



Fungal Diversity Associated with Strawberry Fruit Loss in the Gorakhpur Division, U.P. India

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10.18805/ag.D-6170

ABSTRACT

Background: Strawberry (*Fragaria ananassa* Duch.) is highly nutritious and economically important fruit crop, cultivated worldwide is severely affected by fruit rot disease, leading to significant yield and economic yield loss. Present study aimed to provide the first report of fungal pathogens associated with strawberry fruit rot, which cause significant losses in India. Overall, the research provides valuable insights into the fungal pathogens responsible for strawberry fruit rot in India. The findings contribute to a better understanding of the disease and offer a foundation for effective field control measures against the disease.

Methods: Survey and sampling (triplicate) of infected strawberry fruits were conducted from 2020-2023 across various sites of Gorakhpur division. The data analysis was performed by using R software. Isolation of fungal pathogen was done by Agar Plate methods and standard blotter technique. Morphological characterization was performed based on colony morphology and microscopic characteristics.

Result: More than 350 symptomatic strawberry fruits were collected from 3 different cultivation sites, from which 211 fungal strains were isolated and morphologically identified as 15 different species. Among them, *Botrytis cinerea*, accounted for a higher proportion of all the strains present, comprising 64.6 % loss of fruits; followed by *Colletotrichum* spp. (17.4%), *Rhizopus stolonifer* (9.6%), *Fusarium* spp. (4.6%) and rest of the isolated fungi like, *Phytophthora* spp., *Mucor mucedo*., *Saccharomyces cerevisiae*., *Penicillium* spp., *Aspergillus* spp., *Alternaria* spp., *Cladosporium* spp and *Curvularia* spp caused 3.6% loss of fruits respectively. All isolates were found to be aggressive on both wounded and unwounded strawberry fruit.

Key words: Fruit losses, Fruit rot, Fungal diversity, Strawberry.

INTRODUCTION

Strawberries (*Fragaria ananassa* Duch.), belonging to the Rosaceae family, are globally renowned for their vibrant color and unique flavor. Strawberries are highly nutritious as they contain compounds such as vitamin B6, vitamin K, vitamin A, vitamin E and vitamin C (which is the highest among other fruits at 60 mg/100 g) (Ali *et al.*, 2020, Kuchi and Sharavani, 2019). Previous studies have demonstrated that they also contain carotenoids, phenols and flavonoids, which exhibit high antioxidant activity and have positive health effects (Nowicka *et al.*, 2019). Consuming strawberries regularly can help to reduce the risk of chronic diseases due to their anthocyanin content. Strawberries are significant fruit crop cultivated worldwide on over 370,000 hectares (FAOSTAT, 2021). China is leading strawberry producing country with 3,801,865 tons followed by United State with 1,420,570 tons. Worldwide total strawberry production is 9,125,913 tons per year (Soura *et al.*, 2024). United States alone, the annual strawberry production is valued at more than \$2.3 billion. It is originating from temperate regions such as China, Poland, Iran, Turkey, Germany and Spain. However, there are other varieties (Sweet Charlie, Winter Dawn, Barak, Gili, Hadar and Sabrina) that can be easily grown in mild tropical and subtropical regions (Mahapatra *et al.*, 2020). In India, strawberries are traditionally grown in hilly areas, but they are now being successfully cultivated in flat regions as well (Murthy and Pramanick, 2012), including temperate regions in the north, sub-tropical plains and high-altitude tropical areas. Farmers in tropical and

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How to cite this article: Dwivedi, M., Rai, R.K. and Singh, P. (2024). Fungal Diversity Associated with Strawberry Fruit Loss in the Gorakhpur Division, U.P. India. Agricultural Science Digest. 1-9. doi: 10.18805/ag.D-6170.

Submitted: 27-08-2024 **Accepted:** 22-10-2024 **Online:** 16-12-2024

sub-tropical regions have found success in growing strawberries during the winter months, obtaining their planting materials from the hills. The strawberry crop is highly commercialized and fetches a premium price. In developing countries, postharvest losses are often more severe due to inadequate storage and transportation facilities there are 25-40% loss of untreated strawberry reported due to microbial contamination. Nevertheless, fungal infections pose significant challenges to the fresh fruit industry, causing substantial losses. Numerous factors during pre-harvest, harvest, postharvest and distribution practices can influence the susceptibility of agricultural produce to fungal infection (Petrasch *et al.*, 2019). The nutrient-rich matrix of strawberries provides an ideal environment for spoilage microorganisms, particularly fungi. These fungal growths lead to fruit deterioration, accompanied by the production of harmful metabolites,

which raises concerns regarding food safety (Damyeh *et al.*, 2019; Sharma and Verma, 2019). Although limited studies have focused on identifying spoilage microorganisms specifically on strawberries, the two main types of fruit rot diseases affecting strawberries are grey mold rot, caused by the fungus *Botrytis cinerea* and rhizopus rot, caused by *Rhizopus stolonifer* and *Mucor* species. Grey mold rot is favored by high moisture and humidity levels and can lead to significant economic losses (Feliziani and Romanazzi, 2016). However, during my survey I have found the major problem in the sustainable production of strawberry in Gorakhpur region. The warm temperature and high humidity in Gorakhpur create favorable conditions for the growth of various fungal species, which negatively impact the earnings of farmers and retailers by causing fruit spoilage (Pal *et al.*, 2017). Current study goes beyond this by conducting a comprehensive field study of Gorakhpur division, including an assessment of annual disease incidence in terms of the percentage of fruit spoiling due to rotting pathogens. This study aims to isolate and identify the prevalent fungal pathogens, their distribution patterns and the socioeconomic implications of crop damage; effective strategies can be developed to reduce losses and enhance the sustainability of strawberry production in the region.

MATERIALS AND METHODS

Study area

Gorakhpur division in Uttar Pradesh, India comprises four districts, namely Gorakhpur, Deoria, Maharajganj and Kushinagar (Fig 1). Experiments were conducted in Department of Botany, Deen Dayal Upadhyaya Gorakhpur University, Gorakhpur, U.P., India. The region is located between latitudes 27°05' to 27°25' north and longitudes 83°20' to 84°10' east, with an elevation ranging from 107 meters above sea level. Gorakhpur division experiences a subtropical climate with four distinct seasons. Winters are

cool and foggy with temperature 5°-20°C, spring is mild having temperature 15°-25°C, summers are hot temperature ranging from 30°-35°C and monsoons bring heavy rainfall. The region's soil is primarily sandy loam and alluvial soil, which are well-drained and fertile, rich in organic matter, making them suitable for strawberry cultivation. However, proper management of pests, diseases and ensuring necessary irrigation practices are essential for successful cultivation.

Field surveys, disease symptoms and sampling

Field surveys were conducted during winter seasons of 2020-2023, collected fruit samples from strawberry fields in three different locations within Gorakhpur division. Fig 1 provide details about the topography, soil characteristics and names of study sites/ strawberry farms. The main strawberry varieties planted were Sweet Charlie, Winter Dawn, Festival and Gili. At each collection site, strawberry sample collection area was divided into three transects. From each transect, 20 fruits randomly selected from the strawberry fields. The fruit samples from each transect were then mixed together in a Ziploc bag to create a master sample. In total, three master samples were obtained. To ensure the preservation of the fruit samples, the Ziploc bags containing the samples were placed in an ice box and brought to the laboratory for further investigation and isolation of deteriorating fungal species.

Fungal isolation and maintenance

The deteriorating fungal species from each sample were isolated by using the following methodology:

Agar Plate method: Infected tissue from fruits that exhibited typical symptoms were selected and cut them into small bits measuring about 6mm. The tissue was then surface sterilized using a 0.1% mercuric chloride (HgCl_2) solution and washed three times with sterile distilled water. Next, transfer the surface sterilized tissues to sterile petri plates containing PDA media under aseptic

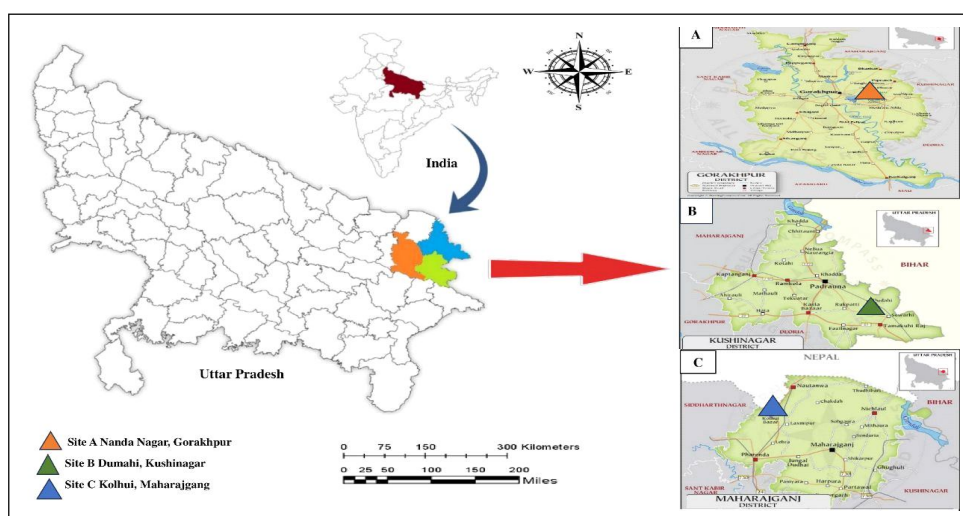


Fig 1: The map showing the study site of the experiments.

conditions. These Petri plates were placed in BOD incubator at $22\pm 2^{\circ}\text{C}$ under 12 h white light for seven days and fungal colonies were later observed using a binocular microscope.

Standard blotter technique: In this method, the surface sterilized fruits were dried between the filter papers and placed on the moistened blotter paper plated onto the Petri plates. The plates were then incubated in BOD incubator at $22\pm 2^{\circ}\text{C}$ under 12 h white light for seven days and fungal growth was observed using a binocular microscope. The fungal colonies distinct in morphology on agar plate were selected based on shape, size, margin, elevation and opacity. Cultures of each isolated fungal species were maintained on PDA slants at $4\pm 1^{\circ}\text{C}$ for further experimentation. Throughout the entire experiment, 7-day-old cultures of each fungus was used.

Morphological characterization

Fungal species were identified using a combination of gross colony morphology and microscopic characteristics. These characteristics were observed using a camera-enabled light microscope (Olympus magnus, Mumbai, India). The identification of the colonies was based on their color, texture of mycelia and type of pigmentation. Additionally, detailed morphological characteristics of the fungi, such as the presence of septation in hyphae, the type of reproductive structure (sporangia/conidia) present in chains or as solitary units and the shape of spores, were determined using established fungal keys (Pitt and Hocking, 2009).

Pathogenicity test

To assess the pathogenicity of a fungus on strawberry fruits, healthy fruits of uniform size were collected from three different cultivars. The fruits were washed with tap water and then disinfected with 0.1% Mercuric chloride solution. After several rinses with distilled sterile water, the fruits were air dried in a laminar air flow. Superficial wounds were created on the fruits surface using a sterile Cork borer. For inoculation, small disks of potato dextrose agar (PDA) with actively growing fungal mycelium were placed on the wounds of all the fruits, while control samples were inoculated with sterile PDA plugs. Inoculated fruits were individually placed in plastic box containing a swab of distilled water to maintain humidity. The boxes were covered with parafilm paper and incubated at room temperature ($22\pm 2^{\circ}\text{C}$). Lesions on the fruits were observed at 7 days post-inoculation (DPI) and photographs were taken at 10 DPI. To fulfill Koch's postulates, diseased tissues were transferred onto PDA. The pathogen was re-isolated and its identification was verified based on colony and conidial characteristics.

The disease incidence of strawberry fruit in Gorakhpur division was determined by analyzing the number of diseased fruit samples in comparison to the total number of observed fruits in the fields. Data was collected when

Per cent disease incidence =

$$\frac{\text{Weight of infected fruits}}{\text{Total weight of fruits assessed}} \times 100$$

visible disease symptoms appeared on ripe fruits. The percentage of disease incidence was calculated using a specific formula.

Statistical analysis

All the data values obtained from the experiments are presented as means of three replicates $\pm$ standard error. Analysis of the data from the field surveys indicated no significant differences at $P>0.001$, between the different sites, with homogeneous error variances, allowing for the data for further analysis. The data analysis was conducted using R software (R Core Team, 2020). Prior to performing ANOVA (Table 1), the data were assessed for CRD Analysis and the ANOVA was carried out if the data met the required assumptions. If the results of the ANOVA test showed a significant difference then Tukey's honestly significant difference (HSD) test was performed to further explore the variations among different sites.

RESULTS AND DISCUSSION

Field surveys and Sample collection

The field survey revealed visible symptoms of fruit rot, observed on the surface of all samples collected (Fig 2). This problem extended beyond just the fruits, as the strawberry plants themselves exhibited wilting, indicating a significant infection by various fungal pathogens that resulted in severe rotting symptoms (Fig 2 C-K). Increasing demand of strawberry consumption in recent years, has led to a rapid expansion of the commercial strawberry cultivation industry, with the planting regions of strawberries growing in number. However, the continuous cropping has also brought about an increase in the severity of diseases affecting the crop (Krüger *et al.*, 2020). In current investigation, the frequency of disease occurrence varied from 30.5% to 44.4% across different locations within Gorakhpur division. The highest disease incidence of 44.4% was documented at Site A Nandanagar, Gorakhpur between March and April (2022-2023), coinciding with the period of increased humidity and high temperatures. In contrast, the lowest disease incidence of 30.5% was noted at Site C Kolhui, Maharajganj during November to December (2020-2021), in the winter season. This variability underscores the influence of environmental factors on the spread of fruit diseases in Gorakhpur division. Variations in disease incidence and strawberry crop susceptibility towards pathogens may also be related to level of genetic resistance present in the cultivars used for cropping. (Keldibekova and Knyazev, 2023). Strawberry cultivars may also respond to different planting seasons and conditions (Gogoi *et al.*, 2023). This causes biochemical variability in fruits which may indirectly affect the response of strawberry against pathogens.

Table 1: Major fungal pathogen isolated from infected strawberry fruits of different study sites.

Survey sites	Survey months	Fungal Isolates															
		<i>Botrytis cinerea</i>	<i>Colletotrichum stamense</i>	<i>Colletotrichum fructicola</i>	<i>Colletotrichum fragariae</i>	<i>Colletotrichum acutatum</i>	<i>Fusarium oxysporum</i>	<i>Fusarium commune</i>	<i>Phytophthora cactorum</i>	<i>Rhizopus stolonifer</i>	<i>Mucor mucedo</i>	<i>Saccharomyces cerevisiae</i>	<i>Sclerotinia sclerotiorum</i>	<i>Sclerotium rolfii</i>	<i>Verticillium dahliae</i>	<i>Penicillium expansum</i>	<i>Penicillium frequentans</i>
Site A	Nov-Dec	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Jan-Feb	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Mar- Apr	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Site B	Nov-Dec	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Jan-Feb	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Mar- Apr	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Site C	Nov-Dec	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Jan-Feb	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	Mar- Apr	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
		+ presence; - absence.															

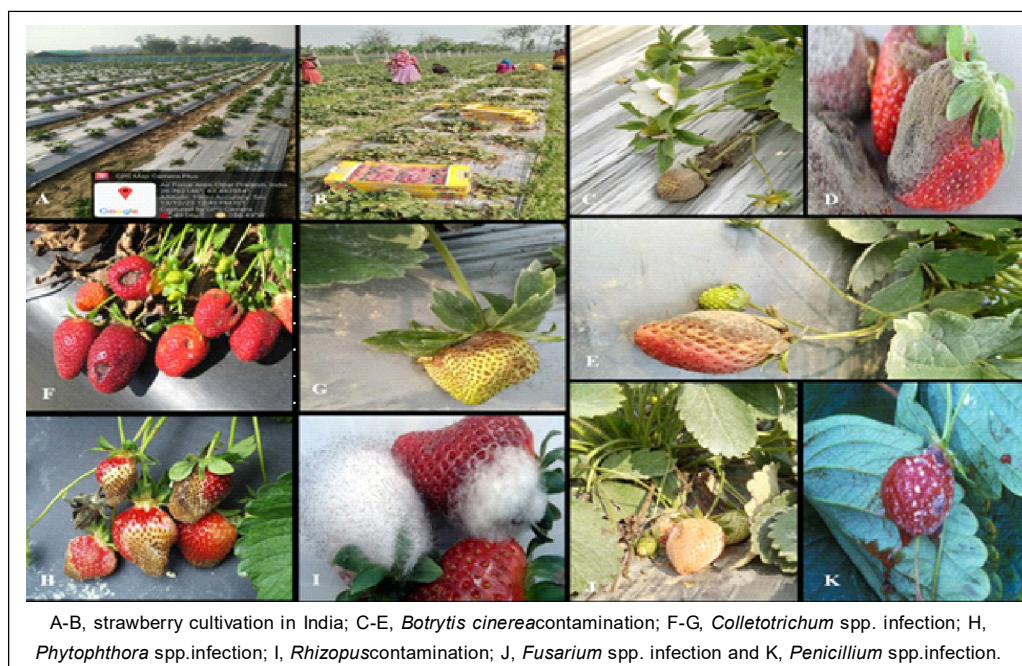


Fig 2: Prevalent Fungal Pathogens Causing Diseases on Strawberry fruits.

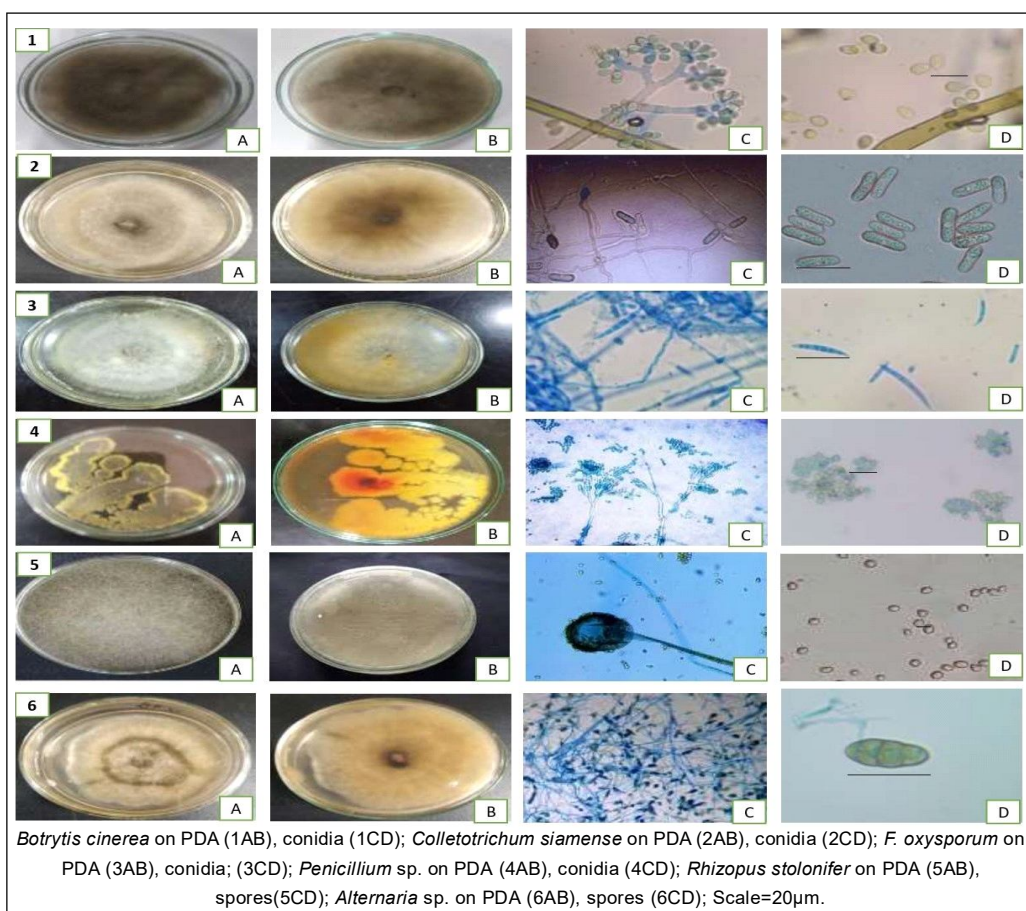


Fig 3: The morphological characteristic of the fungi causing strawberry fruit rot (SFR).

Fungi isolated

A total of 15 distinct fungal strains were isolated from the diseased samples (Fig 3). These strains included the major pathogen responsible for rotting, such as *Botrytis cinerea*, *Colletotrichum* spp., *Phytophthora* spp., *Rhizopus* spp., *Fusarium* spp. and a few other fungi in smaller numbers.

Botrytis cinerea

The mycelium of *Botrytis cinerea* consisted of brownish olive hyphae with septation. This fungus was characterized by abundant hyaline conidia (asexual spores) that were born on grey, branching tree-like conidiophores. The conidia were ellipsoidal or ovoid in shape and possessed a dry and hydrophobic nature (Fig 3 1CD). They were colorless, smooth and produced highly resistant sclerotia in old cultures. The colony of *Botrytis cinerea* exhibited an olive color (Fig 3 1AB). This strain was identified as the causative agent of Botrytis fruit rot.

Colletotrichum spp.

The colonies of *Colletotrichum* spp. were villous, with a gray and neat edge in the middle and a yellow edge in the middle of the back (Fig 3 2AB). The conidia heap appeared dark yellow. The conidia were straight, terete, blunt round at both ends, colourless and smooth (Fig 3 2CD). The appressoria, which were brown and oval or spindle-shaped, had intact margins with some irregular shapes.

Fusarium spp.

Different colony morphologies were observed for all *Fusarium* strains isolated from the strawberry fruits on potato dextrose agar (PDA). *F. oxysporum* exhibited woolly, white to pale aerial hyphae with a loose structure (Fig 3; 3AB). The pedicellate conidia were solitary and not branched. The large conidia were meniscus- and sickle-shaped and mostly had 3-6 septa, while the small conidia were single-celled, ovoid to elliptic and colorless (Fig 3 3CD).

Other fungal species

In addition to the aforementioned pathogenic fungal species, a few other strains were also isolated are mentioned below in Table 2 (Fig 3), nine species were preliminarily identified as *Phytophthora* spp., *Rhizopus* spp., *Mucor mucedo*., *Saccharomyces cerevisiae*., *Penicillium* spp., *Aspergillus* spp., *Alternaria* spp., *Cladosporium* spp and *Curvularia* spp etc. Table 1 exhibited the most frequent pathogen of the strawberry fruit rot is *Botrytis cinerea* in this region, followed by *Colletotrichum* spp., *Fusarium* spp., *Phytophthora*, *Rhizopus*, *Mucor*, *Penicillium*, *Aspergillus* and *Alternaria* and a list of other fungi were also found in traces. Greatest loss of fruit was caused by *Botrytis cinerea*, because the pathogen was present throughout the season in the field. In India, a groundbreaking study has been conducted to assess the disease potential and losses of commercial strawberry cultivars in field conditions, where Botrytis fruit rot is the most prevalent, followed by

Table 2: Percentage of disease estimated during field survey (Nov 2020 - April 2023) of strawberry fruits in Gorakhpur division.

Survey sites	Survey duration (In months)	Total fruit observed in each survey (Replication mean)	Disease incidence during field survey (Nov 2020- April 2023)					
			No. of infected fruits in -2021(Mean $\pm$ Std)	Per cent disease incidence during 2020 -2021	No. of infected fruits in 2021-2022 (Mean $\pm$ Std)	Per cent disease incidence during 2021 -2022	No. of infected fruits in 2022-2023 (Me Std)	Per cent disease incidence during 2022-2023
Site A (Nandanagar)	Nov-Dec	20	7.443 $\pm$ 2.3	37.2%	8.217 $\pm$ 2.2	41.0%	8.107 $\pm$ 1.5	40.5%
Gorakhpur	Jan-Feb	20	7.550 $\pm$ 2.1	37.7%	7.993 $\pm$ 2.0	39.2%	7.883 $\pm$ 1.9	39.4%
	Mar- Apr	20	8.110 $\pm$ 1.8	40.5%	8.107 $\pm$ 1.5	40.5%	8.887 $\pm$ 2.6	44.4%
Site B (Dumhui)	Nov-Dec	20	6.887 $\pm$ 2.6	34.4%	7.107 $\pm$ 2.1	35.5%	6.993 $\pm$ 2.0	34.9%
Kushinagar	Jan-Feb	20	7.107 $\pm$ 2.1	35.5%	6.883 $\pm$ 1.9	34.4%	7.887 $\pm$ 2.6	39.4%
	Mar- Apr	20	7.110 $\pm$ 1.8	35.5%	7.993 $\pm$ 2.0	39.2%	7.993 $\pm$ 2.0	39.2%
Site C (Kolhui)	Nov-Dec	20	6.110 $\pm$ 1.8	30.5%	6.220 $\pm$ 1.8	31.1%	6.883 $\pm$ 1.9	34.4%
Maharajganj	Jan-Feb	20	6.887 $\pm$ 2.6	34.4%	7.220 $\pm$ 1.8	36.1%	6.220 $\pm$ 1.8	31.1%
	Mar- Apr	20	6.993 $\pm$ 2.0	34.9%	7.887 $\pm$ 2.6	39.4%	7.550 $\pm$ 2.1	37.7%

Values are given as the mean of three replicates $\pm$ standard errors. Data are not significant at $P > 0.001$.

Anthrachnose rot, soft rot, Fusarium rot and leather rot. These findings provide valuable insights for strawberry growers, enabling them to make informed decisions on the cultivation of specific cultivars to minimize losses from strawberry fruit rots in India. Strawberry is susceptible to various pathogens, such as fungi, bacteria, viruses and nematodes. Fungi are the most economically damaging pathogens of strawberry crops, as they can infect all parts of the plant and cause destructive loss or even death. Among fungi, *Botrytis cinerea*, an ascomycete, is considered the primary pathogen of strawberry in field and after harvest, causing significant economic losses to the industry worldwide (Garrido *et al.*, 2011). This fungus leads to the development of grey mould in strawberry fruits and dying plant parts and it can also affect the vegetative tissues. In humid conditions, untreated strawberry plants can lose over 80% of their flowers and fruits due to *B. cinerea* infection. *Botrytis cinerea* is a damaging pathogen that thrives on over a thousand different products, predominantly fruits and is particularly destructive to them during storage, leading to substantial financial losses on a global scale (Petraschet *et al.*, 2019). In this study, 211 fungal isolates were obtained from strawberries showing SFR symptoms in different sites of Gorakhpur are the major strawberry

production areas were identified as 15 different species. Among them, *Botrytis cinerea*, *Colletotrichum siamense*, *Colletotrichum fructicola*, *Colletotrichum fragariae*, *Rhizopus stolonifer*, *Fusarium oxysporum*, *Fusarium commune* and other fungi (Table 1) accounted for a higher proportion of all strains, comprising 64.6%, 17.4%, 9.6%, 4.6% and 3.6%, losses of strawberry fruits respectively (Fig 4).

Disease incidence and percentage loss

Table 3 provides an overview of the prevalence of diseased fruits in each site.

It was observed that the percentage of disease incidence ranged from 30.5 % to 44.4% in the selected sites of Gorakhpur division. The highest percentage of disease incidence (44.4%) was recorded at the Site A Nandanagar, Gorakhpur during march to april (2022-2023), which corresponds to the rising temperature as characterized by humidity and high temperatures. Conversely, the lowest percentage of disease incidence (30.5%) was observed at Site C Kolhui, Maharajganj between November to December (2020-2021), during the winter season. This variation highlights the impact of environmental conditions on the prevalence of fruit diseases in Gorakhpur division.

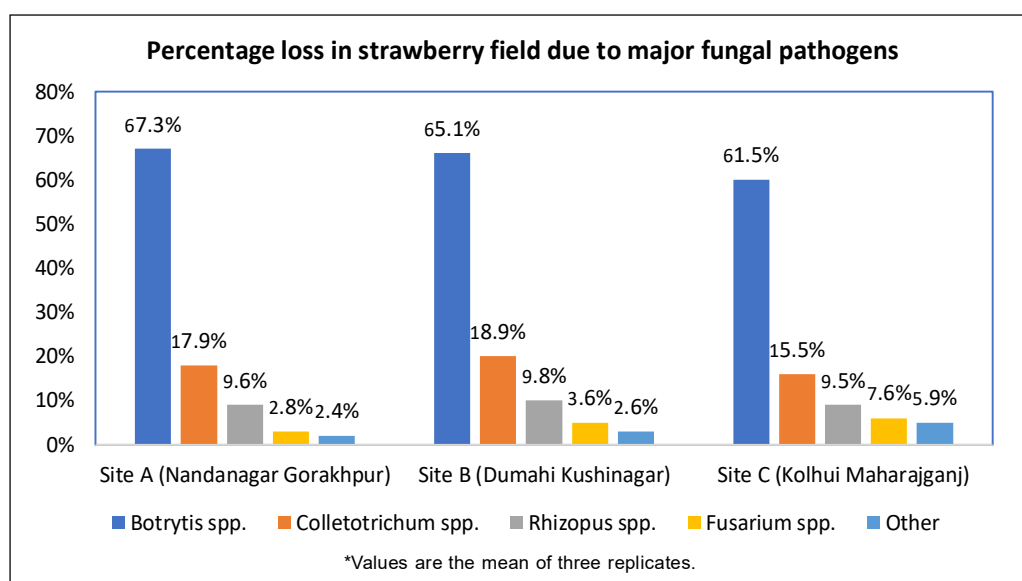


Fig 4: Estimation of percentage loss of strawberry fruits in different cultivation sites.

Table 3: ANOVA Table.

ANOVA analysis duration	Source of variance (SoV)	Df	Sum Sq	Mean Sq	F value	Pr(>F)	SE(m)	CV value	P value
2020-2021	Replication	2	2.251	1.125	0.208	0.818	1.343	33.951	>0.001
	Error	6	32.451	5.409	NA	NA			
2021-2022	Replication	2	6.663	3.332	0.782	0.499	1.192	28.896	>0.001
	Error	6	25.571	4.262	NA	NA			
2022-2023	Replication	2	2.748	1.374	0.388	0.694	1.087	25.691	>0.001
	Error	6	21.265	3.544	NA	NA			

Treatment means are not significantly different. Multiple comparison test is not performed. Df., Degree of freedom, SE., Standard error.

As the result exhibited that the most prevalent pathogen of strawberry fruit rot is *B. cinerea*, present at each study sites in whole season were caused the greatest loss upto 64.6%. The appropriate temperature and high humidity in Gorakhpur create favorable conditions for the growth of *B. cinerea*, which negatively impact the earnings of farmers and retailers by causing fruit spoilage. The result is not varied in different study sites, because all study sites are under Gorakhpur division having same climate and environmental condition. Rysin *et al.* (2015) reported that, *B. cinerea* expands through high moisture and high humidity content (>93%) at moderate to high temperature range from 18° to 24°C. In view of this, in Iran and in Italy reported 25% to 37% losses of strawberry fruits due to gray mould rot of *B. cinerea* (Salami *et al.*, 2010; Ugolini *et al.*, 2014; Tane, 2022). Yield losses of up to 80% strawberry fruit of total production occurs due to gray mould disease because the pathogen gets resistant against the fungicides (Higuera *et al.*, 2019). The 2nd most frequent pathogen of SFR is *Colletotrichum* spp. leads to anthracnose symptoms, characterized by small, sunken, dark lesions on the fruit surface and causing 50% of yield losses in field (Reddy *et al.*, 1998). In current investigation, *Colletotrichum* spp. is responsible for the major loss upto 17.4% estimated in this region. Fruit rot in strawberries typically begin with the infection of the roots and crowns by *Colletotrichum* spp., leading to the spread of the disease to the leaves, petioles, flowers and fruits over time. In addition to the above two major fungal pathogen that have been widely known to infect strawberry and cause fruit rot, other fungal genera have also been linked to fruit rot in strawberry. For instance, reports from strawberry production regions like Spain and North America have highlighted the involvement of *Phytophthora* spp. such as *P. cactorum* and *P. nicotianae* in causing root rot, crown rot and fruit rot. Notably, *P. cactorum* causing rapid fruit collapse, resulting in soft, water-soaked patches known as leather rot, leading to significant yield loss of up to 40% (Higuera *et al.*, 2019). Additionally, *Rhizopus stolonifer* is also devastating pathogen of strawberry fruit, in this study pathogen caused 9.6% of fruit losses in strawberry field. Severely infected fruit may display a cotton-like mycelial growth with black sporangia (Reddy *et al.*, 1998). Apart from *Rhizopus*, *Fusarium* spp., such as *Fusarium oxysporum* and *Fusarium commune*, are the prevalent fruit rot pathogen of strawberry, often entering through wounds and leading to secondary infections and causing significant decay up to 4.6% of fruits. In addition to loss, other deteriorating fungi like, *Mucor mucedo*, *Saccharomyces cerevisiae*, *Penicillium* spp., *Aspergillus* spp., *Alternaria* spp., *Cladosporium* spp and *Curvularia* spp etc. positively contribute to strawberry fruit decay and exhibited 3.6% loss of fruits. The pathogenicity experiments showed that *Botrytis cinerea*, *C. siamense*, *C. fructicola*, *C. fragariae*, *Rhizopus stolonifer*, *Fusarium oxysporum*, *Fusarium commune*, *Phytophthora cactorum* and *Penicillium expansum* caused symptoms of SFR. The

disease incidence of *B. cinerea*, *C. siamense*, *F. oxysporum*, *R. stolonifer* and *Mucor mucedo* was relatively high; the disease incidence reached more than 44.0%, which indicated that these species were the main fungi causing SFR. This study confirmed the diversity of pathogenic fungi for SFR, which included not only *Botrytis*, *Colletotrichum* and *Fusarium*, as previously reported, but also *Rhizopus stolonifer*, *Phytophthora cactorum*, *Penicillium expansum* and *Mucor mucedo*, were also responsible for SFR. The above research findings revealed that the spectrum of pathogenic fungi causing strawberry fruit rot (SFR) in various location of Gorakhpur Division extended beyond *Botrytis cinerea* and *Colletotrichum* spp. It was evident that, in case of regional climates and environments shifted, the composition of pathogenic species also underwent significant changes. Therefore, focusing solely on *Botrytis cinerea* and *Colletotrichum* spp. as the primary pathogen or targeting fruit rot for controlling strawberry fruit rot (SFR) may not be adequate. This study highlighted the diverse pathogens responsible for SFR through a comprehensive analysis encompassing morphology and pathogenicity assessments. The identified SFR pathogenic fungi comprised not only *Botrytis cinerea*, *Colletotrichum* spp., *Rhizopus* spp. and *Fusarium* spp., but also other fungi like *Phytophthora* spp., *Mucor mucedo*, *Saccharomyces cerevisiae*, *Penicillium* spp., *Aspergillus* spp., *Alternaria* spp., *Cladosporium* spp and *Curvularia* spp. This discussion shed light on the range of SFR fungi, offering valuable insights for early detection, prediction and mitigation strategies for strawberry fruit rot.

CONCLUSION

In conclusion, the cultivation of strawberry in Gorakhpur region faces significant challenges due to the prevalence of fungal pathogens leading to fruit spoilage. The warm temperatures and high humidity in the region create favorable conditions for fungal growth, causing substantial postharvest losses for farmers and retailers. These losses not only impact the economic viability of strawberry cultivation but also raise concerns about food safety and security. To address these challenges, it is imperative to implement effective strategies to mitigate fungal infections and improve the sustainability of strawberry production in Gorakhpur. This may include investing in better storage and transportation facilities, adopting improved pre-harvest and postharvest practices and implementing integrated disease management approaches to control fungal pathogens. By identifying and understanding the prevalent fungal pathogens, their distribution patterns and the socioeconomic implications of crop damage, targeted interventions can be developed to reduce losses and ensure the long-term success of strawberry cultivation in the region. Overall, this study provides valuable insights into the challenges faced by strawberry farmers in Gorakhpur and underscores the importance of addressing fungal infections to enhance the quality and yield of

strawberry crops. By implementing evidence-based strategies and fostering collaboration between researchers, farmers and policymakers, we can work towards a more sustainable and resilient strawberry industry in the region.

Conflict of interest

The authors declare no conflict of interest.

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