SYNERGISTIC EFFECT OF *ORIGANUM COMPACTUM* ESSENTIAL OIL AND TETRACONAZOLE ON THE GROWTH OF *FUSARIUM CULMORUM*

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Abstract: Agriculture is a key pillar of Morocco's socio-economic framework, with the cereal sector playing a crucial role. Cereal yields are significantly affected by biotic and abiotic constraints, such as drought and fungal diseases, particularly root rot caused by *Fusarium culmorum*. To mitigate these issues, chemical treatments are commonly used; however, they pose environmental and human health risks.

This study explores an alternative approach by evaluating the synergistic effects of different concentrations of *Origanum compactum* essential oil and tetraconazole on *Fusarium culmorum* growth. Using the poisoned medium method, results showed that adding 0.29 mg/mL of *Origanum compactum* essential oil to tetraconazole achieved complete inhibition of *Fusarium culmorum 1*, while a lower concentration of 0.07 mg/mL was sufficient to inhibit *Fusarium culmorum 2*. This antifungal activity is primarily attributed to the high carvacrol content in the essential oil, known for its strong efficacy against phytopathogens.

Keywords : *Fusarium culmorum*, *Origanum compactum*, Antifungal, concomitant action, tetraconazole.

INTRODUCTION

Antioxidant Cereals and their derivatives hold significant nutritional, social, and economic importance. The average annual per capita cereal consumption exceeds 200 kg, accounting for 25% of household food expenditures. In livestock farming, cereals, straw, and oat bran provide 40% of total forage unit requirements. Additionally, cereals contribute 30% to the Agricultural Gross Domestic Product and represent 45% of total food imports, making their trade a key driver of the national economy (Ait el Mekki, 2006).

Despite this, cereal yields remain relatively low in both rainfed and irrigated agricultural zones. Several factors contribute to this issue, including fragmented land holdings, climatic fluctuations, limited financial resources for farmers, and biotic and abiotic stresses such as pests and fungal diseases (Zidane *et al.*, 2010).

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Fungal diseases such as septoria, rusts, and root rot pose major threats to cereal production, causing significant qualitative and quantitative losses that hinder national cereal cultivation. Common root rot can lead to characteristic discoloration of nodes and roots. This disease is complex and involves multiple soilborne fungi, with *Bipolaris sorokiniana* and *Fusarium culmorum* being the primary causal agents (Zahraoui *et al.*, 2023,2024).

Given the economic significance of agriculture, effective disease management strategies are essential. Control methods include cultural practices, genetic resistance, biological control, and chemical treatments (Pintureau, 2006). Among these, chemical fungicides remain the most widely used solution, playing a crucial role in protecting crops and sustaining agricultural productivity (Chang *et al.*, 2008).

However, concerns about fungicide residues in food products and the emergence of resistant pathogens underscore the need for alternative control strategies. One promising approach involves the use of natural compounds against phytopathogens. Historically, plant diseases have had devastating effects on crop yields, but in recent years, the search for bioactive natural products with antimicrobial properties has gained momentum (Nosrati *et al.*, 2010).

Research suggests that alternative control methods can effectively inhibit root rot pathogens. Plant-derived natural products have emerged as potential sources of new fungicides for managing fungal diseases (Chang *et al.*, 2008). Essential oils and plant extracts have gained significant attention due to their antioxidant properties, bioactive compounds, and antifungal and antimicrobial activities. These properties have led to their application in food preservation, pharmaceuticals, alternative medicine, and natural therapies (Naeini *et al.*, 2010).

In this context, the present study aims to evaluate the synergistic effect of *Origanum compactum* essential oil and tetraconazole in controlling *Fusarium culmorum*–induced root rot while reducing the reliance on synthetic fungicides.

MATERIALS AND METHODS

FUNGAL ISOLATES OF *FUSARIUM CULMORUM*

Fusarium culmorum was isolated from wheat roots exhibiting root rot and identified at the Phytopathology Laboratory of INRA in Settat. Two isolates of *Fusarium culmorum* were tested:

- *Fusarium culmorum* 1 (Fc1): This isolate was obtained from a farmer's field in Oujda, in the Oriental region. It is characterized by strong resistance to chemical treatments, likely due to the excessive use of chemical products.
- *Fusarium culmorum* 2 (Fc2): This isolate is sensitive to chemical treatment. It was isolated from the root rot of wheat cultivated at an agronomic

station in Jemaa Shaim, in the Marrakech-Safi region, where fungicides are used in a more rational manner.

ANTIFUNGALS

Tetraconazole – Chemical Fungicide

Tetraconazole is a chemical fungicide commercially available and used in agriculture. It has been shown to be effective in controlling fungal diseases, providing good protection against root infections and reducing the severity of diseases caused by *Fusarium culmorum*. The concentration used is that indicated on the bottle, which is 125 g/L.

Essential Oil (EO) of *Origanum compactum Benth*

The essential oil of *Origanum compactum Benth* was provided by the company SANTIS-Sarl. It was extracted via hydro distillation from the leaves, stems, and flowers of raw material harvested between April and July 2009 in the Ouazzane-Taounate region.

Chemical analysis by Gas Chromatography (GC), conducted at CNRST, showed that this *Origanum compactum* essential oil is primarily composed of 43.97% Carvacrol, 17.87% p-Cymene, and 11.56% Thymol, making it a highly active antifungal alternative (Zahraoui *et al.*, 2017).

Poisoned-agar Technique

A series of 1/2 dilutions of the tetraconazole and essential oil is prepared and incorporated into Potato Dextrose Agar (PDA) culture medium, in a supercooled state between 40 and 45°C in test tubes. These tubes, containing different concentrations, are then poured into Petri dishes and left to solidify. A 6 mm mycelial disc is placed at the center of each Petri dish. The dishes are incubated at 28°C for 5 to 7 days. Each experiment was repeated twice (Soylu *et al.*, 2010).

DETERMINATION OF TETRACONAZOLE INHIBITION PARAMETERS

Preparation of Dilutions

A series of 1/2 dilutions of tetraconazole is prepared starting from an initial concentration of 1.25 mg/mL, down to 0.0011 mg/mL. Eleven test tubes are used to prepare the successive dilutions, along with a control tube containing only the PDA culture medium. The first tube contains 10 mL of PDA, and 100 µL of tetraconazole is added. A 5 mL volume is taken from this tube and added to the next tube, and so

on. These prepared tubes with different concentrations are then poured into sterile Petri dishes. After solidifying the medium, a 6 mm mycelial disc from each *Fusarium culmorum* isolate is placed at the center of each dish. The Petri dishes are incubated at 28°C for 5 to 7 days. Each experiment was repeated twice.

EVALUATION OF RESULTS

The cultures obtained allow the determination of:

- Minimum Inhibitory Concentration (MIC): This is the lowest concentration that inhibits fungal growth. It is determined by observing the development of the mycelial disc.
- Minimum Fungicidal Concentration (MFC): Based on the MIC results, this is determined by placing the undeveloped mycelial discs in a sterile Petri dish containing only PDA culture medium. After incubation, if fungal growth is observed, the concentration is fungistatic (it stopped growth but did not kill the fungus). If there is no growth, the concentration is fungicidal (it stopped growth and killed the fungus) (Soylu *et al.*, 2010).
- Median Inhibitory Concentration (IC_{50}): This is determined from the dose-response curve equations for the concentrations of tetraconazole and the concomitant activity of essential oils (Kumar *et al.*, 2014).
- Inhibition Rate (IC %): This is calculated using the following formula:

IC (%) = (D(C-) - DT) / D(C-) × 100

Where:

D(C-) = the diametric growth of the colony without fungicide (control).

DT = the diametric growth of the fungus in the presence of a fungicide concentration.

SELECTION OF TESTED CONCENTRATIONS OF *ORIGANUM COMPACTUM BENTH* ESSENTIAL OIL

The previous study (Zahraoui *et al.*, 2017) showed that this essential oil has a variable MIC depending on each *Fusarium culmorum* isolate. Therefore, two different concentrations of the essential oil for each isolate were tested:

Case of *Fusarium culmorum* 1

- 0.29 mg/mL: This concentration is both the minimum inhibitory and fungistatic concentration.
- 0.14 mg/mL and 0.07 mg/mL: These concentrations are below the MIC and allowed fungal development.

A series of 1/2 dilutions of *Origanum compactum* essential oil is prepared from an initial concentration of 9.11 mg/mL to 0.07 mg/mL. Eight test tubes are used to prepare the successive dilutions, with a control tube containing only the PDA culture medium. The first tube contains 10 mL of PDA, and 100 µL of *Origanum compactum* essential oil is added. A 5 mL volume is taken from this tube and added to the next tube, and so on. The two tubes with concentrations of 0.14 mg/mL and 0.07 mg/mL are poured into sterile Petri dishes.

Case of *Fusarium culmorum* 2

The concentrations used are 0.07 mg/mL and 0.035 mg/mL. These two concentrations are below the MIC of 0.14 mg/mL, which is fungicidal (it stopped the growth and killed the fungus). These two concentrations were prepared in the same way as the *Fc2* isolate.

SIMULTANEOUS ACTIVITY OF *ORIGANUM COMPACTUM* ESSENTIAL OIL AND TETRACONAZOLE

Preparation of Dilutions of Both Antifungals

The dilutions prepared differ between isolates due to their varying sensitivity to the tested antifungals.

Case of *Fusarium culmorum* 1

- Three series of 1/2 dilutions of tetraconazole were made from an initial concentration of 1.25 mg/mL down to 0.0012 mg/mL.
- Six series of 1/2 dilutions of *Origanum compactum* essential oil were prepared from an initial concentration of 9.11 mg/mL down to 0.07 mg/mL.

To prepare the concentrations of 0.29 mg/mL, 0.14 mg/mL, and 0.07 mg/mL of *Origanum compactum* essential oil, the tubes with concentrations of 0.57 mg/mL, 0.29 mg/mL, and 0.14 mg/mL of the essential oil were added to tubes with tetraconazole concentrations lower than the MIC, ranging from 0.039 mg/mL to 0.0012 mg/mL. The tubes were then poured into sterile Petri dishes. After the medium solidified, a 6 mm mycelial disc from *Fusarium culmorum* 1 was placed at the center of each dish. The Petri dishes were incubated at 28°C for 5 to 7 days.

Case of *Fusarium culmorum* 2

- Two series of 1/2 dilutions of tetraconazole were made from an initial concentration of 1.25 mg/mL down to 0.0012 mg/mL.
- Seven series of 1/2 dilutions of *Origanum compactum* essential oil were prepared from an initial concentration of 9.11 mg/mL down to 0.035 mg/mL.

To prepare the concentrations of 0.07 mg/mL and 0.035 mg/mL of *Origanum compactum* essential oil, the tubes with concentrations of 0.14 mg/mL and 0.07 mg/mL were added to tubes with tetraconazole concentrations lower than the MIC, ranging from 0.07 mg/mL to 0.0012 mg/mL. The tubes were then poured into sterile Petri dishes. After solidification of the medium, a 6 mm mycelial disc from *Fusarium culmorum* 2 was placed at the center of each dish. The Petri dishes were incubated at 28°C for 5 to 7 days.

Evaluation of Results

The diametric growth measurements of the mycelium were used to calculate the percentages of inhibition as well as the IC_{50}, MIC, and MFC.

Statistical Analysis

The results from the means of two repetitions for IC_{50}, MIC, and MFC parameters were compared using analysis of variance (ANOVA) followed by mean comparison using the Duncan test at a 5% probability, using SPSS 22 software.

RESULTS

TETRACONAZOLE

Fusarium *culmorum*1 (Fc1)

The results of the antifungal activity of tetraconazole on the Fc1 strain showed that concentrations between 1.25 mg/mL and 0.0781 mg/mL completely inhibit the growth of this strain. The two concentrations 1.25 mg/mL and 0.625 mg/mL are fungicidal, whereas the other inhibitory concentrations are fungistatic (Table I). For the lower concentrations from 0.0391 mg/mL to 0.0012 mg/mL, mycelial growth is observed progressively as the tetraconazole concentrations decrease (Figure 1).

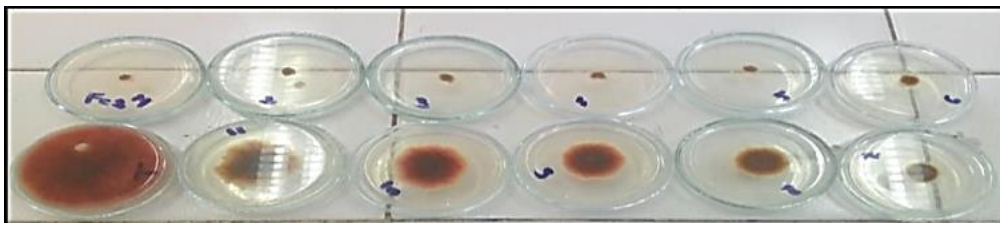

Figure 1. Antifungal activity of tetraconazole on the growth of *Fusarium culmorum* 1.

Table 1

Inhibition parameters of tetraconazole against *Fusarium culmorum* 1

Concentrations (mg/mL)	Mycelium diameter (cm)	Inhibition percentage (%)	Effect
1.25	0	100	Fungicidal
0.625	0	100	Fungicidal
0.3125	0	100	Fungistatic
0.1563	0	100	Fungistatic
0.0781	0	100	Fungistatic
0.0391	0.30	93.33	–
0.0195	0.75	83.33	–
0.0098	1.40	68.89	–
0.0049	1.90	57.78	–
0.0024	2.30	48.89	–
0.0012	2.40	46.67	–
0	4.50	0	–

***Fusarium culmorum*2 (Fc2)**

The inhibition percentages reported in Table II show that the concentrations of 1.25 mg/mL and 0.625 mg/mL have a fungicidal effect, while 0.3125 mg/mL and 0.1563 mg/mL are fungistatic, leading to complete inhibition. A mycelial growth is observed at concentrations ranging from 0.0781 mg/mL to 0.0012 mg/mL (Figure 2).

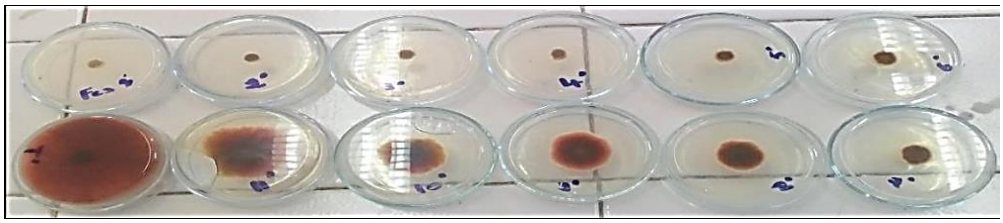

Figure 2. Antifungal activity of tetraconazole on the growth of *Fusarium culmorum*2.

Table 2

Inhibition parameters of tetraconazole against *Fusarium culmorum* 2

Concentrations (mg/mL)	Mycelium diameter (cm)	Inhibition percentage (%)	Effect
1.25	0	100	Fungicidal
0.625	0	100	Fungicidal
0.3125	0	100	Fungistatic
0.1563	0	100	Fungistatic

Concentrations (mg/mL)	Mycelium diameter (cm)	Inhibition percentage (%)	Effect
0.0781	0.10	97.78	–
0.0391	0.35	92.22	–
0.0195	0.60	86.67	–
0.0098	1.25	72.22	–
0.0049	1.80	60	–
0.0024	2.00	55.56	–
0.0012	2.90	35.56	–
0	4.50	0	–

The obtained results for the two strains of *Fusarium culmorum* allow the determination of the following inhibition parameters (table 3).

Table 3

IC50, MIC and MFC of tetraconazole

Strains	**IC_{50} (mg/mL)**	**MIC (mg/mL)**	**MFC (mg/mL)**
Fusarium culmorum (Fc1)	0.0025	0.00781	0.625
Fusarium culmorum (Fc2)	0.0022	0.1563	0.625

The comparison of the inhibition percentages of tetraconazole against the two *Fusarium culmorum* isolates showed that concentrations between 1.25 mg/mL and 0.1563 mg/mL completely inhibit both isolates. For the other concentrations, variations were observed in the inhibition percentages between the two isolates, with a stronger sensitivity for the Fc1 strain.

CONCOMITANT ANTIFUNGAL ACTIVITY OF TETRACONAZOLE AND ESSENTIAL OIL OF *ORIGANUM COMPACTUM*

FUSARIUM CULMORUM1 (FC1)

Essential Oil of *Origanum compactum* at 0.29 mg/mL

The addition of 0.29 mg/mL of essential oil of *Origanum compactum* to the non-inhibitory tetraconazole concentrations showed that these concentrations became inhibitory at 100% with a fungicidal effect (Table 3 and Figure 4).

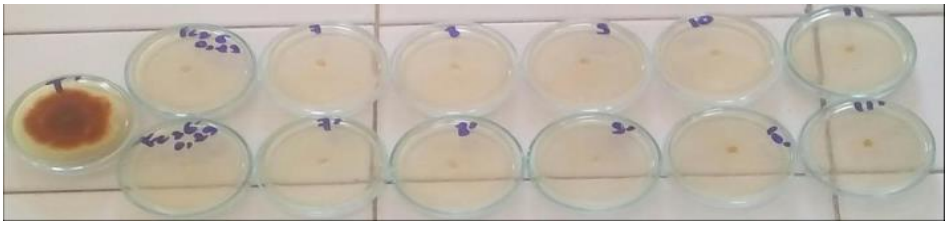

Figure 3. Concurrent activity of tetraconazole and essential oil of *Origanum compactum* at 0.29 mg/mL on Fc1.

The comparison of the activity of tetraconazole alone and tetraconazole combined with the essential oil of *Origanum compactum* at 0.29 mg/mL showed that the combined action is more effective against *Fusarium culmorum*2, with an inhibition rate of 100%, whereas the inhibition rate of tetraconazole decreases progressively with concentrations (Figure 4).

Table 4

Inhibition parameters of the concurrent activity of tetraconazole and essential oil of *Origanum compactum* at 0.29 mg/mL against Fc1

Tetraconazole concentrations (mg/mL)	Mycelium diameter (cm)	Inhibition percentage (%)	Effect
0.0391	0	100	Fungicidal
0.0195	0	100	Fungicidal
0.0098	0	100	Fungicidal
0.0049	0	100	Fungicidal
0.0024	0	100	Fungicidal
0.0012	0	100	Fungicidal
0	4.5	0	–

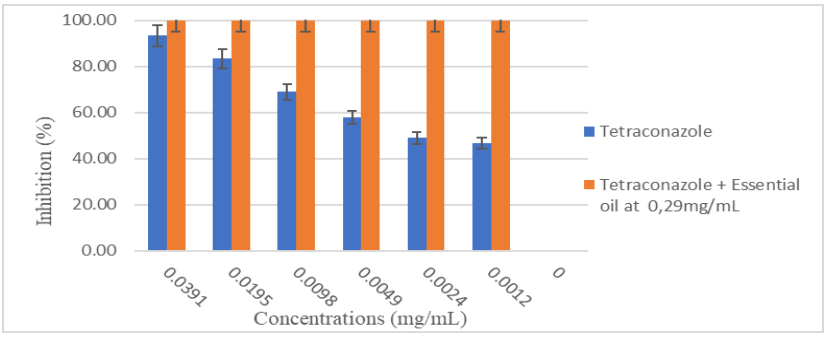

Figure 4. Comparison of the antifungal activity of tetraconazole alone and tetraconazole combined with essential oil at 0.29 mg/mL on Fc1.

Essential Oil of *Origanum compactum* at 0.14 mg/mL

For the Fc1 isolate, concentrations ranging from 0.0391 mg/mL to 0.0049 mg/mL exhibit a fungistatic effect with inhibition percentages of 100%. However, concentrations of 0.0024 mg/mL and 0.0012 mg/mL have inhibition rates of 91.67% and 77.08%, respectively (Table 5 and Figure 5). These results reveal a minimum inhibitory concentration (MIC) of 0.0049 mg/mL, a median inhibitory concentration (IC_{50}) of 0.0008 mg/mL, and a minimum fungicidal concentration (MFC) greater than 0.0391 mg/mL.

Table 5

Inhibition parameters of the simultaneous activity of tetraconazole and *Origanum compactum* essential oil at 0.14 mg/mL on Fc1

Tetraconazole Concentrations (mg/mL)	Mycelium Diameter (cm)	Inhibition Percentage (%)	Effect
0.0391	0	100	Fungistatic
0.0195	0	100	Fungistatic
0.0098	0	100	Fungistatic
0.0049	0	100	Fungistatic
0.0024	0.2	91.67	–
0.0012	0.55	77.08	–
0	2.4	0	–

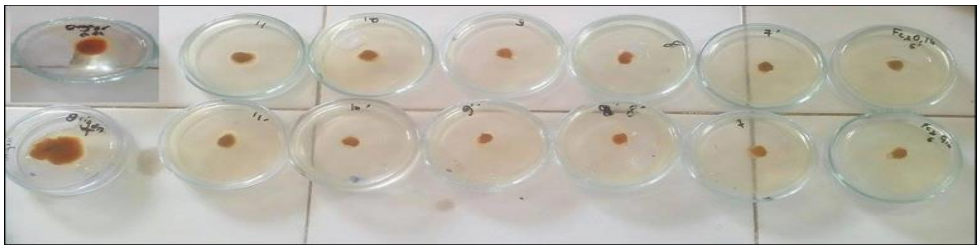

Figure 5. Concomitant activity of tetraconazole and Origanum compactum essential oil at 0.14 mg/mL on Fc1.

The study of the concomitant activity of tetraconazole in the presence of 0.14 mg/mL *Origanum compactum* essential oil showed that the latter increased the effectiveness of tetraconazole at all studied concentrations. The results show that concentrations ranging from 0.00391 mg/mL to 0.0049 mg/mL have inhibition percentages ranging from 93.33% to 57.78%, which were increased to 100% in the presence of the essential oil. Likewise, for the other two concentrations, a clear improvement in activity was observed (Figure 6).

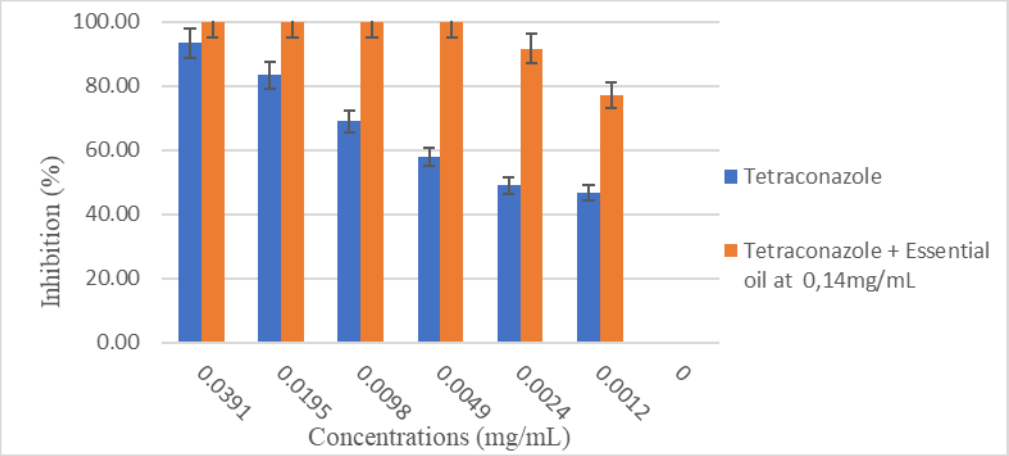

Figure 6. Comparison of the antifungal activity of tetraconazole alone and tetraconazole combined with essential oil at 0.14 mg/mL on Fc1.

Origanum compactum essential oil at 0.07 mg/mL

The combined action of tetraconazole and essential oil at 0.07 mg/mL exerts moderate inhibition on the growth of the Fc1 isolate, with inhibition percentages ranging from 77.08% to 60.42% (Table 6 and Figure 7). These results allowed us to estimate an MIC and MFC greater than 0.0391 mg/mL, and to calculate the IC_{50}, which has a value of 0.0010 mg/mL.

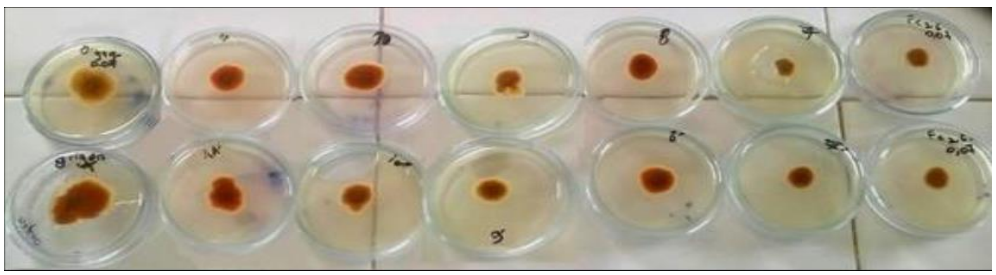

Figure 7. Concomitant activity of tetraconazole and Origanum compactum essential oil at 0.07 mg/mL on Fc1.

The comparative study of tetraconazole and *Origanum compactum* essential oil at 0.07 mg/mL showed that the latter improved the antifungal effectiveness of low concentrations of tetraconazole, namely 0.0049 mg/mL, 0.0024 mg/mL, and 0.0012 mg/mL.

Table 6

Inhibition parameters of the simultaneous activity of tetraconazole and *Origanum compactum* essential oil at 0.07 mg/mL on Fc1

Tetraconazole Concentrations (mg/mL)	Mycelium Diameter (cm)	Inhibition Percentage (%)	Effect
0.0391	0.55	77.08	–
0.0195	0.53	78.13	–
0.0098	0.85	64.58	–
0.0049	0.90	62.50	–
0.0024	0.90	62.50	–
0.0012	0.95	60.42	–
0	2.4	0	–

There is also an interaction between tetraconazole and the essential oil at concentrations 0.0391 mg/mL, 0.0195 mg/mL, and 0.0098 mg/mL. This interaction led to an antagonistic effect that reduced the effectiveness of tetraconazole at these three concentrations (Figure 8).

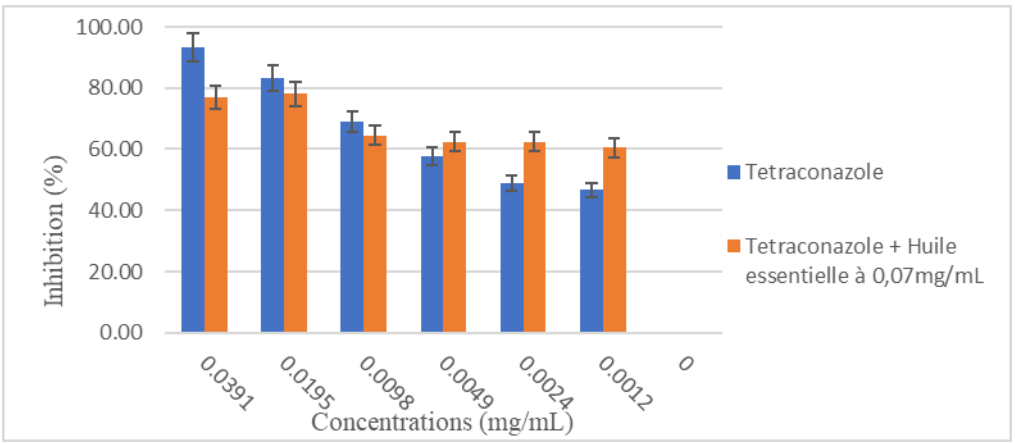

Figure 8. Comparison of the antifungal activity of tetraconazole alone and tetraconazole in the presence of essential oil at 0.07 mg/mL on Fc2.

FUSARIUM CULMORUM2 (FC2)

Origanum compactum essential oil at 0.07 mg/mL

The inhibition percentages reported in Table 6 show that the concentration of 0.0781 mg/mL is fungicidal, while other concentrations exert variable inhibition below 100% (Table 7 and Figure 9).

Table 7

Inhibition parameters of the simultaneous activity of tetraconazole and *Origanum compactum* essential oil at 0.07 mg/mL on Fc2

Tetraconazole Concentrations(mg/mL)	Mycelium Diameter (cm)	Inhibition Percentage (%)	Effect
0.0781	0	100	Fungicide
0.0391	0.78	82.78	–
0.0195	0.88	80.56	–
0.0098	1.1	75.56	–
0.0049	1.75	61.11	–
0.0024	1.85	58.89	–
0.0012	2.7	40	–
0	4.5	0	–

The median inhibitory concentration is 0.002 mg/mL, and both the minimum inhibitory concentration and minimum fungicidal concentration are 0.0781 mg/mL (Figure 10).

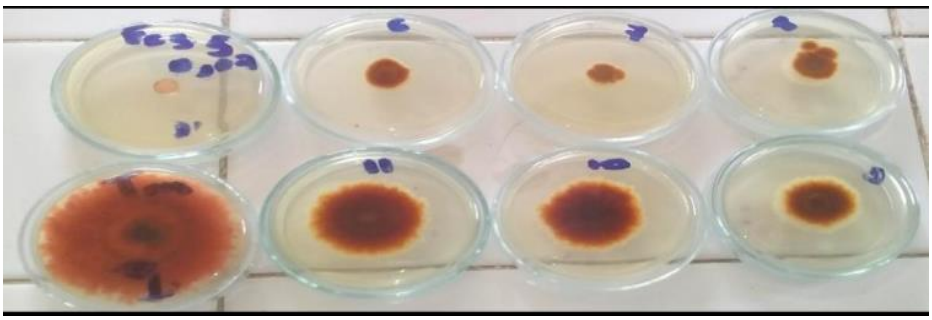

Figure 9. Concomitant activity of tetraconazole and *Origanum compactum* essential oil at 0.07 mg/mL on Fc2.

Analysis of Figure 10 showed that *Origanum compactum* essential oil at 0.07 mg/mL exerts significant antifungal activity in combination with tetraconazole concentrations ranging from 0.0049 mg/mL to 0.0012 mg/mL.

The antagonistic interaction is only observed at the concentrations 0.0391 mg/mL and 0.0195 mg/mL.

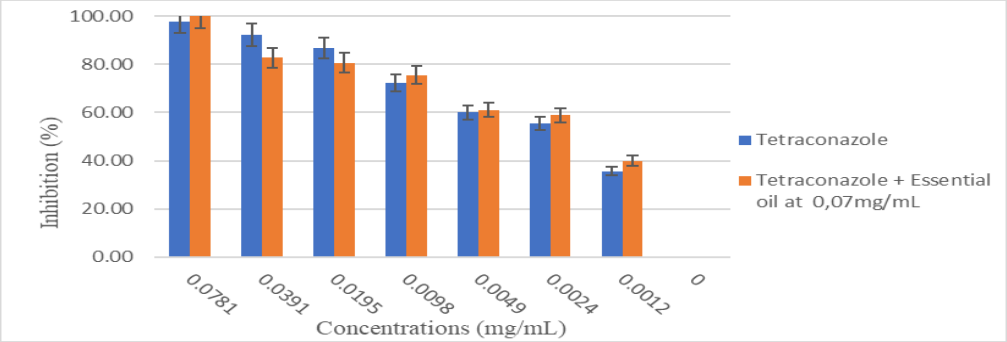

Figure 10. Comparison of the antifungal activity of tetraconazole alone and tetraconazole in the presence of essential oil at 0.07 mg/mL on Fc2.

Origanum compactum essential oil at 0.035 mg/mL

The combined action of tetraconazole and essential oil at 0.035 mg/mL showed a progressive decrease in antifungal activity, represented by inhibition percentages ranging from 94.44% to 40% (Table 8 and Fig. 11).

Table 8

Inhibition parameters of the simultaneous activity of tetraconazole and *Origanum compactum* essential oil at 0.07 mg/mL on Fc2

Tetraconazole Concentrations (mg/mL)	Mycelium Diameter (cm)	Inhibition Percentage (%)	Effect
0.0781	0.25	94.44	–
0.0391	0.75	83.33	–
0.0195	1.2	73.33	–
0.0098	1.4	68.89	–
0.0049	1.7	62.22	–
0.0024	2.15	52.22	–
0.0012	2.7	40	–
0	4.5	0	–

The inhibition percentages reported in Table 8 allowed us to calculate the inhibition parameters, with an IC_{50} of 0.0023 mg/mL and concentrations for both the minimum inhibitory and fungicidal concentrations greater than 0.0781 mg/mL.

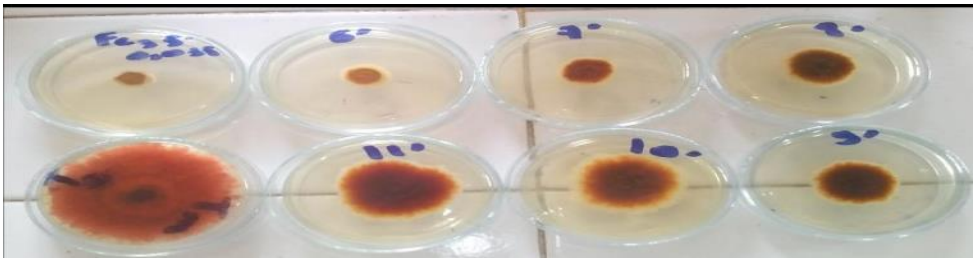

Figure 11. Concomitant activity of tetraconazole and *Origanum compactum* essential oil at 0.035 mg/mL on Fc2.

The activity of tetraconazole in the presence of essential oil at 0.035 mg/mL showed an interaction that reduced the inhibitory power of tetraconazole at concentrations between 0.0781 mg/mL and 0.0098 mg/mL. This essential oil slightly increased the inhibition of mycelial development of the Fc2 strain compared to tetraconazole at concentrations ranging from 0.0049 mg/mL to 0.0012 mg/mL for the other concentrations (Figure 12).

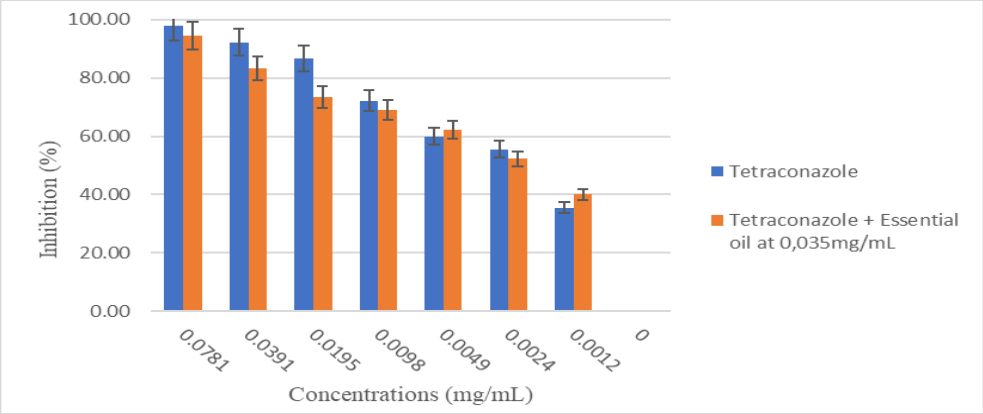

Figure 12. Comparison of the antifungal activity of tetraconazole alone and tetraconazole in the presence of essential oil at 0.035 mg/mL on Fc2.

STATISTICAL ANALYSIS OF ANTIFUNGAL ACTIVITY PARAMETERS

The statistical analysis of the values of IC_{50}, MIC, and MFC (table 9) showed that, for the Fc1 strain, there is no significant difference between the IC_{50} of the concomitant activity of tetraconazole in the presence of the three concentrations of *Origanum compactum* essential oil (0.29mg/mL, 0.14mg/mL, and 0.07mg/mL). These IC_{50} values remain lower than the IC_{50} of tetraconazole alone. These conclusions also hold true for the MIC and MFC, with a significant difference observed between the concentrations 0.29mg/mL and 0.14mg/mL.

For the Fc2 isolate, the IC_{50} analysis showed no significant difference between the two combined activities of tetraconazole and the concentrations of 0.07mg/mL and 0.035mg/mL of *Origanum compactum* essential oil, and tetraconazole. However, for the MIC and MFC, a significant difference was noticed between tetraconazole and *Origanum compactum* essential oil at 0.07mg/mL, which was more active than tetraconazole alone.

Table 9

Comparison of Treatments with Respect to IC_{50}, MIC, and MFC (mg/mL)

	Strains					
	Fc1			**Fc2**		
Treatments	**IC_{50}**	**MIC**	**MFC**	**IC_{50}**	**MIC**	**MFC**
Tetraconazole	0,0025[b]	0,0781[c]	0,625[b]	0,0022[a]	0,1563[b]	0,625[b]
Tetraconazole + Essential Oil at 0,29	<0,0012[a]	<0,0012[a]	0,0012[a]	–	–	–
Tetraconazole + Essential Oil at 0,14	0,0008[a]	0,0049[b]	>0,0391	–	–	–
Tetraconazole + Essential Oil at 0,07	0,0010[a]	>0,0391	>0,0391	0,0021[a]	0,0781[a]	0,0781[a]
Tetraconazole + Essential Oil at 0,035	–	–	–	0,0023[a]	>0,0781	>0,0781

NB: Values in the same column followed by the same letter are not significantly different according to Duncan's test at a 5% probability level.

DISCUSSION

For the two *Fusarium culmorum* isolates tested, the study of the concurrent activity of tetraconazole and different concentrations of essential oil from *Origanum compactum* showed that the addition of this essential oil improves the effectiveness of tetraconazole. The concentrations of 0.29 mg/mL, 0.14 mg/mL, 0.07 mg/mL, and 0.035 mg/mL of essential oil added reduced the IC_{50}, MIC, and MFC values of tetraconazole. Antagonistic interactions were observed during this study, particularly at the concentrations of 0.07 mg/mL for the Fc2 strain and 0.07 mg/mL and 0.035 mg/mL for the Fc3 strain.

A comparative study on the mycelial growth of *Fusarium culmorum* proved that the essential oil of *Origanum compactum* inhibits the development of the fungus at low concentrations, unlike the essential oils of thyme and clove, which require higher concentrations (Dobre and Niculiga, 2012).

The essential oil of *Origanum compactum*, which contains carvacrol, thymol, and p-cymene, was shown to be more toxic than *Origanum acutidens*, which is only rich in carvacrol, on several *Fusarium* strains, including one of *Fusarium culmorum*. These compounds alter and lyse fungal cells (Kordali *et al.*, 2008). Another study showed that phenolic compounds such as carvacrol and thymol disrupt the structure of fungal cell membranes (Akthar *et al.*, 2014). The high content of carvacrol and thymol gives this essential oil its antifungal properties (Belkamel *et al.*, 2013).

It has also been demonstrated that the mechanism of toxicity of phenols against fungi is based on the inactivation of fungal enzymes containing the SH group in their active sites (Fadel *et al.*, 2013).

The results of the interaction between *Origanum compactum* essential oil and tetraconazole align with those of the study by Catinca *et al.*, (2014), which highlighted the existence of an interaction between cardamom essential oil and amoxicillin or ciprofloxacin. This interaction can be either additive or antagonistic depending on the concentration ratio between the essential oil and the antibiotics. This study also showed that at high antibiotic concentrations, the interaction was antagonistic, whereas at lower concentrations, the interaction was additive.

The study by Van Vuuren *et al.* (2009) demonstrated that the combined action of several essential oils and conventional antimicrobials, such as Amphotericin B and ciprofloxacin, can be either additive or antagonistic depending on the concentrations used. These interactions are due to the chemical complexity of essential oils.

The concurrent activity of cumin essential oil and fluconazole shows the existence of a synergistic interaction between the conventional antifungal and the compounds in this essential oil (Patil, 2015).

CONCLUSION

The findings of this study highlight the significant potential of ***Origanum compactum*** essential oil as a natural antifungal agent. The observed inhibition of ***Fusarium culmorum*** growth suggests that essential oils could play a crucial role in plant disease management. Their use as an alternative or an additive to synthetic fungicides presents multiple advantages, including reducing the reliance on chemical treatments, minimizing environmental impact, and lowering toxicity risks for humans and ecosystems.

Moreover, integrating essential oils into crop protection strategies could contribute to sustainable agricultural practices by promoting eco-friendly disease

control methods. However, further research is necessary to explore the mechanisms underlying their antifungal properties, optimize their application methods, and evaluate their long-term effectiveness in real agricultural settings. This approach could pave the way for innovative and safer plant protection solutions, reducing the adverse effects associated with conventional fungicides.

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