Check for updates

RESEARCH ARTICLE

REVISED Evaluation of possible biological control of *Fusarium concentricum* sp. using plant extracts and antagonistic species of microbes *in vitro*

[version 2; peer review: 1 approved, 2 approved with reservations]

Mohammed Faruk Hasan, Mohammed Asadul Islam, Biswanath Sikdar

Department of Genetic Engineering and Biotechnology, University of Rajshahi, Rajshahi, 6205, Bangladesh

v2 **First published:** 03 Dec 2020, **9**:1394
https://doi.org/10.12688/f1000research.27098.1
Latest published: 27 Apr 2022, **9**:1394
https://doi.org/10.12688/f1000research.27098.2

Abstract

Background: *Fusarium concentricum* is one of the most devastating fungi responsible for fruit and vegetable crops rot worldwide. The present study was designed to find an ecofriendly control measure for pathogenic *Fusarium concentricum*, using suitable bioagents.
Methods: Medicinal plant extracts were evaluated or their antifungal activities against *Fusarium concentricum* using the poisoned food and serial dilution methods. Antagonistic potency of some nonpathogenic microbes was also assessed on *Fusarium concentricum* using the dual culture method.
Results: Highest inhibition of growth of *Fusarium concentricum* was observed with 68.1% (0.389 mg per 90 mm Petri plate) of mycelia on *Coccinia grandis* plant leaf extract, in comparison to the control grown with 100.0% (1.22 mg/dish). The tested plants extract showed MIC values rages of 80-150 µg/ml on the isolated fungus. The highest inhibition of radial growth was observed using *Trichoderma viride* on *Fusarium concentricum* (46.01% inhibition).
Conclusions: The findings of present study would be benevolent for antifungal drug development to control *Fusarium concentricum* causing fruit and vegetable rot.

Keywords

Fusarium sp., Plants extract, Non-pathogenic microbes, Antagonisms, Biocontrol

Open Peer Review

Approval Status ✔ ? ?

	1	2	3
version 2 (revision) 27 Apr 2022	✔		
version 1 03 Dec 2020	? view	? view	? view

1. **Natarajan Amaresan**, C. G. Bhakta Institute of Biotechnology, Uka Tarsadia University, Surat, India
2. **Shaikh Jamal Uddin**, Khulna University, Khulna, Bangladesh
3. **Mustafa M. El-Zayat**, Mansoura University, Mansoura, Egypt

Any reports and responses or comments on the article can be found at the end of the article.

This article is included in the Plant Science gateway.

This article is included in the Agriculture, Food and Nutrition gateway.

Corresponding author: Biswanath Sikdar (sikdar2014@gmail.com)

Author roles: Hasan MF: Conceptualization, Investigation, Methodology, Visualization, Writing – Original Draft Preparation; **Islam MA**: Data Curation, Formal Analysis; **Sikdar B**: Conceptualization, Funding Acquisition, Methodology, Supervision, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: This research work was supported by the Ministry of Education, Government of the People's Republic of Bangladesh (2019-2021) [No.3900.0000.09.06.217/2BS 91/170].
The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Copyright: © 2022 Hasan MF *et al*. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Hasan MF, Islam MA and Sikdar B. **Evaluation of possible biological control of *Fusarium concentricum* sp. using plant extracts and antagonistic species of microbes *in vitro* [version 2; peer review: 1 approved, 2 approved with reservations]** F1000Research 2022, **9**:1394 https://doi.org/10.12688/f1000research.27098.2

First published: 03 Dec 2020, **9**:1394 https://doi.org/10.12688/f1000research.27098.1

REVISED **Amendments from Version 1**

Changes to the manuscript include:

Indicated the name of species as *Fusarium concentricum*

Sated the photos with high resolution

MIC values and graph have been added in results section

Uses and active constituents of the selected plants have been added in introduction section

Antifungal activities of the selected plants have been included in the discussion section.

Any further responses from the reviewers can be found at the end of the article

Introduction

Fusarium species are a large genus of hyaline filamentous mold fungi, responsible for fruit and vegetable crop rot (Al-Najada & Gherbawy, 2015; Ziedan *et al*., 2018). *Fusarium* species are deeply invasive and can cause hematogenously disseminated infections with high mortality in neutropenic patients (Dignani & Anaissie, 2004). Numerous species of *Fusarium* contribute to yield loss and reduced quality to varying degrees by infection with some mycotoxins (O'Donnell *et al*., 2009). They also cause decay of various fruit in storage and postharvest conditions (Whiteside *et al*., 1988). The fruit rot caused by *Fusarium* incurs enormous yield losses and is often observed in fields and markets (Baria *et al*., 2015). The application of antagonistic agents and plant extracts in agriculture are becoming a major focus of plant protection research. Different preservatives or fungicide treatments are frequently applied to manage fruits diseases and decay, which is an alarming health concern (Munhuweyi *et al*., 2020). Since most chemical fungicides are highly toxic to humans and animals and they frequently cause water and soil pollution (Al-Najada & Gherbawy, 2015). Moreover, continuous and indiscriminate use is leading to the development of fungicide resistant strains of pathogens (Tafinta *et al*., 2013). There are some previous reports on plants extract based controlling techniques in *Fusarium* fungi causing diseases for some crops and plants (Baria *et al*., 2015; Ziedan *et al*., 2018).

In Bangladesh, fruit rot is a destructive disease caused by *Fusarium* species on pre-harvest and postharvest fruits. To the best of our knowledge, there is no previous research on biological control of this pathogenic fungus. Therefore, the main objective of the study was to find an ecofriendly control system of *Fusarium concentricum* to decrease fruit and vegetable rot in Bangladesh.

Methods

Collection of *Fusarium concentricum*

A pure culture of *Fusarium* sp. was previously isolated and identified (Accession No. MT856371) from postharvest *Citrus reticulata* fruit rot (Hasan *et al*., 2020a). The culture was preserved at the Department of Genetic Engineering and Biotechnology, University of Rajshahi (Rajshahi, Bangladesh).

In vitro assessment of antifungal potential of selected plants

For antifungal activities screening, six healthy, mature medicinal plants, *Allium sativum, Zingiber officinale, Coccinia grandis, Brassica juncea, Ocimum tenuiflorum* and *Hibiscus rosa-sinensis* were collected from Mirzapur, Binodpur and Kajla village, Motihar, Rajshahi, Bangladesh.

Collected plants were washed with water to remove dust from the plants' surface and dried in room temperature. Plant extract preparation and fractionation was performed according to the method by Kader *et al.* (2018). Different parts of selected plants- bulb of *Allium sativum*; rhizome of *Zingiber officinale*; leaves of *Coccinia grandis*, *Brassica juncea*, O*cimum tenuiflorum*; and flowers of *Hibiscus rosa-sinensis* plants were cut into small species and ground by blender to form fine powder. The dried powder of the plants (100gm of each plant) were rinsed in methanol (500ml) using a conical flask, and were incubated in a shaking incubator with occasional shaking for fourteen days. The liquid contents were pressed through Markin cloth followed by filtration using Whatman no. 1 filter paper. Obtained filtered liquids were dehydrated *in vacuo* to leave a blackish and sticky mass. The extracts were collected in vials and preserved in a refrigerator at 4°C.

The inhibitory effect of different plant extracts was measured by following the poisoned food technique (Balamurugan, 2014). For this, 20µg of each plant extract was added to 20ml of potato dextrose agar (PDA) to fill a 90mm size Petri plate and mixed well. After solidification, seven day old 6 mm size fungal plug was placed in the center of the Petri plates. The Petri plates were incubated at 35°C for seven days in static condition.

Determination of minimum inhibitory concentration (MIC)

The required MIC values of selected plants extracts were measured through the serial dilution technique (Hussain *et al.*, 2010). The extract of 1 gm of plant-based methanol leaves was dissolved into 1 ml methanol to obtain this same stock solution. Fungal suspension was taken into each test tube and added 50–150µg of plants extracts. The tested tubes were then incubated for 3 days. The progression of the isolated fungus was detected in tubes in which the strength of the substances was just below the inhibition level.

In vitro antagonistic test

For evaluation of antagonistic effects, six non-pathogenic pure microbe cultures, *Escherichia coli (pathogenic)*, *Rhizobium phaseoli, Rhizobium leguminosarum, Neofusicoccum mangifera, Trichoderma viride* and *Pestalotiopsis sp.* were used against *Fusarium concentricum*. The pure microbe cultures were kindly provided by Dr. Md. Salah Uddin, Associate Professor and Director, Microbiology Lab., Department of Genetic Engineering and Biotechnology, University of Rajshahi, as the part of collaboration.

To assess the antagonistic effects, the dual culture technique was used, as previously described (Vethavalli & Sudha, 2012). A mycelial disc of 6 mm diameter was cut from the periphery of both antagonist cultures and the test pathogen and placed on a Petri plate with PDA media. For the control, only the test pathogen was placed in the centre of a Petri plate. The Petri plates were incubated at 35°C in darkness.

Data analysis

The inhibition percentage of mycelial growth= [(Gc-Gt)/Gc] × 100; Where, Gc = Mycelial growth in terms of colony diameter in control set, Gt= Mycelial growth in terms of colony diameter in treatment set. The inhibition percentages of *Fusarium* species growth were calculated using the following formula: Inhibition percentage (%) =100× (dc– dt)/dc; Where, dc = radial growth of pathogen in control, dt = radial growth of pathogen in dual culture. Mean values were compared through least significant different test using SAS software, version 9.4M5 (SAS Inc., Cary, NC, USA). All the experiment and test were replicated thrice.

Results

In vitro screening of extracts presenting antifungal activity

All plant extracts showed a degree of growth inhibition of the tested fungus at the same concentrations. The highest inhibition of growth of the isolates was observed at 68.1% of mycelium on *Coccinia grandis*, which was followed by 64.1% on *Allium sativum*, in comparison to the control culture (100.0%). *Hibiscus rosa-sinensis* showed the lowest inhibition of mycelium with 29.6% against the fungal isolate in comparison to the control. The results are presented in Figure 1 and Figure 2.

Determination of minimum inhibitory concentration (MIC)

The tested plants extracts showed the MIC values ranges from 80-150µg/ml against the fungus *Fusarium concentricum* growth. Highest MIC value of isolated fungus was 80µg/ml on *Coccinia grandis* plant extract and lowest MIC value of isolated fungus was 150 µg/ml on *Hibiscus rosa-sinensis* plant extract (Figure 3).

In vitro antagonistic assay

The highest percentage inhibition of radial growth was observed with *Trichoderma viride* (46.01%) against *Fusarium concentricum* which was followed by 43.33% and 32.05% on *Escherichia coli* and *Rhizobium phaseoli*, respectively (Figure 4). The antagonistic agent *Rhizobium leguminosarum* did not show any inhibitory activity against the isolated fungus (Figure 4). The control group also did not show any inhibition of radial growth of *Fusarium concentricum*.

Discussion

Fruit rot caused by *Fusarium* species is very common in Bangladesh. The main objective of the present study was to study biological control measures for this fungus. Plant extracts are now a superior choice to control different plant pathogens, as reported by several previous studies (Hasan *et al*., 2020b; Kareem & Al-Araji, 2017; Parveen *et al*., 2014). In our study, we found that the plant extracts *Allium sativum, Zingiber officinale,* and *Coccinia grandis* have significant inhibitory effects on mycelial growth of *Fusarium concentricum*. Hosen & Shamsi (2019) also found significant antifungal activity using *Allium sativum* (53.85%), *Ocimum sanctum* (48.72%) and *Zingiber officinale* (49.35%) against *Fusarium solani* and *F. oxysporum*. Our current

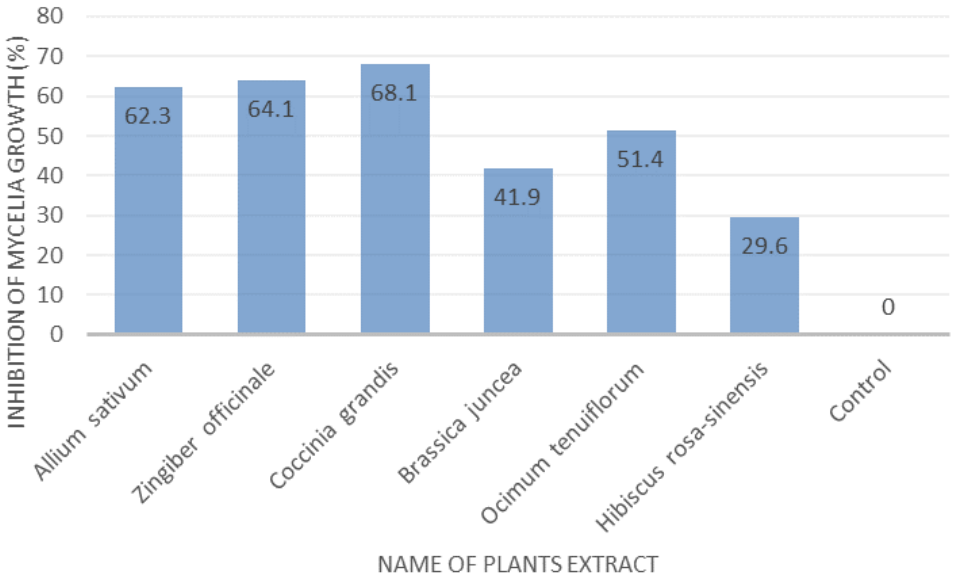

Figure 1. Effect of different methanol plant extracts on percentage inhibition of mycelial growth of *Fusarium concentricum*. Mycelia were collected after seven days of incubation on potato dextrose agar at 35°C in darkness. 90 mm Petri plates were used to culture the tested fungus.

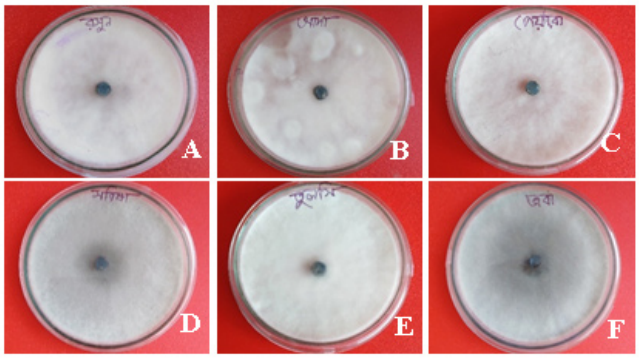

Figure 2. Petri dish images showing the effect of plant extracts on inhibition of mycelial growth of *Fusarium concentricum*. (**A**) *Allium sativum,* (**B**) *Zingiber officinale,* (**C**) *Coccinia grandis,* (**D**) *Brassica juncea*, (**E**) *Ocimum tenuiflorum,* and (**F**) *Hibiscus rosa-sinensis.* Mycelia were collected after seven days of incubation on potato dextrose agar at 35°C in darkness.

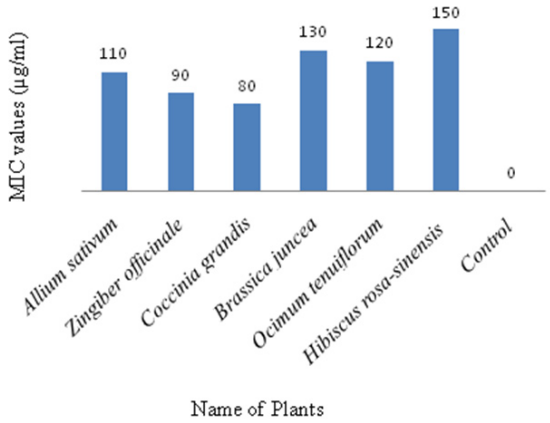

Figure 3. Minimum inhibitory concentration (MIC) of methanol extract showing the effect of plant extracts on inhibition of mycelial growth of *Fusarium concentricum*. (**A**) *Allium sativum*, (**B**) *Zingiber officinale*, (**C**) *Coccinia grandis*, (**D**) *Brassica juncea*, (**E**) *Ocimum tenuiflorum*, and (**F**) *Hibiscus rosa-sinensis*. Mycelia were collected after seven days of incubation on potato dextrose at 35°C in darkness.

results were also supported by data from Khatun *et al.* (2020) and Kareem & Al-Araji (2017). In our present study, *Coccinia grandis* plant leave extract showed a promising MIC capability to control *Fusarium concentricum* fungus. *Piper betle* plant extracts showed a significant antifungal potency on *Fusarium oxysporum* (Gnanasekaran *et al.*, 2015). For the confirmation of antagonist effects on the radial growth of the pathogen in dual culture have been previously reported by Akhtar *et al.* (2010). By contrast, Nasrin *et al.* (2018) reported 87% inhibition potency on mycelium growth of *Fusarium oxysporum* f. sp. *lycopersici* by *Calotropis proceraon* plant extract. In our study, *Trichoderma viride*, *Escherichia coli*, *Rhizobium phaseoli* and *Alternaria sp.* showed significant antagonistic activity against *Fusarium concentricum*. *Trichoderma viride* showed 45.88% growth inhibition on *Fusarium merismoides* fungi in a study by Hosen & Shamsi (2019), which support our present findings. Nasrin *et al.* (2018) also reported 82% inhibition radial growth by *Trichoderma sp.* against *Fusarium oxysporum* f. sp. *lycopersici.* In contrast, Bashar & Chakma (2014) reported that volatile substances produced by *T. viride*, *A. niger*, *A. flavus* and *A. fumigatus* showed 29.75, 20.15, 15.78 and 12.25% growth inhibition, respectively, on *F. oxysporum*.

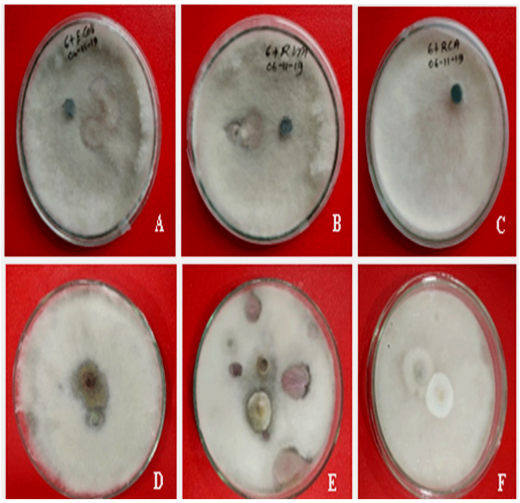

Figure 4. Growth inhibition effects of microbial agents on Fusarium concentricum. (**A**) *Escherichia coli*, (**B**) *Rhizobium phaseoli*, (**C**) *Rhizobium leguminosarum*, (**D**) *Neofusicoccum* mangifera, and (**E**) *Trichoderma viride* and (**F**) *Pestalotiopsis sp.*, Mycelia were collected after seven days of incubation on potato dextrose agar at 35°C in darkness.

In the current investigation, there are some limitations. Although this study showed that some plant extracts and nonpathogenic microbes could control the fungal stain, the number of plant extracts and microbes was limited. In addition, we used only methanol solvent for extraction and did not use other extractions. Moreover, we performed only *in vitro* techniques for antifungal potency screening and did not use any *in vivo* techniques. Therefore, we need to perform further studies to detect ecofriendly control this devastating fungal stain in the future.

Conclusions

We evaluated different biological control measures for the devastating *Fusarium* fungi. Various medicinal plants extracts and non-pathogenic microbes showed promising inhibitory activities on *Fusarium concentricum in vitro*. These identified control measures of *Fusarium concentricum* shows the importance of further research on *Fusarium* taxonomy to decline the risk of *Fusarium concentricum* caused fruit rot in Bangladesh.

Data availability

Underlying data

Figshare: Effect of plant extracts on inhibition of mycelial growth of the *Fusarium* species. https://doi.org/10.6084/m9.figshare.13096262 (Hasan, 2020a). This project contains the images of Petri plates for each treatment condition.

Figshare: Effect of antagonistic agents on inhibition of mycelial growth of the *Fusarium* species. https://doi.org/10.6084/m9.figshare.13096325 (Hasan, 2020b). This project contains the images of Petri plates for each treatment condition.

Figshare: Effects of different plants extract by methanol on inhibition percentages of mycelial growth of the Fusarium species, https://doi.org/10.6084/m9.figshare.13134953 (Hasan, 2020c).

Figshare: Effect of antagonistic agents on inhibition of mycelial growth of the Fusarium species, https://doi.org/10.6084/m9.figshare.13134962 (Hasan, 2020d).

Data are available under the terms of the Creative Commons Attribution 4.0 International license (CC-BY 4.0).

Acknowledgements

A previous version of this article is available on Figshare: https://doi.org/10.6084/m9.figshare.13012517 (Hasan, 2020e).

References

Akhtar J, Kumar V, Tiu KR, *et al.*: **Integrated management of banded leaf and sheath blight disease of maize.** *Plant Disease Research.* 2010; **25**(1): 35–38.
Reference Source

Al-Najada AR, Gherbawy YA: **Molecular Identification of Spoilage Fungi Isolated from Fruit and Vegetables and Their Control with Chitosan.** *Food Biotechnol.* 2015; **29**(2): 166–184.
Publisher Full Text

Balamurugan S: ***In vitro* antifungal activity of *Citrus aurantifolia* Linn plant extracts against phytopathogenic fungi *Macrophomina phaseolina*.** *International Letters of Natural Sciences.* 2014; **13**: 70–74.
Publisher Full Text

Baria TT, Patil RK, Patel JK: **Ecofriendly Management of Fusarium Fruit Rot of Citrus.** *The Bioscan.* 2015; **10**(4): 1807–1811.
Reference Source

Bashar MA, Chakma M: ***In vitro* control of *Fusarium solani* and *F. oxysporum* the causative agent of brinjal wilt.** *Dhaka Univ J Biol Sci.* 2014; **23**(1): 53–60.
Publisher Full Text

Dignani MC, Anaissie E: **Human fusariosis.** *Clin Microbiol Infect.* 2004; **10 Suppl 1**: 67–75.
PubMed Abstract | Publisher Full Text

Gnanasekaran P, Salique SM, Panneerselvam A, *et al.*: ***In vitro* Biological Control of *Fusarium oxysporum* f. sp. *Cubense* by using Some Indian Medicinal Plants.** *Int J Current Res and Review.* 2015; **3**(11): 107–116.
Reference Source

Hasan M: **Effect of plant extracts on inhibition of mycelial growth of the Fusarium species.** *Figshare.* Figure. 2020a.
http://www.doi.org/10.6084/m9.figshare.13096262

Hasan M: **Effect of antagonistic agents on inhibition of mycelial growth of the *Fusarium* species.** *Figshare.* Figure. 2020b.
http://www.doi.org/10.6084/m9.figshare.13096325

Hasan M: **Effects of different plants extract by methanol on inhibition percentages of mycelial growth of the Fusarium species.** *figshare.* Dataset. 2020c.
http://www.doi.org/10.6084/m9.figshare.13134953.v1

Hasan M: **Effect of antagonistic agents on inhibition of mycelial growth of the Fusarium species.** *figshare.* Dataset. 2020d.
http://www.doi.org/10.6084/m9.figshare.13134962.v1

Hasan M: **Evaluation of Possible Biological Control of Fusarium sp. by Plants Extract and Antagonistic Species In Vitro.** *figshare.* Preprint. 2020e.
http://www.doi.org/10.6084/m9.figshare.13012517

Hasan MF, Islam MA, Sikdar B: **First report on molecular identification of *Fusarium* species causing fruit rot of mandarin (*Citrus reticulata*) in Bangladesh [version 1; peer review: 1 approved].** *F1000Res.* 2020a; **9**: 1212.
Publisher Full Text

Hasan MF, Islam MA, Sikdar B: **PCR and Sequencing Base Detection of Gummosis Disease on *Citrus aurantifolia* Caused by *Lasiodiplodia theobromae* and Evaluation of Its Antagonisms.** *J Adv Microbiol.* 2020b; **20**(3): 77–90.
Publisher Full Text

Hosen MD, Shamsi S: ***In vitro* antagonism of *Trichoderma verede* and *Aspergillus spp*. against a pathogenic seed borne fungus of sesame.** *J Bangladesh Acad Sci.* 2019; **43**(1): 17–23.
Publisher Full Text

Hussain A, Wahab S, Zarin I, *et al.*: **Antibacterial Activity of the Leaves of *Coccinia indica* (W. and A) Wof India.** *Advances in Biological Research.* 2010; **4**(5): 241–248.
Reference Source

Kader SMA, Hasan M, Ahmed S, *et al.*: **Antioxidant, Antibacterial and Cytotoxic activities of Ethanol extract and its different fractions of *Sterculia cordata* leaves.** *Discovery Phytomedicine.* 2018; **5**(3): 26–33.
Publisher Full Text

Kareem HJ, Al-Araji AM: **Evaluation of *Trichoderma Harzianum* Biological Control Against *Fusarium Oxysporum* F. Sp. *Melongenae*.** *Journal of Science.* 2017; **58**(4B): 2051–2060.
Reference Source

Khatun MJ, Khalequzzaman KM, Naher MS, *et al.*: **Management of Fusarium Wilt of Tomato by Botanicals and Biocontrol Agents and Their Effect on Yield.** *Bangladesh J Bot.* 2020; **49**(1): 71–74.
Publisher Full Text

Munhuweyi K, Mpai S, Sivakumar D: **Extension of Avocado Fruit Postharvest Quality Using Non-Chemical Treatments.** *Agronomy.* 2020; **10**(2): 212.
Publisher Full Text

Nasrin L, Podder S, Mahmud MR: **Investigation of Potential Biological Control of *Fusarium Oxysporum* f.sp. *Lycopersici* by Plant Extracts, *Antagonistic* sp. and Chemical Elicitors *In Vitro*.** *Fungal Genom Biol.* 2018; **8**(1): 155.
Publisher Full Text

O'Donnell K, Sutton DA, Rinaldi MG, *et al.*: **Novel multilocus sequence typing scheme reveals high genetic diversity of human pathogenic members of the *Fusarium incarnatum-F. equiseti* and *F. chlamydosporum* species complexes within the United States.** *J Clin Microbiol.* 2009; **47**(12): 3851–3861.
PubMed Abstract | Publisher Full Text | Free Full Text

Parveen S, Wani AH, Ganie AA, *et al.*: **Antifungal activity of some plant extracts on some pathogenic fungi.** *Archives of Phytopathology and Plant Protection.* 2014; **47**(3): 279–284.
Publisher Full Text

Tafinta IY, Shehu K, Abdulganiyyu H, *et al.*: **Isolation and Identification of Fungi Associated with the Spoilage of Sweet Orange (*Citrus Sinensis*) Fruits In Sokoto State.** *Nigerian Journal of Basic and Applied Science.* 2013; **21**(3): 193–6.
Publisher Full Text

Vethavalli S, Sudha SS: ***In vitro* and *in silico* studies on biocontrol agent of bacterial strains against *Fusarium oxysporum* f. sp. *lycopersici*.** *Research in Biotechnology.* 2012; **3**(2): 22–31.
Reference Source

Whiteside J, Bennett J, Holtzblatt K: **Usability engineering: our experience and evolution**. In: M. Helander (Ed.), *Handbook of human-computer interaction*. New York, North Holland, 1988; 791–817.
Publisher Full Text

Ziedan ESH, Khattab AA, Sahab AF: **New fungi causing postharvest spoilage of cucumber fruits and their molecular characterization in Egypt.** *J Plant Prot Res.* 2018; **58**(4): 362–371.
Publisher Full Text

Open Peer Review

Current Peer Review Status: ✔ ? ?

Version 2

Reviewer Report 13 May 2022

https://doi.org/10.5256/f1000research.132272.r136047

© **2022 Amaresan N.** This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

✔ **Natarajan Amaresan**
C. G. Bhakta Institute of Biotechnology, Uka Tarsadia University, Surat, Gujarat, India

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Microbial Diversity, Plant-Microbe interaction, Phytoremediation

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Version 1

Reviewer Report 15 December 2021

https://doi.org/10.5256/f1000research.29932.r96967

© **2021 M. El-Zayat M.** This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

? **Mustafa M. El-Zayat**
[1] Unit of Genetic Engineering and Biotechnology, Mansoura University, Mansoura, Egypt
[2] Unit of Genetic Engineering and Biotechnology, Mansoura University, Mansoura, Egypt

This research dealt with an important topic in the safe control of the fungus *Fusarium* sp. that causes damage to many types of fruits without the use of chemicals that represent a danger to the environment and health.

- The author should indicate which species of *Fusarium* has been used.

- It is too difficult to extract 100 grams of the plant using only 500 ml methanol.
- It has been mentioned that *E. coli* is not pathogenic while it is pathogenic strain.
- The photos need to be somehow more clear because its resolution is low.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Yes

Are sufficient details of methods and analysis provided to allow replication by others?
Partly

If applicable, is the statistical analysis and its interpretation appropriate?
No

Are all the source data underlying the results available to ensure full reproducibility?
Partly

Are the conclusions drawn adequately supported by the results?
Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Microbiology, Ecology, Nanotechnology, Phytochemistry, Fermentation technology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Reviewer Report 26 July 2021

https://doi.org/10.5256/f1000research.29932.r89306

© 2021 Uddin S. This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

?

Shaikh Jamal Uddin
[1] Pharmacy Discipline, Life Science School, Khulna University, Khulna, Bangladesh
[2] Pharmacy Discipline, Life Science School, Khulna University, Khulna, Bangladesh

Authors presented the antifungal activity of some plant extract as a eco-friendly control measure for pathogenic *Fusarium* species using the poisoned food method. They found some plant extracts showed promising (>65%) inhibitory activity against *Fusarium* sp. and suggested to use as bioagents to control fungal affected fruit and vegetable rot.

However, the authors can improve the article if they consider and revise the below points:

1. Authors did not mention any MIC values. It will be good if they included MIC values of each extracted.

2. In introduction section author might include a table on the selected plant with their traditional uses and list of active constituents.

3. In the discussion section, there is a lacking of discussion between antifungal activity of selected plant extracts and their traditional uses and reported phytoconstituents.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Yes

Are sufficient details of methods and analysis provided to allow replication by others?
Yes

If applicable, is the statistical analysis and its interpretation appropriate?
Yes

Are all the source data underlying the results available to ensure full reproducibility?
Yes

Are the conclusions drawn adequately supported by the results?
Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Pharmacological evaluation of ethnomedicinal plants, natural products chemistry and drug discovery

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Reviewer Report 13 April 2021

https://doi.org/10.5256/f1000research.29932.r80330

© 2021 Amaresan N. This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

?

Natarajan Amaresan
[1] C. G. Bhakta Institute of Biotechnology, Uka Tarsadia University, Surat, Gujarat, India
[2] C. G. Bhakta Institute of Biotechnology, Uka Tarsadia University, Surat, Gujarat, India

The authors are reporting some interesting results which will be of significance to the readership however, the manuscript lacks indepth research.

The effect of the bacteria and botanical extracts should be tested in the pot studies for their effectiveness. *In vitro* studies alone will not warrant their suitability to be used as bioinoculants for management of pathogens.

Without the *in vivo* studies the work appears as shallow research. The authors may be encouraged to do pot studies and update the manuscript.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Partly

Are sufficient details of methods and analysis provided to allow replication by others?
Partly

If applicable, is the statistical analysis and its interpretation appropriate?
Yes

Are all the source data underlying the results available to ensure full reproducibility?
Yes

Are the conclusions drawn adequately supported by the results?
Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Microbial Diversity, Plant-Microbe interaction, Phytoremediation

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com

F1000Research