Online ISSN: 2682-2628
Print ISSN: 2682-261X

IJCBR

International Journal of Cancer and Biomedical Research

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EACR EGYPTIAN ASSOCIAN FOR CANCER RESEARCH
Since 2014

International Journal of Cancer & Biomedical Research
(IJCBR) https://jcbr.journals.ekb.eg

IJCBR is an Int. journal published by the Egyptian Society of Cancer Research (EACR, established in 2014, http://eacr.tanta.edu.eg) and sponsored by the Egyptian Knowledge Bank (EKB: www.ekb.eg).

IJCBR has been approved by the Supreme Council of Universities, Egypt with score 7 (http://egjournal.scu.eg). The journl is cited by google scholar and registered by Publons (https://publons.com). The journal has recently been evaluated in 2020 by Nature Springer with a good standing.

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INTERNATIONAL JOURNAL OF CANCER AND BIOMEDICAL RESEARCH (IJCBR)

RESEARCH ARTICLE

Evaluation of the anti-fusarium effect of ethanolic extracts of *Cinnamoum zeilanicum*, *Berberise vulgaris* and *Caluna vulgaris*

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ABSTRACT

Background: Mycotoxins such as Fumonisin B **(FB)**, are low-molecular-weight natural products produced as secondary metabolites by filamentous fungi. The mycotoxins, FB, contaminates grain products and acts as a virulence factor for host cells. **Aim**: This study aimed to evaluate the effect of ethanolic crude extracts of Cinnamuom zeilanicum (CZ), Berberise vulgaris (BV) and Calluna vulgaris (CV) on the growth of corn seed's fungi and /or mycotoxin production. **Materials and methods**: Firstly, corn seeds were collected and cultured on water agar media and the grown fungi were inoculated into Sabouraud's dextrose media, then the toxins were identified. Secondly, the dried plants of CZ, BV and CV were phytochemically screened for alkaloids, phlobatannins, saponnins, flavonoids, steroids, terpenoids and cardiac glycosides. The fungus purified DNA was sequenced, and BLAST alignment was used to identify the fungi and mycotoxin types. Furthermore, the effect of different plant extract treatments on fungi growth was assessed. Finally, toxin production was assessed using HPLC. **Results**: The obtained results confirmed that Fusarium isolate was Fusarium sporotrichioides. Among the three tested extracts, Cinnamuom zeilanicum extract was the most potent inhibitor for Fumonisin B production followed by Berberise vulgaris and Calluna vulgaris. **Conclusion**: Cinnamuom zeilanicum extract could be used during corn seed long-term packaging and storage to inhibit (FB) production and toxicity.

Keywords: Corn seeds, Fumonisin B1, Fusarium sporotrichioides, Mycotoxins, zearalenone

Editor-in-Chief: Prof. M.L. Salem, PhD - Article DOI: 10.21608/JCBR.2020.30493.1039

ARTICLE INFO

Article history

Received: May 17, 2020
Revised: July 2, 2020
Accepted: July 28, 2020

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INTRODUCTION

Mycotoxins are toxic compounds that are naturally produced by certain species of fungi, these metabolites constitute a toxicologically and chemically heterogeneous assemblage that is grouped only because the members can cause disease and death in humans and other vertebrates. The majority of human mycoses are caused by opportunistic fungi (Janbon et al., 2019).

The food supply chain is a crucial element for all livestock production systems, according to the European Feed Manufacturers' Federation, within the European Union-, approximately 475 million tons of feedstuffs and forages are consumed by livestock each year. Fungi on crops produce mycotoxins in the field, during handling, and in storage (Wu, 2004). Mycotoxins exhibit a variety of biological effects such as liver and kidney toxicity, central nervous system abnormalities, estrogenic responses and others (Omotayo et al., 2019). Mycoses are frequently acquired via inhalation of spores from an environmental reservoir or by unusual growth of commensal species that is normally resident on human skin or the gastrointestinal tract. The majority of mycotoxicoses result from eating contaminated foods, or exposure through consumption of contaminated food (Omotayo et al., 2019).

Fumonisins (B1 and B2) are cancer-promoting metabolites of Fusarium proliferatum and Fusarium verticillioides that have a long-chain hydrocarbon unit (similar to that of sphingosine and sphinganine) which plays a role in their toxicity (Braun et al., 2018). Fumonisin B1 (FB1) is the most toxic and appears to promote tumours in rats and cause equine leukoencephalomalacia and porcine pulmonary edema (Alwan.,2011). In humans, Fusarium species cause a broad spectrum of infections, including superficial (such as keratitis and onychomycosis), locally invasive, or disseminated infections, with the last occurring almost exclusively in severely immune-compromised patients (Nucci and Anaissie, 2007). Fumonisins appear to inhibit sphingosine synthesis and may promote carcinogenesis by acting as indirect or direct antagonists to sphingosine's cytostatic action (Kamle et al., 2019).

Many drugs are currently available in Western medicine, originally isolated from plants, plant-derived compounds represent an untapped source of safe, effective, and environmentally friendly antimicrobials. The properties of various plant-derived antimicrobials that target cellular viability of microbes have been adequately discussed previously (Anand et al., 2019). Flavonoids with the most potent complement inhibitory activity found in those plants are afzelin and quercitrin (De-Eknamkul et al., 2015). The combination of ethanolic extract of cinnamon bark and honey against Propionibacterium acnes and Staphylococcus epidermidis showed additive activity with a fractional inhibitory concentration index (FICI) value of 0.625. Therefore, the combination of cinnamon bark extract and honey has potential activity against acne-causing bacteria (Julianti et al., 2017). Berberise vulgaris extract and decoctions are traditionally used for their activities against a variety of microorganisms including bacteria, viruses, fungi, protozoa, helminthes, in Ayurvedic, Chinese, and Middle-Eastern folk medicines (Rahimi-Madiseh et al., 2017). Caluna vulgaris has been used in ethnopharmacology as antiseptic, antibacterial, cholagogue, diuretic, expectorant, antirheumatic, antioxidant, antitumor and anti-inflammatory agent (Rodrigues et al.,2018). Therefore, our study aimed to explore the potential anti-fungal effect of Cinnamuom zeilanicum, Berberise vulgaris and Calluna vulgaris crude extracts as well as their effect on mycotoxin production in fungi isolated from corn seeds.

MATERIALS AND METHODS

Materials

Corn seeds were obtained from Department of Microbiology, Faculty of Science, Mansoura University, Egypt. Seeds were collected, stored at 4˚C and then kept for one week at room temperature before being used. Berberis vulgaris roots, Calluna vulgaris shoot and Cinnamuom zeilanicum parks were obtained from Botany Department, Faculty of Science, Alexandria University, Egypt. Ethanol (95%) was purchased from El-Nasr Pharmaceutical Chemicals Co., Egypt, and agarose from Sigma-Aldrich, USA. Easy-RED^{TM} total RNA Extraction kit from INtRON Biotechnology, Korea. Yeast Extract from Oxoid, England, Agar from Scharlau, Spain and Folin Reagent from Lobachemie, India. All other chemicals and reagents were of highest quality available commercially.

Methods

Isolation of fungus from corn seeds. Isolation of the fungus was performed as described in Aryal (2015). In brief, corn seeds with very poor storage conditions, were separately grown on water-agar media (20 grams agar dissolved in one litre distilled water and supplemented with 10% NaCl for 10 days at 25^{o}C. The grown fungus was isolated and transferred into potato dextrose agar (PDA) media (200 g potato, 20 g dextrose and 20 g agar in one-litre distilled water) and kept at 25^{o}C for 12 days.

Extraction of plants active ingredients (Redfern et al., 2016). For each plant, the dried collected part was exhaustively defatted with petroleum ether for three days at room temperature then dried in air. One litre absolute ethanol was added to 250 g of this dried plant and was shacked for three days at 60 rpm and 37ºC. Ethanolic extract was collected by filtration and concentrated till dryness using a rotary evaporator to obtain a crude extract powder.

Recovery and detection of mycotoxins. For aflatoxin and ochratoxine determination, AflaTest was used (Trucksess and Scott, 2008). briefly, contaminated corn seeds (50 gm) were ground, blended with five grams of sodium chloride and 100 ml methanol:water (80:20) for one minute. Blended mixture was then filtered and 10 ml of the filtrate was diluted with 40 ml distilled water (or PBS for Ochratoxin) then filtered on microfiber filters, 10 ml of the filtrate was allowed to pass through AflaTest® column with a flow rate of 1-2 drops/second until air comes through column (Trucksess and Scott, 2008). Then washed with 10 ml distilled water (or 0.1 % Tween – PBS for Ochratoxin) a second wash was performed using 10 ml distilled water. Finally, 1.5 ml of methanol was added using a syringe to the column with a flow rate of 1 drop /second and the elute was collected in a cuvette and was read in fluorimeter with excitation and emission wavelengths of 365 nm and 440 nm, respectively.

Fungus identification. Fungus (100 mg) was frozen in liquid nitrogen then ground. The powder was then transferred into a 1.5 ml microcentrifuge tube containing 350 µl of Lysis solution (Strelau et al., 2011). A Sample was vortexed for 10-20 seconds to mix thoroughly then 50 µl of Lysis solution B and 20 µl RNaseA were added. The sample was then incubated for 10 min at 65°C. Precipitation solution (130 µl) was added and mixed by inverting the tube 2-3 times. Samples were incubated 5 min on ice and centrifuged for 5 min at 14,000 rpm 4°C. The supernatant was collected and transferred to a clean microcentrifuge tube. Then (400 µl) gDNA Binding Solution and 96% ethanol (400 µl) were added to supernatant and mixed well.

The Thermo Scientific GeneJET Plant Genomic DNA Purification Mini Kit was used for the purification of high-quality genomic DNA. The Internal Transcribed Spacer (ITS) regions of fungal ribosomal DNA (rDNA) were amplified using PCR using the following primers; ITS_1 (Forward primer): 5'TCCGTAGGTGAACCTGCGG3', ITS4 (Reverse primer): 5'TCCTCCGCTTATTGATATGC3'. The amplified DNA was purified and cleaned up using silica membrane column. The purified (ITS) was sequenced using the BigDye ™ Terminator v3.1 Cycle Sequencing Kit (Matousková et al., 2015) according to the manufacturer instructions.

Finally, the Blast sequence analysis tool was used to identify the obtained sequence and was submitted to GenBank.

Qualitative screening. Dried plants were screened for alkaloids, phlobatannins, saponnins, flavonoids, steroids, terpenoids and cardiac glycosides according to the methods described in Ayoola et al. (2008) and the determination of plant extracts phenolic compounds were performed by HPLC was performed as described by Seal (2016).

Antifungal effects of tested plants' extracts. An inoculum of the grown fungus was cultured in 35 ml of Sabouraud's dextrose media (20 g glucose, 10 g yeast extract and 10 g peptone in one litre distilled water) at 25°C for 12 days as then the fungi were grown at the surface of the media and produced mycotoxines in the liquid media. The floating fungi were separated from liquid media and identified, while the media was used for measuring toxin concentration using HPLC and for the in vivo tests. The antifungal effect of plants' extracts was done according to Huynh et al. (1984). Equal amounts of F. sporotrichioides (about 3 inoculums) were mixed with 35ml Sabouraud's dextrose media as (+ve control) In another flask, different concentrations of Dimethyl sulfoxide, DMSO, (0.5, 1, 1.5 and 2%) were added to 35 ml Sabouraud's dextrose media as (-ve control) and then different concentrations of each tested plant extracts (0.5, 1, 1.5 and 2% were dissolved in DMSO (1%) and added to 35ml Sabouraud's dextrose media which was treated with F. sporotrichioides (about 3 inoculums) and all flasks were left for ten days until complete growth. The effect of different plant treatments on fungi growth was documented by photography, and F. sporotrichioides toxin production was assessed by using HPLC assay (Liu et al.2005).

Statistical analysis

Data were collected and analyzed using IBM SPSS software package version 20. Quantitative data were described using mean and standard deviation. Comparison between the studied groups was analyzed using F-test (ANOVA), that

followed by Student Newman-Keuls test. The differences were considered statistically significant at p values < 0.05.

RESULTS

Fungi sequence alignment

The Internal Transcribed Spacer (ITS) region of fungal ribosomal DNA from our strain was sequenced and the obtained sequence (Supplementary Figure S1) was given accession number (KX808233.1) by gene bank, it was classified as Fusarium sporotrichioides with a 93% identity to Fusarium proliferatum **(**Supplementary Figure S2**).** The phylogenetic tree of obtained nucleotide sequence is shown in Figure 1.

```
  1 ggaactactc aatggtgggg cagtctagag gatggagcag gaggaaatag tagccgcgga
 61 ggacccccgc tcccggtaaa acgggacggc ccgccagagg acccctaaac tctgtttcta
121 tatgtaactt ctgagtaaaa ccataaataa atcaaaactt tcaacaacgg atctcttggt
181 tctggcatcg atgaagaacg cagcaaaatg cgataagtaa tgtgaattgc agaattcagt
241 gaatcatcga atctttgaac gcacattgcg cccgccagta ttctggcggg catgcctgtt
301 cgagcgtcat ttcaaccctc aagcccagct tggtgttggg actcgcgagt caaatcacgt
361 tccccaaatt gatttgcggt cacgtccagc ttccatagcg tcatcgtcaa accctagtta
421 cttgtaaacg tcgcggccac acccataaac ccaacttagg aaagttgacc agggatcggg
481 aatcaaaacc cgctcaaaga aacctccacg agccgagata gcatccgttc gtcatattgc
541 taagctccgc agcagggtga acagacgact cctcctcttg ctgagctccg cctccggttc
601 aacgggtccc ccctcagatt acttaaaccc tcctcaagtt agacacttga agttaacaac
661 atattaaaat cagactttaa cccggcacct ttggattcgg cctcaataaa taagcatcat
721 aatgcgaatc aataatgagt aattaaaaaa ttatagtaaa caacaaaact ttaaaaccaa
781 ttgcaccgcc aattttatgg ggggcatcct ggattataag agatttaccc ccacccccct
841 agtggttgag acaaacggaa ataggaaact cccccaacta aaatgggggg gaagttaaaa
901 tttatttgg
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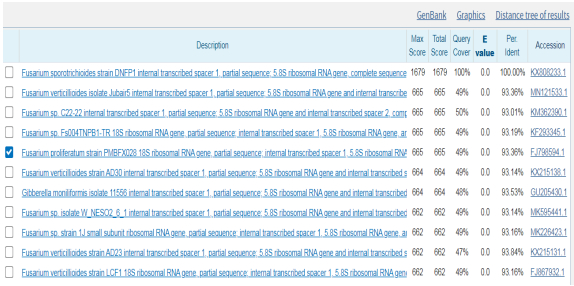
Supplementary Figure S1. Nucleotide sequence of the Internal Transcribed Spacer (ITS) region of fungal ribosomal DNA coding for 18S ribosomal RNA.



	Description	Max Score	Total Score	Query Cover	E value	Per. Ident	Accession
☐	Fusarium sporotrichioides strain DNFP1 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene, complete sequence	1679	1679	100%	0.0	100.00%	KX808233.1
☐	Fusarium verticillioides isolate Jubair5 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribe	665	665	49%	0.0	93.36%	MN121533.1
☐	Fusarium sp. C22-22 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed spacer 2, comp	665	665	50%	0.0	93.01%	KM362390.1
☐	Fusarium sp. Fs004TNPB1-TR 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene, ar	665	665	49%	0.0	93.19%	KF293345.1
☑	Fusarium proliferatum strain PMBFX028 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA	665	665	49%	0.0	93.36%	FJ798594.1
☐	Fusarium verticillioides strain AD30 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed s	664	664	49%	0.0	93.14%	KX215138.1
☐	Gibberella moniliformis isolate 11556 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed	664	664	48%	0.0	93.53%	GU205430.1
☐	Fusarium sp. isolate W_NESO2_6_1 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed	662	662	49%	0.0	93.14%	MK595441.1
☐	Fusarium sp. strain 1J small subunit ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene, a	662	662	49%	0.0	93.16%	MK226423.1
☐	Fusarium verticillioides strain AD23 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed s	662	662	47%	0.0	93.84%	KX215131.1
☐	Fusarium verticillioides strain LCF1 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5.8S ribosomal RNA gene	662	662	49%	0.0	93.16%	FJ867932.1

Supplementary Figure S2. GeneBank alignment data

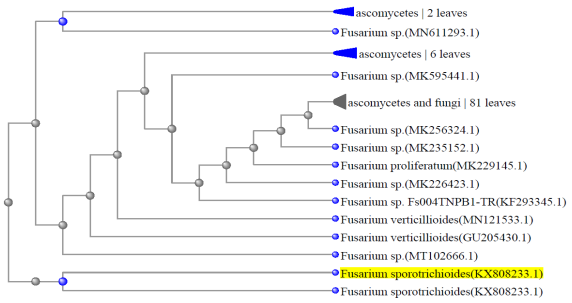


Figure 1. The phylogenetic tree of the obtained nucleotide sequence of *Fusarium sporotrichioides* (accession number: KX808233.1). This tree was produced using BLAST pairwise alignments.

Qualitative phytochemical screening of the plant extracts

Table 1 shows that all tested plant extracts contained alkaloids, flavonoids and terpenoids. Cinnamuon zeilanicum and calluna vulgaris extracts contained tannins and phobataninns. Berberis vulgaris and Cinnamon zeilanicum extracts contained Saponins and Cardiac glyceroids.

Quantitative phytochemical screening

HPLC analysis of the polyphenolic compounds of different plants ethanolic extracts and their concentrations (Table 2). The characteristic component(s) for each extract was detected like, Cinnamic Acid and Cinnamaldhyde for C. zeilanicum (7.910 and 44.92 mg/g extract, respectively), Berberine (0.62 mg/g extract) for B. vulgaris and 3,4 Dicaffeoylquinic acid as well as 3,5- Dicaffeoylquinic Acid for C. vulgaris (5.472 and 52.942 mg/g extract, respectively). Furthermore, it was found that 4,5-Dicaffeoyl Quinic Acid, Quercetin, Phenol, 2-Methyl-5-(1-Methylethyl) and Benzaldhyde were present in C. zeilanicum only. C. zeilanicum contained the highest concentration of chlorogenic acid, caffeic acid and lacked gallic acid, retinol and tanic acid. B. vulgaris had highest concentration of gallic acid (90.7530 mg/g extract) while, C. vulgaris had highest levels of 3,5-Dicaffeoylquinic Acid, 3,4 Dicaffeoylquinic acid and Retinol (8.90 mg/g extract).

Effect of plant crude extracts on fungus growth

Neither C. vulgaris L, nor B. vulgaris ethanolic extracts caused an inhibition in the fungi growth at any of the tested concentrations, however. C. zeilanicum ethanolic extract inhibited Fusarium sporotrichioides growth at concentrations starting from 1% (Figure 2).

Toxin concentration after treatment with the three plant extracts

Deoxynivalenol and aflatoxins were not detected in growth media of F. sporotrichioides, On the other hand, zearalenone produced by F. sporotrichioides was highly reduced by treatment with 2% Cinnamuom zeilanicum (0.568±0.005 ng/ml) and moderately reduced by 2% *Berberise vulgaris* (1.562±0.009 ng/ml) while *Calluna vulgaris* had no significant effect.

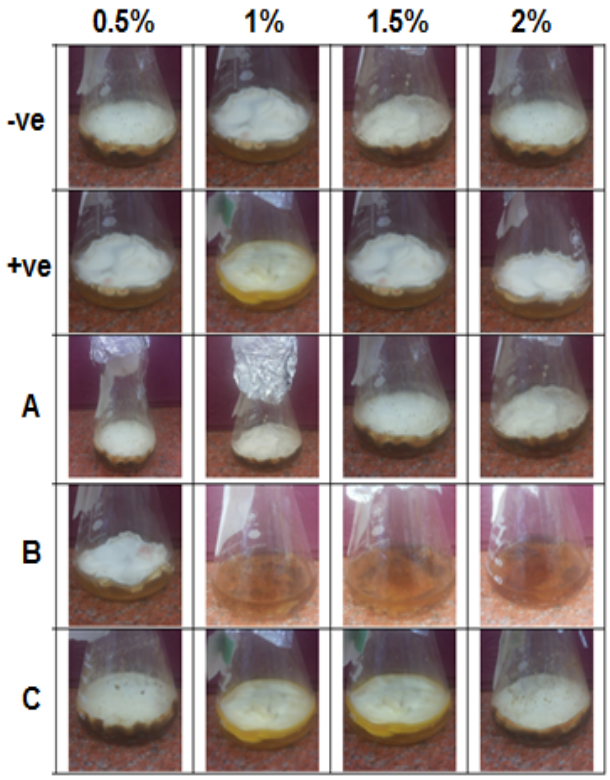


Figure 2. Antifungal effects of different concentrations (0.5%, 1.0%, 1.5%, 2.0%) of plant extracts on *Fusarium sporotrichioides*; Negative control (with DMSO) and positive control (media only) are shown; (A) *Berberis Vulgaris; (B) Cinnamoum Zeilanicum; (C) Calluna Vulgaris.*

on zearalenone level (1.952±0.008 ng/ml) compared to control (1.944±0.01 ng/ml). The three tested extracts induced a decrease in the production of fumonisin B1 significantly compared to control, the highest inhibitory effect was for C. zeilanicum (0.625±0.01 ng/ml) followed by B. vulgaris (1.10±0.03 ng/ml) and finally C. vulgaris (2.46±0.009 ng/ml) (Table 3).

DISCUSSION

In this study, fungi were isolated from corn seeds. Ethanolic crude extracts of Berberis vulgaris, Calluna vulgaris and Cinnamuon zeilanicum were used as antifungal compounds, as we assessed the antifungal effects on the toxin production and different toxin concentrations. We have isolated 909 nucleotides of the 18S ribosomal RNA from our fungi strain. This sequence was aligned with the GenBank sequences using BLAST online tool (https://blast.ncbi.nlm.nih.gov/Blast.cgi). Our sequence showed 93% identity with Fusarium proliferatum, indicating that we have isolated a new Fusarium strain which was given the accession number (KX808233), version 1 (KX808233.1) by GenBank and named Fusarium sporotrichioides.

Table 1. Qualitative phytochemical screening of the three tested ethanolic plants extracts.

	alkaloids	tanins	phobataninns	saponnins	flavonoids	steroids	terpenoids	Cardiac glyceroids
Berberis vulgaris roots	+	-	-	++	+	-	+	++
Cinnamon zeilanicum shoots	+	+	+	+	+	-	+	++
Calluna vulgaris park	+	+	+	-	+	-	+	-

Table 2. Polyphenolic compounds concentrations in different plant ethanolic extracts.

Compounds concentration (mg/g extract)	***C. vulgaris* shoots**	***B. vulgaris* shoots**	***C. zeilanicum* park**
Chlorogenic acid (Phenolic acid)	4.0763	000.201	11.752
Caffeic acid (Phenolic acid)	3.207	000.159	4.733
3,4 Dicaffeoylquinic acid	5.472	-	0.873
3,5- Dicaffeoylquinic Acid	52.942	-	4.890
Gallic Acid (Phenolic Acid)	61.264	90.7530	-
Retinol (Flavonoid)	8.90	000.0005	-
Tanic Acid (Tannin)	4.714	21.590	-
4,5-Dicaffeoyl Quinic Acid	-	-	0.962
Cinnamic Acid	-	-	7.910
Quercetin	-	-	10.61
Phenol, 2-Methyl-5-(1-Methylethyl)	-	-	38.75
Total Phenolic Content	-	-	86.27
Cinnamaldhyde	-	-	44.92
Benzaldhyde	-	-	11.30
Berberine	-	0.62	-

Table 3. Toxins concentration and treatment effect.

Treatment	Deoxynivalenol (ng/ml)	Zearalenone (ng/ml)	Fumonisin B1 (ng/ml)	Aflatoxin (ng/ml)
Control untreated media	ND	1.944±0.01^{c}	3.42±0.02^{d}	ND
Cinnamuom zeilanicum (2%)	ND	0.568±0.00^{a}	0.625±0.01^{a}	ND
Berberise vulgaris (2%)	ND	1.562±0.00^{b}	1.10±0.03^{b}	ND
Calluna vulgaris (2%)	ND	1.952±0.00^{c}	2.46±0.009^{c}	ND

ND: Not detectable. Values were calculated as the mean ± standard deviation, within the column, mean with different letters (a,b,c and d) are statistically significant, where mean with a is the lowest mean while mean with c or d is the highest one, at $p<0.05$

Our results showed that all tested plant extracts contained alkaloids, flavenoids and terpenoids. HPLC analysis of the polyphenolic compounds of plant ethanolic extracts showed that C. zeilanicum contained the highest concentration of chlorogenic acid, Berberis vulgaris had the highest concentration of gallic acid. While, Calluna vulgaris had the highest levels of 3, 5-Dicaffeoylquinic acid, 3, 4 Dicaffeoylquinic acid. Quercetin was one of several naturally-occurring dietary flavonoids detected in the cinnamon crude extract. The presence of flavanoids in our extracts may be responsible for its antioxidant and thus protective activity. Numerous studies have suggested that flavonoids commonly function as antioxidants and may protect plants against oxidative stress caused by suboptimal environmental conditions. Kasote et al. (2015) showed that plants have an innate ability to synthesize non-enzymatic antioxidants. Essential oils from different plants were previously used in the prevention of fungi and mycotoxins accumulation in cereals (Magan et al., 2010). Filip et al. (2011) determined that a high concentration of flavonoids was detected in ethyl acetate extract from C. vulgaris. these findings provided a scientific basis for the traditional use of this plant in the treatment of tuberculosis. Another study showed that species belonging to the family of Ericaceae are rich in phenolics (Vučić et al., 2013). Antibacterial effects of different extracts of C. vulgaris showed that phenolic compounds and flavonoids may be responsible for the growth inhibition of bacterial strains (Vučić et al., 2014) but in comparison with our study, it didn't show any effect against the fungal growth.

The interest in possible health benefits of flavonoids has increased due to their powerful antimicrobial activities (Özçelik et al., 2011). This is in agreement with our results especially for B. vulgaris and C. zeilanicum. Several studies have shown that phenolic acids that are associated with the carbohydrates of cell walls have inhibitory effect on rumen microorganisms (Iason et al., 1993; Murray et al., 1996). Tannin phenolics affect digestion and hence, intake and diet selection by ruminants (McArthur et al., 1993). All concentrations of Calluna vulgaris L. (Ericaceae) (0.5%, 1%, 1.5% and 2%) and Berberis vulgaris showed no inhibitory effect toward fungi growth. On the other hand, only Cinnamuom zeilanicum was extremely potent inhibitor for Fusarium sporotrichioides growth as its high concentrations (1%, 1.5% and 2%) completely inhibited the fungi growth.

Our data of different toxin concentrations shows that Deoxynivalenol and Aflatoxin were not detected in the growth media of F. sporotrichioides (new strain). On the other hand, Zearalenone level produced by F. sporotrichioides was highly reduced by treatment with 2% Cinnamuom zeilanicum and moderately reduced by 2% Berberise vulgaris while Calluna vulgaris had almost no effect on Zearalenone level. Our data shows that the three tested extracts decreased the production of Fumonisin B1, the highest effect was for Cinnamuon zeilanicum followed by Berberise *vulgaris* and finally *Calluna vulgaris.* This finding might be because the essential oil from cinnamon bark is rich in trans-cinnamaldehyde with antimicrobial effects against animal and plant pathogens, food poisoning, spoilage bacteria and fungi as it inhibits cell wall biosynthesis, membrane function and specific enzyme activities. In agreement with our results, different studies found that flavonols exhibit numerous biological and pharmacological effects, including antioxidant, chelation, anticarcinogenic, cardioprotective,

bacteriostatic, and secretory properties (Tabrez et al., 2013; Kumar and Pandey, 2013). In the last few years, essential oils from different plants were used in the prevention of fungi and mycotoxins accumulation in cereals (Magan et al., 2010). This is in agreement with Isaac and Abu-Tahon (2014) who revealed that cold distilled water extracts of Ocimum basilicum and Eucalyptus globulus were the most effective ones for inhibiting the growth of F. oxysporum f. sp. lycopersici. Butanolic and ethanolic extracts of the tested plants inhibited the pathogen growth to a higher extent than water extracts. Butanolic extract of O. basilicum completely inhibited the growth of F. oxysporum f. sp. lycopersici. Butanolic extracts (2.0%) also had a strong inhibitory effect on hydrolytic enzymes; β-glucosidase, pectin lyase and protease of F. oxysporum f. sp. lycopersici. These findings clearly accords with our results suggesting that different alkaloids, phenolics and flavonoid compounds detected in our tested natural crude extracts are responsible for antifungal and for the reduction of the toxins production.

CONCLUSION

We have isolated a new strain of fusarium named *F. sporotrichioides.* Furthermore, the ethanolic crude extract of *Cinnamon zeilanicum* showed a high inhibitory effect against fungi growth and different toxin production. Therefore, we recommended the usage of *Cinnamuom zeilanicum* as a potent inhibitor for *Fusarium sporotrichioides* growth and for decreasing fusarium B1 production.

Implications

Our findings indicate that natural extracts specially *Cinnamuom zeilanicum ethanolic extract might be used effectively for the inhibition of Fusarium sporotrichioides growth and mycotoxin production during* long-term storage of agricultural crops such as corn. A finding *that might be of great economic, environmental and medical implications*.

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Egyptian Association for Cancer Research (EACR)
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EACR is an NGO society that was declared by the Ministry of Social Solidarity (Egypt) No. 1938 in 19/11/2014 based on the initiative of Prof. Mohamed Labib Salem, the current Chairman of EACR. EACR aims primarily to assist researchers, in particular young researchers in the field of cancer research through workshops, seminars and conferences. Its first international annual conference entitled "Anti-Cancer Drug Discovery" was successfully organized in April 2019 (http://acdd.tanta.edu.eg). Additionally, EACR aims to raise the awareness of the society about the importance of scientific research in the field of cancer research in prediction, early diagnosis and treatment of cancer. EACR is also keen to outreach the scientific community with periodicals and news on cancer research including peer-reviewed scientific journals for the publication of cutting-edge research. The official scientific journal of EACR is "International Journal of Cancer and biomedical Research (IJCBR: https://jcbr.journals.ekb.eg) was successfully issued in 2017 and has been sponsored by the Egyptian Knowledge Bank (EKB: www.ekb.eg).

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International Journal of Cancer & Biomedical Research
(IJCBR) Online ISSN 2682-2628

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