



DETERMINATION OF THE EFFECT OF CHINABERRY TREE (*Melia azedarach* L.) LEAF AND FRUIT EXTRACTS ON GERMINATION OF MAIZE (*Zea mays* L.) SEEDS INFECTED WITH *Fusarium verticillioides*

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Abstract: Maize (*Zea mays* L.) is one of the important and basic cereal crops in the world. *Fusarium verticillioides* is one of the most important pathogens causing root, stalk, ear rot, and seedling blight diseases in maize. In recent years, the restriction of the use of most chemical fungicides has led to the importance of research on plant resistance inducers. Different parts of the *Melia azedarach* L., such as bark, leaf, seeds, and root have very strong medicinal value, and the seeds of this species can also be used in the production of fungicides and pesticides in agricultural products. In this study investigated the effects of *Melia azedarach* leaf and fruit extracts on Kale (F1) corn seeds infected with *Fusarium verticillioides*. Six different applications (Positive Control, Negative Control, Fruit Extract, Leaf Extract, Fruit Extract + GA3, Leaf Extract + GA3) were established in a randomized plot design with three replications. According to the results, the highest value for germination rate on the 3rd day was 67.78% in the negative control group, and the highest value for germination rate on the 7th day was obtained in the positive control 77.77% and negative control 74.44%. In terms of *Fusarium* contamination rates, the highest values on the 3rd and 7th days were found to be 36.67% and 50.00% in the fruit extract group, respectively. Root length was determined as the highest value at 47.03 mm in the leaf extract group and the lowest value at 16.00 mm in the fruit extract + GA3 group. In shoot length, leaf extract gave the best results at 57.13 mm and leaf extract + GA3 as 49.23 mm. The findings show that *Melia azedarach* extracts have positive effects on germination and growth in *Fusarium*-infected seeds. It is thought that this natural solution may provide positive contributions to maize cultivation.

Keywords: *Zea mays* L., germination, *Fusarium verticillioides*, *Melia azedarach* L.

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Received: June 11, 2025

Accepted: July 10, 2025

Published: July 15, 2025

Cite as: Cengiz R, Yıldız Z. 2025. Determination of the effect of chinaberry tree (*Melia azedarach* L.) leaf and fruit extracts on germination of maize (*Zea mays* L.) seeds infected with *Fusarium verticillioides*. BSJ Agri, 8(4): 556-562.

1. Introduction

Maize (*Zea mays* L.) is the third most widely cultivated cereal crop globally, following wheat and barley. With the rapid growth of the global population and the ongoing effects of climate change, increasing agricultural production and productivity have become increasingly important (Öner, 2023). In addition to being consumed directly as a food source, maize also has extensive applications in the industrial sector. It is utilized in the production of numerous products, including starch, ethanol, sorbitol, dextrin, beer, ice cream, ink, and cosmetics (Du Plessis, 2003). Native to Central America and Mexico, maize spread to other parts of the world after the discovery of the Americas and was introduced to Türkiye in the 1600s (Dallar, 2017). Currently, maize is cultivated on approximately 9.5 million hectares of land in Türkiye, yielding an annual production of around 9 million tons (TUIK, 2025).

Maize is an annual crop that thrives in warm climates and exhibits considerable genetic diversity. Factors such as optimal temperature, soil structure, irrigation, and fertilization are critically important for its cultivation.

However, environmental conditions like frost, poor soil quality, and humidity can adversely affect yield in maize-growing regions. Moreover, maize is highly susceptible to fungal diseases, which can result in root and stalk rot, seedling blight, and ear rot, ultimately reducing both yield and quality (Emeklier, 2018).

Currently, approximately 80% of all plant diseases are caused by fungal pathogens. Among these, *Fusarium verticillioides* is a major pathogenic agent that affects maize, as well as other plants such as tomatoes, bulbous crops, sweet potatoes, cucurbits, bananas, cotton, and ornamentals (Miller et al., 1994). This pathogen causes root, stalk, and ear rot as well as seedling blight in maize, leading to yield losses, poor grain quality, and the production of harmful mycotoxins that pose risks to both human and animal health (Logrieco et al., 2002). Warm temperatures and adequate moisture provide ideal conditions for fungal growth and infection (Miller et al., 1994). *Fusarium*-induced ear rot in maize is a widespread fungal disease observed across Europe, the Americas, Africa, Asia, and Oceania. The two primary fungal agents responsible for maize ear rot globally are



Fusarium graminearum and *Fusarium verticillioides*. Stalk rot, caused by soil-borne fungi, blocks xylem vessels, leading to wilting and ultimately plant death (Okungbowa and Shittu, 2014).

The interaction between *Fusarium verticillioides* and maize seeds has long been a controversial topic among plant pathologists and seed producers. This pathogen, which causes stalk and root rot in the field and ear rot post-harvest, has led to significant losses for farmers. As a mycotoxigenic pathogen capable of systemic infection through seeds, *Fusarium verticillioides* can reduce maize seed germination and vigor to varying degrees. In recent years, restrictions on the use of chemical fungicides have led to an increased focus on research into plant resistance inducers. Pre-sowing treatments and seed coating techniques represent promising strategies to improve the germination of diseased or low-vigor seeds (Aguado et al., 2019).

Gibberellins (GAs) are a large group of tetracyclic diterpenoid carboxylic acids, although only a few of them function as growth hormones in plants. Among them, GA₃ has been shown to exert beneficial effects on seed germination, leaf expansion, stem elongation, and the development of flowers and fruits. Due to their role in promoting plant growth, gibberellins are considered essential throughout the plant life cycle. They also support developmental phase transitions. Recent studies have increasingly focused on their roles in plant responses to abiotic stress and adaptation. GA₃ is known to interact with other phytohormones across numerous developmental and stimulus-response processes (Wani et al., 2016).

In recent years, restrictions on the use of chemical fungicides have sparked growing interest in exploring natural antifungal properties in plants and environmentally friendly control methods (Aguado et al., 2019). *Melia azedarach* L. contains natural compounds with antifungal, antibacterial, and insecticidal properties. Its seeds and leaves have shown potential for use as botanical fungicides in agricultural settings. *Melia azedarach* L. is a large tree belonging to the *Meliaceae* family. Native to Asia, it is now also found in parts of Northern Australia, Africa, the Americas, and Southern Europe. It is known by various names worldwide, including Chinaberry tree, Persian lilac, Indian lilac, and White cedar (Ntalli et al., 2010).

Melia azedarach L. is a deciduous, perennial tree that can reach heights of up to 45 meters and is highly drought-resistant (Seth, 2004). Its bark is smooth and greenish-brown when young, turning gray and fissured with age. The dark green leaves are compound, 20–40 cm long, and composed of opposite leaflets. The fragrant flowers range in color from white to lilac and purple, and are borne in axillary clusters on slender stems. The fruits are small, round, yellow drupes containing hard, dark brown to black seeds. These plant parts have been reported to possess antiviral, antibacterial, antifungal, antioxidant, and insecticidal properties due to their chemical constituents (Sharma and Pal, 2013).

This study aims to investigate the antifungal effects of *Melia azedarach* L. fruit and leaf extracts on maize seeds infected with *Fusarium verticillioides*, to develop an environmentally friendly and sustainable approach to disease management.

2. Materials and Methods

This study was conducted in 2023-24 at the Biotechnology and Tissue Culture Laboratories of the Faculty of Agriculture, Sakarya University of Applied Sciences. The experiment was designed as a randomized plot trial with three replications. The experimental materials included seeds of the Kale (F1) maize cultivar and a *Fusarium verticillioides* spore suspension (5×10^5 spores mL⁻¹), both obtained from the Sakarya Maize Research Institute (Figure 1). Fruits and leaves of the *Melia azedarach* L. were supplied by the Atatürk Horticultural Central Research Institute (Figure 2). Six treatment groups were established for the experiment as Positive Control (germination water), Negative Control (*Fusarium* + distilled water), Fruit Extract (*Fusarium* + fruit powder extract), Leaf Extract (*Fusarium* + leaf powder extract), Fruit Extract + GA₃ (*Fusarium* + fruit powder extract + gibberellic acid), Leaf Extract + GA₃ (*Fusarium* + leaf powder extract + gibberellic acid).

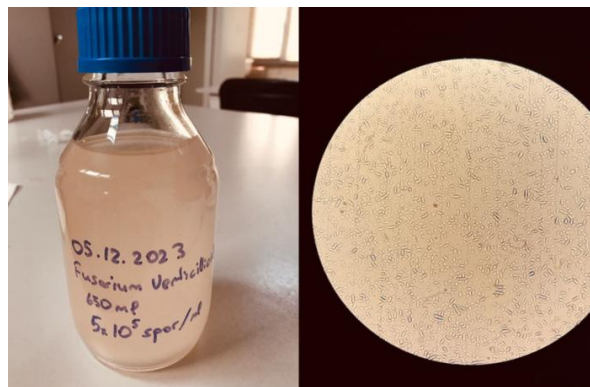


Figure 1. *Fusarium verticillioides* (5×10^5 spores mL⁻¹) suspension and microscopic image.



Figure 2. Pre-drying view of *Melia azedarach* L. leaves and fruits.

Preparation of *Melia azedarach* Leaf and Fruit Extracts
The fruits of *Melia azedarach* were separated from their

seeds and dried together with the leaves in an oven at 70 °C for 24 hours. After drying, the plant materials were ground into powder. The leaves were extracted using ethanol, while the fruits were extracted with methanol. The leaf extract was prepared by mixing 10 parts of leaf powder with 40 parts of ethanol in a 50 mL Falcon tube. Similarly, the fruit extract was prepared by mixing 10 parts of fruit powder with 40 parts of methanol in another 50 mL Falcon tube. The tubes were placed on an orbital shaker for 48 hours to ensure thorough extraction. Following this, the solvent was evaporated at room temperature to remove ethanol and methanol (Figure 3). The resulting dry extracts were re-dissolved in distilled water to prepare the final solutions for application.

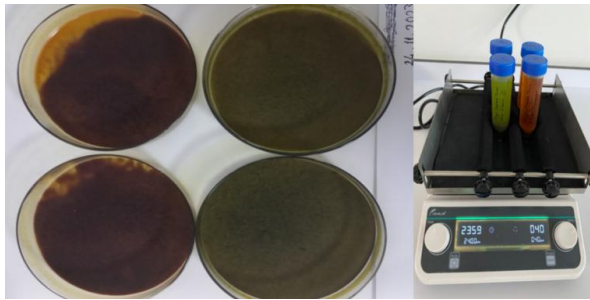


Figure 3. Preparation of *Melia azedarach* L. leaf and fruit extracts

2.1. Surface Sterilization of Maize Seeds

A 0.5% sodium hypochlorite (NaClO) solution was prepared and used to sterilize the maize seeds by immersing them in a 1000 mL beaker containing the solution for 15 minutes. After sterilization, the seeds were transferred onto blotting paper to dry and then made ready for use in the experimental trials.

2.2. Experimental Treatments

Surface-sterilized and dried seeds were placed in Petri dishes, 30 seeds per dish, on filter paper, and then covered with another layer of filter paper. *Melia azedarach* leaf and fruit extracts, prepared in distilled water, were transferred into spray bottles and applied directly to the seeds. The seeds were then dried in an oven at 35 °C for 30 minutes and placed back into petri dishes.

For the positive control, a 0.5% sodium hypochlorite solution was used as germination water. The negative control group consisted of seeds inoculated with *Fusarium verticillioides* (5×10^5 spores/mL) and treated only with distilled water. In treatment groups involving *Melia azedarach* extracts, the seeds were first sprayed with the extracts and then inoculated with the *Fusarium* suspension. All Petri dishes were sealed with stretch film to prevent airflow and maintain the integrity of the fungal infection, and labeled accordingly.

The prepared Petri dishes were incubated for 7 days under controlled conditions: 23 °C, 65% relative humidity, and complete darkness. Observations and measurements were conducted following the rules of the International Seed Testing Association (ISTA), evaluating the following parameters: germination rate (%) on days 2, 3, and 7; *Fusarium* contamination rate (%) on days 3 and 7; root length (mm); and shoot length (mm) (Figure 4).

2.3. Statistical Analysis

The data obtained from the experiment were analyzed using the MSTAT-C statistical software package. After performing analysis of variance (ANOVA), Duncan's multiple range test was applied to compare the significance levels of the treatments.

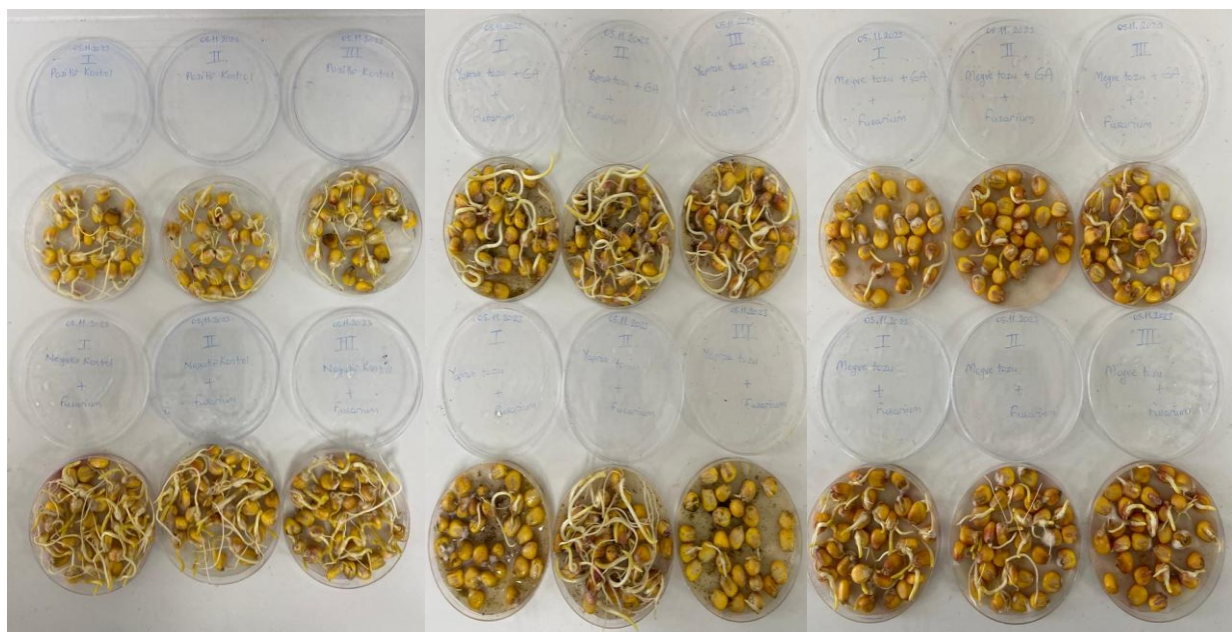


Figure 4. View of experimental treatments on day 7.

3. Results

3.1. Germination Rates (%)

In this study, germination rates on days 2, 3, and 7; *Fusarium* contamination rates on days 3 and 7; as well as

root and shoot lengths, were all found to be statistically significant at the 1% level. The mean values for these parameters and the results of Duncan's multiple range test are presented below (Table 1).

Table 1. Germination rates (%) on days 2, 3, and 7

Treatment	Germination rates (%)		
	Day 2	Day 3	Day 7
Positive Control	23.33a	58.88ab	77.77a
Negative Control	28.88a	67.78a	74.44a
Fruit Extract	0.000b	41.11ab	55.56ab
Leaf Extract	0.000b	34.44b	35.56b
Fruit Extract + GA ₃	1.112b	37.78b	37.78b
Leaf Extract + GA ₃	2.223b	47.78ab	48.89b
LSD (1%)	9.69**	27.11**	24.33**

** Significant at 1%.

Treatments involving fruit and leaf extracts, as well as their combinations with GA₃, exhibited significantly lower germination rates compared to the positive and negative control groups. These findings are consistent with previous studies. For example, Monteiro et al. (2024) reported germination rates ranging from 65.1% to 69.5% with garlic and ginger bio-extracts, while clove extract reduced germination to just 3.0% due to its toxic effects.

Yıldız et al. (2020) observed that nettle extract reduced maize germination from 40–45% down to 20%, with similar inhibitory effects reported for kale extract. These findings suggest that some plant extracts may contain allelopathic or toxic compounds detrimental to the germination process.

De Oliveira et al. (2024) examined microbial seed inoculations and found that a combination of *Azospirillum brasilense* and *Bacillus subtilis* yielded a 90% germination rate, while applying four microorganisms simultaneously reduced germination to 67.5%. This

highlights the importance of microbial composition and compatibility in priming.

Kadir et al. (2023) emphasized that hydropriming improved maize germination to 96.7%, whereas the control group remained below 82%, further confirming the beneficial role of seed priming.

The current study revealed that *Melia azedarach* leaf and fruit extracts exhibited allelopathic effects by reducing germination rates compared to controls. Even in combination with GA₃, germination remained substantially lower. These results suggest that the chemical composition of extracts, environmental conditions, and the applied priming/inoculation method all influence germination performance.

3.2. *Fusarium* Contamination Rates (%)

Contamination levels caused by *Fusarium verticillioides* were assessed on days 3 and 7 following the application of various extract treatments. Results are presented in Table 2.

Table 2. *Fusarium verticillioides* contamination rates (%)

Treatment	Contamination rates (%)	
	Day 3	Day 7
Positive Control	0.000c	0.000b
Negative Control	16.67bc	50.00a
Fruit Extract	36.67a	50.00a
Leaf Extract	26.67ab	40.00a
Fruit Extract + GA ₃	33.33ab	37.78a
Leaf Extract + GA ₃	34.44a	48.89a
LSD (1%)	16.96**	20.50**

** Significant at 1%.

Monteiro et al. (2024) found that garlic and ginger bio-extracts significantly suppressed *Fusarium* contamination, while clove extract had a minimal effect. Garlic extract, in particular, was highlighted for its ability to reduce contamination by up to 90%.

De Oliveira et al. (2024) also reported that *Trichoderma harzianum* was highly effective in reducing *Fusarium* infection, while combinations involving *Bacillus subtilis* and *Bacillus megaterium* were less effective under certain conditions.

Ashraf et al. (2020) demonstrated that silver nanoparticles synthesized using *Melia azedarach* leaf extract suppressed *Fusarium oxysporum* by 79–98%, suggesting strong antifungal potential and the utility of nanotechnology in pathogen management.

Han et al. (2023) emphasized the potential of GA₃ to promote germination and enhance plant resistance against pathogens such as *Fusarium*. However, in the present study, GA₃-based combinations were less effective than expected, likely due to interactions with the extracts or environmental variability.

Martínez-Álvarez et al. (2016) introduced a *Bacillus cereus* B25 spore-based formulation as an effective biocontrol agent against *F. verticillioides*, underscoring the need for advanced formulation strategies. In contrast, the direct application of plant extracts in our study yielded comparatively limited suppression, indicating that efficacy may depend on both formulation and

method of application.

Our findings suggest that *Fusarium* contamination levels vary based on the antifungal potential of the extracts and their combinations with GA₃. The complete absence of contamination in the positive control confirms the effectiveness of sterilization. However, the limited effectiveness of the fruit extract in pathogen control could be due to insufficient antifungal compounds. Leaf extract applications showed a ~10% lower contamination rate compared to the negative control.

3.3. Root and Shoot Length (mm)

Root and shoot lengths were measured on day 7, and the data were presented in Table 3. Dilmenler (2021) observed a reduction in shoot length from 49.00 mm (control) to 21.00 mm under salt stress (18 dS m⁻¹). Sezen and Küçük (2023) reported that a 1% dose of *Microcystis viridis* and *Aphanizomenon gracile* mixture increased shoot length from 48.00 mm to 67.68 mm.

Table 3. Root and shoot lengths (mm) by treatment

Treatment	Root Length (mm)	Shoot Length (mm)
Positive Control	28.67b	17.73c
Negative Control	27.43b	25.90b
Fruit Extract	18.80c	24.40b
Leaf Extract	47.03a	57.13a
Fruit Extract + GA ₃	16.00c	23.10b
Leaf Extract + GA ₃	23.83bc	49.23a
LSD (1%)	8.142**	8.385**

** Significant at 1%.

Zahedifar (2013) found that shoot length in salinity-stressed maize increased to 53.00 mm with 1.5% potassium nano-chelate but declined at higher concentrations due to toxicity. Yıldız et al. (2020) reported that nettle extract reduced shoot length by over 50%, from 40–45 mm to 20–25 mm, with similar results observed for kale extract.

The current study confirms that *Melia azedarach* leaf extract significantly promoted shoot growth, whereas fruit extracts and GA₃ combinations had a more limited effect. These outcomes are consistent with the literature, which suggests that shoot growth depends on environmental stressors, toxicity levels, and the chemical composition of the applied materials. The positive impact of the leaf extract is likely linked to beneficial phytochemicals enhancing shoot development.

Dilmenler (2021) reported a decline from 24.30 mm to 11.20 mm for root length under salt stress. Sezen and Küçük (2023) observed an increase from 23.50 mm to 37.60 mm with 2% cyanobacterial application. Shah et al. (2021) found root length improved from 32.10 mm to 42.00 mm under 60 ppm TiO₂ application. Zahedifar (2013) achieved 74.02 mm root length with 1.5% nano-chelate but observed toxicity at 3%. Yıldız et al. (2020) noted complete inhibition of root growth with 20% nettle extract.

4. Conclusion

This study examined the antifungal effects of *Melia azedarach* L. leaf and fruit extracts on seeds of the Kale maize variety infected with *Fusarium verticillioides*, as well as their influence on germination parameters. The results indicated that *Melia azedarach* L. extracts had a suppressive effect on the germination rate of infected seeds. However, among those seeds that did germinate, noticeable improvements were observed in various growth parameters. The findings indicate that leaf extract treatments promoted root and shoot development and improved germination in infected seeds, likely due to bioactive phytochemicals present in the extract. *Melia azedarach* L. may serve as a promising natural alternative in the management of *Fusarium* pathogens.

Nonetheless, further research is warranted to evaluate the effects of *Melia azedarach* across different maize cultivars and under varying environmental conditions. Additionally, studies should be expanded to explore how such natural solutions can be integrated into agricultural systems as environmentally friendly alternatives to chemical control methods.

The extracts and bioactive compounds of *Melia azedarach* hold potential as eco-friendly agents in

biological control strategies. However, more extensive research and field applications are required to validate and optimize their use. It is also recommended that long-term environmental impacts and potential side effects of such botanical interventions be carefully assessed.

In conclusion, identifying the most effective application methods and appropriate dosages of *Melia azedarach* extracts is essential to enhance agricultural productivity while preserving ecological balance. The conscious and effective utilization of such natural alternatives may contribute significantly to developing sustainable agricultural practices.

Author Contributions

The percentages of the authors' contributions are presented below. All authors reviewed and approved the final version of the manuscript.

	R.C.	Z.Y.
C	90	10
D	90	10
S	90	10
DCP	10	90
DAI	70	30
L	20	80
W	60	40
CR	80	20
SR	90	10
PM	60	40
FA	90	10

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The authors declare no conflict of interest. This research received no external funding.

Ethical Consideration

The article does not require an ethics committee decision.

Acknowledgments

We would like to thank the Sakarya Maize Research Institute for providing the maize variety (Kale F1) and *Fusarium verticillioides* solution used in the study, and the Atatürk Horticultural Central Research Institute for providing the *Melia azedarach* L. plant material.

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