BIODIVERSITAS
Volume 25, Number 3, March 2024
Pages: 1089-1096

ISSN: 1412-033X
E-ISSN: 2085-4722
DOI: 10.13057/biodiv/d250323

Mycoflora associated with stored rice from storage facilities in Makassar and Maros, Indonesia

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Manuscript received: 12 November 2023. Revision accepted: 22 March 2024.

Abstract. *Brugman E, Mario MB. 2024. Mycoflora associated with stored rice from storage facilities in Makassar and Maros, Indonesia. Biodiversitas 25: 1089-1096.* Rice storage is a postharvest process that aims to maintain food availability, seed supply, and price stability. One of the main problems encountered in the storage process is the potential for fungal infection and contamination, leading to postharvest loss. This research aims to evaluate the mycoflora associated with stored rice from storage facilities in Makassar and Maros and their contamination level. Fungal isolation was done by spread plating with serial dilution and direct culture method. The fungal identification was made based on morphological characters, including colony, sexual, and asexual structures. The fungal contamination was determined by colony counting, contamination percentage, and species occurrence. The result showed that all samples tested positive for fungal contamination, with an average contamination level of 83.89% (Makassar) and 86.11% (Maros). The highest colony densities in the samples from Makassar and Maros were 9.0 × 103 CFU/g and 14.2 × 103 CFU/g, respectively. The average fungal contamination in the samples from Makassar was 3.67×10^3 CFU/g, and Maros was 8.32×10^3 CFU/g. The mycoflora associated with the rice grain samples from Makassar was *Rhizoctonia solani* (11.34%), *Aspergillus* spp. (52.18%), *Fusarium* spp. (10.87%), *Rhizopus* sp. (15.22%), and unidentified cultures (10.39%). Meanwhile, the mycoflora associated with the stored rice from Maros are *Curvularia* sp. (15.22%), *Alternaria* sp. (8.7%), *Aspergillus* spp. (17.39%), *Fusarium* spp. (32.61%), *Rhizopus* sp. (15.22%), and *Bipolaris oryzae* (10.87%).

Keywords: Food safety, postharvest loss, storage fungal

INTRODUCTION

Food is an essential source of nutrients and energy for human life and health. Most of the Indonesian population relies on rice as a single staple food. Indonesia's weekly rice consumption reached 1.6 kg per capita in 2022. This value varies by district, with the lowest weekly per capita consumption at 0.174 kg in Puncak district and the highest at 2.574 kg in South Nias district. In South Sulawesi, the average weekly rice consumption is 1.81 kg per capita, slightly higher than the national weekly rice consumption per capita (Statistics Indonesia 2023). Although Indonesia is the fourth largest rice-producing country in the world, the government still needs to import rice to fulfill the large per capita consumption every year. In 2022, Indonesia imported 429.207,2 tons of rice from several countries such as India, Thailand, Vietnam, Pakistan, Myanmar, Japan, and others (Statistics Indonesia 2022).

There are several approaches to increasing rice availability, one of which is reducing postharvest losses. The total national rice postharvest loss in 2018 was estimated to reach 8.14 million tons (Falatehan et al. 2021). Postharvest losses can occur during various stages, such as harvesting, threshing, distribution, storage, processing, packaging, and marketing to post-consumers (Saba and Ibrahim 2018). One of the main problems encountered in the storage process is the potential for fungal infection and contamination, leading to postharvest loss. Postharvest disease can be caused by field infection (infected) or postharvest contamination (infested). Infection in the field is latent, and the disease only begins to develop in a condition that favors the fungal pathogen to grow; meanwhile, postharvest contamination occurs during storage and is caused by the storage fungi (Tripathi et al. 2021).

Storage fungi are widespread and prevalent in storage facilities. Storage fungi can live as both saprophytes and parasites. They contaminate the grain surface and can further develop on grains without free water. Storage fungi thrive at relative humidity of 70-80% and seed moisture content >16% (Mohapatra et al. 2017; Daou et al. 2021). Storage fungi generally are members of the genera *Aspergillus* and *Penicillium.* Several studies reported common mycoflora associated with rice including *Aspergillus flavus*, *Aspergillus niger*, and *Aspergillus* spp. (Bagus et al. 2017); *Aspergillus* spp., *Penicillium* spp., *Fusarium* spp., *Rhizopus* spp., *Alternaria* spp., *Mucor* spp. (Ali et al. 2018); *A. flavus*, *Fusarium proliferatum* (Phan et al. 2021); *A. flavus*, *A. parasiticus* (Qi et al. 2022); *Bipolaris* spp., *Fusarium* spp., *Macrophomina phaseolina*, *Aspergillus* spp., *Curvularia* spp., *Botryodiplodia* spp. (Ackaah et al. 2023); *A. niger*, *Talaromyces pinophilus* (Laut et al. 2023); and *Alternaria alternata*, *F. moniliforme*, *Rhizopus nigricans*, *Helminthosporium oryzae*, *Pyricularia grisea* (Kumar et al. 2023).

The contamination of fungi poses a great risk to food safety, as several storage fungi are known to produce

numerous mycotoxins. Mycotoxins are toxic secondary metabolites produced by various fungal species. These mycotoxins can persist in food commodities, posing health risks to humans and animals (Daou et al. 2021). The fungal genera *Aspergillus*, *Fusarium*, *Talaromyces*, and *Penicillium* are well-known producers of mycotoxins and are responsible for synthesizing several well-described mycotoxins. Among these mycotoxins, Aflatoxins (AFTs), Fumonisins (FBs), Ochratoxin A (OTA), and Zearalenone (ZEN) are particularly concerning, and they can indeed contaminate rice, making them the most significant chemical hazards in food (Qi et al. 2022; Santos et al. 2022). Proper storage and processing practices are crucial for mitigating the risk of mycotoxin contamination in rice and maintaining the safety of other food commodities (Mannaa and Kim 2017; Laut et al. 2023).

Continuous rice demand needs to be supported by adequate rice storage facilities. Proper storage facilities provide safe storage conditions, prevent contamination from pests, dust, and environmental factors, reduce the risk of microbial growth and mycotoxin formation, maintain the nutritional quality of rice, and extend its shelf life. Additionally, it assures consumers of safe handling practices and fosters confidence in the safety of the food supply chain. Food safety is an essential aspect of food security. Ensuring food safety is a shared responsibility involving governments, businesses, and consumers working together to safeguard the food supply. This study aims to evaluate the mycoflora associated with stored rice in South Sulawesi and their contamination level in different storage facilities by comparing the conventional storage facility managed by the public and the modern storage facility operated by the government. The results provide potentially important data for the strategic guidance for local rice grain storage to better control fungal contamination and improve grain quality.

MATERIALS AND METHODS

Sample collection and moisture content analysis

Rice grain samples were collected from a public storage facility in Maros Regency and a government rice storage facility managed by the Indonesia Logistics Bureau (BULOG) in Makassar, South Sulawesi. The public storage facility in Maros represents conventional storage, and the storage facility in Makassar represents modern storage. The samples were not necessarily of the same variety and were not stored under similar conditions. Samples from the government storage were taken from six sides of the staple. On each side of the staple, there were five subsample points. The subsamples obtained from the five points were composited into 50 g. Unhusked rice grain samples from the public storage in Maros were collected similarly. The illustration of sampling points is shown in Figure 1. The samples were then submitted to the Plant Disease Laboratory, Universitas Hasanuddin, for moisture content analysis using rice moisture meter RIKA TS-7B (Tokyo RIKA). Each sample was analyzed three times to maintain the accuracy.

Fungal isolation and morphological characterization

Isolation of mycoflora from rice grain samples was carried out using serial dilution. One gram of each sample was grounded and added into 10 mL of sterile aqua dest. Based on the preliminary test with three replicable plates, it was determined that the serial dilutions in this study were up to 1/1000th for most countable Colony Forming Units (CFUs). Each dilution series was cultured in Potato Dextrose Agar (PDA) media added with rifampicin (150 ppm). The solid media modification by rifampicin addition was used to eliminate potential bacterial growth (Grum-Grzhimaylo et al. 2016). The dilution series was repeated three times for each sample. The isolation was done by pouring 0.5 mL of the suspension into PDA plates in three replications for each sample and dilution series. The PDA plates were then incubated at room temperature (28±2°C) for five days. The fungal growth was observed daily. Different morphologies of growing fungi represent one representative colony. Each colony representative was isolated and purified in new PDA plates for morphological characterization. Morphological characterization was done by observing the colony pattern, color, and microscopic structures, including the hyphae, spores, conidia, conidiophore, and other observable structures. The identification was carried out based on the Pictorial Atlas of Soil and Seed Fungi (Watanabe 2010) and several other related studies. The identification was done using four replicates of each colony representative to consider the consistency of the morphological characteristic.

Mycoflora was isolated from rice seed samples using the agar plate method (ISTA 2006) on PDA plates to determine the contamination percentage. The grain samples were first surface-sterilized using 2% sodium hypochlorite (NaOCl) for two minutes and rinsed twice with sterile distilled water. The seed samples were then air-dried on filter paper in Laminar Airflow. A total of ten grains were isolated on PDA plates in three replications. The plates were then incubated at room temperature (28±2°C) for five days. The fungal colonies growing around the grains were documented.

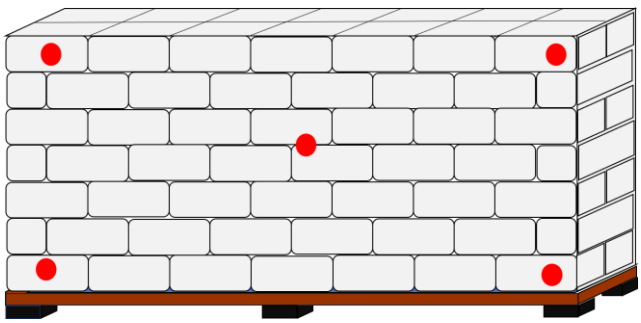

Figure 1. Sampling points on each staple

Data analysis

Each plate showing fungal growth from the 10^3 dilution series was selected for colony counting. The average number of Colony-Forming Units (CFU) from three replications was counted. Colony counting for CFU/g was calculated using the method from Pitt and Hocking (1997). The contamination percentage was calculated from the number of infected seeds isolated on PDA plates. The infected seed was determined by observing the emergence of the fungal colony around the seed. The formula for calculating the contamination percentage is as follows:

% Contamination = (The number of infected seeds/total number of seeds tested) × 100

The percentage incidence of individual pathogens was determined using the following formula:

% Occurrence of species = (The number of colonies of a species/total number of colonies) × 100

RESULTS AND DISCUSSION

Morphological characterization

Observation of mycoflora colonies in stored rice grain from Makassar resulted in 5 colony representatives and 8 colony representatives in the samples from Maros. The morphological characters of each colony representative are presented in Table 1, Figures 2 and 3.

Table 1. The morphology characteristic and conidia/spores size of representative isolates

Sample name	Colony	Hypae	Conidia/Spore	Conidia/Spore size					
				Length (µm)			Width/Diameter (µm)		
				Min	Max	Mean	Min	Max	Mean
Makassar CR1	Velvety, white to cream	Septate, rhizoid	-	-	-	-	-	-	-
Makassar CR3	Powdery, white to brown	Septate, filamentous	Yellow-brown, spherical to elliptical in shape, arranged in chain-like structures	-	-	-	2.06	5.05	3.81
Makassar CR3	Cottony, white	Septate, hyaline	Hyaline, sickle-shaped, septate, produced in cluster	2.97	3.86	3.33	25.28	34.94	29.53
Makassar CR4	Cottony, white to grey or dark brown	Non-septate, hyaline	Dark brown to black color, globose	7.11	14.87	11.68	10.71	16.58	13.57
Makassar CR5	Powdery, white to yellow-green	Septate, filamentous	Olive green, spherical to elliptical in shape, arranged in chain-like structures	-	-	-	3.50	6.52	4.93
Maros CR1	Powdery, yellow-green with white margin	Septate, branched	Olive green, globose	-	-	-	4.52	7.58	6.22
Maros CR2	Velvety, cream to brown	Septate, branched	Brown to dark brown, septate, curved or sigmoid shape	38.51	52.17	43.49	14.30	18.91	16.45
Maros CR3	Velvety, greyish-black	Septate, branched	light brown to brown, septate, elliptical in shape	53.22	84.01	72.67	14.52	20.62	16.89
Maros CR4	Cottony, greyish-white	non-septate	Dark brown to black color, globose	6.84	14.22	11.43	7.57	15.45	11.32
Maros CR5	Cottony, white to pale white	septate, hyaline	Hyaline, kidney-shaped microconidia	4.57	8.76	5.88	2.10	3.57	2.92
Maros CR6	Powdery, dark brown to black	Septate, filamentous	dark brown to black, globose	-	-	-	2.61	3.56	3.02
Maros CR7	Cottony, white to yellow	septate, hyaline	Hyaline, sickle-shaped macroconidia	24.98	50.56	40.03	4.00	6.22	4.86
Maros CR8	Velvety, cream to brown	Septate, branched,	Deep brown to black, septate,	23.59	37.59	30.38	11.68	15.25	13.13

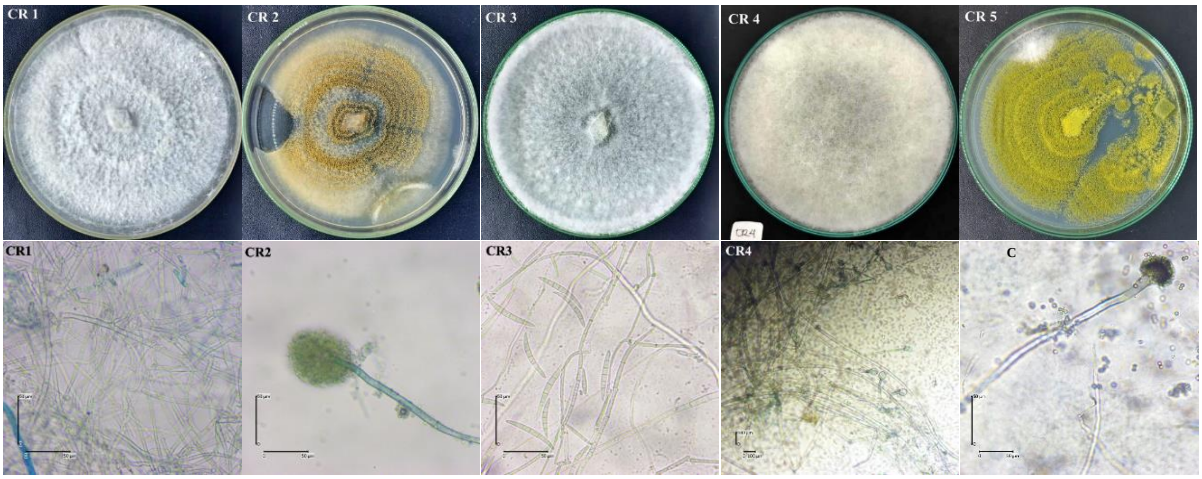

Figure 2. The colony and microscopic structures of representative isolates cultured from the Makassar sample

In Figure 2, CR1 has a white-to-cream velvety colony and filamentous septate hyphae. There are no conidia or spores observed. The hypae had branches forming 45 degrees angles; more mature branches will be perpendicular and thickened. Based on these characteristics, CR1 is identified as *Rhizoctonia solani* (Moni et al. 2016; Desvani et al. 2018). CR2 and CR5 have the same colony pattern; both isolates acquired the white color of the colony at first, but after two days of isolation, CR2 produced yellow-brown conidia while CR5 had olive green conidia. The microscopic observation of both isolates showed thick, discolored conidiophores and globular vesicles; phialide and conidia are arranged in the vesicle, forming chain-like structures. Based on these characteristics, CR2 and CR5 are identified as *Aspergillus* spp. (Thathana et al. 2017). CR3 has a white-colored and cottony colony with hyalin and septate hypae. The hyphae are mostly branched. Abundant sickle-shaped macroconidia were also observed. The macroconidia is hyaline, slender, with 5-6 septate. There was no microconidia observed. Chlamydospores are arranged in a chain of over four spores. Based on these characteristics, CR 3 is identified as *Fusarium* sp. CR4 has a fast-growing colony; it only took two days to grow fully in the Petri dish. The colony is cottony and white at first, turning grey or dark brown as the reproductive structure develops. Sporangiophore is aerial and bears sporangia at the tip. Sporangia are globose, with a dark brown to black color. Zygospore was also observed in two replicates. Based on these characteristics, CR 4 is categorized as *Rhizopus* sp. (Kwon et al. 2011).

Based on the morphological structures shown in Figure 3, CR1 and CR6 have the same reproductive structures, including the unbranched conidiophores with septate hyphae. The vesicles are globose, and the conidia have a flask-like appearance. Both isolates exhibit a rapid growth rate. The colonies are powdery with different colors. CR1 has olive green conidia, and the CR6 has dark brown to black conidia. Both CR1 and CR6 are identified as *Aspergillus* spp. CR2 has a cream-colored colony in the early growth and turns dark brown to black after a few days. Hyphae are septate and branched. The conidia are typically curved or boomerang-shaped, with multiple cells, mostly 4-celled with a darker brown color. Based on its morphological appearance, CR2 is identified as *Curvularia* sp. (dos Santos et al. 2018). CR3 has a greyish-black colored velvety colony with septate and branched hyphae. Conidiophore is simple and bears conidia apically. The color of conidia ranged from light brown to brown and brown to dark brown. The conidia are ellipsoidal and have a presence sector. CR3 is then identified as *Bipolaris oryzae* (da Silva et al. 2022). CR4 has a fast-growing cotton colony. The microscopic structure observed is the sporangiophore, deep-brown columella, brown sporangiospores, and the rhizoid at the base of the sporangiophore. CR4 is identified as *Rhizopus oryzae* (Kwon et al. 2011). CR5 and CR7 are identified as *Fusarium* sp. as they produce oval to kidney-shaped microconidia (CR5) and sickle-shaped macroconidia with septation (CR7) (Hafizi et al. 2013; Bag et al. 2022). CR8 has a cream-to-brown colored colony at early growth and turns to deep brown later. CR8 has branched conidiophores, is elongated, multicellular, and has a characteristic spindle shape. The conidia have a brown or black appearance and are produced in clusters at the ends of the conidiophores. The septa within the conidia are dark and easily visible, providing a distinctive appearance. CR8 is identified as *Alternaria* sp. (Quintana et al. 2017; He et al. 2021).

The fungal contamination level on stored rice

Fungal isolation from rice grain samples collected from Maros and Makassar storage facilities was done using the spread-plate method. The samples were grounded and diluted up to 10^3. Each plate showing fungal growth from the 10^3 dilution series was selected for colony counting. The average number of the colony (CFU/g), contamination percentage, and moisture content are presented in the following tables (Tables 2 and 3).

The result of colony counting showed that mycoflora prevailed in all samples. The highest colony density in the Makassar sample was 9×10^3 CFU/g, and the lowest was 0.5×10^3 CFU/g (Table 1); meanwhile, for the Maros sample, the highest was 14.2×10^3 CFU/g, and the lowest was 4.0×10^3 CFU/g (Table 2). This contamination level is still under the maximum limit of fungal contamination standard for grains in Indonesia as per the recommendation of the National Standardization Agency, which is around 1×10^4 CFU/g. The average moisture content level ranges from 12-13%. The occurrence of fungal contaminant species is shown in the Figures 4 and 5:

Table 2. The fungal contamination and moisture content of stored rice from Makassar

Sample code	Average CFU/g	Average contamination (%)	Average moisture content (%)
Makassar 1	2.0×10^3	60.00	13.013
Makassar 2	2.5×10^3	86.67	13.153
Makassar 3	9.0×10^3	86.67	12.780
Makassar 4	3.5×10^3	96.67	13.127
Makassar 5	4.5×10^3	96.67	13.067
Makassar 6	0.5×10^3	76.67	13.100
Average	3.67×10^3	83.89	13.04

Table 3. The fungal contamination and moisture content of stored rice from Maros

Sample code	Average CFU/g	Average contamination (%)	Average moisture content (%)
Maros 1	4.0×10^3	93.33	13.013
Maros 2	11.0×10^3	66.67	13.153
Maros 3	10.0×10^3	100.00	12.780
Maros 4	4.0×10^3	83.33	13.127
Maros 5	14.2×10^3	90.00	13.067
Maros 6	6.7×10^3	83.33	13.100
Average	8.32×10^3	86.11	13.67

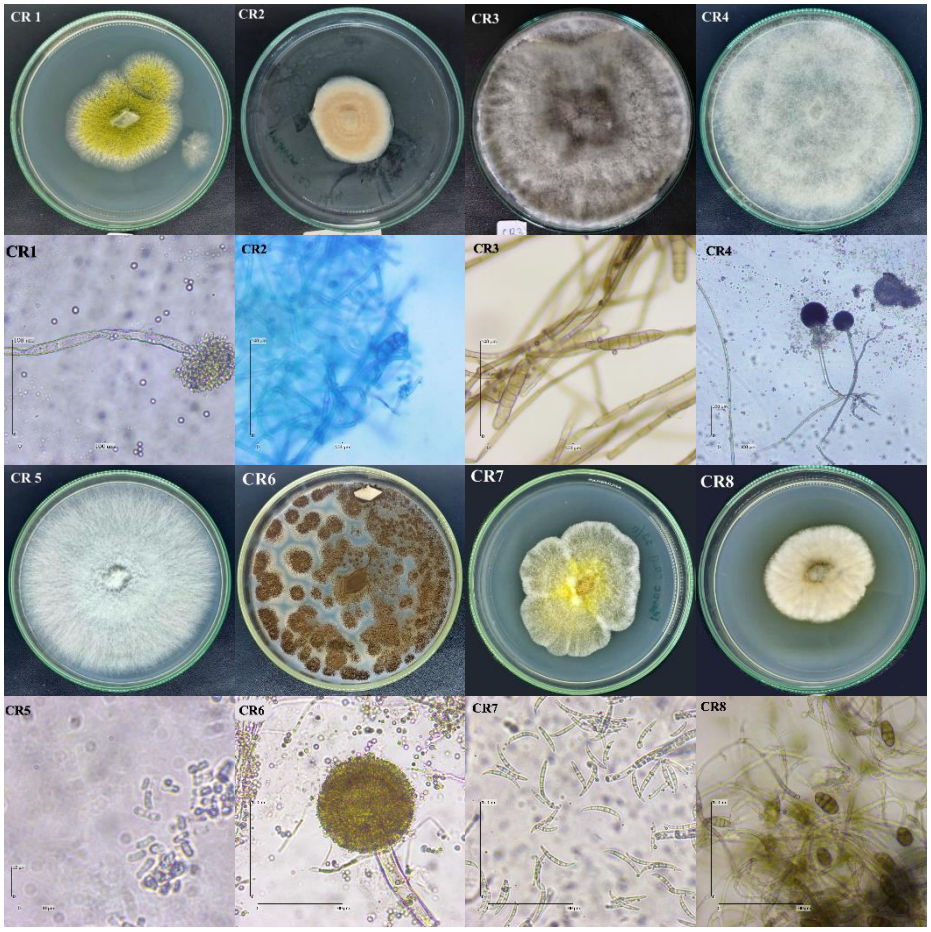

Figure 3. The colony and microscopic structures of representative isolates cultured from the Maros sample

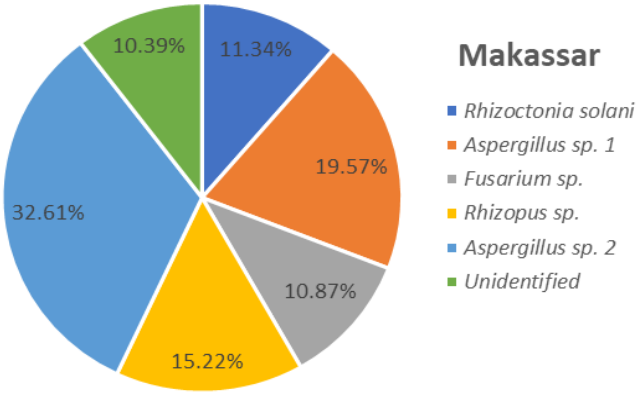

Figure 4. The species occurrence percentage of fungal associated with stored rice from Makassar

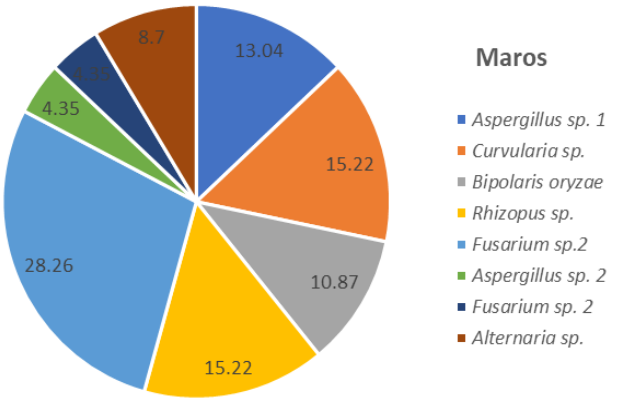

Figure 5. The species occurrence percentage of fungal associated with stored rice from Maros

Based on the percentage of species occurrence from Makassar (Figure 3), we found that the species with the highest occurrence is *Aspergillus* sp. 2 (32.61%), and the lowest is *Fusarium* sp. (10.87%). Meanwhile, for the samples from Maros (Figure 4), the mycoflora with the highest occurrence is *Fusarium* sp. 1 (28.26%), and the lowest is *Aspergillus* sp. 2 and *Fusarium* sp. 2 (4.35%). The mycoflora diversity of samples from Maros is higher than samples from Makassar; this result is in line with the fungal contamination level from both facilities.

Discussion

Storage fungi pose a great risk to postharvest loss and food safety, causing loss of germination, hot spot, color, flavor and decreased nutritional value (Mohapatra et al. 2017). This study collected the stored rice grain samples

from different storage facilities (Maros and Makassar). The storage facility in Maros is managed by local growers (public); meanwhile, the government manages the storage facility in Makassar. The average fungal contaminant in the stored rice samples from Maros (8.32×10^3 CFU/g) is higher than the samples from Makassar (3.67×10^3 CFU/g). However, the level of contamination in both types of storage facilities is still below the recommended maximum level of contamination for grains in Indonesia, regulated by the National Standardization Agency, which is 1×10^4 CFU/g. This result is possibly influenced by the different storage conditions and management. Rice storage in Makassar is considered a modern storage with standard roofs, walls, floor, ventilation, drainage channels, lighting, and entry-exit routes. Rice stored at the Makassar facility is required to comply with the national standard criteria, including milling degree (min. 95%), moisture content (max. 14%), grain appearances, and several other quality components. In addition, regular monitoring is carried out routinely every 14 days to check the rice quality component. Fumigation is also done as Pest Control Quality every three months. Meanwhile, the storage in Maros is considered conventional storage. According to our observation, the ventilation and organization of staple do not meet the required standards, as there was no space for air circulation between the staples and there is lack of regular monitoring to check the rice quality. Inadequate ventilation can lead to moisture build-up and temperature fluctuation within the storage area, affecting microbial activity. Stored grains continue to respire, consuming oxygen and releasing heat and moisture. In environments with poor ventilation, the heat and moisture released during respiration can become trapped, contributing to the formation of hot spots.

The contamination level of rice samples from both locations is high, with more than 60% of grain samples contaminated. Moisture content is known to be the primary factor in determining the kinds of fungi that invade stored grain and the degree to which they invade it (Paderes 1996). In the present study, the moisture content of stored rice grain ranges from 12-13%; this level is acceptable for short-term storage of less than one year. The circulation of rice stock in Makassar storage usually takes 6-12 months. Different storage periods need different levels of moisture content for safe storage. Some fungi can invade and overgrow other species, which may result in differences in growth rates between fungal species. Different fungal species require a certain moisture limit for their growth, which determines the degree to which they dominate (Rani et al. 2013; Mannaa and Kim 2017). Moisture content level also contributes to mycotoxin production by storage fungi. Higher water activity encourages fungi to produce mycotoxins (Daou et al. 2021).

The types of fungi associated with stored rice in this study are *Rhizoctonia solani*, *Bipolaris oryzae*, *Aspergillus* sp., *Fusarium* sp., *Rhizopus* sp., *Curvularia* sp., and *Alternaria* sp. A similar study on the characterization of fungi associated with rice grains from Lahore, Pakistan documented that the fungal contaminant is *Aspergillus flavus*, *Penicillium* spp., *Aspergillus fumigatus*, *Alternaria* spp., *Aspergillus niger*, and *Fusarium* spp. (Shanakht et al. 2014). Major fungal genera, including both pathogenic and saprophytic species, such as *Aspergillus flavus*, *Aspergillus niger*, *Penicillium* spp., *Fusarium* spp., *Alternaria* spp., *Curvularia* spp., *Rhizopus* spp., *Cercospora* spp., *Trichoderma* spp., *Chaetomium* spp., *Pyricularia oryzae*, *Helminthosporium oryzae*, *Sarocladium oryzae* are reported to be associated with rice grain discoloration in Eastern India (Raghu et al. 2020). Another study on the analysis of fungal community during rice storage through high throughput Internal Transcribed Spacer (ITS) showed that the main fungi communities are *Aspergillus* sp., *Fusarium* sp., *Rhizopus* sp., *Gibberella* sp., *Tilletia* sp., and *Penicillium* sp. (Li et al. 2022). Most of these fungi are known as storage fungi. Storage fungi predominantly grow and proliferate on stored grains, seeds, or other food materials in storage facilities. *Aspergillus* spp., *Penicillium* spp., and *Curvularia* spp. are known to be storage fungi; meanwhile, *Fusarium* spp. and *Alternaria* spp. are more known as field fungi (Mannaa and Kim 2017). The potential for storage fungi infestation and infection are serious concerns for food safety, as some storage fungi are known to be capable of producing mycotoxins (Mohapatra et al. 2017; Thathana et al. 2017; Qi et al. 2022; Laut et al. 2023). Major mycotoxin produced by storage fungi includes Aflatoxin B1, B2, G1, G2; Aflatoxin M1; patulin; Trichothecenes; Ochratoxin A; Zearalenone; and Fumonisin B1, B2, B3; and Ergot. Exposure to mycotoxin in animals and humans can lead to a toxic effect known as mycotoxicosis. Mycotoxicosis can be nephrotoxic, immunosuppressive, carcinogenic, and teratogenic. A related study found that the mycotoxin contamination profile of Pakistani rice poses a potential risk for the development of aflatoxin-induced Hepatocellular Carcinoma (HCC) in Pakistanis (Majeed et al. 2018). Generally, the major factors that affect mycotoxin production are temperature, active water, relative humidity, pH, fungal strain, and substrate (Daou et al. 2021).

Interestingly, in the present study, we found *R. solani* in the samples from Makassar and *B. oryzae* in the samples from Maros. *Rhizoctonia solani* is a known pathogenic fungus causing sheath blight on rice. *Rhizoctonia solani* can survive for extended periods in the soil, and infected plant debris or soil particles can adhere to the surface of seeds, potentially leading to transmission during planting or germination (Singh et al. 2019). *Bipolaris oryzae* is a seed-borne pathogen known as the causal agent of brown spots in rice; it is a hemibiotrophic parasite that can infect and remain dormant in the seeds, and the dormant inoculum will be reactive during seed germination (Sunder et al. 2014; Ackaah et al. 2023; Thuy et al. 2023). So far, there is no report of mycotoxin produced by these pathogens or any other risk related to human health. *R. solani* and *B. oryzae* are mainly field fungi that infect plants during production.

The storage facility is very important in minimizing the potential of storage fungi infestation and infection. Several factors affect fungal growth and their ability to germinate in storage, including moisture content, temperature, and active water; thus, they need to be monitored and controlled over the entire storage period. Long-term storage

requires lower moisture content. The required moisture content for rice grain/seed stored for 2 to 3 weeks is 14-18%, for 8 to 12 months is 12-13%, and for more than one year is less than 9% (IRRI 2010). Generally, storage fungi are capable of growing at a relative humidity of 70 to 90% and an optimum temperature of 25-35°C. Other factors that also determine safe storage are the physical condition and nutrient composition of the grain, inter-granular air level, microbial interactions between different species in a bulk system such as bacteria and yeasts, usage of inhibiting materials such as chemical preservatives, storage time, and the hygienic conditions (Mannaa and Kim 2017). Effective temperature and humidity monitoring in storage facilities are recommended for early-stage detection and effective intervention. Regular monitoring is recommended, and this may include visual assessments for any signs of damage, pest attack, or mold growth, in addition to microbiological and chemical assessments through periodically collecting representative samples from the bulk and assessing their quality parameters, including temperature, moisture content, and fungal contamination (Daou et al. 2021).

In conclusion, the mycoflora associated with stored grain in South Sulawesi is dominated by storage fungi, including *Aspergillus* spp., *Fusarium* spp., and *Curvularia* sp.; plant pathogenic fungi, including *R. solani*, *B. oryzae*, and *Alternaria* sp.; and cosmopolitan fungi of *Rhizopus* sp. The average contaminant from all samples is 5.995×10^3 CFU/g, which is still considered safe based on national standards on the maximum limit of fungal contamination for grains. Proper storage management practice is recommended to minimize the fungal contamination of the stored rice.

ACKNOWLEDGEMENTS

The authors would like to thank Muh. Ridha Taqwa Tang, Asri Ainun Amaliah, Mukhti Muhammad, and Ahmad Arisandi Jamal for their help in sample collection.

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