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Article

Effect of Brassica Species Biofumigants on Wheat Plants Infected with *Fusarium pseudograminearum*

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Abstract

Fusarium head blight (FHB), is a serious fungal disease of wheat, worldwide causing yield losses and grain contamination with mycotoxins that jeopardize food and feed safety. This study will look into the effectiveness of using biofumigation as a safe alternative for wheat Fusarium head blight control in order to avoid using fungicides that are destructive to human health, plants, and soil.

The biofumigation treatments with three brassica crops (Cabbage – Cauliflower – Mustard and the fungicide treatment celest fs 10 % had significant effects on the growth characteristics of the two wheat cultivars Giza 171 and Misr 2. After measuring the crop characteristics of the two cultivars, wheat was noted that Giza 171 was the most affected by the fungal infection, while the cultivar Misr 2 was the least affected. All brassicas-biofumigation treatments (incorporation of brassica plants into soil) were very effective in enhancing wheat growth parameters and reduced fusarium head blight indexes compared with untreated control. This study found that biofumigation was a very effective treatment for fusarium head blight disease in wheat, so we recommend it as a very safe alternative to conventional fungicides that cause major damage to human and animal health, as well as soil and water.

Key words: *Fusarium* – *Brassica* – Biofumigation – Wheat - *pseudograminearum*

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1.Introduction

Wheat (*Triticum aestivum* L.) is an important cereal crop for food security around the world. It's cultivated all over the world (Singroha et al., 2017). According to the FAO's 2020 report, 215 million hectares of wheat generated 774.8 million metric tons of wheat with an average yield of 3.6 metric tons per hectare, In Egypt wheat generated 8.9 million metric tons in 2020/2021 (https://www.fao.org/faostat).

It is in charge of delivering carbohydrates and other essential nutrients, accounting for more than 40% of all nutritional requirements.

Wheat is not only an important nutritional cereal crop in Egypt, but it is also recognized as a key to food security. Unfortunately, wheat is frequently infected by a variety of diseases, resulting in substantial losses in yield quality and quantity. There is a considerable gap between wheat grain yield and country consumption. (Ouda & Zohry, 2017).

In Egypt. as a result, conserving each grain from wheat production is considered a pressing necessity. Better treatment of fungal-incited diseases, which can cause production losses of 15 –20 % per year, is a significant component in solving this problem. Several fungal infections are currently contributing to these losses in wheat (Figueroa et al., 2018). Fusarium head blight (FHB) is one of the most common and destructive diseases, capable of severely reducing crop output as well as quality. FHB outbreaks were first recorded in England in 1884, when it was known as "wheat scab" (Bai & Cai, 2018). It is a devastating disease of cereal crops that affects cereal production all over the world (Bai & Cai, 2018). Fusarium head blight (FHB) is caused by several Fusarium species, *F. graminearum*, *F.pseudograminearum*, *F. avenaceum* and *F. culmorum* (Ali & Mahmoud, 2019 and Mylonas et al., 2020).

This study was undertaken in order to evaluate the effects of biofumigation with various Brassica species on certain growth and agronomic characters of two wheat cultivars infected with FHB causal agent under greenhouse conditions at Matrouh governorate, Egypt.

2.Materials and Methods

2.1. Plant material

Three Brassica plants, cabbage (*Brassica oleracea var. oleracea*), cauliflower (*Brassica oleracea var. botrytis*) and mustard (*Sinapis alba*) were used in the current study. Seeds were obtained from Mecca trade company, Egypt. Grains of wheat Cultivar Giza 171 and Misr 2 were used for the biofumigation experiment, Grains of wheat Cultivars Giza 171 and Misr 2 were obtained from Agriculture Research Centre, Giza, Egypt.

2.2. Soil infestation

Plastic pots (disinfested with 5% sodium hypochlorite solution) 30 cm in diameter containing 5 kg autoclaved sandy soil. Soil infestation was carried out one week before planting brassica crops. Inoculum of the isolated fungus *Fusarium pseudograminearum* was prepared by adding 1 cm disc of the tested fungus in 250 ml conical flasks containing 100 gm of autoclaved barley and incubated at 28 ± 2 C° for 3-4 weeks. The sterilized pots filled with the autoclaved soil were infested with each of the tested fungal inoculum at the rate of 5 % of soil weight. Wheat seeds were planted in the infested soil after 24 hours from incorporation of brassica crops.

2.3. Preceding biofumigants planting and treatments application

Three types of cruciferous crops, cabbage) *Brassica oleracea var. oleracea*), cauliflower (*Brassica oleracea var . botrytis*)and mustard (*Sinapis alba*),were selected as a soil biofumigants. Biofumigants as well as the fungicide (Celest 10% FS) were applied to the sterilized soil. Cabbage and cauliflower seeds were sown in the nursery then transplanted into the pot after 6 weeks. Mustard seeds were directly sown in the pot on 19 August in both seasons. Agricultural best practices were followed.

The preceding Brassica crops were directly incorporated into the soil at maturity stage after 80 days from planting, on 8 November. Plants were chopped and incorporated into the soil to a depth of no more than 10 cm, resulting in a fine tilth. Then, after irrigating the soil in the pots to its field capacity, the soil surface was tightly covered with a transparent plastic film for 21 days to keep the gases produced by the biodegradation of the organic matter from influencing the outcome.

on the other hand, wheat seeds were treated before planting with Celest 10 % FS, Fludioxonil as its active ingredient as separate treatment. The application rate was 100 ml /100 kg seed as recommended by (Aveling, et al. 2013).

Wheat grains from the Giza 171 and Misr 2 cultivars were sown 24 hours later from incorporation of brassica crops on 30 November in both seasons. Plants in the control group were left untreated and uncovered, but were irrigated to field capacity in a similar way. The experiment was repeated twice on 2019/2020 and 2020/2021 seasons (**Fig. 2).**

2.4. Experimental design and Statistical analysis

The greenhouse experiment comprised of four treatments in two cultivars (Giza171- Misr 2) namely, Cabbage, Cauliflower, mustard and the fungicide Celest.in addition to two untreated control (infested and non-infested). The treatments were arranged in a randomized complete block design in four replicates (RCBD). Data were statistically analyzed as completely randomized block design (RCBD), by using Genstat12th edition computer program. The least significant difference (L.S.D) at 0.05 level was used for comparing the differences between means **(**GenStat, 2009).

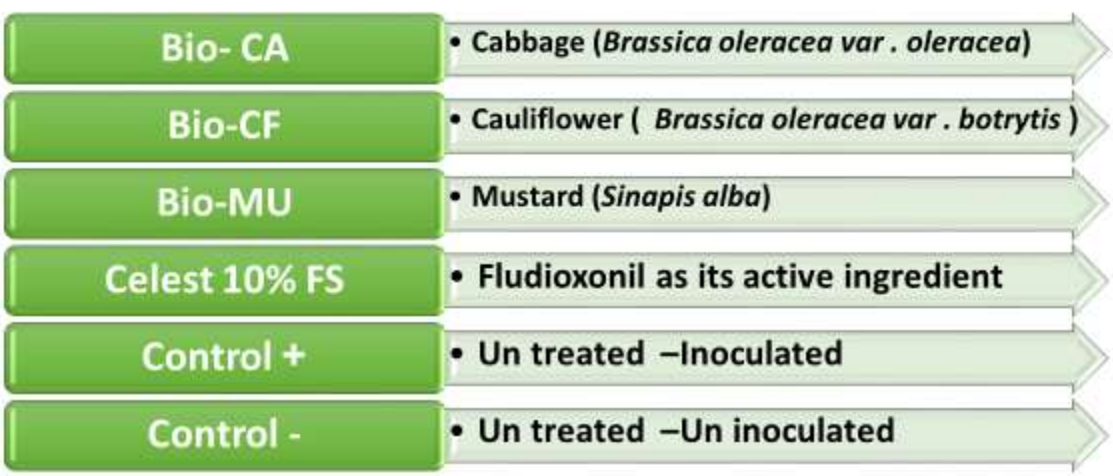

Figure. (1): The greenhouse experiment treatments abbreviations

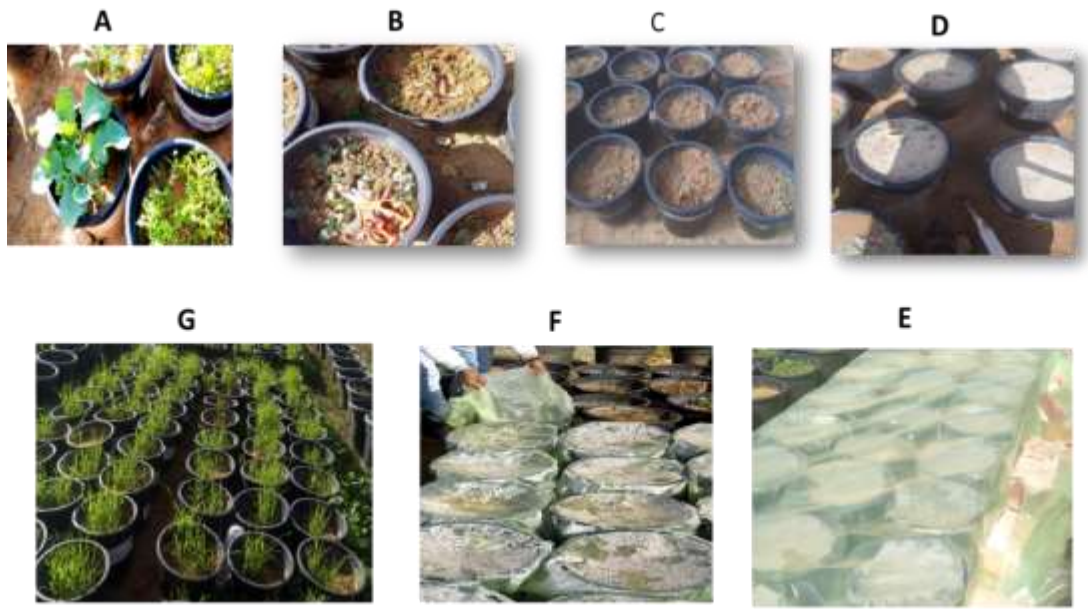

Figure. (2): Biofumigants application processing

A-Biofumigants plants in the pots under greenhouse conditions, **B-**Macerating biofumigants tissues at maturity stage, **C-** Incorporation biofumigants into the soil, **D-**Irrigating the soil to its field capacity, **E-** covering the soil with a transparent plastic film (2 layers), **F-** Removing the transparent plastic film after 21 days. **G-** Wheat planting under greenhouse conditions.

3.Results and Discussion

The data in the following section shows how biofumigation and fungicide Celest fs 10 % affect wheat plant growth parameters.

3.1. Plant height (cm):

The effect of biofumigation with *Brassica* spp., and fungicide on plant height of the two wheat cultivars was revealed in **Table (1)** in both seasons (2019/2020) and (2020/2021), there was no significant differences between biofumigation with mustard and Celest FS 10 % treatments resulted in the highest means of plant height compared with control ,in the first season biofumigation with mustard on both cultivars Giza 171 and Misr 2 respectively resulted in (89.12 and 82.75 cm) while in the second season resulted in (89.00 and 82.31cm). Celest fs 10 % treatment on Giza 171 cultivar resulted in (89.12 and 86.69 cm) in both seasons 2019/2020 and 2020/2021, respectively. While Celest fs 10 % treatment on Misr 2 cultivar resulted in (82.81 and 83.00 cm) in both seasons.

Table 1. The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on plant heights (cm) of wheat plants cultivated in soil infested with *Fusarium pseudograminearum*

Treatments	Season 2019/2020		Season 2020/2021	
	Giza171	Misr 2	Giza171	Misr 2
Control +	70.87	70.06	72.50	71.19
Control -	92.25	84.50	93.75	84.25
Bio.CA	87.19	79.00	86.38	80.75
Bio.CF	83.37	77.25	83.44	78.81
Bio.MU	89.12	82.75	89.00	82.31
Celest Fs 10%	89.12	82.81	86.69	83.00
L.S.D 0.05	0.89		0.96	

Values are means of plant heights. (%). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-) plants without fungus treatments. Bio.CA, Bio.CF, and Bio.MU indicate biofumigation with cabbage, cauliflower and mustard, respectively.

3.2. Number of spikes m^{-2}:

Table (2) shows the effect of biofumigation with the three Brassica spp., as well as fungicide, on the number of spikes m^{-2} of the two wheat cultivars.

In both season (2019/2020) and (2020/2021), biofumigation with mustard, cabbage and Celest fs 10 % treatments resulted in the highest means of number of spikes m^{-2}, in the first season biofumigation with mustard on Giza 171 cultivar resulted in (431.50/m^2) while biofumigation with mustard on Misr 2 cultivar resulted in (499.20/m^2), Celest Fs 10 % treatment resulted in (445.30/m^2 and 535.80/m^2) on Giza 171 and Misr 2 cultivars, respectively. In the second season biofumigation with mustard resulted in (439.30/m^2 and 507.80/m^2) on Giza 171 and Misr 2 cultivars, respectively. Investigating the effect of treatments on number of panicle of cultivars indicated that Celest 10 % was the highest significant effect in both 2019/2020 and 2020/2021 seasons.

Table 2. The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on number of Spikes (m^{-2}) of wheat plants cultivated in soil infested with *Fusarium pseudograminearum*.

Treatments	**Season 2019/2020**		**Season 2020/2021**	
	Giza171	**Misr 2**	**Giza171**	**Misr 2**
Control+	339.50	427.80	360.80	430.00
Control -	455.80	508.20	454.80	506.20
Bio.CA	416.50	520.20	412.80	489.50
Bio.CF	389.00	509.80	378.50	469.00
Bio.MU	431.50	499.20	439.30	507.80
Celest Fs 10%	445.30	535.80	463.30	509.20
L.S.D 0.05	21.74		13.21	

Values are means of number of spikes m^{-2}. (%). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-)plants without fungus treatments. Bio.CA, Bio.CF, and Bio. MU indicate biofumigation with cabbage, cauliflower and mustard, respectively.

3.3. Number of grains/spike:

Biofumigation with mustard and Celest FS10% treatments resulted in the highest means of number of grains/spike, in the first season biofumigation with mustard resulted in (47.00 and 51,00 grains / spike) on Giza 171 and Misr 2 cultivars, respectively while Celest Fs 10% treatment resulted in (50.50 and 54.75 grains / spike) on Giza 171 and Misr 2 cultivars, respectively. In the second season biofumigation with mustard on Giza 171 and Misr 2 resulted in (49.25 and 51.00 grains / spike), while Celest Fs 10 % treatment resulted in (49.75 and 54.00 grains / spike) on Giza 171 and Misr 2 cultivars, respectively. **(Table 3).**

Table.3. The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on number of grains/spike of wheat plants cultivated in soil infested with *Fusarium pseudograminearum.*

Treatments	season 2019/2020		season 2020/2021	
	Giza171	Misr 2	Giza171	Misr 2
Control (+)	28.50	32.50	26.25	32.25
Control (-)	51.00	54.00	51.50	53.50
Bio.CA	44.50	48.50	45.50	48.50
Bio.CF	40.25	45.00	41.00	44.50
Bio.MU	47.00	51.00	49.25	51.00
Celest Fs 10%	50.50	54.75	49.75	54.00
L.S.D 0.05	1.46		2.48	

Values are means of numbers grains/spike. (%). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-) plants without fungus treatments. Bio.CA, Bio.CF, and Bio.MU indicate biofumigation with cabbage, cauliflower and mustard, respectively.

3.4. 1000 grain weight (g):

The effect of biofumigation with *Brassica* spp., and fungicide on 1000 grain weight of the two wheat cultivars was revealed in **Table (4)** In both season (2019/2020) and (2020/2021), biofumigation with mustard and Celest Fs 10 % treatments resulted in the highest means of 1000 grain weight, in the first season biofumigation with mustard resulted in (75.93 and 74.98 g) on both cultivars Giza 171 and Misr 2 respectively. While Celest Fs 10 % treatment resulted in (80.33 and 79.40 g). In the second season biofumigation with mustard resulted in (75.98 and 75.93 g) on both cultivars Giza 171 and Misr 2 respectively. While Celest Fs 10 % treatment resulted in (80.60 and 77.43 g).

Table .4 The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on 1000 grain weight of wheat plants cultivated in soil infested with *Fusarium pseudograminearum*.

Treatments	Season 2019/2020		Season 2020/2021	
	Giza171	Misr 2	Giza171	Misr 2
Control (+)	66.38	65.28	66.93	65.68
Control (-)	81.50	77.25	81.00	76.50
Bio.CA	74.48	74.15	74.70	73.13
Bio.CF	73.23	72.93	73.45	71.78
Bio.MU	75.93	74.98	75.98	75.93
Celest Fs 10%	80.33	79.40	80.60	77.43
L.S.D 0.05	0.72		0.51	

. Values are means of 1000 grain weight (g). (%). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-)plants without fungus treatments. Bio.CA, Bio.CF, and Bio.MU indicate biofumigation with cabbage, cauliflower and mustard, respectively.

3.5. Harvest index (%):

Table (5) shows the effect of biofumigation with the three *Brassica spp*. and fungicide on the Harvest Index (%) of the two wheat cultivars.

In both seasons (2019/2020) and (2020/2021), biofumigation with mustard and Celest Fs 10 % treatments resulted in the highest means of harvest index (%), in the first season biofumigation with mustard on Giza 171 and Misr 2 cultivars respectively resulted in (29.50 and 29.00 %), while Celest FS 10 % treatment resulted in (32.50 and 35.00 %). In the second season biofumigation with mustard resulted in (30.00 and 29.00 %). While Celest fs 10 % treatment resulted in (32.50 and 34.50 %) on Giza 171 and Misr 2 cultivars, respectively.

Table 5. The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on harvest index % weight of wheat plants cultivated in soil infested with *Fusarium pseudograminearum*.

Treatments	season 2019/2020		season 2020/2021	
	Giza171	Misr 2	Giza171	Misr 2
Control +	23.00	25.00	23.50	24.75
Control -	32.00	35.50	32.25	34.25
Bio.CA	27.25	27.75	27.75	27.75
Bio.CF	25.50	26.50	25.50	26.50
Bio.MU	29.50	29.00	30.00	29.00
Celest Fs 10%	32.50	35.00	32.50	34.50
L.S.D 0.05	1.20		1.52	

Values are means of harvest index (%). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-) plants without fungus treatments. Bio.CA, Bio.CF, and Bio.MU indicate biofumigation with cabbage, cauliflower and mustard, respectively.

3.6. Grain yield (g/pot):

The efficacy of biofumigation with three species of Brassica and fungicide on grain yield (g/pot) of two wheat cultivars was evaluated.

Table (6) In both season (2019/2020) and (2020/2021), biofumigation with mustard and Celest fs 10 % treatments resulted in the highest means of grain yield (g/pot), in the first season biofumigation with mustard resulted in (78.87and 83.63 g/pot) on both cultivars Giza 171 and Misr 2. While Celest FS10 % treatment resulted in (80.04 and 85.60 g/pot). In the second season biofumigation with mustard resulted in (78.07and 84.80g/pot). While Celest fs 10 % treatment resulted in (80.54 and 86.09 g/pot) on both cultivars Giza 171 and Misr 2, respectively.

Table .6. The effect of interaction between wheat cultivars (Giza171 and Misr 2) & biofumigation with three *Brassica spp*, fungicide on grain yield (g/pot) of wheat cultivars Giza171and Misr 2.

Treatments	**season 2019/2020**		**season 2020/2021**	
	Giza171	**Misr 2**	**Giza171**	**Misr 2**
Control +	81.74	87.89	82.79	87.95
Control -	43.23	54.07	41.89	53.67
Bio.CA	75.27	80.89	74.73	81.05
Bio.CF	72.77	75.26	71.76	75.07
Bio.MU	78.87	83.63	78.07	84.80
Celest Fs 10%	80.04	85.60	80.54	86.09
L.S.D 0.05	1.03		0.97	

Values are means of grain yield (g/pot). Each value is mean of four replicates. Control (+) plants treated with the fungus and control (-)plants without fungus treatments. Bio.CA, Bio.CF, and Bio.MU indicate biofumigation with cabbage, cauliflower and mustard, respectively. Each value is mean of four replicates.

The biofumigation treatments resulted in considerable increase in plant height when compared to plants that were not treated, these results are consistent with Sarhan et al. 2020, they found that the reduction in damping off due by biofumigation treatments was reflected in the chickpea yield components as increased plant height under field conditions.

According to Oka et al., 2007, green manure application in the soil, is advantageous not only for disease management but also for boosting plant growth and productivity. Several other studies found that using *Brassica* spp. as seed meal or green manure suppresses soil-borne pathogenic fungi, enhancing plant growth and yield by the release of volatile biocidal compounds, primarily isothiocyanates (ITCs), produced by hydrolyzed Brassica spp. in the soil (Smolinska et al., 2003; Matthiessen & Kirkegaard, 2006 ; Mazzola et al., 2007).

According to the results, the treatment with bio-fumigation resulted in a significant increase in the weight of 1000 grains as compared to the non-treated control. These results are comparable with those of Hansen & Keinath 2013, who found that brassica treatments enhanced pepper yields as significantly as some other treatments. Sarhan et al., 2020, the biofumigation of chickpea plants with mustard and canola seed meals was studied against soil-borne pathogens *F. oxysporum*, *S. sclerotiorum*, and *R. solani.* The released volatile biocidal compounds, primarily isothiocyanates (ITCs), derived from the hydrolyzed *Brassica* spp. in the soil, increased yield in vitro, greenhouse, and field conditions. In contrast, Hartz et al., 2005 found that using overwintering mustard cover crops had no consistent effects on soilborne disease control or tomato fruit productivity in six field studies. Green manure application in the soil, according to Oka et al., 2007, is beneficial not only for disease management but also for enhancing plant growth and productivity.

According to Matthiessen & Kirkegaard 2006, indirect effects on the pathogen related with changes in antagonistic organism populations, as well as impacts of compounds released from the tissues, are among the reasons for suppression effects from crucifer residue incorporation.

According to the results, the treatments with biofumigants resulted in a significant increase in grain yield, this increase was due to the increase in wheat yield components which were as follows, Number of spikes m^{-2} , number of grain / spike and 1000 grain weight in both cultivars in two seasons but the cultivar Misr 2 resulted in the highest means of grain yield and its components .

Conclusion

When compared to the untreated control, all brassicas-biofumigation treatments (incorporation of brassica plants into soil) were very effective in improving wheat growth metrics under greenhouse conditions. Biofumigation may represent a practical strategy for growing wheat crop in soil infected with FHB.

All biofumigation treatments applied following the removal of brassicas crops resulted in significant increases in wheat yield. Biofumigation may represent a practical strategy for growing wheat crop in soil infested with FHB. This technique may be applied with other plant disease management tactics such as crop rotation and the use of resistant varieties, in both organic and conventional agriculture systems, providing a very safe alternative for traditional fungicides which may cause significant harm to human and animal health.

Further work is needed at the field level in order to assess the role of biofumigation in improving wheat growth parameters in FHB infested soil under field conditions.

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تأثيرالتبخير الحيوي بواسطة بعض أصناف العائلة الصليبية علي نباتات القمح المصابة بفطر *Fusarium pseudograminearum*

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الملخص العربي

يعتبر مرض لفحة سنابل القمح (السنبلة البيضاء) ، مرض فطري خطير يصيب محصول القمح و يتسبب في خسائرفادحة في المحصول كماً ونوعاً في جميع أنحاء العالم بالإضافة إلي تلوث الحبوب بالسموم الفطرية التي تعرض سلامة الغذاء والأعلاف للخطر. تختلف علامات وأعراض مرض السنبلة البيضاء تبعًا لمرحلة الإصابة. تنتج العدوى في مرحلة مبكرة اللون البني في أول اثنين أو ثلاثة من السلاميات القاعدية. يلاحظ وجود الرؤوس البيضاء (السنابل المصابة) في الحقل أثناء الإصابة الشديدة ويمكن رؤيتها بوضوح عندما لا تزال نباتات القمح خضراء قبل النضج التام لها.

بحثت هذه الدراسة في فعالية استخدام التبخير الحيوي كبديل آمن لمكافحة مرض السنبلة البيضاء في القمح من أجل تجنب استخدام مبيدات الفطريات التي تضر بصحة الإنسان والنبات والتربة ,بعد قياس خصائص محصول الصنفين لوحظ أن جيزة 171 كانت الأكثر تأثراً بالعدوى الفطرية الطبيعية ، بينما الصنف مصر 2 كان الأقل تأثر(الأكثر مقاومة).

كانت جميع معاملات التبخير الحيوي بالصليبيات (دمج نباتات العائلة الصليبية في التربة) فعالة جدًا في تعزيز معايير نمو القمح وتقليل مؤشرات مرض السنبلة البيضاء مقارنةً بنباتات القمح المنزرعة في تربة غير معاملة وبالأخص المعاملة بالخردل .

وجدت هذه الدراسة أن التبخير الحيوي كان علاجًا فعالاً للغاية للسيطرة علي مرض السنبلة البيضاء في القمح ، لذلك توصي الدراسة به كبديل آمن لمبيدات الفطريات التقليدية التي تسبب أضرارًا جسيمة لصحة الإنسان والحيوان ، وكذلك التربة والمياه و انه لمن الأهمية بمكان تعزيز الوعي بضرورة استبدال مبيدات الفطريات الكيميائية الضارة بتدابير مكافحة آمنة بديلة.

الكلمات المفتاحية :

القمح - التبخير الحيوي - الصليبيات – السنبلة البيضاء – الأيزوثيوسينات - *Fusarium- pseudograminearum*