, *Ectomycorrhiza* *Gliocladium* *Trichoderma* *Cladosporium* and *Gliocladium et al*⁴².

Chemical Seed Treatment:

Fungicides:

Fungicides dominate the market with 68% share followed by insecticides at 11%. The range of products on world markets today are quite different from those in the early part of the century when organo-mercurials, dithiocarbamates (thiram) and heterocyclic compounds (captan) acted by direct contact with the pathogens were the primary products used in seed treatment. Organo-mercurials are now banned in most countries because of toxicity to animals and humans. The use of captan and thiram has been restricted in some countries nevertheless, the latter materials, however, are the main stay of seed treatment chemistry for many crop species. They have a broad spectrum of activity, are easily applied, and are relatively inexpensive. On a worldwide basis, systemic products that contain carboxin or triadimenol occupy 40% of the fungicide seed treatment market. Oxine-Cu, pencycuron, captan, benomyl, TBZ and mercury hold 30% of the market, and another 51 products constitute the remaining 30. The change in the landscape of fungicide seed treatment chemistry can be attributed to the need to replace of older chemistry because of environmental concern and the discovery of systemic compounds that provided new opportunities to control foliar and systemic pathogens by seed treatment. metalaxyl, which is effective against Phycomycetes such as *Phytophthora* and downy mildews; and ethirimol and triadimenol for control of the powdery mildews. Other system chemistry that has potential for use seed treatments includes iprodione and imazalil^{43,44}.

In-vivo studies of Fungicides, Botanicals, and Bio- control agents:

Evaluation of Fungicides against mycoflora:

Basavarajaiah *et al*, evaluated efficacy of eight fungicides against *A. carthami* *in vitro* following poisoned food technique and found complete inhibition of fungal growth with thiram 500 $\mu\text{g ml}^{-1}$ and triphenyl tris hydroxide 500 $\mu\text{g ml}^{-1}$. Siddaramaiah *et al*, tested nine fungicides for eradication of *A. carthami* from heavily infected safflower seeds and found that seed treatment with captan and RH 2161 was highly effective in eradicating the test pathogen completely. Ayyavoo and Shanmugam evaluated five fungicides against *A. carthami* and it was observed that the treatment with dithane Z-78 (0.1 per cent) was able to reduce the incidence of the disease by 16 per cent over others. Srinivas *et al*. reported that spraying carbendazim was the most effective in controlling blight of sunflower caused by *A. alternata*. Krishna *et al*. evaluated six fungicides at seven different concentrations *in vitro* for their efficacy against *A. carthami* following poison food technique and the results indicated that Aureo-fungin was found most effective which carbendazim was less effective in inhibiting the test pathogen. Shivankar *et al*., reported that the seed treatment with carbendazim at 0.1 per cent recorded the highest germination shoot length (9.37 cm) in *A. alternata* infected wheat seeds. In *in vitro* evaluation of eight fungicides against *A. alternata* causing leaf blight of turmeric, propiconazole was found to be superior in inhibiting the growth of the fungus while ziram a non systemic fungitoxicant found to be the best in inhibiting the growth of fungus. The other isolates were sensitive to both fungicides with LD50 values similar or lower than those presented by *A. solani*. These results suggested that successful integrated control programme could be implemented when *Chaetomium globosum* is used in combination with Dithane M 45 and when *Cladosporium cladosporioides* and *Rhodotorula* were used in combination with

daconil. Wadibhasme *et al.*, tested six non-systemic and three systemic fungicides *in vitro* against *A. helianthi* by poisoned food technique. They found that dithane M-45 was the most effective followed by fytalon and dithane Z-78.

Evaluation of Bio-agents against mycoflora:

The uses and expectations of biological seed treatments are greater today due to the impact of environmental regulations that have either banned or restricted the use of oiler seed dressing fungicides such as organomercurial compounds. Biological seed treatment provides economical and relatively non-polluting delivery systems for protective materials as compared to other field application systems. Bioprotectants like *T. viride*, *T. harzianum* and *Pseudomonas fluorescens* applied to seeds may not only protect seeds but also may colonize and protect roots and increase the plant growth. Leifert *et al.* reported that *Serratia* sp. and *Pseudomonas* sp. showed antagonism against *B. cinerea* and *A. brassicicola* *in vitro* they were also tested *in vivo* and certain isolates of *Pseudomonas* CL42, 66, 82 were also proved to be effective *in vitro*. Seed treatment or spraying with spore suspensions of *T. viride* on growing plants controlled *Alternaria linicola* on linseed. Sawant *et al.* reported that combination of seed treatment and soil application with *T. viride* was more effective in reducing the incidence of early blight of tomato caused by *A. solani*. Babu *et al.* tested certain fungal antagonists *in vitro* against growth of *A. solani* causal agent of leaf blight of tomato and the results showed that *T. harzianum* followed by *T. viride* were significantly effective in inhibiting mycelial growth of test pathogen. Seed treatment with *T. viride* eliminated seed borne infection of *A. alternata*, *R. bataticola*, *R. solani* and *C. lunata* associated with pigeon pea by with significant tease in seed germination, vigor index and fresh weight of seedling over untreated control. Gaikwad and Behere evaluated seed treatment with *T. harzianum* and *A. fumigatus* as biocontrol agents @ 8.72×10^7 spores ml^{-1} and 9.26×10^7 spore ml^{-1}

respectively against *F. oxysporum* fsp. *carthami* and found reduction of disease incidence by 100 per cent on susceptible safflower cv. Tara under glass house conditions. Rajeswari *et al.*, studied the efficacy of seed treatments with bioagents and fungicides against seed mycoflora of safflower were evaluated and the results indicated that seed treatments will *T. viride* (6 g kg^{-1}), mancozeb (2.5 g kg^{-1}) and neem oil (10 ml kg^{-1}) not only enhanced seed quality and but also effective in reducing total seed mycoflora and seedling mortality.

Evaluation of plant-extract against mycoflora:

Biological screening of higher plants has shown that many of these plants contain highly potent inhibitors of plant pathogens. Some of these inhibitors provide complete protection against the diseases and in many cases the antipathogenic activity was obtained with crude extracts. The nature of inhibitors characterized from higher plants was found to be different. Annapurna *et al.* found that aqueous leaf and fruit extracts of neem was found to be effective in inhibiting the growth of *A. padwickii* in rice seeds. Neem oil was also effective in checking the growth of *A. alternata* causing post harvest rotting of tomato. Sundriyal reported that floral extracts of *Lantana camera* inhibited spore germination and germ tube growth of *A. solani* *in vitro* while conidial germination was completely inhibited after five hours exposure. Lal *et al.* tested various plant extracts against *A. alternata* and observed that extracts of *Achyranthus* sp. was found to be most effective in inhibiting the mycelial growth by 61.9 per cent followed by *Azadiracta indica*. Sheno *et al.* reported that *Pongamia glabra* extract was effective against *A. alternata* causal agent of tobacco. Spore germination, mycelial growth and sporulation of *A. helianthi* were inhibited by *P. glabra* extract *in vitro*. Extracts of turmeric rhizome was found to be inhibitory to the growth of *A. alternata*. Amaresh reported that, among plant extracts tested neem leaf extract (5 per cent), *Ocimum canum* leaf extract (5 per cent) and

Bougainvillea leaf extracts (5 per cent) were found to be effective in controlling *Alternaria* blight. Meena *et al.*, and Yadav, advocated that foliar sprays of aqueous bulb extract of *Allium sativum* (garlic) @ 1% and leaf extract (1.5 to 2.0%) of *Azadirachta indica* (neem) and *Eucalyptus globules* had been reported to effectively manage *Alternaria* blight on leaves and pods and could be eco-friendly substitute for chemical fungicide mancozeb in management of mustard diseases.

Purpose and significance of seed treatment:

Seed treatment is an age old practice for plant protection. It has enormous benefits in comparison with traditional crop protection methods. It not only improves the seed quality and manages the seed borne pathogens, but also improves stand quality. protects seed from seed and soil borne pathogens, improves seed shape for planting. increase yield and return on investment, improve seed storability, helps in fixing nitrogen and enhances uptake of nutrients. Seed treatment also reduces active ingredient loading into the environment and decreases the effect on non target organisms and environment safety. Using a seed treatment reduces the area in contact with a crop protection product from 10,000m² for foliar application or 500m for furrow application to only 50m². It is important to go for seed treatment when field is for seed production; low test eight or older seed; planting in unfavourable germination conditions (dry soil or cold soil); planting into fields with a history of stand establishment problems: planting to precise populations; replanting will not be feasible if first planting fails, seed is expensive, seed is infected or infested, and yield potential of field is high. Seed treatment helps in seed disinfestations by killing of spores, mycelia, or propagules of microorganisms on seed surface or in seed disinfection by eliminating the pathogen that has penetrated deep into living cells of seed (e.g. smut or bunt), or protect the seed from soil borne pathogens (damping-off). A systemic fungicide may provide post-emergence protection.

Discussion and future aspect:

Present study showed that there was large variation in the diversity and severity of fungal contamination to seed-borne fungi in seed samples. Among seed-borne fungi, *Aspergillus niger* and *Aspergillus flavus* had the most severity rate compared to others. Scientific research and inventions have always been the thrust of mankind and is largely responsible for the standard of living he has today. Natural resources of a country are of primary importance for the economic development. From present review we are able to identify and control seed borne pathogenic fungi and bacteria still now science is unable to identify those pathogenic microorganisms like bacteria, virus etc. So more research work is needed to detect and control them. If we can detect them, then it will be easier to decrease infections caused by them. At last but not least people should remain conscious or alert about this and they should have enough knowledge about this infection.

CONCLUSION

During the past few years, pathogens in oilseed crops have been recognized as major forces causing economic losses, with identification of certain important ones based on their symptoms, etiology and also ecological zones. Recent research has helped by developing new resistant varieties and other effective management strategies. This review describes the causal organisms, symptoms and management of diseases of oilseed crops like castor, groundnut, safflower, sesame and sunflower. Cultural practices for managing certain diseases have been pinpointed. Critical stages for growth of some foliar diseases, namely rust, early and late leaf spot of groundnut, blight and mildew of sunflower, fusarial wilt of safflower and castor, have been identified. Recommendations are given on controlling various diseases by chemical, botanical and other effective and eco-friendly methods. Oil is an essential household commodity required for food and daily use. Certain oils are used as therapeutic agents and are in much demand for their conversion into energy or potential biodiesel. Losses to the

tune of 20% in certain oilseed crops need our utmost attention. Various fungal diseases of groundnut, sesame and castor are described. Disease management with fungicides and other available methods are illustrated. . Institutions have to cooperate with researchers and scientists to carry on this type of research work. Then we will be able to identify those mycoflora and control them.

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