Chemical Control on Contamination Caused by Three Molds in Edible Mushroom Production

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This experiment aimed to test the effectiveness of four antifungal chemicals in controlling mold contamination in edible mushroom production. The antifungal chemicals were terbinafine hydrochloride, prochloraz, azoxystrobin, and sodium dichloroisocyanurate. The inhibitory effects of the chemicals were evaluated for inhibition on *Cladosporium* sp., *Aspergillus niger*, and *Neurospora* sp. The mycelia of the three molds and *Morchella sextelata* were cultured individually and co-cultured on plates with different concentrations of these chemicals, and then the mycelial growth was observed. By comparing the growth areas under the same conditions, the appropriate concentrations of each chemical were determined. The results indicated that terbinafine hydrochloride and prochloraz significantly inhibited the mycelial growth of all three mold species at certain concentrations, whereas their impact on the mycelial growth of *M. sextelata* was not significant. These results suggest that these two chemicals are effective in controlling the mycelial growth of the three molds, potentially increasing the yield and quality of *M. sextelata* and reducing mold contamination during storage and transportation.

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Keywords: Cladosporium sp.*; Aspergillus niger; Neurospora* sp.*; Terbinafine hydrochloride; Prochloraz*

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INTRODUCTION

Edible mushrooms are well marketed worldwide due to their fast growth, high nutritional value, unique flavor, and environmental friendliness (Hassett *et al*. 2015; Scholtmeijer *et al*. 2023; Chowdhury *et al*. 2024). In recent years, the edible mushroom industry in China has rapidly developed under the policy support of the rural revitalization and health industry demands (Li 2018). In 2022, the production of fresh edible mushrooms in China reached 42 million tons, with a total output value of 380 billion yuan (China 2024), ranking the first producer in the world and providing strong support for global agricultural economic development and food security (Bao *et al*. 2022).

However, molds can easily grow on mushrooms in the cultivation, storage, and processing stages, leading to food contamination, quality reduction, health risk, and potential economic loss (Adeoye *et al*. 2023). Currently, the most common methods involve removal or the use of broad-spectrum fungicides for treatment, but there is no dedicated agent that is both environmentally friendly and effective at inhibiting molds' growth (Bian *et al*. 2021). Additionally, there has been no attempt to apply human medications in agricultural production.

Cladosporium sp. is a saprotrophic fungus that can infect various substrates used in mushroom cultivation. It decomposes lignocellulose and reduces mushroom production

(Jin 2012; Bensch *et al.* 2015; Virginia *et al*. 2020; Park *et al.* 2008). As an opportunistic pathogen, it threatens human health by causing various skin and other human illnesses, such as allergic rhinitis and asthma (Yew *et al*. 2016; Marcelo *et al*. 2015).

Aspergillus niger, which is found widespread on grains and in soil, is one of the main contributors to agricultural mycotoxin contamination. It grows rapidly and results in degradation of lignin and hemicellulose (Zhao *et al*. 2024). This fungus is a major pathogen causing post-harvest rotting of edible mushrooms, especially during the storage and transportation stages of nitrogen-rich mushroom spawn bags and fruiting bodies (Song *et al*. 2013; Maribel *et al*. 2014).

Neurospora species are widely present in the natural environment and commonly associated with the cultivation materials like cottonseed hulls and corn cobs used for edible mushroom production (Kuo *et al*. 2014). At the suitable temperatures, such as 25 to 30 °C, its growth rate far exceeds that of edible mushrooms (Yin and Du 2024). Due to its rapid growth, it can infest an entire mushroom spawn bag in a few days. *Neurospora* species can spread the airborne conidia, rapidly dispersing throughout the cultivation greenhouse (Zhu 2013; Liu and Dong. 2022). Among common pathogens infecting edible fungi, *Neurospora* species cause the most severe damage to mushroom production, including yield reduction and complete crop failure (Feng *et al*. 2019; Zhang 2022).

M. sextelata is a highly valuable, soil-cultivated edible fungus (Zhang *et al*. 2024). It is well-known for its unique flavor and nutritional properties, making it an important mushroom in both economic and scientific research (Du *et al*. 2015). It is particularly prized for high protein content, essential amino acids, polysaccharides, and vitamins. These nutritional components contribute to their potential health benefits, such as anticancer properties and immune system enhancement (Han *et al*. 2019; Meng *et al*. 2019). In China, the cultivation area of morels reached 16,466 hm^2 in 2022. Unfortunately, approximately one-quarter of this cultivated area was affected by fungal diseases, resulting in significant losses (Tu *et al*. 2024).

As morels are valuable mushrooms and susceptible to fungal infections, *M. sextelata* was chosen as the experimental material in this study. The results obtained from this study will contribute to the development of control methods for mushroom disease prevention and management.

This study is aimed at effectively screening chemicals from human-use medications and farm chemicals to control edible mushroom contamination and diseases. Through co-cultivating with *M. sextelata* and the molds, it was compared with the mycelial growth of molds and edible mushrooms to find out which chemicals are of the most effective inhibition on the molds, but very little or no effect at inhibition on the mushrooms, while still being safe for humans and the environment.

EXPERIMENTAL

Materials

M. sextelata, used in this study as the testing mushroom, was provided by the Edible Fungi Engineering Center of Tongren Polytechnic College, Tongren City, Guizhou Province, China. *Cladosporium* sp., *A. niger*, and *Neurospora* sp. were isolated and purified from the mushroom spawn bags in the Cultivation Greenhouse. Terbinafine hydrochloride was purchased from Shanghai McLean Biochemical Technology Co., Ltd., China. Each milliliter of terbinafine hydrochloride contains the main ingredient Terbinafine

hydrochloride 0.01 grams and excipients: ethanol, 1,2-propanediol, purified water. Prochloraz was purchased from Qingdao Haina Biotechnology Co., Ltd, China. The concentration is 45%. Azoxystrobin was purchased from Shandong Zouping Pesticide Co., Ltd. Its active ingredient content is 0.25g/L. Sodium dichloroisocyanurate was purchased from China National Pharmaceutical Group Chemical Reagents Co., Ltd., China. Terbinafine hydrochloride is a medication used for human antifungal infections and can be taken orally (Tundisi *et al*. 2023; Zhang *et al*. 2024). Prochloraz, azoxystrobin, and sodium dichloroisocyanurate are agricultural chemicals registered officially in China, used in edible mushroom or vegetable production for inhibiting fungal diseases (Zhang and Bian 2013; Ministry 2017).

Methods

Identification of the pathogenic fungi and morel

Three common pathogenic fungi were isolated from edible mushroom spawn bags and subsequently purified for culturing. Both morphological and phylogenetic approaches were utilized to identify these pathogenic fungi and morel.

A fungal genomic DNA extraction kit was employed to extract the total DNA of the pathogenic fungi and morel. Using the total DNA as a template, the internal transcribed spacer (ITS) sequences were amplified with the universal primers ITS1/ITS4 to acquire the total fungal DNA. The amplified products were dispatched to Shengong Bioengineering (Chengdu) Co., Ltd., China for sequencing. A phylogenetic tree was constructed based on their ITS gene sequences, as shown in Fig. 1.

Based on the phylogenetic development tree, the following inferences can be made: A1 showed the highest homology with *Morchella sextelata* (MG431334.1). A2 showed the highest homology with *Cladosporium sphaerospermum* (NR111222.1). A3 showed the highest homology with *Aspergillus niger* (PP837990.1). A4 showed the highest homology with *Neurospora terricola* (OP597951.1). Combining these phylogenetic relationships with morphological observations (shown in Fig. 2), the identifications are as follows: A1 was identified as *M. sextelata*, A2 was identified as *Cladosporium* sp., A3 was identified as *A. niger*, and A4 was identified as *Neurospora* sp..

Screening of chemicals

After filtering the chemicals, 300 μL of the solution was transferred and spread onto a PDA plates. An equal amount of sterile blank plate was used as a control. Using a puncher, 6-mm diameter blocks from the PDA plates covered with the colonies of *M. Sextelata*, *Cladosporium* sp., *A. niger*, and *Neurospora* sp. were punched and inoculated onto the plates smeared with the chemical liquids and control plates. The pure culture and confrontation culture with *M. sextelata* were conducted simultaneously, with three replicates for each treatment. All the testing plates were cultured at 25 °C and observed for the colony growth.

Investigation of concentration

The test chemicals were diluted to 10, 100, 1000 and 10000 times from their original concentration to form gradient of dilutions. Each dilution was prepared to screen if chemicals effectively inhibit the mycelial growth of the molds and very little or no effect on the mycelial growth of *M. sextelata*. The same method was used in the chemical screening test to conduct pure culture and confrontation culture for observing the mycelial growth potential.

Data statistics

Once the plates were fully covered with the mycelia, a transparent paper with grid squares having dimensions 1 mm^2 was placed on the back of the plates to measure the mycelial growth area. Stata/SE 15.0 software (Stata Corp LLC, College Station, TX, USA) was used for the data processing and analyzing.

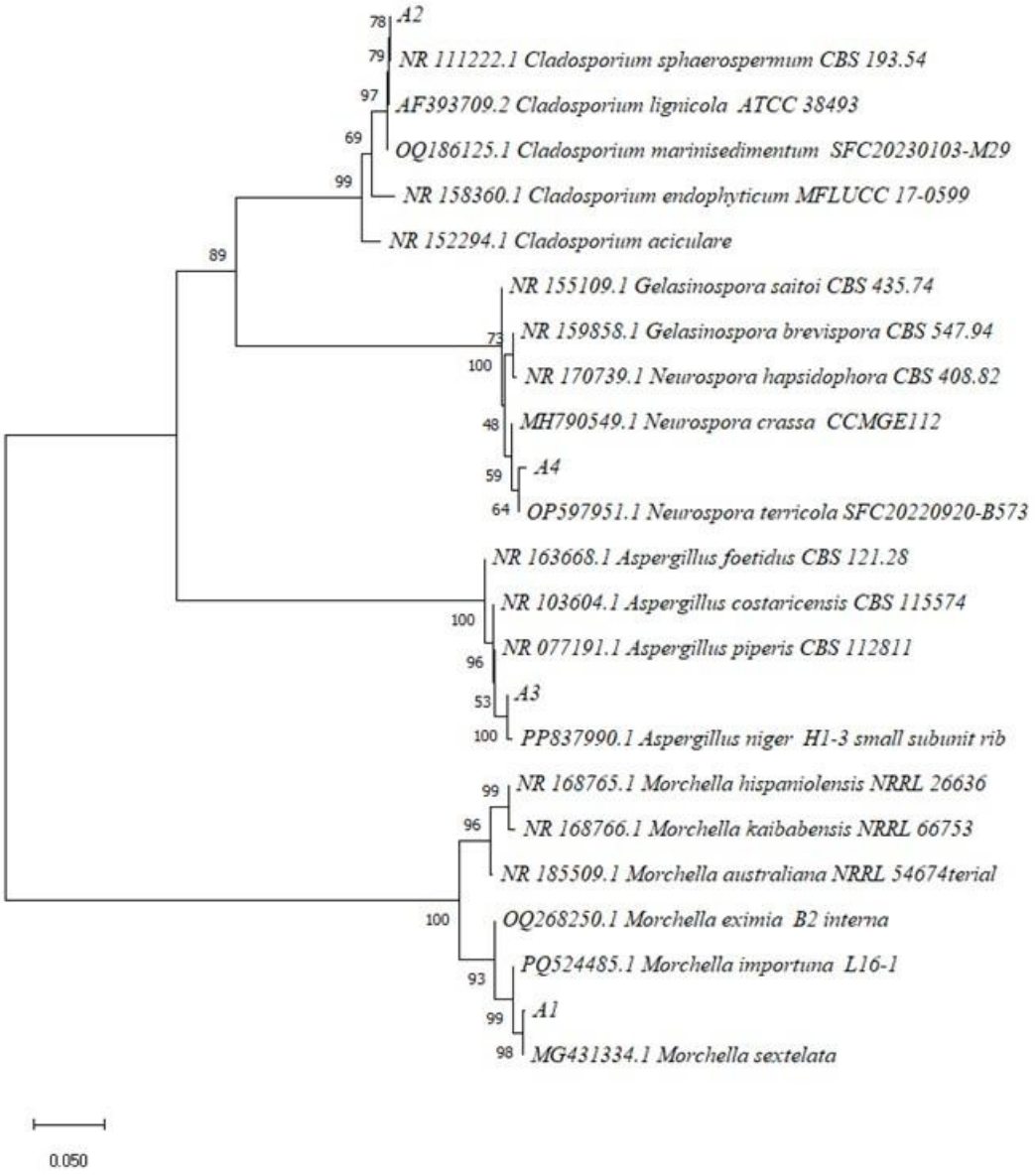

Fig. 1. Phylogenetic tree based on ITS sequence

RESULTS AND DISCUSSION

Morphological Identification of Pathogenic Fungi

Three purified fungi, namely *Cladosprium* sp., *A niger, and Neurospora* sp., were identified with microscopy, as shown in Fig. 2.

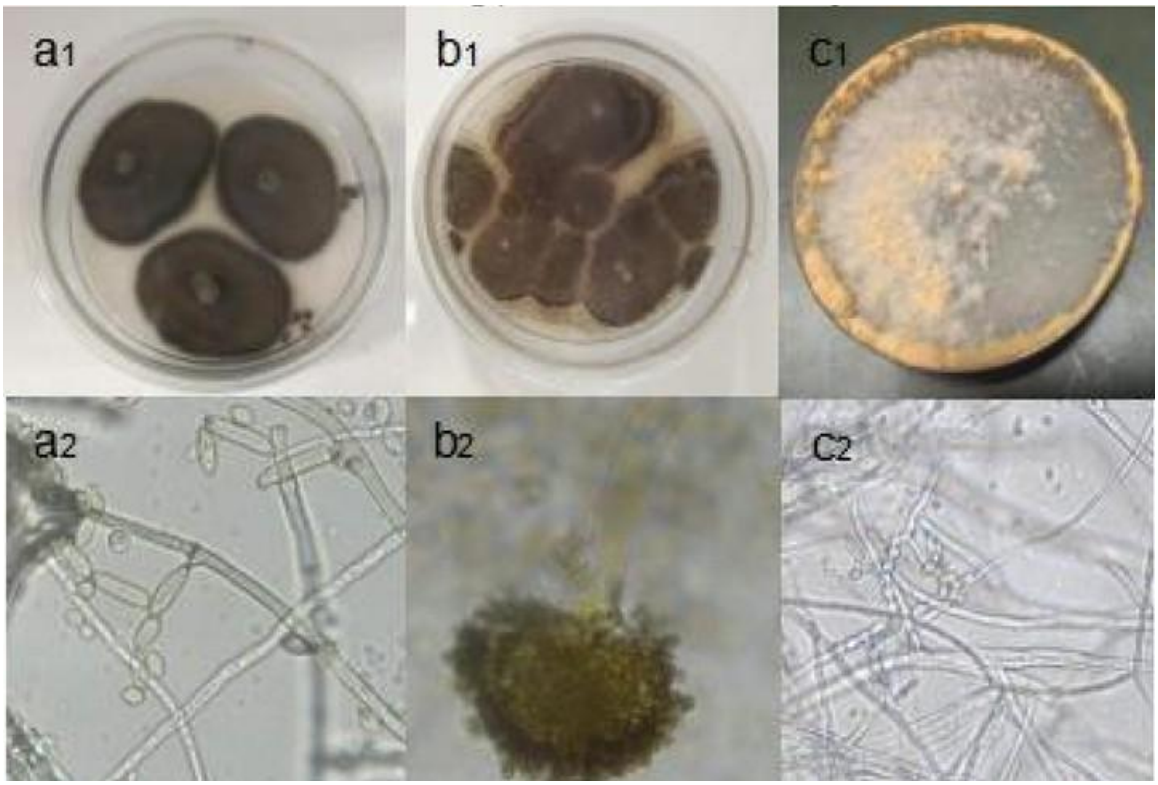

Fig. 2. Microscopic images of three molds
Note: a1-a2 *Cladosporium* sp., b1-b2 *A. niger*, c1-c2 *Neurospora* sp.

Screening Suitable Chemicals

Based on Table 1, in comparison with the control group (CK), the inhibitory effects of terbinafine hydrochloride and prochloraz on the growth of the three molds and *M. sextelata* showed highly significant differences ($P < 0.01$). In contrast, the overall differences between azoxystrobin and sodium dichloroisocyanurate were not significant. According to Table 2, under confrontation culture conditions, compared to *M. sextelata*, terbinafine hydrochloride and prochloraz significantly restrained the growth of *Cladosporium* sp., *A. niger*, and *Neurospora* sp. Specifically, terbinafine hydrochloride showed highly significant differences ($P < 0.01$) in inhibiting *M. sextelata* and *Cladosporium* sp., and significant differences ($P < 0.05$) in inhibiting *A. niger* and *Neurospora* sp. Under prochloraz treatment, there were highly significant differences ($P < 0.01$) in the inhibition of *M. sextelata* and the three molds. For azoxystrobin and sodium dichloroisocyanurate treatments, the inhibition of the three molds was not significant in comparison to *M. sextelata*. *Neurospora* sp. grew faster than *M. sextelata* in the confrontation culture ($P < 0.01$).

The mycelial growth of *Cladosporium* sp., *A. niger*, and *Neurospora* sp. was slow on the plates smeared with terbinafine hydrochloride and prochloraz solutions. *M. sextelata* was also inhibited by terbinafine hydrochloride and prochloraz solutions, but the mycelia remained robust. The confrontation culture showed that mycelia of *M. sextelata* significantly grew better than the ones of the three molds under the treatment of the two chemicals. Therefore, these chemicals could be used for mold control in mushroom production. Using these chemicals in edible mushroom production can effectively inhibit mold growth while having minimal impact on the growth of the edible mushroom,

maintaining economic losses at an acceptable level. In contrast, azoxystrobin showed only some inhibitory effect on *Cladosporium* sp., but has no effect on the other two molds, while sodium dichloroisocyanurate did not significantly inhibit the three molds.

Table 1. Results of Pure Plate Cultures with Different Chemicals

Chemicals	C	A	N	M
Th	0.67 ± 0.12^{Aa}	0.70 ± 0.10^{A}	14.00 ± 1.00^{Aa}	6.40 ± 0.53^{A}
Pr	0.63 ± 0.06^{Aa}	0.77 ± 0.15^{B}	2.50 ± 0.50^{Aa}	23.77 ± 2.25^{B}
Az	21.50 ± 1.50^{Ba}	55.40 ± 0.00^{Ca}	55.40 ± 0.00^{Bb}	40.53 ± 1.96^{C}
Di	25.17 ± 3.75^{Bab}	55.40 ± 0.00^{Ca}	55.40 ± 0.00^{Bb}	55.40 ± 0.00^{Da}
CK	29.67 ± 2.02^{Bb}	55.40 ± 0.00^{Ca}	55.40 ± 0.00^{Bb}	55.40 ± 0.00^{Da}

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). Th: Terbinafine hydrochloride, Pr: Prochloraz, Az: Azoxystrobin, Di: Sodium dichloroisocyanurate, C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*. CK is the control group

Table 2. Results of Plate Confrontation Cultures with Different Chemicals

Chemi-cals	M--C		M--A		M--N		Confrontation Culture P Value		
							M/C	M/A	M/N
Th	7.33 ± 0.58	0.62 ± 0.03	5.33 ± 0.58	2.83 ± 0.76	6.33 ± 1.53	3.33 ± 1.53	0.0013	0.0189	0.0175
Pr	16.67 ± 1.15	0.73 ± 0.06	15.33 ± 3.06	1.53 ± 1.29	14.00 ± 1.00	0.73 ± 0.23	0.0009	0.0051	0.0006
Az	32.90 ± 1.80	22.50 ± 1.80	25.83 ± 2.55	29.57 ± 2.55	14.83 ± 1.26	40.59 ± 1.26	0.0189	0.1665	0.0016
Di	28.75 ± 1.89	26.67 ± 1.89	28.20 ± 2.29	27.20 ± 2.29	13.07 ± 1.79	42.33 ± 1.79	0.2203	0.3709	0.0025

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). Th: Terbinafine Hydrochloride, Pr: Prochloraz, Az: Azoxystrobin, Di: Sodium dichloroisocyanurate. C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*

Terbinafine hydrochloride is a human medicine officially approved by the Chinese government. It can be applied directly (without further dilution) to prevent and control mold infections (Sui *et al*. 2023).

Prochloraz is a fungicide officially approved by the Chinese government. It is characterized by its high efficiency, broad-spectrum activity, and low toxicity. Molds cannot absorb it internally. For human safety, this fungicide should not be taken orally by humans and should be diluted more than 1000 times when used in mushroom cultivation (Hu *et al*. 2024).

It is recommended to spray these chemicals on the surface of the mushroom spawn bags or the soil surface to minimize direct contact with the mushroom mycelia.

Screening the Optimal Concentration

Table 3 shows that a 100-fold dilution of terbinafine hydrochloride exhibited a significant inhibitory effect on *Cladosporium* sp. and *Neurospora* sp. under pure culture

conditions ($P < 0.05$), while a 1000-fold dilution and lower concentrations achieved a highly significant inhibitory effect on *A. niger* ($P < 0.01$). Additionally, dilutions above 10-fold did not show a noticeable inhibitory effect on *M. sextelata*. The results from the confrontation culture in Table 4 indicated that, compared to the effect on *M. sextelata*, a 1000-fold dilution of terbinafine hydrochloride showed an extremely significant difference in inhibitory effect on *Cladosporium* sp. ($P = 0.0008$), and a 100-fold dilution of terbinafine hydrochloride also showed an extremely significant difference in inhibitory effect on *A. niger* ($P = 0.0005$). However, terbinafine hydrochloride did not show a significantly better inhibitory effect on *Neurospora* sp. in comparison to *M. sextelata* (shown in Fig. 3).

Under pure culture conditions, terbinafine hydrochloride exhibited inhibitory effects on all three types of molds and *M. sextelata*. In confrontation culture, terbinafine hydrochloride showed a significant difference in its inhibitory effect on *M. sextelata* and *A. niger*, but there was no significant advantage in its effect on *Neurospora* sp. Therefore, it was recommended to use terbinafine hydrochloride diluted 10 to 1000 times to control *Cladosporium* sp. and *A. niger* during edible mushroom cultivation, while dilutions within 100 times should be used for controlling *Neurospora* sp. outside the mushroom spawn bags.

Table 3. Pure Culture Results of Terbinafine Hydrochloride at Different Concentrations (original concentration 0.01g/mL)

Dilution Levels	C	A	N	M
10×	0.77 ± 0.21^{Aa}	0.7 ± 0.10^{Aa}	17.47 ± 2.25^{Aa}	7.00 ± 1.00^{A}
100×	2.00 ± 1.00^{Aa}	2.00 ± 0.00^{Ba}	32.90 ± 1.83^{Ab}	54.93 ± 0.81^{Ba}
1000×	6.00 ± 2.00^{Aab}	16.00 ± 7.21^{ABb}	55.40 ± 0.00^{Bc}	55.40 ± 0.00^{Ba}
CK	7.00 ± 2.00^{Ab}	55.40 ± 0.00^{C}	55.40 ± 0.00^{Bc}	55.40 ± 0.00^{Ba}

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*. CK is the control group

Table 4. Confrontation Culture Results of Terbinafine Hydrochloride at Different Concentrations (original concentration 0.01g/mL)

Dilution Levels	M--C		M--A		M--N		Confrontation Culture P Value		
							M/C	M/A	M/N
10×	1.77 ± 0.45^{A}	0.80 ± 0.00^{Aa}	7.00 ± 1.00^{Aa}	1.33 ± 0.58^{Aa}	7.50 ± 1.32^{Aa}	6.67 ± 1.53^{A}	0.0327	0.0117	0.0997
100×	32.23 ± 6.73^{Ba}	$3.00 \pm 2.00A^{Ba}$	25.00 ± 1.00^{Bb}	4.33 ± 0.58ABb	$7.17 \pm 1.76A^{ab}$	48.23 ± 1.76^{Ba}	0.0141	0.0005	0.0012
1000×	48.60 ± 2.16^{Bb}	6.00 ± 0.92^{Bb}	$33.43 \pm 7.49A^{Bb}$	15.83 ± 3.71^{Bc}	11.70 ± 1.35^{Ab}	43.70 ± 1.35^{Ba}	0.0008	0.0138	0.0012

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*

According to Table 5, under pure culture conditions, the 10,000-fold dilution of prochloraz showed significant inhibitory effects on all three molds ($P < 0.05$). As for *M. sextelata*, only within 1,000-fold dilutions showed inhibitory effects. Data from confrontation culture Table 6 revealed that the inhibitory effects of the 10,000-fold dilution of prochloraz between molds and *M. sextelata* reached highly significant differences ($P < 0.01$). This suggested that the 10,000-fold dilution of prochloraz could effectively inhibit *Cladosporium* sp., *A. niger*, and *Neurospora* sp., but its inhibitory effect on *M. sextelata* was not pronounced (shown in Fig. 4).

It should be recommended to use diluted prochloraz 10,000 times to control the three molds, *i.e.*, *Neurospora* sp., *Cladosporium* sp., and *A. niger*.

Table 5. Pure Culture Results of Prochloraz at Different Concentrations (Original Concentration 45%)

Dilution levels	C	A	N	M
100×	0.60 ± 0.00^{Aa}	0.60 ± 0.00^{ABa}	0.87 ± 0.23^{Aa}	4.37 ± 0.71^{A}
1000×	0.73 ± 0.23^{Aab}	1.33 ± 0.58^{Aa}	1.00 ± 0.00^{Aa}	20.00 ± 1.32^{B}
10000×	1.20 ± 0.72^{ABa}	1.50 ± 0.50^{ACb}	31.13 ± 8.83^{ABb}	55.40 ± 0.00^{Ca}
CK	7.00 ± 2.00^{ACb}	55.40 ± 0.00^{D}	55.40 ± 0.00^{Bc}	55.40 ± 0.00^{Ca}

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata.* CK is the control group

Table 6. Confrontation Culture Results of Prochloraz at Different Concentrations (original concentration 45%)

Dilution levels	M--C		M--A		M--N		Confrontation Culture P Value		
							M/C	M/A	M/N
100×	1.97 ± 0.15^{A}	0.60 ± 0.00^{Aa}	1.43 ± 0.51^{A}	0.60 ± 0.00^{Aa}	2.77 ± 0.68^{A}	0.60 ± 0.00^{Aa}	0.0021	0.0533	0.0157
1000×	17.67 ± 0.58^{BCa}	0.60 ± 0.00^{Aa}	11.67 ± 0.58^{B}	1.57 ± 0.75^{Ab}	14.33 ± 0.58^{BCa}	0.60 ± 0.00^{Aa}	0.0002	0.0001	0.0003
10000×	35.63 ± 4.73^{Cb}	0.93 ± 0.12^{Ab}	35.13 ± 3.20^{C}	1.20 ± 0.69^{Aab}	31.30 ± 4.39^{Cb}	10.67 ± 1.53^{B}	0.0031	0.0017	0.0055

Note: Capital letters indicate a highly significant difference ($P < 0.01$), while lowercase letters indicate a significant difference ($P < 0.05$). C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*

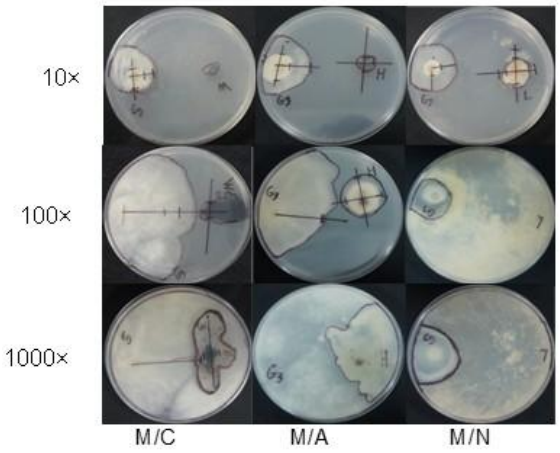

Fig. 3. Confrontation cultures of *M. sextelata* and three molds under terbinafine hydrochloride at different dilutions. Note: C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata.* On the left sides of plates were *M. sextelata* and on the right sides were molds

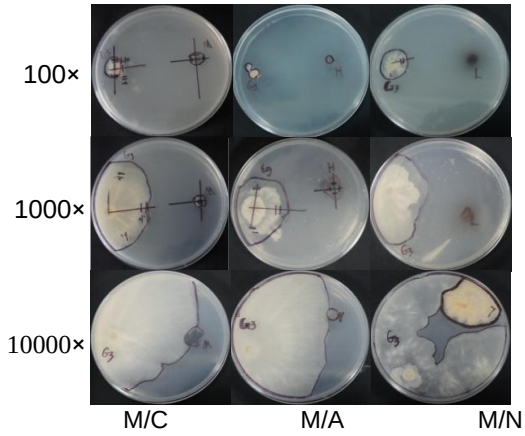

Fig. 4. Confrontation cultures of *M. sextelata* and three molds under prochloraz at different dilutions. Note: C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata.* On the left sides of plates were *M. sextelata* and on the right sides were molds.

Tables 3 and 5 show that terbinafine hydrochloride and prochloraz were effective chemicals to control the molds. Terbinafine hydrochloride is an acrylamide-based antifungal chemical, primarily used to treat fungal infections of the skin and other clinical applications. It is a low-toxicity, high-efficiency, and safe drug (Pharmacopoeia 2020; Yang *et al*. 2022). Currently, there are no reports on the use of terbinafine hydrochloride for the molds control in edible mushroom cultivation. Prochloraz is a broad-spectrum, highly effective, and low-toxicity imidazole fungicide. It is widely used due to its significant effectiveness in controlling storage diseases of fruits and vegetables, such as penicillium mold, anthracnose, and brown rot (Obianom and Sivakumar 2018; Hu *et al*. 2024). Prochloraz-manganese chloride complex (a complex of prochloraz and manganese) is highly praised in Western countries for its excellent antimicrobial effects (Bian *et al*. 2021). No morphological abnormalities were observed in the study. Which indicated that prochloraz also exhibited good inhibitory effects against various molds, including *Neurospora* sp. However, further research is needed to observe whether there are any

physiological changes. Therefore, direct contact with the mushroom mycelia should be minimized when applying these chemicals.

Under pure culture conditions, both terbinafine hydrochloride and prochloraz exhibited significant inhibitory effects on the three molds. However, under confrontational culture conditions (Tables 4 and 6), the inhibition effect of terbinafine hydrochloride dilution on *M. sextelata* was significantly higher than that on *Neurospora* sp., making it unsuitable for controlling *Neurospora* sp. infections. Nevertheless, a 1000-fold dilution of terbinafine hydrochloride could be effectively used for controlling *Cladosporium* sp. and *A. niger*. Meanwhile, using a 10000-fold dilution of prochloraz, *M. sextelata* was no longer inhibited, and the inhibition effects on the three molds reached highly significant levels. The effective control concentration of the chemicals on *Cladosporium* sp. and *A. Niger* still has room to further increase the dilution ratio. In the condition of maintaining ideal inhibitory effectiveness on the molds but no or low inhibition (economically within an acceptable level) on mushrooms, a study of the extreme dilution concentrations of terbinafine hydrochloride and prochloraz will be needed (shown in Figs. 5 and 6).

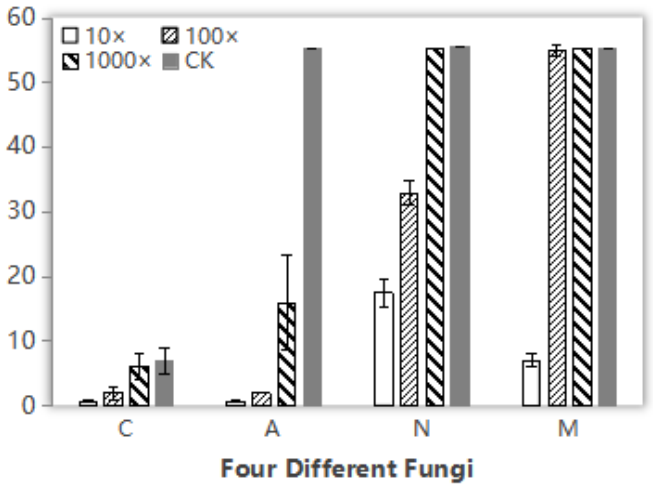

Fig. 5. Growth vigor of four fungal mycelia under terbinafine hydrochloride
Note: C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*

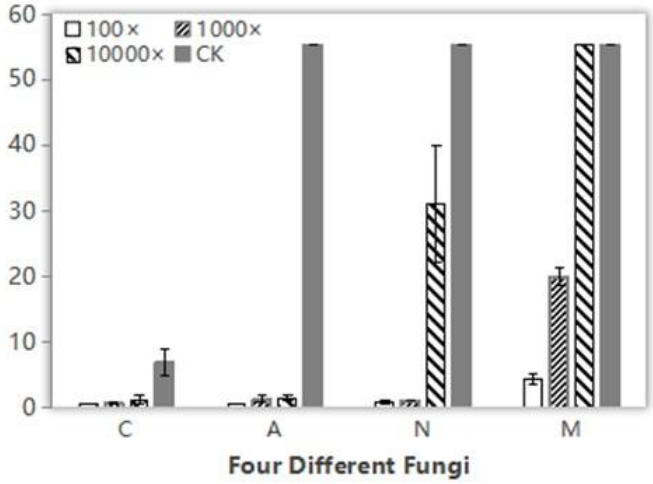

Fig. 6. Growth vigor of four fungal mycelia under prochloraz
Note: C: *Cladosporium* sp., A: *A. niger*, N: *Neurospora* sp., M: *M. sextelata*

Due to the significant harm and high control difficulty posed by *Neurospora* sp., it is crucial to combine chemical treatments with cultivation management. This includes pre-disinfection, timely removal of surrounding waste, and early application of treatments upon detecting infections to effectively control mold growth.

CONCLUSIONS

1. The 1000-fold dilution of terbinafine hydrochloride is recommended to control the diseases caused by *Cladosporium* sp. and *A. niger*.
2. The 100-fold dilution of terbinafine hydrochloride can be used to control the mold infection caused by *Neurospora* sp. on the mushroom spawn bags.
3. The 10000-fold dilution of Prochloraz is strongly recommended to control the molds caused by *Cladosporium* sp., *Aspergillus niger,* and *Neurospora* sp.

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