Cross Current International Journal of Medical and Biosciences
Abbreviated Key Title: Cross Current Int J Med Biosci
ISSN: 2663-2446 (Print) & Open Access
DOI: 10.36344/ccijmb.2024.v06i02.003

Volume-6 | Issue-2 | Mar-Apr, 2024 |

Original Research Article

Study of Toxicity Resistance of *Fusarium incarantum* Isolated from Broad Bean by Arum maculate Leaf Extract in Adult Male Rabbits

Hussein A. R. Abbood[1*], Hana Attiya Salman[1]
[1]Department of Biology, College of Education for Pure Sciences, Kirkuk University, Kirkuk, Iraq

***Corresponding author:** Hussein A. R. Abbood
| **Received:** 27.02.2024 | **Accepted:** 06.04.2024 | **Published:** 17.04.2024 |

Abstract: Fungus isolates were obtained from the roots and leaves of the infected broad bean from Hawija district of Kirkuk city and isolated on the medium of PDA by examining its morphological and phenotypic characteristics of the fungus and the shape of conidia. In this study, 16 male rabbits were brought from the local markets of the city of Kirkuk and divided into 4 groups, each group consisting of 4 rabbits, the first group includes a control group, the second group is an infected group, the third group is an infected group that was treated with 50 mg of plant leaf extract, and an infected group was treated with 100 mg of leaf extract, after which Oxidative stress factors levels were checked. Oxidative stress factors showed high levels of MDA in infected rabbits compared to normal rabbits ($P < 0.05$), while GSH and catalase levels appeared lower compared to MDA in infected rabbits ($P < 0.05$), while in the group of rabbits treated with leaf extract 50 mg and 100 mg) showed few changes in concentrations MDA, GSH and Catalase compared to normal rabbits ($P < 0.05$).

Keywords: Fusarium incarnatum; Arum maculatum; MDA; GSH; Catalas.

INTRODUCTION

Arum maculatum belongs to the Araceae family and is a flowering plant species found in woodlands. It grows and is widely found in various regions of Europe, the Eastern Caucasus, and Turkey. Its leaves appear in the spring, and its leaves reach a length of 7 cm to 20 cm [1].

Above the male flowers of this plant, there is a ring of hair that forms an insect trap. Insects, particularly owl-midges like Psychoda phalaenoides, are attracted to the bulb due to their fecal odor and the temperature around it, which can be up to 15 °C warmer than the surrounding environment [2].

The root-tuber of *A. maculatum* can grow to be quite large and serves as a storage organ for starch. In mature plants, the tuber can be located as deep as 400 mm below the surface of the soil the plant relies on birds to disperse its seeds by consuming the berries. When *A. maculatum* is in its seedling stage, it develops small light green leaves that lack the glossy appearance of mature leaves. At approximately 5 months of age, the leaves begin to grow larger and become more shiny. By the time the plant reaches one year, all its leaves are wilted and shiny at the end, and in the summer it blooms and grows [3].

The roasted root of the cuckoo plant, known as Portland sago, was once traded and consumed. It was used to make salop, a popular drink before tea or coffee, and served as a substitute for arrowroot [4]. However, it is important to note that improper preparation can make it highly toxic. The leaves are fermented with yogurt and then simmered for several hours to remove toxins. *Fusarium incarnatum* is a pathogenic fungus belonging to the genus *Fusarium* of the family Nectriaceae. It is commonly found in natural environments and forms the *Fusarium incarnatum*-equiseti species complex (FIESC) [6], which is associated with more than 40 species. This complex is distributed in subtropical and temperate regions worldwide, leading to numerous reports of plant diseases [7]. *Fusarium incarnatum* produces various mycotoxins, including zearalenone from the trichothecene group, which can cause diseases in both plants and animals [8]. Under laboratory conditions, cotton-like fungi form colonies on PDA and develop chlamydospores, conidia, and macroconidia that produce polyspores [9]. While FIESC colonies resemble *Fusarium incarnatum*, they may have slight differences in color and texture depending on the species present

Quick Response Code

Journal homepage:
https://www.easpublisher.com/

Citation: Hussein A. R. Abbood & Hana Attiya Salman (2024). Study of Toxicity Resistance of *Fusarium incarantum* Isolated from Broad Bean by Arum maculate Leaf Extract in Adult Male Rabbits. *Cross Current Int J Med Biosci*, *6*(2), 45-50.

within the complex [10]. *Fusarium incarnatum* is a widespread pathogenic fungus that significantly affects crop yields worldwide. It can also infect animals and humans, causing a range of diseases [11]. Cases of wheat scab caused by FIESC have been documented in Mexico, and there have been reports of *Fusarium* infection in humans with polytetrafluoroethylene dialysis grafts [12].

MATERIALS & METHODS

2.1. To collect and isolate fungi from plant samples

Samples of plants showing symptoms such as leaves and stems were collected in the Hawija area of Kirkuk, Iraq. In the laboratory, these samples undergo a series of procedures. Wash first with tap water, then with sterile distilled water. To start, the samples were thoroughly rinsed with sterile distilled water to remove any traces of the sterilizing solution. Once this was completed, filter paper was used to dry the plant parts. A 9 cm diameter Petri dish was then prepared with the standard PDA culture medium. Each plate accommodated four plant parts, with a total of three plates assigned to each plant part. To inhibit the growth of fungi in the culture medium, the dishes were placed in an incubator with a temperature set at 25 ± 2 °C for a period of 5 to 7 days. To achieve pure fungal isolates, the fungal colonies that developed underwent a purification process. By using sterilized cork borers that were treated with alcohol and exposed to a flame, the fungus surrounding the plant parts was delicately eliminated. The isolated portion was then transferred to a Petri dish containing PDA medium. The plates were once again placed in the incubator under the same previously mentioned conditions to ensure the growth of uncontaminated fungal isolates [13].

2.2. Morphological identification of isolated fungi

Morphological characteristics of the isolated fungi were recorded by aseptically removing 5 mm diameter disks from pure colonies after 5 days of growth. The sections were then placed in the center of a Petri dish containing PDA and incubated. Observe fungi and record their morphological characteristics using a binocular microscope and the naked eye. Record the shape, size, edges, brightness, transparency, and color of isolated fungal colonies. To determine the color of the filaments and spore structures of fungi, it is necessary to examine all developing fungi under a light microscope. In the process of this analysis, classification is facilitated by meticulously observing and recording a multitude of details, including shape, color, size, and other pertinent microscopic attributes. The phenotypic and morphological traits of the fungi that have been identified and isolated are comprehensively documented. In order to maintain the fungal cultures, sections are collected from the edges of colonies using sterile loop carriers and used to inoculate solid medium slants. These slants are then stored in a refrigerator at a temperature of 4°C until they are needed for future experiments [14, 15].

Preparation of conidial suspension

To quantify the conidia, a light microscope is utilized, following the formula: Conidial concentration = Z × 4 × 10^6 / n. In this equation, Z represents the total count of conidia, while n represents the total number of small squares [16].

Extraction technique for *Arum maculatum* leaves

Considering the traditional usage of this plant by the general public, water was chosen as the extraction solvent. In the water fermentation method (FW: Fermented in Water), fresh plant material was combined with water at 100 °C (leaf water ratio: 1:2), sealed, and left at room temperature for 24 hours. Filtration was performed after the designated time period [17].

Animal model selection

16 adult males were purchased locally from markets in Kirkuk city, with an average weight of 1.5 kg and an age range of 1 year. All animals had free access to food and water and strict observation protocols were implemented to prevent possible infections.

The experimental setup

The experimental setup involved the utilization of male rabbits, which were divided into four groups, each consisting of four rabbits. The first group, known as the control group, was provided with regular food and served as the control for the study. The second group, referred to as the infection group, was subjected to intraperitoneal injection of *Fusarium incarnatum* conidia. Unfortunately, these rabbits succumbed to the infection.

In the treatment group receiving a dosage of 50 mg, rabbits were administered intraperitoneal injections of *Fusarium incarnatum* conidia, followed by a three-week course of root extract treatment before being sacrificed. Similarly, in the treatment group receiving a dosage of 100 mg, rats were intraperitoneally injected with *Fusarium incarnatum* conidia, underwent a three-week treatment regimen with root extract, and were then euthanized.

Sample collection

To collect a sample, anesthesia is administered, and a blood sample is obtained through a cardiac puncture, which is then placed in a test tube. Once the blood has coagulated, the tubes containing the samples are centrifuged at 5000 rpm for 10 minutes to separate the serum. Simultaneously, the lungs are promptly extracted and homogenized with NaCl. The resulting supernatant and serum are carefully extracted and stored in a frozen state until they are ready to be utilized [18].

Oxidative stress factors

MDA (malondialdehyde), determined by thiobarbituric acid (TBA) according to method [19] and glutathione (GSH) by using DTNB, estimated catalase according to method [20].

Statistical Analysis

To evaluate the significance of the variability between the treatment and control groups, the statistical analysis involved utilizing the analysis of variance (ANOVA) test through the Minitab statistical program.

RESULTS AND DISCUSSION

Fungal Isolation and Morphological Characteristics

The physical characteristics of the fungus obtained from vetch are used for its identification. Pure colonies grown on PDA medium vary in color from cream to golden brown and have a rough, cotton-like texture along the edges (Figure 1). After 4 to 5 days, the colonies completely cover the plate and are cream to light brown on the reverse side. Three types of spores were observed: macroconidia, which were 16 in number.85 - 25.33 x 4.34 - 4.09 µm, Microconidia, measuring 7.52 - 12.62 x 2.47 to 4.22 µm and Chlamydospores range from 9.77 to 10.11 x 5.57 - 7.62 µm (Figure 1). These results are consistent with previous research [21, 22].

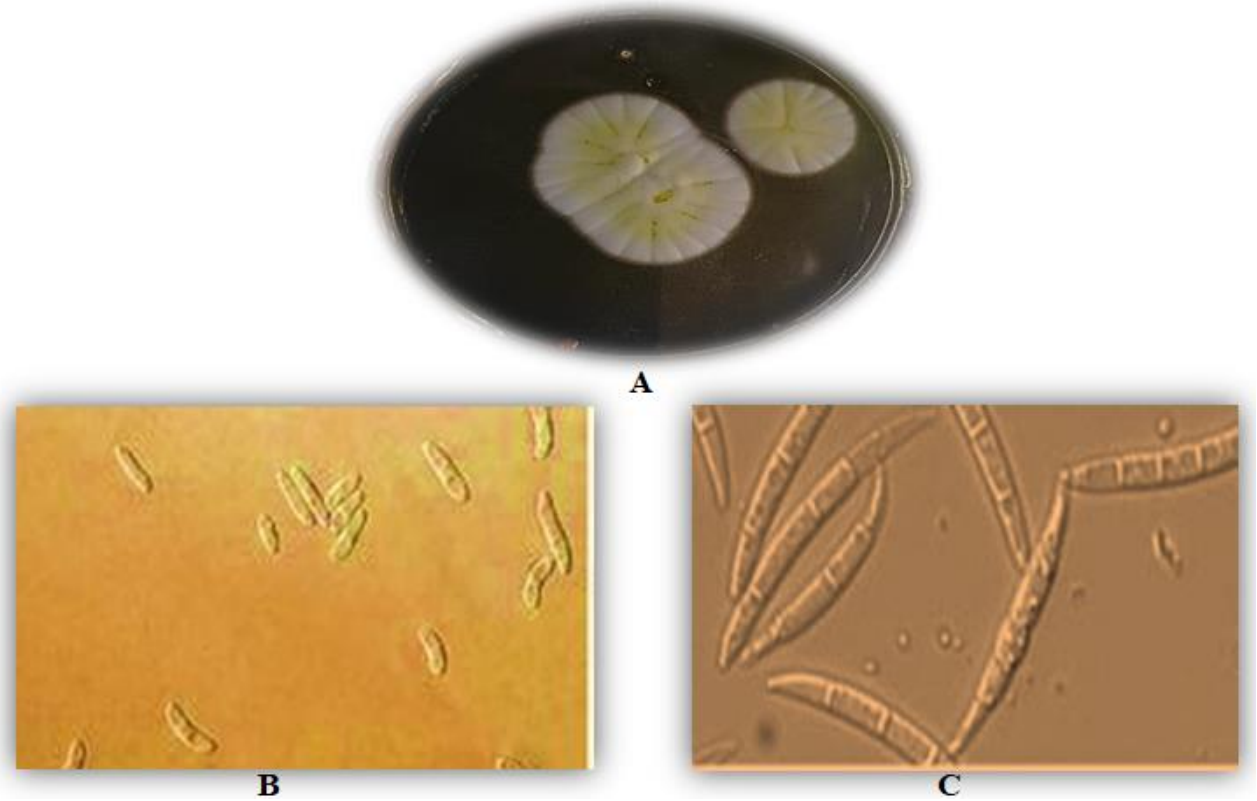
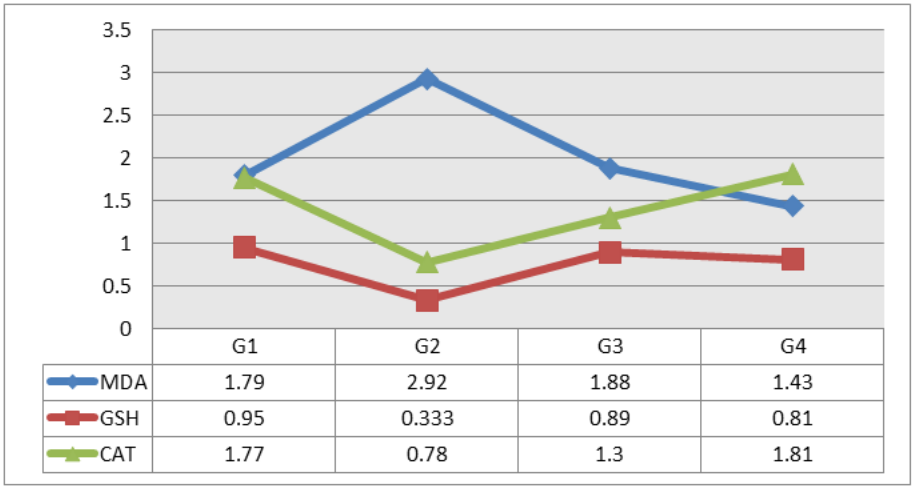


Figure 1: A- *Fusarium incarnatum*; B- Microconidia; C- Macroconidia

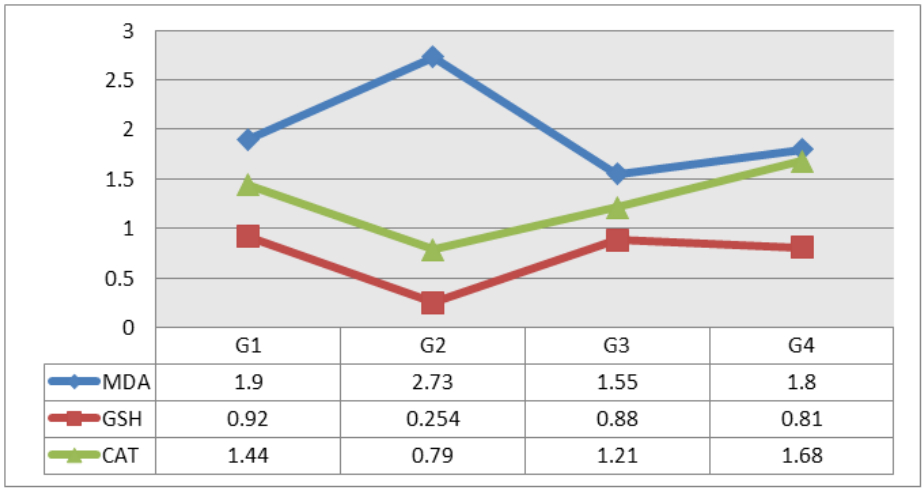
Oxidative stress factors

The MDA values of rabbits injected with *Fusarium incarnatum* (serum: 2.92 ± 0.93 and lung: 2.73 ± 0.75) were significantly increased ($P < 0.05$) compared with normal rabbits (serum: 1.79 ± 0.15 and lung: 1.9 ± 0.89). The MDA values of the groups treated with alocasia leaf extract (50 mg and 100 mg) did not change significantly compared with normal rabbits ($P < 0.05$). The GSH values of rabbits injected with *Fusarium incarnatum* (serum: 0.333 ± 0.083 and lung: 0.254 ± 0.086) were significantly increased ($P < 0.05$) compared with normal rabbits (serum: 0.95 ± 0.2 and lung: 0.92 ± 0.53). There was no significant change in GSH levels in the groups treated with *Arum maculatum* leaf extract (50 mg and 100 mg) compared with normal rabbits ($P < 0.05$). Catalase levels in rabbits injected with *Fusarium incarnatum* (serum: 0.78 ± 0.77 and lung: 0.79 ± 0.082) were significantly lower and significantly increased ($P < 0.05$). Catalase concentrations in groups treated with *Arum maculatum* leaf extract (50 mg and 100 mg) showed insignificant changes ($P < 0.05$) compared with normal rabbits, as shown in Figure 2 and 3.



	G1	G2	G3	G4
MDA	1.79	2.92	1.88	1.43
GSH	0.95	0.333	0.89	0.81
CAT	1.77	0.78	1.3	1.81

Figure 2: Oxidative stress factors levels in groups of study (blood serum)



	G1	G2	G3	G4
MDA	1.9	2.73	1.55	1.8
GSH	0.92	0.254	0.88	0.81
CAT	1.44	0.79	1.21	1.68

Figure 3: Oxidative stress factors levels in groups of study (lung)

Fusarium species are important plant pathogens in many species. plants around the world [23, 24]. *Fusarium* species are considered to be major pathogens of protea plants worldwide [25]. *Fusarium* is a filamentous fungus that occurs in soil and air throughout the world, particularly in tropical and temperate regions [26]. They are primarily plant pathogens but sometimes cause serious infections in livestock. The main site of entry of *Fusarium* in lung transplant patients is inhalation of *Fusarium* conidia into the respiratory tract, followed by skin and mucosal lesions [27-29]. *Fusarium* species are ubiquitous in the environment and Fusarium conidia have been detected in outdoor air samples [27]. One study18 found that Fusarium was more common than *Aspergillus* in air samples. *Fusarium* species have also been isolated from indoor water storage systems and outdoor water bodies. Water-related activities such as outdoor swimming and water sports may cause Fusarium conidia to aerosolize, allowing them to enter the air and more easily spread through the respiratory tract [29, 30]. In most cases of disseminated fusarium in lung transplant patients, the lungs are affected [30]. Pulmonary involvement is associated with increased mortality even after controlling for immune status. Pulmonary lesions of *Fusarium pneumonia* include nodular and cavitated lesions as well as nonspecific alveolar or cavitated lesions. interstitial penetration [31]. Clinical manifestations are often nonspecific and include dyspnea, dry cough, and pleuritic chest pain. The toxic effects of mycotoxins can lead to oxidative stress (OS) and free radical production. Based on [32] [33]. Dysfunction of the antioxidant system leads to an increase in the number of free radicals that damage DNA, proteins, and lipids [34]. Cells typically produce balanced amounts of free radicals and antioxidants. External parameters can promote the development of oxidative stress and excessive production of free radicals [35], leading to an imbalance in cellular homeostasis mechanisms. Oxidative stress may occur due to damage to the antioxidant system and excessive production of free radicals [36]. Many studies have been conducted to investigate the effects of various mycotoxins on antioxidant enzymes in poultry and pigs. The results of [37] showed that consumption of AFB1 in chickens induced oxidative stress in the spleen, as evidenced by decreased levels of antioxidant enzymes (e.g., GSH-Px,

GR, CAT) as well as malondialdehyde (MDA) and GSH levels [38] MDA represents the most important indicator for monitoring oxidative stress resulting from the process of lipid peroxidation and oxidative damage caused to tissues by reactive oxygen species (ROS) [39]. The decrease in the level of glutathione is due to several reasons, including an increase in its consumption rate, which is one of the most important non-enzymatic antioxidants in removing free radicals and their products, as it is transformed from the active form to the inactive form Glutathione disulfide. The sulfur group in the composition of GSH is a good reducing agent, as a hydrogen atom easily blows away. This is due to the weakness of the bond between sulfur and hydrogen (S-H) and the strength of the bond between carbon and hydrogen (C-H) in free radicals. Therefore, it protects cell membranes from free radical damage. One of the reasons for the low level of GAH is a deficiency in the raw materials for its construction, especially the coenzyme (reduced form) NADPH produced. From the pathway, pentaphosphate sugar, which is the catalyst for the action of the GSred enzyme, which works to restore the active form of glutathione from the inactive form [40].

REFERENCE

1. Stace, C. (2005). New Flora of the British Isles (2nd ed.). Cambridge University Press. p. 778.
2. Lack, A. J., & Diaz, A. (1991). "The pollination of Arum maculatum L.- a historical review and new observations" . Watsonia. 18: 333–342. Archived from the original on 2009-01-09. Retrieved 2012-02-15.
3. List of berry-producing plants". British Trust for Ornithology. 7 September 2012. Retrieved 19 May 2021.
4. Arum maculatum (Adam and Eve, Arum, Bobbins, Cuckoo-Pint, Cuckoo Plant, Lords-and-Ladies, Starch Root, Wake Robin)". North Carolina State University Extension Gardener Plant Toolbox. Retrieved 19 May 2021.
5. Raju, K. P., Goel, K., Anandhi, D., Pandit, V. R., Surendar, R., & Sasikumar, M. (2018). Wild tuber poisoning: Arum maculatum:–A rare case report. *International journal of critical illness and injury science*, *8*(2), 111-114. doi: 10.4103/IJCIIS.IJCIIS_9_18. PMC 6018264.
6. Pramunadipta, S., Widiastuti, A., Wibowo, A., Suga, H., & Priyatmojo, A. (2022). Development of PCR-RFLP Technique for Identify Several Members of Fusarium incarnatum-equiseti Species Complex and Fusarium fujikuroi Species Complex. *The plant pathology journal*, *38*(3), 254–260. doi: 10.5423/PPJ.NT.12.2021.0184.
7. Villani, A., Proctor, R. H., Kim, H. S., Brown, D. W., Logrieco, A. F., Amatulli, M. T., Moretti, A., & Susca, A. (2019). Variation in secondary metabolite production potential in the Fusarium incarnatum-equiseti species complex revealed by comparative analysis of 13 genomes. *BMC genomics*, *20*(1), 314. https://doi.org/10.1186/s12864-019-5567-7
8. Avila, C. F., Moreira, G. M., Nicolli, C. P., Gomes, L. B., Abreu, L. M., Pfenning, L. H., ... & Del Ponte, E. M. (2019). Fusarium incarnatum-equiseti species complex associated with Brazilian rice: Phylogeny, morphology and toxigenic potential. *International Journal of Food Microbiology*, *306*, 108267. doi: 10.1016/j.ijfoodmicro.2019.108267.
9. Song, M., Yun, H. Y., & Kim, Y. H. (2014). Antagonistic Bacillus species as a biological control of ginseng root rot caused by Fusarium cf. incarnatum. *Journal of ginseng research*, *38*(2), 136-145. 10.1016/j.jgr.2013.11.016
10. Wang, M. M., Chen, Q., Diao, Y. Z., Duan, W. J., & Cai, L. (2019). Fusarium incarnatum-equiseti complex from China. *Persoonia-Molecular Phylogeny and Evolution of Fungi*, *43*(1), 70-89. doi: 10.3767/persoonia.2019.43.03.
11. Leyva-Mir, S. G., García-León, E., Camacho-Tapia, M., Villaseñor-Mir, H. E., Leyva-Madrigal, K. Y., Mora-Romero, G. A., & Tovar-Pedraza, J. M. (2022). Occurrence of the Fusarium incarnatum-equiseti species complex causing Fusarium head blight of wheat in Mexico. *Plant Disease*, *106*(10), 2755. doi:10.1094/PDIS-11-21-2467-PDN.
12. Riddell, J., 4th, Woodside, K. J., Leavitt, M. A., Newton, D. W., & Punch, J. D. (2010). Fusarium incarnatum/equiseti hemodialysis graft infection. *Infectious disease reports*, *2*(2), e14. https://doi.org/10.4081/idr.2010.e14.
13. Razaq, N. J., & Abass, M. H. (2021). First report of Cladosporium cladosporioides, C. oxysporum, and C. uredinicola as potential pathogens on tomato shoots system in Iraq. *Applied Nanoscience*, *13*(2), 1065-1072.
14. Ellis, M. B. (1971). Dematiaceous hyphomycetes. Commonwealth Mycological Institute, Kew, p 608.
15. Wang, M. M., Chen, Q., Diao, Y. Z., Duan, W. J., & Cai, L. (2019). Fusarium incarnatum-equiseti complex from China. *Persoonia-Molecular Phylogeny and Evolution of Fungi*, *43*(1), 70-89.
16. Faraj, M. K. (1990). "Regulation of mycotoxin formation in Zea mays. Ph.D. thesis. De- partment of bioscience and biotechnology," university of Strathelyde, Glascow, U. K.
17. Comlekcioglu, N., Çolak, S., & Aygan, A. (2021). A study on the bıoactıvıty of plant extracts obtaıned from Arum maculatum leaves by dıfferent extractıon technıques. *Hrvatski časopis za prehrambenu tehnologiju, biotehnologiju i nutricionizam*, *16*(1-2), 41-46.
18. Saleh, A. H., & Abbood, H. A. (2018). Study the Potential Effect of Rheum Palmatum Root Extract Against the Toxicity of A. fumigatus in Adult Male Rabbits. *Al-Mustansiriyah Journal of Science*, *29*(1). Doi: http://doi.org/10.23851/mjs.v29i1.83.
19. Mahmood, N. A. (2010). Glutathion-S-transferase Enzyme and Malondialdehyde (MDA) in Colorectal Cancer and in Healthy Control. *Iraqi Journal of Cancer and Medical Genetics*, *3*(1), 21-26.
20. BM, N. Y., & Mahmood, F. S. (2013). Investigating the influence of emitted Cadmium from crude oil combustion on glutathione level in workers at Al-

Qudis power plant. *Baghdad. J. Sci*, *55*(4), 1792-1801.
21. Wonglom, P., & Sunpapao, A. (2020). Fusarium incarnatum is associated with postharvest fruit rot of muskmelon (Cucumis melo). *Journal of Phytopathology*, *168*(4), 204-210.
22. Ismail, S. I., Noor Asha, N. A., & Zulperi, D. (2021). First report of Fusarium incarnatum-equiseti species complex causing leaf spot on rockmelon (Cucumis melo) in Malaysia. *Plant disease*, *105*(4), 1197.
23. Shan, L., Haseeb, H. A., Zhang, J., Zhang, D., Jeffers, D. P., Dai, X., & Guo, W. (2019). A loop-mediated isothermal amplification (LAMP) assay for the rapid detection of toxigenic Fusarium temperatum in maize stalks and kernels. *International journal of food microbiology*, *291*, 72-78.
24. Srivastava, S., Kadooka, C., & Uchida, J. Y. (2018). Fusarium species as pathogen on orchids. *Microbiological research*, *207*, 188-195.
25. Swart, L., Denman, S., Lamprecht, S. C., & Crous, P. W. (1999). Fusarium wilt: A new disease of cultivated Protea in Southern Africa. *Australasian Plant Pathology*, *28*(1), 156-161.
26. Nelson, P. E., Dignani, M. C., & Anaissie, E. J. (1994). Taxonomy, biology, and clinical aspects of Fusarium species. *Clinical microbiology reviews*, *7*(4), 479-504.
27. Nucci, M., & Anaissie, E. (2002). Cutaneous infection by Fusarium species in healthy and immunocompromised hosts: implications for diagnosis and management. *Clinical Infectious Diseases*, *35*(8), 909-920.
28. Nucci, M., & Anaissie, E. (2007). Fusarium infections in immunocompromised patients. *Clinical microbiology reviews*, *20*(4), 695-704.
29. Mattiuzzi, G. N., Alvarado, G., Giles, F. J., Ostrosky-Zeichner, L., Cortes, J., O'brien, S., ... & Estey, E. H. (2006). Open-label, randomized comparison of itraconazole versus caspofungin for prophylaxis in patients with hematologic malignancies. *Antimicrobial agents and chemotherapy*, *50*(1), 143-147.
30. O'Donnell, K., Sutton, D. A., Rinaldi, M. G., Magnon, K. C., Cox, P. A., Revankar, S. G., ... & Robinson, J. S. (2004). Genetic diversity of human pathogenic members of the Fusarium oxysporum complex inferred from multilocus DNA sequence data and amplified fragment length polymorphism analyses: evidence for the recent dispersion of a geographically widespread clonal lineage and nosocomial origin. *Journal of Clinical Microbiology*, *42*(11), 5109-5120.
31. Nucci, M., Anaissie, E. J., Queiroz-Telles, F., Martins, C. A., Trabasso, P., Solza, C., ... & Souza, C. A. (2003). Outcome predictors of 84 patients with hematologic malignancies and Fusarium infection. *Cancer*, *98*(2), 315-319.
32. Adhikari, M., Negi, B., Kaushik, N., Adhikari, A., Al-Khedhairy, A. A., Kaushik, N. K., & Choi, E. H. (2017). T-2 mycotoxin: toxicological effects and decontamination strategies. *Oncotarget*, *8*(20), 33933-33952.
33. Wang, X., Wu, Q., Wan, D., Liu, Q., Chen, D., Liu, Z., ... & Yuan, Z. (2016). Fumonisins: oxidative stress-mediated toxicity and metabolism in vivo and in vitro. *Archives of toxicology*, *90*, 81-101.
34. Assi, M. (2017). The differential role of reactive oxygen species in early and late stages of cancer. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *313*(6), R646-R653.
35. Young, I. S., & Woodside, J. V. (2001). Antioxidants in health and disease. *Journal of clinical pathology*, *54*(3), 176-186.
36. Halliwell, B., & Whiteman, M. (2004). Measuring reactive species and oxidative damage in vivo and in cell culture: how should you do it and what do the results mean?. *British journal of pharmacology*, *142*(2), 231-255.
37. Chen, J., Chen, K., Yuan, S., Peng, X., Fang, J., Wang, F., ... & Geng, Y. (2016). Effects of aflatoxin B1 on oxidative stress markers and apoptosis of spleens in broilers. *Toxicology and industrial health*, *32*(2), 278-284.
38. Rajput, S. A., Sun, L., Zhang, N., Khalil, M. M., Gao, X., Ling, Z., ... & Qi, D. (2017). Ameliorative effects of grape seed proanthocyanidin extract on growth performance, immune function, antioxidant capacity, biochemical constituents, liver histopathology and aflatoxin residues in broilers exposed to aflatoxin B1. *Toxins*, *9*(11), 371.
39. Namik, M. F., Al-Janaby, M. S., & Abbas, S. K. (2018). Assessment of Albumin, Malondialdehyd Levels and Superoxide Dismutase Activity in Pregnant Women in Kirkuk City. *Kirkuk Journal of Science*, *13*(4), 253-265. DOI: http://doi.org/10.32894/kujss.2018.13.4.18.
40. Ali, J. M., Hammed, R., & Raoof, A. (2017). Evaluation of oxidative stress state in patients with Renal failure. *Kirkuk Journal of Science*, *12*(1).