



## Screening of some filamentous fungi for cadmium tolerance in aquatic environments

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**Abstract:** Heavy metals (HMs), such as cadmium (Cd), ingress into the human body via the food chain through animal and plant agrochemicals, inducing epigenetic modifications, DNA damage, genetic mutations, and carcinogenesis. This investigation aimed to scrutinize and identify fungi tolerant to Cd toxicity in aqueous solutions using the poisoned food technique. Thirty fungal strains were cultured under three distinct concentrations of Cd (0, 100, and 300 mg/L), with subsequent quantitative evaluation of mycelial growth. Cluster analysis delineated two fungal groups comprising 19 and 11 strains. Group II demonstrated superior performance across most evaluated traits compared to Group I, with exceptions noted for biomass at 0 mg/L and stress tolerance index (STI) at 300 mg/L Cd. Consequently, Group II was deemed the superior cohort. Within Group II, *Epicocum nigrum* exhibited the highest biomass production (2.17 and 1.35 g/L, respectively) at both 0 and 100 mg/L Cd concentrations, alongside the highest STI (2.18 and 0.43, respectively) at Cd levels of 100 and 300 mg/L. Conversely, *Clonostachys rogersoniana* displayed the highest biomass production (0.46 g/L) at a Cd concentration of 300 mg/L, coupled with the lowest percentage of inhibition (PI) (-11.35 and -31.40%, respectively) at Cd toxicity levels of 100 and 300 mg/L. Hence, the 11 strains within Group II, particularly *E. nigrum* and *C. rogersoniana*, exhibit promise for further

investigation concerning their efficacy in Cd removal from aqueous solutions.

**Keywords:** Cluster analysis, fungi, heavy metal toxicity, inhibition rate, stress tolerance index.

### INTRODUCTION

Environmental pollution presents a significant global health challenge. The challenges posed by HM pollutants are further compounded by diverse industrial and agricultural activities on a worldwide scale (Xu et al., 2011). Effective strategies are imperative for removing HMs, either through separation and concentration or recovery and reuse. Among HMs, Cd, Ni, Cr, Pb, Hg, As, Fe, Cu, and Zn are harmful to humans due to their high toxicity, tendency to accumulate, and stability (Sud et al., 2008).

Cadmium (Cd), renowned for its high toxicity and mobility among HMs, is a by-product of Zn mining, smelting, and refining. Its exceptional significance arises from being 20 times more toxic to plants compared to the other metals. It exhibits an estimated average biological half-life of 18 years in the environment and 10 years within the human body (Salt et al., 1995). Extensive agricultural soils worldwide are contaminated with low to moderate Cd concentrations due to prolonged usage of phosphate fertilizers and sewage sludge (Vassilev et al., 2003). The typical Cd concentration in soil is approximately 1 µg/g, with toxicity thresholds ranging from 3 to 8 µg/g and critical limits in plants ranging from 5 to 30 µg/g (Kubier et al., 2019). HMs such as Cd permeate the human body through the food chain via animal and plant agrochemicals, leading to epigenetic alterations, DNA damage, genetic mutations, and carcinogenesis (Knasmüller et al., 1998). The human threshold for Cd absorption is 0.1 mg/kg per day (Satarug et al., 2017), with associated risks including kidney failure, hypertension, hepatic and pulmonary damage, bronchitis, atherosclerosis, genetic mutations, and cancer (Brahman et al., 2013; Davis et al., 2003).

Various methods including chemical, physical, electrochemical, and biological approaches have been employed to remove HM (Imre-Lucaci et al., 2011). Presently, biological absorption stands out as the most

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practical and effective method for decontaminating HM-laden wastewater and reducing metal concentrations therein. Biological absorption encompasses the uptake of ions by microorganisms, encompassing microbial leaching, bioaccumulation, bioabsorption, and enzymatic transformation (Soleimanifar et al., 2012). Microbial mechanisms involved in HM bioremediation play pivotal roles in anaerobic respiration reduction, detoxification, uptake, dissolution, and accumulation (Siddiquee et al., 2015). Genetic engineering and chemical modification enhance HM removal by modifying cell surface components and augmenting microorganism adsorption capacities (Zafar et al., 2007).

Microorganisms with high HM tolerance are typically selected for bioremediation, with numerous microbial species, including algae, bacteria, yeasts, and fungi, serving as bio-based adsorbents with significant absorption potential (Mungasavalli et al., 2007). Fungi, owing to their distinctive properties such as cation and anion binding to fungal cell walls, exhibit superior HM absorption capabilities (Chojnacka, 2010). Consequently, both living and deceased fungal cells possess notable capacities for absorbing toxic metals from aqueous solutions (Wang & Chen, 2009) and are frequently employed in HM recovery processes (Kacprzak & Malina, 2005). Notably, *Penicillium*, *Aspergillus*, and *Rhizopus* fungi have been extensively researched for HM removal and biotreatment in contaminated environments (Gomes et al., 2014; Rezaei et al., 2022; Bahari Saravi et al., 2022). For instance, a study explored the efficacy of three *Trichoderma* species (*T. asperellum*, *T. harzianum*, and *T. tomentosum*) in removing Cd ions, indicating significant potential for Cd ion reduction and bioremediation across varied pH and concentration conditions (Mohsenzadeh & Shahrokhi, 2014). Additionally, Yaghoobian et al. (2019) investigated the biological removal of Cd from aqueous solutions using filamentous fungi (*Piriformospora indica* and six strains of *Trichoderma* species), highlighting the high Cd tolerance and absorption capacity of *Trichoderma* spp., particularly *T. simmonsii*. The researchers suggested *Trichoderma* species, especially *T. simmonsii* (UTFC 10063), as potent bio-remediation agents in Cd-contaminated aqueous solutions (Yaghoobian et al., 2019). Furthermore, the bioremediation potential of *Phlebia brevispora* in removing Pb, Cd, and Ni was examined, demonstrating its applicability for HM bioremediation in industrial wastewater (Sharma et al., 2020). Previous studies have underscored the influence of various factors, including species type, HM type and concentration, pH, temperature, and biosorbent nature, on fungi-mediated HM bioremediation (Zaghir & Kakhki, 2021). For example, recent research revealed significant

increases in residual metal concentrations and absorption percentages at all levels by altering Cd and Ni concentrations (Zaghir & Kakhki, 2021). The exploration of novel fungal species holds immense potential for enhancing the efficacy, specificity, and sustainability of bioremediation strategies for HM removal from contaminated environments. This study aims to screen and identify fungi tolerant to cadmium toxicity in aqueous solutions.

## MATERIALS AND METHODS

To investigate the tolerance of 30 fungal strains to Cd toxicity, a laboratory experiment was conducted at Sari Agricultural Sciences and Natural Resources University (SANRU), Mazandaran, Iran.

### Fungal strains

The fungal strains used in this study were obtained from the Department of Plant Pathology and Genetics and the Agricultural Biotechnology Institute of Tabarestan (GABIT), SANRU, Sari, Iran. The strain names used are listed in Table 1. Fungal strains were grown on the sterile potato dextrose agar (PDA, Merck) medium at 28°C for 10 d.

### Fungal strains screening

To screen fungal strains for tolerance to Cd toxicity and to determine the appropriate concentration of Cd, 30 strains of different fungi were cultured in three concentrations of Cd (0, 100, and 300 mg/L) from the source of Cd chloride ( $\text{CdCl}_2 \cdot 2.5\text{H}_2\text{O}$ ). The Cd tolerance of the fungi was quantitatively assessed using the poisoned food technique (Dhingra and Sinclair, 1995). Potato dextrose broth (PDB, Merck) medium was prepared and distributed in a volume of 25 ml in 50 ml sterilized Erlenmeyer flasks. The desired concentration of Cd was achieved by adding appropriate quantities of  $\text{CdCl}_2 \cdot 2.5\text{H}_2\text{O}$  stock solution to the culture medium. The flasks were inoculated with 4-mm agar discs of 10-day-old cultures of fungal strains. The samples were then placed in a shaking incubator for 15 days at 28°C and a rotation speed of 120 rpm (Yaghoobian et al., 2019). Three replicate flasks were used per concentration. Then, the dry weight, inhibition rate, and stress tolerance index (STI) of the fungal strains at different Cd concentrations were assessed.

### Determination of biomass

Mycelial biomass (g/L of the culture medium) was used as a measure of fungal growth. After 15 days of incubation, the fungi were examined by measuring the dry weight of the biomass. To achieve this, biomass was harvested by filtering through filter paper (Whatman no.42), washed three times with deionized water, and incubated at 65 °C for 24 h. The

mycelial dry weight was measured using a digital scale with a sensitivity of 0.0001.

#### Calculation of fungal growth inhibition rate

The growth inhibition rate was expressed as the percentage reduction in fungal biomass compared with that in the control group. The inhibition rate was calculated using the equation described by Yaghoobian et al. (2019) (Equation 1), where the percentage of inhibition (PI) was determined using the fungal biomass (mg/L) in the control (0 mg/L Cd), culture media (C) and the fungal biomass (mg/L) in the Cd-amended culture media (T):

$$PI = \left[ \frac{(C-T)}{C} \right] \times 100 \quad (1)$$

#### Calculation of stress tolerance index

The stress tolerance index (STI) was calculated based on the dry biomass of the fungal strains under both stress and control conditions, utilizing the following equation:

$$STI = \frac{(Y_n \times Y_s)}{(\bar{Y}_n)^2} \quad (2)$$

where  $Y_n$ , biomass of fungal strains in control group (mg/L);  $Y_s$ , biomass of fungal strains under Cd toxicity (mg/L); and  $\bar{Y}_n$ , average biomass of all fungal strains under control conditions (Wen et al, 2023).

#### Statistical analysis

The obtained data were analyzed using the one-way ANOVA and means of treatments were compared by LSD test at the  $P \leq 0.05$  level by statistical analysis system (SAS) version 9.4. Based on the traits, a hierarchical clustering analysis using Euclidean distances and Ward's minimum variance method was used.

In this study, various fungi were cultured across three Cd concentrations: 0, 100, and 300 mg/L. Sari Agricultural Sciences and Natural Resources University (SANRU), Sari, Iran.

## RESULTS

In this study, various fungi were cultured across three Cd concentrations: 0, 100, and 300 mg/L. These microorganisms exhibited significant tolerance, surviving even at elevated Cd concentrations. However, distinct growth patterns and tolerance levels were observed among different species (Fig 1 & Table 2).

*B. dotidae* showed a notable decrease (94%) in biomass as Cd concentration increased to 300 mg/L.

*C. rogersoniana* exhibited a unique response, with a noticeable biomass increase (37%) from 0 to 300 mg/L Cd, albeit lower than most isolates under control conditions. *P. lilacinus* recorded maximum biomass (4.891 g/L) at 0 mg/L Cd but decreased at 100 mg/L (0.247 g/L) before slightly increasing at 300 mg/L (0.453 g/L). *B. dotidae* initially decreased in growth at 100 mg/L Cd but later increased at 300 mg/L.

*B. dotidae* exhibited the highest inhibition at Cd concentrations of 100 mg/L (98.11%) and 300 mg/L (94.64%). In contrast, *C. rogersoniana* displayed the lowest inhibition (-11.35% and -31.40%, respectively). *P. lilacinus* and *T. lixii* were identified as the most resistant and sensitive to Cd exposure, respectively (Table 2). After clustering analysis at 100 and 300 mg/L Cd levels, two distinct groups emerged: Group I comprising 19 fungal strains, and Group II comprising 11 fungal strains (Fig 2). Wilks' Lambda test confirmed significant differences between these groups ( $P < 0.001$ ), with a grouping accuracy of 96.7% (Table 3).

In inter-group comparisons, Group I produced more biomass under Cd-free conditions but exhibited higher inhibition rates when exposed to Cd (PI = 74.33% and 81.01% at 100 and 300 mg/L, respectively). Conversely, Group II displayed higher biomass production in the presence of Cd and a higher STI at 100 mg/L Cd. *E. nigrum* in Group II showed the highest biomass production and STI at both 0 and 100 mg/L Cd concentrations (Table 4). *C. rogersoniana* exhibited the highest biomass production at 300 mg/L Cd and the lowest PI at both Cd toxicity levels (Table 2).

This comprehensive analysis underscores the diverse responses of fungal strains to Cd toxicity, critical for understanding their potential applications in bioremediation or industrial processes.

## DISCUSSION

Thirty strains of various filamentous fungi underwent evaluation for biomass production under cadmium (Cd) toxicity utilizing the poisoned food technique. Our findings indicate a decrease in biomass production across all fungi with increasing Cd concentrations, highlighting species-specific tolerance variations to Cd toxicity. Similar observations were made by Liaquat et al. (2020), who investigated the growth of isolated fungi under elevated concentrations of Cd, Cr, and Pb. Previous studies have also documented the adverse effects of Cd and other HMs on fungal biomass production. However, numerous studies have demonstrated fungi's capability to adapt and flourish in high metal concentrations.

Cluster analysis identified two distinct groups of fungi, with Group II exhibiting superior performance

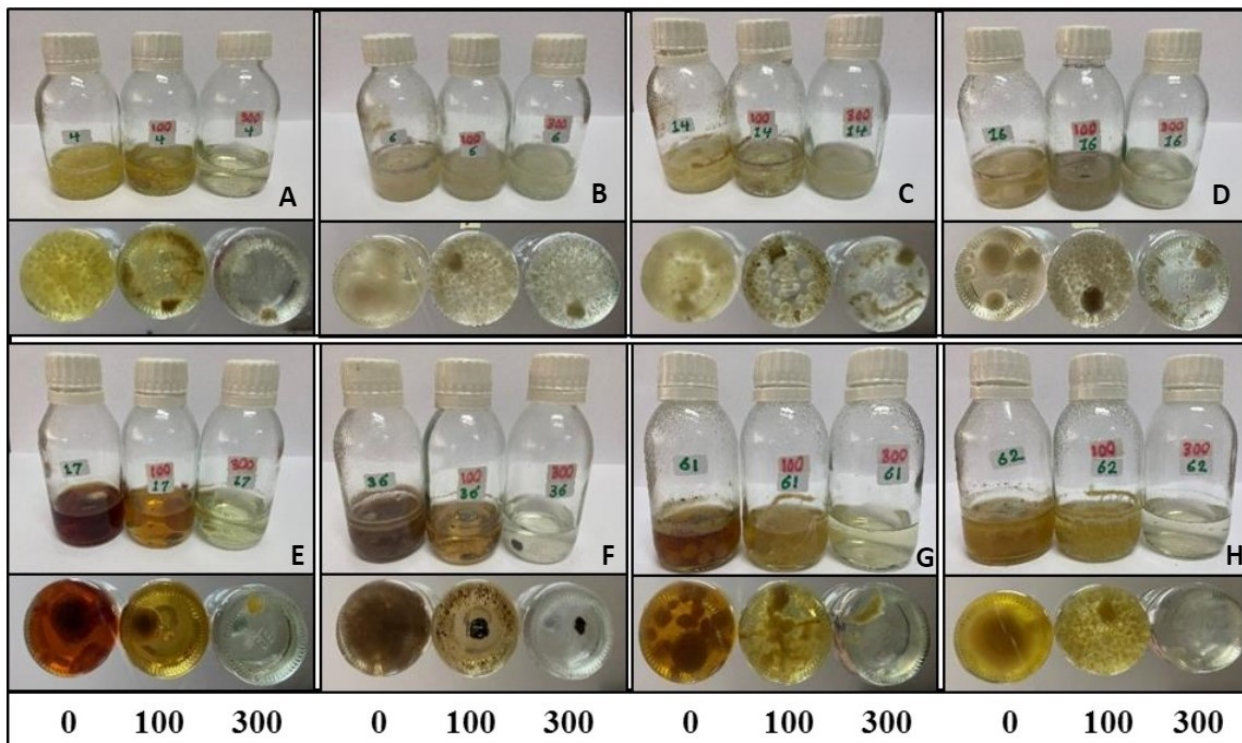
across most examined traits, except for biomass under Cd-free conditions and stress tolerance index (STI) at 300 mg/L Cd concentration. Consequently, Group II was deemed the superior group.

Within the 11 fungal strains comprising Group II, *Epicoccum nigrum* displayed the highest biomass production at both 0 and 100 mg/L Cd concentrations,

along with the highest STI when exposed to Cd levels of 100 and 300 mg/L. Conversely, *Clonostachys rogersoniana* exhibited the highest biomass production at a Cd concentration of 300 mg/L, concurrently displaying the lowest phytotoxicity index (PI) at both Cd toxicity levels.

**Table 1.** List of fungal strains examined in this study, sourced from the Department of Plant Pathology and the Agricultural Biotechnology Institute of Tabarestan (GABIT), Sari Agricultural Sciences and Natural Resources University (SANRU), Sari, Iran.

NO	Fungi strains name	NO	Fungi strains name
1	<i>Acrostalagmus luteoalbus</i>	16	<i>Paecilomyces marquandii</i>
2	<i>Alternaria tenuissima</i>	17	<i>Paecilomyces lilacinus</i>
3	<i>Aspergillus terreus</i>	18	<i>Paraconithyrium brasiliense</i>
4	<i>Botyophaeria dotidae</i>	19	<i>Penicilium pinophyllum</i>
5	<i>Ceriporia alachuana</i>	20	<i>Phoma medicagenis</i>
6	<i>Cladosporium perangustum</i>	21	<i>Phoma rabiei</i>
7	<i>Clonostachys rogersoniana</i>	22	<i>Phomopsis nobolis</i>
8	<i>Epicocum nigrum</i>	23	<i>Scytalidium thermophilum</i>
9	<i>Fusarium oxysporum</i>	24	<i>Trichoderma citrinoviride</i>
10	<i>Hypoxyylon investines</i>	25	<i>Trichoderma harzianum</i>
11	<i>Hypoxyylon sabmonticlosum</i>	26	<i>Trichoderma lixii</i>
12	<i>Lentisnigiacnus tigrinus</i>	27	<i>Trichoderma longibrachiatum</i>
13	<i>Metarhizium anisopliae</i>	28	<i>Trichoderma simmonsii</i> (UTFC10061)
14	<i>Montagnula sp.</i>	29	<i>Trichoderma simmonsii</i> (UTFC10063)
15	<i>Nigrospora sphaeria</i>	30	<i>Xylaria sinerea</i>



**Fig 1.** Growth pattern of fungal strains (A: *P. marquandii*, B: *P. lilacinus*, C: *C. rogersoniana*, D: *P. brasiliense*, E: *E. nigrum*, F: *H. investines*, G: *T. longibrachiatum*, H: *T. simmonsii*) in different cadmium concentration (0, 100, and 300 mg/l) in broth culture, 15 days after inoculation.

**Table 2.** Effect of cadmium concentrations on biomass production, percentage of inhibition (PI) and stress tolerance index (STI) of fungal strains in the broth culture.

Cluster groups	Fungi strain	Dry weight (g/l)			PI (%)		STI	
		0	100	300	Cadmium (mg/l)		100	300
					100	300		
I	<i>Scytalidium thermophilum</i>	0.515 <sup>ijk</sup>	0.176 <sup>de</sup>	0.135 <sup>e-j</sup>	65.40 <sup>a-h</sup>	75.59 <sup>a-f</sup>	0.070 <sup>cd</sup>	0.058 <sup>hij</sup>
	<i>Trichoderma lixii</i>	0.291 <sup>k</sup>	0.104 <sup>e</sup>	0.070 <sup>j</sup>	64.16 <sup>a-h</sup>	76.06 <sup>a-f</sup>	0.022 <sup>cd</sup>	0.015 <sup>l</sup>
	<i>Lentisnigiacnus tigrinus</i>	0.841 <sup>hij</sup>	0.227 <sup>de</sup>	0.131 <sup>e-j</sup>	62.60 <sup>a-h</sup>	83.30 <sup>a-d</sup>	0.119 <sup>cd</sup>	0.089 <sup>g-j</sup>
	<i>Trichoderma simmonsii (UTFC10063)</i>	0.528 <sup>ijk</sup>	0.202 <sup>de</sup>	0.105 <sup>hij</sup>	60.39 <sup>b-h</sup>	79.06 <sup>a-f</sup>	0.076 <sup>cd</sup>	0.039 <sup>ij</sup>
	<i>Xylaria sinerea</i>	1.210 <sup>fgh</sup>	0.509 <sup>bcd</sup>	0.183 <sup>d-j</sup>	55.26 <sup>c-i</sup>	82.64 <sup>a-d</sup>	0.474 <sup>bcd</sup>	0.158 <sup>c-i</sup>
	<i>Hypoxylon investines</i>	0.873 <sup>ghi</sup>	0.344 <sup>cde</sup>	0.124 <sup>fj</sup>	58.68 <sup>c-h</sup>	86.00 <sup>a-d</sup>	0.211 <sup>cd</sup>	0.081 <sup>hij</sup>
	<i>Aspergillus terreus</i>	2.118 <sup>bcd</sup>	0.646 <sup>bc</sup>	0.179 <sup>d-j</sup>	69.24 <sup>a-g</sup>	91.59 <sup>ab</sup>	1.001 <sup>b</sup>	0.281 <sup>cde</sup>
	<i>Penicillium pinophyllum</i>	0.444 <sup>ijk</sup>	0.075 <sup>e</sup>	0.111 <sup>e-j</sup>	71.95 <sup>a-g</sup>	67.21 <sup>a-f</sup>	0.015 <sup>d</sup>	0.033 <sup>ij</sup>
	<i>Trichoderma logibrachiatum</i>	0.457 <sup>ijk</sup>	0.124 <sup>e</sup>	0.169 <sup>d-j</sup>	73.06 <sup>a-f</sup>	63.75 <sup>c-g</sup>	0.043 <sup>cd</sup>	0.059 <sup>hij</sup>
	<i>Cladosporium perangustum</i>	1.159 <sup>fgh</sup>	0.337 <sup>cde</sup>	0.259 <sup>b-f</sup>	71.06 <sup>a-g</sup>	77.50 <sup>a-f</sup>	0.290 <sup>cd</sup>	0.220 <sup>d-g</sup>
	<i>Trichoderma simmonsii (UTFC10061)</i>	0.387 <sup>jk</sup>	0.101 <sup>e</sup>	0.093 <sup>ij</sup>	73.29 <sup>a-f</sup>	75.75 <sup>a-f</sup>	0.028 <sup>cd</sup>	0.026 <sup>ij</sup>
	<i>Nigrospora sphaeria</i>	0.532 <sup>ijk</sup>	0.201 <sup>de</sup>	0.145 <sup>d-j</sup>	70.05 <sup>a-g</sup>	72.61 <sup>a-f</sup>	0.104 <sup>cd</sup>	0.065 <sup>hij</sup>
	<i>Ceriporia alachuana</i>	1.306 <sup>fg</sup>	0.264 <sup>de</sup>	0.242 <sup>b-g</sup>	76.95 <sup>a-c</sup>	79.65 <sup>a-c</sup>	0.240 <sup>cd</sup>	0.230 <sup>def</sup>
	<i>Phoma medicagenis</i>	1.264 <sup>fgh</sup>	0.269 <sup>de</sup>	0.344 <sup>ab</sup>	78.41 <sup>a-c</sup>	72.95 <sup>a-f</sup>	0.247 <sup>cd</sup>	0.322 <sup>bcd</sup>
	<i>Phomopsis nobolis</i>	2.520 <sup>b</sup>	0.313 <sup>cde</sup>	0.233 <sup>b-h</sup>	87.00 <sup>a-d</sup>	90.34 <sup>abc</sup>	0.558 <sup>bc</sup>	0.415 <sup>bc</sup>
	<i>Phoma rabiei</i>	1.828 <sup>de</sup>	0.182 <sup>de</sup>	0.137 <sup>d-j</sup>	90.30 <sup>abc</sup>	92.64 <sup>a</sup>	0.252 <sup>cd</sup>	0.188 <sup>d-h</sup>
	<i>Alternaria tenuissima</i>	2.235 <sup>bcd</sup>	0.184 <sup>de</sup>	0.264 <sup>b-e</sup>	91.20 <sup>abc</sup>	87.29 <sup>a-d</sup>	0.289 <sup>cd</sup>	0.412 <sup>bc</sup>
	<i>Paecilomyces lilacinus</i>	4.891 <sup>a</sup>	0.247 <sup>de</sup>	0.453 <sup>a</sup>	95.05 <sup>ab</sup>	90.75 <sup>abc</sup>	0.911 <sup>b</sup>	1.638 <sup>a</sup>
	<i>Botryphaeria dotidae</i>	2.388 <sup>bc</sup>	0.083 <sup>e</sup>	0.137 <sup>d-j</sup>	98.11 <sup>a</sup>	94.64 <sup>a</sup>	0.067 <sup>cd</sup>	0.256 <sup>de</sup>
	II	<i>Paecilomyces marquandii</i>	0.307 <sup>k</sup>	0.277 <sup>de</sup>	0.196 <sup>d-j</sup>	10.80 <sup>ikl</sup>	36.50 <sup>g</sup>	0.063 <sup>cd</sup>
<i>Fusarium oxysporum</i>		0.205 <sup>k</sup>	0.186 <sup>de</sup>	0.119 <sup>e-j</sup>	19.14 <sup>i-l</sup>	39.30 <sup>g</sup>	0.034 <sup>cd</sup>	0.019 <sup>l</sup>
<i>Hypoxylon sabmonticlosum</i>		0.366 <sup>k</sup>	0.356 <sup>cde</sup>	0.164 <sup>d-j</sup>	1.10 <sup>kl</sup>	64.14 <sup>b-g</sup>	0.104 <sup>cd</sup>	0.057 <sup>hij</sup>
<i>Trichoderma citrinoviride</i>		0.449 <sup>ijk</sup>	0.201 <sup>de</sup>	0.192 <sup>d-j</sup>	46.81 <sup>e-j</sup>	51.51 <sup>fg</sup>	0.059 <sup>cd</sup>	0.060 <sup>hij</sup>
<i>Trichoderma harzianum</i>		0.478 <sup>ijk</sup>	0.206 <sup>de</sup>	0.208 <sup>c-i</sup>	53.75 <sup>d-i</sup>	51.56 <sup>fg</sup>	0.074 <sup>cd</sup>	0.071 <sup>hij</sup>
<i>Acrostalagmus luteoalbus</i>		1.369 <sup>ef</sup>	0.319 <sup>cde</sup>	0.198 <sup>d-j</sup>	51.95 <sup>d-i</sup>	60.35 <sup>d-g</sup>	0.285 <sup>cd</sup>	0.103 <sup>fj</sup>
<i>Paraconithyrium brasiliense</i>		1.218 <sup>fgh</sup>	0.429 <sup>b-d</sup>	0.334 <sup>abc</sup>	36.59 <sup>g-k</sup>	54.05 <sup>efg</sup>	0.265 <sup>cd</sup>	0.234 <sup>def</sup>
<i>Metarhizium anisopliae</i>		1.954 <sup>cd</sup>	0.793 <sup>b</sup>	0.198 <sup>d-j</sup>	45.65 <sup>e-j</sup>	88.10 <sup>abc</sup>	0.913 <sup>b</sup>	0.270 <sup>de</sup>
<i>Epicocum nigrum</i>		2.172 <sup>bcd</sup>	1.353 <sup>a</sup>	0.271 <sup>bcd</sup>	37.95 <sup>fj</sup>	87.51 <sup>a-d</sup>	2.176 <sup>a</sup>	0.432 <sup>b</sup>
<i>Montagnula sp.</i>		0.303 <sup>k</sup>	0.210 <sup>de</sup>	0.079 <sup>ij</sup>	31.94 <sup>h-k</sup>	71.50 <sup>a-f</sup>	0.048 <sup>cd</sup>	0.016 <sup>l</sup>
<i>Clonostachys rogersoniana</i>	0.336 <sup>k</sup>	0.381 <sup>cde</sup>	0.459 <sup>a</sup>	-11.35 <sup>l</sup>	-31.40 <sup>h</sup>	0.097 <sup>cd</sup>	0.119 <sup>fj</sup>	
<b>Significant</b>		**	**	**	**	**	**	**

\*\* Significant at the 1% probability levels. The same letter in each column indicates non-significant different according to LSD at 5% of probability.

**Table 3.** Detection function test for fungal strains under different concentrations of Cd by using Wilks' Lambda.

Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
I	0.290	30.327	7	0.000

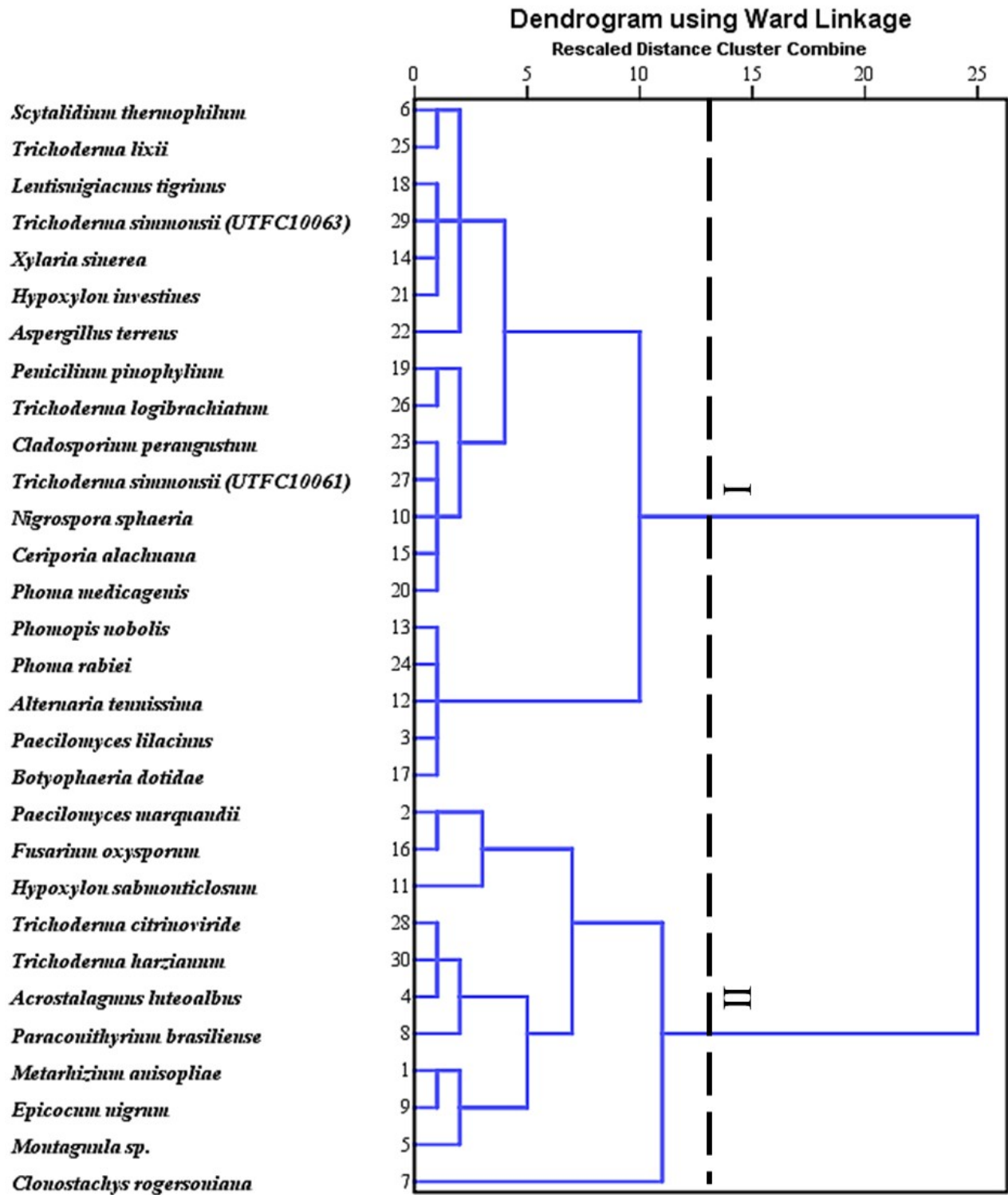


Fig. 2. The dendrogram obtained from the cluster analysis of fungal strains at 100 and 300 mg/L levels of Cd by using *Ward's minimum variance method*.



**Table 4.** Inter-group mean comparison of growth and tolerance traits of fungal strains under different concentrations of Cd (mg/l).

Groups	Dry weight (g/l)			PI (%)		STI	
	0	100	300	100	300	100	300
I	1.357 <sup>a</sup>	0.241 <sup>b</sup>	0.185 <sup>b</sup>	74.327 <sub>a</sub>	81.013 <sub>a</sub>	0.264 <sub>b</sub>	0.241 <sup>a</sup>
II	0.832 <sup>b</sup>	0.428 <sup>a</sup>	0.220 <sup>a</sup>	29.483 <sub>b</sub>	52.103 <sub>b</sub>	0.374 <sub>a</sub>	0.130 <sub>b</sub>
Sig.	**	*	ns	**	*	*	*

ns, \* and \*\*, non-significant and significant at 5% and 1% probability level, respectively.

In each column, the means with the same letter (s) did not differ significantly based on the LSD test at the 5% level of probability.

These findings suggest that the 11 strains in Group II, particularly *E. nigrum* and *C. rogersoniana*, merit further investigation regarding their potential in Cd removal from aqueous solutions. However, studies by Zafar et al. (2007) and Yagoubian et al. (2019) have indicated that there may not be a direct correlation between metal tolerance and the bioabsorption capacity of fungal isolates. Consequently, additional research is needed to explore this aspect comprehensively.

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## غربالگری قارچ‌های رشته‌ای متحمل به فلز سنگین کادمیوم در محیط‌های آبی

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**چکیده:** فلزات سنگین مانند کادمیوم (Cd) از طریق زنجیره غذایی وارد بدن انسان می‌شوند و باعث تغییرات اپی‌ژنتیک، آسیب به DNA، جهش‌های ژنتیکی و سرطان‌زایی می‌شوند. این پژوهش با هدف غربالگری و شناسایی قارچ‌های متحمل به سمیت کادمیوم در محلول‌های آبی با استفاده از روش غذای مسموم اجرا شد. بنابراین، ۳۰ جدایه قارچ در سه غلظت متفاوت کادمیوم (۰، ۱۰۰، ۳۰۰ میلی‌گرم در لیتر) کشت شده و میزان رشد میسلیوم به صورت کمی مورد ارزیابی قرار گرفت. در تجزیه و تحلیل خوشه‌ای (کلاستر)، دو گروه قارچ به ترتیب با ۱۹ و ۱۱ جدایه شناسایی شدند. گروه دوم در بیشتر صفات مورد بررسی، به جز زیست‌توده در غلظت صفر و شاخص تحمل تنش (STI) در غلظت ۳۰۰ میلی‌گرم در لیتر کادمیوم، بهتر از گروه اول عمل کرد. در نتیجه گروه دوم به عنوان گروه برتر انتخاب شد. در میان جدایه‌های گروه دوم، سویه *Epicocum nigrum* بالاترین تولید زیست‌توده (به ترتیب ۲/۱۷ و ۱/۳۵ گرم در لیتر) را در غلظت‌های صفر و ۱۰۰ میلی‌گرم در لیتر کادمیوم و همچنین بالاترین STI (به ترتیب ۲/۱۸ و ۰/۴۳) را در سطوح ۱۰۰ و ۳۰۰ میلی‌گرم در لیتر کادمیوم نشان داد. در حالی که *Clonostachys rogersoniana*، بالاترین تولید زیست‌توده (۰/۴۶ گرم در لیتر) را در سطح ۳۰۰ میلی‌گرم در لیتر کادمیوم و کمترین درصد مهار (PI) (به ترتیب ۱۱/۳۵- و ۳۱/۴۰- درصد) در هر دو سطح ۱۰۰ و ۳۰۰ میلی‌گرم در لیتر کادمیوم را نشان داد. بنابراین، ۱۱ سویه گروه دوم به ویژه *E. nigrum* و *C. rogersoniana*، پتانسیل ارزیابی بیشتر برای توانایی آن‌ها در حذف کادمیوم از محلول‌های آبی را دارند.

**کلمات کلیدی:** تجزیه خوشه‌ای، قارچ، سمیت عنصر سنگین، نرخ مهار، شاخص تحمل تنش