

**Original Article*****In vitro* Antifungal activity of aqueous and ethanol extracts of *Taraxacum officinale* leaf against seven toxigenic Fungi**

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ABSTRACT

This study evaluated the *in vitro* antifungal activity of aqueous and ethanol leaf extracts of *Taraxacum officinale* (dandelion) against seven toxigenic fungal species commonly associated with food and feed contamination. Using the disc diffusion method, both extracts demonstrated concentration dependent inhibition (100–400 mg/mL) against all tested isolates, including *Aspergillus flavus*, *A. fumigatus*, *A. parasiticus*, *Fusarium graminearum*, *F. proliferatum*, *F. verticillioides*, and *Penicillium chrysogenum*. *F. verticillioides* was the most susceptible isolate to both extracts, exhibiting the largest zones of inhibition (24.33 ± 0.03 mm for aqueous and 24.80 ± 0.06 mm for ethanol at 400 mg/mL), while *A. fumigatus* was the least susceptible. Although the commercial antifungal control produced significantly larger inhibition zones than either plant extract, the results confirm that *T. officinale* leaf extracts possess measurable antifungal properties, supporting their potential as bio-fungicidal agents for controlling mycotoxin-producing fungi in agricultural commodities.

Keywords: *Taraxacum officinale* (or Dandelion), Antifungal activity, Toxigenic fungi and Leaf extracts

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INTRODUCTION

Food and feed contamination by toxigenic fungi is a serious and long-recognized health concern, particularly in stored agricultural commodities. The main genera involved include *Aspergillus*, *Fusarium*, *Claviceps*, *Penicillium*, *Stachybotrys*, and *Alternaria*, many of which produce mycotoxins that cause

diseases collectively termed “mycotoxicoses”. Although mycotoxins are not essential for fungal growth, some secondary metabolites contribute to virulence, development, and pathogenicity [1, 2]. Even at low concentrations, mycotoxins can adversely affect humans and animals, with their proliferation favoured by environmental factors such as

humidity and temperature, as well as substrate composition [3].

Botanicals, including essential oils, spices, herbs, and crude plant extracts are increasingly explored as environmentally friendly, safer, and more affordable alternatives to synthetic fungicides for controlling fungi and mycotoxins in food and feed [4]. They can act synergistically as protectants against fungal and mycotoxin contamination and may stimulate natural defense pathways in plant tissues. Many botanicals contain phytochemicals with pharmacological properties, and recent studies have investigated their use as bio-fungicides and nutraceuticals to reduce toxigenic fungi and mycotoxin contamination [5].

Taraxacum officinale (dandelion) is a species of particular interest due to its rich nutrient profile (vitamins, minerals, polyphenols, flavonoids, fatty acids) and reported anti-inflammatory, antioxidant, anti-tumour, and antimicrobial activities. Plants and their extracts can compromise the toxic and genotoxic effects of mycotoxins [6] through multiple mechanisms: antioxidants scavenge free radicals and protect cell membranes; phytochemicals can disrupt fungal cell membrane permeability, inhibit cytoplasmic and mitochondrial enzymes, interfere with cell wall synthesis, and alter cellular osmotic and redox balance. Additionally, plant compounds may induce xenobiotic detoxification and biotransformation pathways, inhibit Phase I carcinogen-activating enzymes while inducing Phase II detoxification enzymes. Such bioactive plant compounds have been used as additives to prevent fungal growth and aflatoxin contamination, thereby reducing mutagenic and carcinogenic risks [7].

Despite growing interest, safety, toxicity, and quality concerns remain, including reports of mycotoxin contamination in medicinal plant materials and evidence that some traditional plant extracts can be toxic or mutagenic. Consequently, rigorous screening of toxicological properties is necessary when exploiting plant extracts for antifungal and nutraceutical applications [8]. The aim of the study is to assess the *in vitro* antifungal activity of aqueous and ethanol leaf extracts of *Taraxacum officinale* against seven toxigenic fungal species, as part of evaluating their potential as bio-fungicidal and nutraceutical agents.

MATERIALS AND METHODS

Preparation of plant extracts

The leaves were thoroughly washed with tap water to remove dirt, then dried under shade with optimal ventilation. After complete air-drying at room temperature, the leaves were ground and pulverized into powder using a pestle and mortar. Extraction followed a slightly modified method previously described by Okereke *et al.* [9]. Briefly, 150 g of the pulverized leaf powder was soaked in 1,000 mL of distilled water for 48 hours to prepare the aqueous extract. The mixture was filtered through muslin cloth and Whatman No. 1 filter paper, and the filtrate was dried in a water bath at 50°C until all water evaporated. The dried extract was stored at room temperature until further analysis.

Phytochemical screening

The aqueous leaf extract of *T. officinale* was qualitatively and quantitatively screened for naturally occurring biochemically active compounds using methods outlined by Banu and Catherine [10], with adaptations.

Test fungi

Seven toxigenic fungal species (presumably from genera such as *Aspergillus*, *Fusarium*, *Penicillium*, and *Alternaria*, were used as test organisms. Only colonies confirmed as toxigenic fungi were included in the study.

In vitro antifungal activity assay

Antifungal activity of the *T. officinale* leaf extract was evaluated using the disc diffusion method as described by Yaseen *et al.* [11]. All procedures were performed under strict aseptic conditions, and tests were conducted in triplicate to minimize experimental error. The diameter of the zone of inhibition around each disc was measured in millimeters. A commercially available antifungal drug was used as a positive control to compare the efficacy of the *T. officinale* aqueous extract.

Determination of minimum inhibitory concentration (MIC)

The MIC was determined using the standard methodology documented by Awang *et al.* [12]. The MIC is defined as the lowest concentration of extract that visibly inhibits fungal growth after incubation.

Determination of minimum fungicidal concentration (MFC)

The MFC was determined using the standard method described by Dahham *et al.* [13]. The MFC is the minimum concentration required to completely kill the fungi, assessed by sub-culturing from MIC wells/tubes onto fresh agar and observing for growth.

Data analysis

Data were analyzed using appropriate statistical software. Results were expressed as mean $\pm$ standard deviation. One-way analysis of variance (ANOVA) was used to compare means across treatment groups, with pairwise comparisons performed using Student's t-test. Differences were considered statistically significant at $p \leq 0.05$.

RESULTS

Table 1 shows that aqueous *Taraxacum officinale* leaf extract inhibited all seven fungal isolates, but its effect was generally concentration-dependent: inhibition zones increased as extract concentration rose from 100 to 400 mg/mL. However, the control produced the largest zone for every fungus, indicating that the control treatment had stronger antifungal activity than the plant extract under these test conditions. Thus, *F. verticillioides* was the most susceptible isolate to the extract, whereas *A. fumigatus* was the least susceptible at the highest extract concentration. For example, in *A. flavus*, the sequence e, d, c, b, a show that every increase in treatment level produced a statistically significant increase in inhibition. The same pattern occurs for *A. parasiticus*, *F. verticillioides*, and *P. chrysogenum*. The comparatively larger zones for the *Fusarium* species suggest that these isolates were more responsive to the aqueous leaf extract than the tested *Aspergillus* and *Penicillium* isolates. Differences may reflect variation in cell-wall structure, growth rate, sporulation, detoxification mechanisms, or sensitivity to particular plant compounds.

Table 1. Mean Diameter Zones of Inhibition (mm) of Different Concentrations of Aqueous *Taraxacum officinale* Leaf Extract Against Potential Toxigenic Fungi

Fungal Isolates	Extract Concentrations (mg/mL)				
	100	200	300	400	Control
<i>A. flavus</i>	10.73±0.29 ^e	12.13±0.18 ^d	13.67±0.35 ^c	15.10±0.36 ^b	17.40±0.25 ^a
<i>A. fumigatus</i>	11.70±0.26 ^c	12.63±0.23 ^b	13.23±0.09 ^b	13.87±0.20 ^b	20.73±0.26 ^a
<i>A. parasiticus</i>	12.67±0.12 ^e	13.43±0.19 ^d	14.60±0.21 ^c	15.73±0.32 ^b	18.30±0.12 ^a
<i>F. graminearum</i>	17.10±0.12 ^c	18.33±0.22 ^b	18.50±0.23 ^b	18.97±0.07 ^b	21.80±0.50 ^a
<i>F. proliferatum</i>	18.07±0.09 ^c	19.47±0.18 ^b	20.30±0.06 ^b	21.03±0.19 ^a	22.20±0.96 ^a
<i>F. verticillioides</i>	20.60±0.12 ^e	21.90±0.36 ^d	23.33±0.07 ^c	24.33±0.03 ^b	25.70±0.12 ^a
<i>P. chrysogenum</i>	11.37±0.18 ^e	12.67±0.19 ^d	13.50±0.23 ^c	14.37±0.18 ^b	17.63±0.22 ^a

Value are presented as mean ± standard error (SE) of three replicates. Means carrying different superscript letters within the same row are significantly different according to Duncan's multiple range test (DMRT) at $p \leq 0.05$.

Table 2 demonstrates that the ethanol extract of *Taraxacum officinale* leaves inhibited all seven tested fungi. Inhibition generally increased as the extract concentration rose from 100 to 400 mg/mL, although the control remained more effective than the extract for every isolate. The largest inhibition was recorded against *F. verticillioides*, while *A. fumigatus* showed the smallest inhibition at the highest extract concentration. *A. flavus*, *A. fumigatus*, *A. parasiticus*, *F. verticillioides*, and *P. chrysogenum* have different letters at all five treatment levels.

Therefore, each concentration, including the control, differs significantly from the others within those rows. In *F. graminearum*, the 300 and 400 mg/mL treatments is not statistically significant. *F. proliferatum*, the 300, 400 mg/mL was statistically comparable to the control for this isolate, despite the control having the numerically largest mean. The very small standard errors, particularly for *A. parasiticus* at 100 mg/mL and *F. verticillioides* at 400 mg/mL, indicate low variation among the three replicates.

Table 2. Mean Diameter Zones of Inhibition (mm) of Different Concentrations of Ethanol *Taraxacum officinale* Leaf Extract Against Potential Toxigenic Fungi

Fungal Isolates	Extract Concentrations (mg/mL)				
	100	200	300	400	Control
<i>A. flavus</i>	11.27±0.07 ^e	12.53±0.18 ^d	13.97±0.22 ^c	15.47±0.23 ^b	17.40±0.25 ^a
<i>A. fumigatus</i>	12.03±0.12 ^e	12.97±0.17 ^d	13.63±0.22 ^c	14.30±0.15 ^b	20.73±0.26 ^a
<i>A. parasiticus</i>	12.90±0.00 ^e	13.80±0.06 ^d	15.03±0.19 ^c	16.23±0.17 ^b	18.30±0.12 ^a
<i>F. graminearum</i>	17.50±0.15 ^d	18.50±0.15 ^c	18.87±0.03 ^b	19.43±0.03 ^b	21.80±0.50 ^a
<i>F. proliferatum</i>	18.33±0.15 ^c	19.83±0.03 ^b	20.80±0.06 ^a	21.50±0.15 ^a	22.20±0.96 ^a
<i>F. verticillioides</i>	21.17±0.09 ^e	22.37±0.28 ^d	23.83±0.06 ^c	24.80±0.06 ^b	25.70±0.12 ^a
<i>P. chrysogenum</i>	11.93±0.09 ^e	13.17±0.19 ^d	14.03±0.19 ^c	15.13±0.15 ^b	17.63±0.22 ^a

Value is presented as mean ± standard error (SE) of three replicates. Means carrying different superscript letters within the same row are significantly different according to Duncan's multiple range test (DMRT) at $p \leq 0.05$.

DISCUSSION

The findings demonstrate that both aqueous and ethanol extracts of *T. officinale* leaves exhibit broad-spectrum antifungal activity against all seven toxigenic fungal isolates tested. Inhibition was consistently concentration dependent, with zones of inhibition increasing significantly as extract

concentrations rose from 100 to 400 mg/mL for most isolates. This pattern aligns with previous reports that dandelion extracts contain bioactive phytochemicals including flavonoids, phenolic acids, sesquiterpene lactones, and polyphenols that disrupt fungal cell membranes, inhibit enzymatic activity, and interfere with cell wall synthesis [5].

Ethanol extracts generally produced slightly larger inhibition zones than aqueous extracts across most fungal species, likely due to better solubility and extraction efficiency of hydrophobic bioactive compounds (e.g., flavonoids and terpenoids) in ethanol. This observation is consistent with other studies showing that alcoholic extracts of *Taraxacum* species often demonstrate superior antimicrobial activity compared to water-based extracts [14].

Notably, *Fusarium* species (*F. graminearum*, *F. proliferatum*, and *F. verticillioides*) were markedly more susceptible to both extracts than *Aspergillus* and *Penicillium* species. *F. verticillioides* consistently showed the largest inhibition zones, while *A. fumigatus* was the most resistant isolate. This differential sensitivity may reflect variations in cell wall composition, growth rates, sporulation patterns, or detoxification mechanisms among fungal genera. For instance, *Fusarium* species may possess cell wall structures or metabolic pathways that render them more vulnerable to the specific phytochemicals present in dandelion leaf extracts. The comparatively lower activity against *Aspergillus* species, particularly *A. fumigatus*, is consistent with earlier reports that dandelion extracts show inconclusive or weak activity against certain *Aspergillus* strains. This suggests that while *T. officinale* extracts may be effective against some toxigenic fungi, their utility as standalone antifungal agents may be limited against more resistant species.

In all cases, the commercial antifungal drug used as a positive control produced significantly larger inhibition zones than either plant extract at any concentration

tested. For example, against *F. verticillioides*, the control yielded 25.70 ± 0.12 mm compared to 24.33 ± 0.03 mm (aqueous) and 24.80 ± 0.06 mm (ethanol) at 400 mg/mL. This indicates that while *T. officinale* extracts possess antifungal properties, they are less potent than conventional synthetic antifungals under the tested conditions [15]. However, the moderate activity observed supports the potential use of dandelion extracts as complementary or adjunct treatments, particularly in contexts where synthetic fungicides are undesirable due to environmental, safety, or cost concerns. Botanicals are increasingly explored as safer, more affordable alternatives that can act synergistically with other control measures and stimulate natural defense pathways in plant tissues.

Given that the tested fungi are known producers of mycotoxins (e.g., aflatoxins, fumonisins, trichothecenes), the antifungal activity of *T. officinale* extracts suggests potential applications in reducing fungal contamination and mycotoxin accumulation in stored agricultural commodities. Plant-derived compounds can mitigate mycotoxin risks through multiple mechanisms, including direct antifungal action, antioxidant protection, and induction of xenobiotic detoxification pathways (Powers *et al.*, 2019). Nevertheless, the study also highlights important caveats. Safety, toxicity, and quality concerns remain regarding the use of plant extracts, as some traditional botanicals have been reported to contain mycotoxins or exhibit toxic/mutagenic properties. Rigorous toxicological screening is therefore essential before *T. officinale* extracts can be recommended for widespread nutraceutical or bio-fungicidal applications.

This study was limited to in vitro evaluation using disc diffusion assays; further research should include determination of minimum inhibitory concentration (MIC) and minimum fungicidal concentration (MFC) values to quantify potency more precisely. Additionally, chemical characterization of the active compounds in both aqueous and ethanol extracts would help identify the specific phytochemicals responsible for antifungal activity.

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