

A Study on *Mycoflora* of Cumin Seeds

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ABSTRACT

For the purpose of performing the study, cumin seeds were obtained from several sources. From tested cumin seeds, several fungus species were recovered. In seed-borne fungus, the results showed suppressive of *Fusarium* spp. and *Aspergillus*. The majority of storage fungi that attack stored seeds have caused a drop in the germination rate. When seed-borne mycoflora are present in cumin seeds, transformation occurs, output is reduced, there is a bad smell, and the seeds become hydrolytically rancid. Other metabolic alterations brought on by a seed mycoflora attack are also noticeable during respiration.

KEYWORDS: *Cumin (Cuminum cyminum); Mycoflora · seed-borne fungi*

INTRODUCTION

A member of the *Apiaceae* family, cumin is a perennial plant crop with Egyptian origins. *Cuminum cyminum* L. As a crop grown during the winter, it is widely cultivated. The vegetation is either seasonal or perennial herbs, and is a plant that is grown around the world for the purpose of recovering its essential oil (Nasir & Ali, 1972). According to Peter and Nybe (2002), the primary countries where it is grown include India, Egypt, Libya, Iran, Pakistan, and Mexico. When it comes to seasoning dishes, cumin oil is very useful in curries and other Asian-inspired dishes. In some cases, it is also used to flavour beverages and make soaps fragrant. Only trace amounts of cumin aldehyde, which has a powerful aroma and is employed in the creation of synthetic floral fragrances like cassie, are used.

Cuminum cyminum L. is utilised as an antibacterial, antispasmodic, stimulant, and preservative (Romeilan, 2010). primarily utilised in pickles, chutney, pickle powder, and seasoning (Farell, 1985). Although spices are typically resistant to microbial deterioration, toxic moulds like *Aspergillus* sp., *Penicillium* sp., and *Fusarium* sp. may grow on cumin seeds if the moisture content is too high (Abou-donia, 2008). Fungi are normal component of food mycoflora and cause spoilage and mycotoxin production.

It is well known fact that several fungi causes considerable damage to spices under storage condition, and also decrease in germination ability, discolouration of parts, loss in weight and production of toxin and it depends upon the type of fungi present, the composition of food, storage and handling (Mandeel, 2005). The most frequent fungal pathogen of cumin seed is from genera *Aspergillus* and *Penicillium* (Koci et al., 2007).

A thorough and in-depth survey of the fungus from *Cuminum cyminum* L. was conducted in the current investigation. The current research was done using cumin seeds that were gathered from several cumin growing belts of Jalore districts in Rajasthan, India's between August 2016 and March 2017

MATERIALS AND METHODS

The *Cuminum cyminum* L. (Cumin) seeds utilised in this study came from local farmers in a number of villages in the Jalore District and were purchased from several marketplaces in both pack and loose form. In order to transfer the various samples to the lab, they were all placed in clear polythene bags. According to Essono et al. (2007), the moisture content is quantified and the mean moisture percentage is computed.

Isolation of different fungal was made from the collected samples of these medicinal plants, by following the Standard Blotter Technique as well as by Agar Plate Method as recommended by ISTA (1966). Physical examination of each seed sample was critically done to look for the presence and absence of mycelia mats, damage, cracks, discolouration, galls, sclerotia and general health condition of the seed sample. For analysis of the samples, for the fungi present on seed surface, 100 apparently healthy looking seeds from each sample were randomly sorted out and were placed in Petri dishes containing PDA medium and also on moist blotting papers. 10 g of abnormal looking seeds were taken in conical flask containing 100 ml of sterilized distilled water. The flask was subsequently shaken for 15 min in an electric shaker. After sometime the water from each flask was decanted and centrifuged for 15 min. The deposited sediment was divided in to 2 parts; one part of it was examined with a drop of distilled water

under microscope for the presence of spores and mycelia fragments. To the other part of the sediment was added 1 ml sterilized distilled water and placed on PDA medium to find out the viability of the mycelia fragments and spores.

For studying the *endophytically carried seed fungi*, 100 seeds from each sample were surface sterilized for 1 min in 0.1 % HgCl₂ solution. They were then rinsed with sterilized distilled water. The seeds were then placed on three layered moistened blotter paper and on agar plate, 10 seeds per plate. The platings were performed in sterilized chamber to avoid the chances of external contamination. Germination was ascertained by counting the number of germinated seeds after 5 days of incubation. Fungal growth from the seeds was examined under stereo-binocular. The fungi were isolated, purified, sub-cultured, cultivated and maintained on PDA medium for further studies.

Experiments were conducted to ascertain the exact location of pathogen in different parts and to determine the site of infection of the storage as well as field fungi by the method of Vidhyasekharan et al. (1970) and Srivastava and Gupta (1984). Freshly harvested healthy seeds were taken and were surface sterilized with 0.1% HgCl₂ solution and then washed with several changes of sterilized water. The seeds were divided into different lots. They were thoroughly mixed with 1 week old cultures of different fungi and one lot was kept as control. Then the seeds were thoroughly dried under shade to remove excess moisture. The seeds were placed inside the plastic cups which were already sterilized with rectified spirit and the mouths of the cups were closed with a cloth to prevent contamination. The cups were placed inside a desiccators maintained at 75% RH with a saturated solution of Sodium Chloride. After 30 days of storage, the seed samples were drawn and component plating was done to locate the seed-borne fungi led in various parts of the seed. 50 seeds in each treatment were allowed to swell in sterilized water for 2-4 hour and then each seed was dissected aseptically with a sterile scalpel to separate the seed coat, and embryo. These parts were then plated in agar medium with needles and forceps. The corresponding parts from the untreated were also plated and kept as control. After 5 days of incubation at 25±2° C under the alternating condition of 12 hr light and dark period, the development of incubated seed-borne fungi in the different parts of the seeds was observed.

Aspergillus flavus, *Alternaria alternata*, *Drechslera tetramera*, *Penicillium pinophilum* and *Fusarium moniliforme* were frequently encountered seed-borne fungi, which were isolated from freshly harvested and stored seeds. Experiments were conducted to study the production of toxic substances by these seed-borne fungi. For this, fungi were grown in Czapek's Dox's liquid media, 30 ml of the medium was poured in conical flasks and was sterilized at 15 lb pressure for 15 min. Later each flask was inoculated and subsequently incubated at 25± 2° C for a period of 7, 14, 21 days. The cultures were harvested at the end of the incubation period and filtered through Whatman No. 42 filter paper. The filtrates so obtained were used to assay the toxin following the methods of Vidhyasekharan et al. (1970) by assessing percentage inhibition of seed germination, root and shoot elongation.

RESULTS AND DISCUSSIONS

Seeds carry a large number of fungi in field as well as in storage. The intensity of seed infection varies from weak to heavy and depending upon this the extent of invasion of seed-tissues by the pathogen also varies. In heavily infested seeds, the endosperm and embryonal infection was so high that the seeds failed to germinate. In fresh seeds, germinability is normally quite high and seed deterioration ultimately leads to loss of germination (Bilgrami, 1983).

The fungi cause decrease in germination percentage of seeds during storage due to invasion of the embryo. These storage fungi change the nutritional properties of the seed, by making it nutritionally poor or by secreting certain toxic substances unfavourable to the seed and the consumers. Data revealed in the table, a total of 19 fungal species were isolated from cumin seeds viz. *Alternaria alternate*, *Alternaria brassicae*, *Aspergillus candidus*, *Aspergillus flavus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Aspergillus terreus*, *Cunninghamella* sp., *Curvularia lunata*, *Fusarium oxysporum*, *Mucor* sp., *Penicillium chrysogenum*, *Penicillium citrinum*, *Penicillium janthinellum*, *Penicillium notatum*, *Penicillium* sp., *Rhizopus arrhizus*, *Rhizopus nigricans*, *Rhizopus stolonifer*.

Table.1 List of isolated fungi and their incidence percentage on the seeds of different samples of cumin seeds (*Cuminum cyminum L.*)

S. No.	Fungal Species	Percentage incidence of different species of fungi on cumin seeds All values in Percentage (%)
1	<i>Alternaria alternata</i>	3.15
2	<i>Alternaria brassicae</i>	6.30
3	<i>Aspergillus candidus</i>	9.45
4	<i>Aspergillus flavus</i>	12.60
5	<i>Aspergillus fumigatus</i>	3.15
6	<i>Aspergillus niger</i>	18.90
7	<i>Aspergillus terreus</i>	6.30
8	<i>Cunninghamella sp.</i>	6.30
9	<i>Curvularia lunata</i>	6.30
10	<i>Fusarium oxysporum</i>	3.15
11	<i>Mucor sp.</i>	6.30
12	<i>Penicillium chrysogenum</i>	3.15
13	<i>Penicillium citrinum</i>	3.15
14	<i>Penicillium janthinellum</i>	3.15
15	<i>Penicillium notatum</i>	3.15
16	<i>Penicillium sp.</i>	3.15
17	<i>Rhizopus arrhizus</i>	3.15
18	<i>Rhizopus nigricans</i>	3.15
19	<i>Rhizopus stolonifer</i>	3.15

As per the results of the study on cumin seeds, *Aspergillus niger* incidence was highest (18.90%) and lowest of (3.15%) *Alternaria alternata*, *Aspergillus fumigatus*, *Fusarium oxysporum*, *Penicillium chrysogenum*, *Penicillium citrinum*, *Penicillium janthinellum*, *Penicillium notatum*, *Penicillium sp.*, *Rhizopus arrhizus*, *Rhizopus nigricans*, *Rhizopus stolonifer*.

A significant amount of the annual production is affected by fungi because there are insufficient post-harvest preservation techniques (Seema and Basu, 2003). Specifically during the handling, storage, and transportation of spices, cumin seeds may become contaminated with fungi (Dimic et al., 2008). To effectively protect human health, it is important to improve the circumstances of cumin seeds throughout processing, storage, and shipping as well as maintain continuous mycological and mycotoxicological control before food preparation.

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