Research Article

Monika Grzanka*, Łukasz Sobiech, Jakub Danielewicz, Joanna Horoszkiewicz-Janka, Grzegorz Skrzypczak, Zuzanna Sawinska, Dominika Radzikowska, Stanislaw Świtek

Impact of essential oils on the development of pathogens of the *Fusarium* genus and germination parameters of selected crops

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**Abstract:** Fungal pathogens can significantly reduce the potential yield of agricultural crops, especially cereals. One of the most dangerous are pathogens of the *Fusarium* genus. They contribute to the infestation of plants, reduction of yields, and contamination of agricultural crops with mycotoxins, which are harmful to human beings and animal health. The absence of active substances, the problem of pathogen resistance to fungicides, and the pressure of society to limit the use of chemical plant protection products are the most important issues in agriculture. This has resulted in research aimed at finding natural methods to control plant pathogens gaining importance. One of them is the use of essential oils. In laboratory experiments, clove essential oil and pine essential oil were used. The influence of different concentrations of the above-mentioned substances on the development of the mycelium of *Fusarium* species (*F. equiseti*, *F. poae*, *F. culmorum*, and *F. avenaceum*) was analyzed and the germination of wheat and maize seeds infected with the pathogens of the genus *Fusarium* was assessed. Clove oil significantly inhibited the growth of mycelium of the *Fusarium* species and reduced germination parameters than pine oil.

**Keywords:** plant disease, seed soaking, wheat, maize

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**1 Introduction**

Wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.) are among the most important crops in the world [1]. The potential yield of these plants is affected by disease occurrence [2–4]. The harm due to fungal diseases affecting crops is not limited to decrease in yield. Mycotoxins (the secondary metabolites of fungi) may appear in food. Pathogens of the genus *Fusarium* are an important problem in wheat and maize cultivation [5,6]. The most known mycotoxins produced by the fungi belonging to the above-mentioned genus include deoxynivalenol (DON – produced mainly by *F. culmorum* and *F. graminearum*), nivalenol (NIV – produced mainly by *F. culmorum*, *F. cerealis*, and *F. graminearum*), zearalenone (ZEA mainly produced by *F. graminearum*, *F. cerealis*, and *F. culmorum*), fumonisin B1 (mainly produced by *F. proliferatum* and *F. verticillioides*), and T-2 toxin (mainly produced by *F. langsethiae*, *F. sporothricoides*, and *F. poae*) [7]. These mycotoxins have negative effects on human and animal health [8–10].

*Fusarium* fungi are detected, *inter alia*, in soil and on crop residues [11,12]. They are also found on the surface and inside the grain of cereals [13]. One of the ways to control pathogens of the genus *Fusarium* during the cultivation of wheat and maize is seed treatments [14,15]. Another method is soaking the grain in different solutions [16]. *Fusarium* species which can be often found in wheat and maize plantations include, among others, *F. culmorum*, *F. equiseti*, *F. avenaceum*, and *F. poae* [17–19]. One of the diseases that can be caused by fungi of the genus *Fusarium* is seedling blight [20,21]. These pathogens also contribute to the occurrence of *Fusarium* stem rot, leaf spot caused by *Fusarium*, *Fusarium* head blight, and *Fusarium* stalk rot [22–24].

Farmers will need to increase crop production, either by increasing the amount of agricultural land to grow crops or by enhancing productivity on existing agricultural lands by adopting new methods of plant protection [25].
It has to be mentioned here that the presence of pests reduces yield significantly [26]. At the same time, more and more attention is now paid on the responsible use of chemical products that end up in the environment and on food safety [27]. The selection of weeds, pathogens, and pests resistant to the used active substances of plant protection products becomes another problem [28–30]. Natural alternatives to synthetic agents are needed [31–33]. However, biopesticides constitute only a small part of the plant protection market [34].

Essential oils are examples of plant protection products of natural origin [35]. In some research papers, authors have described essential oils as potential herbicides [36]. Other studies indicate the possibility of their use for repelling pests [37]. Work is also underway to use them as insecticides [38]. The results of the experiments also indicate the potential possibility of using them as fungicides [39, 40]. Some research found essential oils useful in the control of Fusarium pathogens [41].

Essential oils are volatile substances obtained by various methods from many parts of plants, including flowers, fruits, stems, bark, leaves, and roots [42]. They contain many bioactive compounds that have antimicrobial and antioxidant properties [43–46], and show bacteriostatic activity [47]. Essential oils have, in their composition, large amounts of terpenoids, occurring in the form of sesquiterpenes and for the most part in the form of monoterpenes [48]. Monoterpenes belong to the group of isoprenoids [49]. In plants, essential oils have various functions like, among others, protect against pests and parasites, function as signaling devices for insects, and inhibit the growth of other plant species [50]. They are insoluble in water, but they dissolve in ether, alcohol, and fixed oils [51]. Essential oils are commonly used in aromatherapy [52]. They find their use as preservatives and as active substances in cosmetics [53].

The aim of the study was to determine the effect of selected essential oils and their efficacy in Fusarium genus mycelium growth control and their influence on the germination of wheat and maize seeds infected with Fusarium pathogens.

## 2 Methods

### 2.1 Effects of essential oils on in vitro fungal growth

The research material consisted of two essential oils: clove (Eugenia caryophyllus (Spreng.) Bullock & SG Harriso) and pine (Pinus sylvestris L.) (from a commercial source – Etja, Elbląg, Poland) mixed with ethoxylated rapeseed oil (Rokacet R217, PCC group, Brzeg Dolny, Poland) in a 4:1 ratio. For the experiment F. equiseti, F. culmorum, F. poae, and F. avenaceum cultures isolated from wheat kernels of the highest pathogenicity, selected in greenhouse tests, were used. (Cultures are part of the collection of Department Mycology.) The essential oils were tested in four doses: $5 \times 10^3$, $10 \times 10^3$, $15 \times 10^3$, and $20 \times 10^3$ ppm. The tested essential oils were added to a sterile PDA medium cooled to $45^\circ C$ in such amounts to obtain the appropriate concentrations. Around 18 ml of the medium was poured into each petri dish with a diameter of 90 mm. Discs of individual fungus cultures with a diameter of 4 mm were placed on the solidified medium in Petri dishes in their central part. The control combination was pure PDA medium (no essential oil added). The plates were incubated at 20°C under controlled conditions in a binder chamber. Assessment was made by measuring the linear growth of the mycelium. The experiment was performed in two series, each time in three replications and incubated at 20°C for 10 days. The results are presented as the mean value. Statistical analysis was performed and photographic documentation was made (Figures 1–4). The data obtained in the experiment were subjected to analysis of variance (ANOVA), and then to Tukey’s protected LSD test with a probability level of 0.05.

### 2.2 Rolled towel test

The research was carried out in the laboratory of the Department of Agronomy at the Poznań University of
Life Sciences. The rolled towel test uses the same oils mixed with ethoxylated rapeseed oil as in the experiment carried out on Petri dishes.

In the experiment, the influence of the two essential oils on the energy and germination capacity of grains, the length of shoots and roots, and the health of maize (*Zea mays* L.) seedlings (PR39H3 variety) and winter wheat (*Triticum aestivum* L.) seedlings (Arkadia variety) was assessed. In the rolled towel test, which was set up with the use of filter paper, seeds inoculated with fungi of the genus *Fusarium* were used. Seed inoculation was carried out according to the Jaber and Enkerli methodology [54]. Fungal conidia were harvested under sterile conditions by gently scraping the surface of 21-day-old sporulating cultures of *Fusarium: F. culmorum, F. equiseti, F. avenaceum,* and *F. poae.* The resulting mycelia and spores were then filtered through several layers of sterile cheese cloth into a sterile glass bottle containing 250 ml sterile distilled water plus 0.1% of surfactant. The conidial suspension of each fungal strain was then homogenized with a magnetic stirrer for 5 min (spore concentration – 4 million/ml).

For plant inoculation, surface-sterilized seeds were soaked in the conidial suspension of *Fusarium* spp. fungal strain [55]. Bottles containing the soaking seeds were kept for 24 h in the dark at 25°C. After this time, the rolled towel test was started. The control sample consisted of seeds that had not been soaked in any more solution. In the next combination, the seeds were soaked for 8 min in distilled water, the remaining ones in essential oil solutions of various doses (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm). The test was performed in triplicate for each species, and 25 seeds were used in each replication. The rolls were placed in a thermostatic cabinet, ensuring constant humidity (70%) at a temperature of 21°C. After 4 days from the beginning of the experiment, the germination energy of seeds was determined. After 7 days from the beginning of the research, the germination capacity of seeds, the length of the shoots and seedling roots were assessed. On the basis of the collected results, the vigor index was determined: vigor index = [seedling length (cm) × germination (%)]. Additionally, the infection on the surface of grains and seedlings by fungal pathogens was visually assessed. The data obtained in the experiment were subjected to ANOVA, and then to Tukey’s protected LSD test with a probability level of 0.05.

**Ethical approval:** The conducted research is not related to either human or animal use.

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**Figure 2:** *Fusarium culmorum* cultures on medium supplemented with essential oils: control: (1) clove essential oil, (2–5) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm); pine essential oil (6–9) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm).

**Figure 3:** *Fusarium poae* cultures on medium with the addition of essential oils: control sample: (1) clove essential oil, (2–5) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm); pine essential oil (6–9) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm).

**Figure 4:** *Fusarium avenaceum* cultures on the medium with the addition of essential oils: control: (1) clove essential oil, (2–5) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm); pine essential oil (6–9) (5 × 10³, 10 × 10³, 15 × 10³, and 20 × 10³ ppm).
3 Results

3.1 Effects of essential oils on in vitro fungal growth

The use of clove essential oil in all the tested concentrations inhibited the development of the tested pathogens in 100% (Table 1). When pine essential oil was added, its efficacy depended on the dose of the substance and the species of fungus. The increase in the concentration of the essential oil led to an increase in its efficacy (Figures 1–4). The lowest efficacy of this compound was observed for F. avenaceum mycelium growth control. In the case of F. equiseti, F. culmorum, and F. poae, none of the doses of pine essential oil resulted in inhibition of mycelium growth at the level of statistical significance equal to the combinations in which the clove essential oil was used.

3.2 Rolled towel test

The highest statistically significant level of germination energy of maize seeds was observed for the control object, combinations in which the seeds were soaked in distilled water and in the combinations where the lowest concentration of clove oil and 5 × 10^3, 10 × 10^3, and 15 × 10^3 ppm of pine oil. The highest values of germination of maize seeds were observed in the untreated objects (controls), combinations in which the seeds were soaked in distilled water and solutions of 10 × 10^3 and 15 × 10^3 ppm of pine oil were added. The above-mentioned observations were statistically confirmed. Soaking the seeds in solutions of 15 × 10^3 and 20 × 10^3 ppm of clove oil contributed to the complete inhibition of germination of corn seeds (Table 2).

The highest statistically significant level of maize seedling root length was recorded for the control (untreated objects) and combinations in which the seeds were soaked in distilled water and solutions of 15 × 10^3 and 20 × 10^3 ppm of pine oil. The longest maize seedlings shoot and the highest vigor index level was observed in the control (untreated) sample, combinations in which seeds were soaked in distilled water, and in the combinations where pine oil in concentrations 10 × 10^3, 15 × 10^3, and 20 × 10^3 ppm was tested. Soaking the seeds in clove essential oil contributed the most to lowering the vigor index value. The above-mentioned observations were statistically confirmed (Table 3).

The highest statistically significant level of germination energy of winter wheat seeds was observed in control objects (untreated), combinations in which seeds were soaked in distilled water and combinations where pine oil was tested in concentrations 15 × 10^3 and 20 × 10^3 ppm. The highest values of germination capacity of winter wheat seeds were observed in the untreated objects (controls), combinations in which the seeds were soaked in distilled water and solutions of 10 × 10^3, 15 × 10^3, and 20 × 10^3 ppm of pine oil were added. Soaking winter wheat seeds in solutions containing clove oil completely inhibited their germination (Table 4).

The highest statistically significant level of root length was found in winter wheat seedlings, in a combination where seeds were soaked in distilled water. The longest shoots of seedlings and the vigor index were recorded for combinations where the seeds were soaked in distilled water and solutions of 20 × 10^3 ppm of pine oil. Inhibition of seed germination in combinations in which clove

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**Table 1: Mycelium development after the application of essential oils**

| No. | Treatment              | Dose (ppm) | F. equiseti (cm) | F. culmorum (cm) | F. poae (cm) | F. avenaceum (cm) |
|-----|------------------------|------------|-----------------|-----------------|-------------|-------------------|
| 1   | Control                | —          | 4.60 ± 0.00⁹    | 4.60 ± 0.00⁹    | 4.60 ± 0.00⁹ | 4.60 ± 0.00⁹      |
| 2   | Clove essential oil    | 5 × 10³    | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹ | 0.00 ± 0.00⁹      |
| 3   |                        | 10 × 10³   | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹ | 0.00 ± 0.00⁹      |
| 4   |                        | 15 × 10³   | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹ | 0.00 ± 0.00⁹      |
| 5   |                        | 20 × 10³   | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹    | 0.00 ± 0.00⁹ | 0.00 ± 0.00⁹      |
| 6   | Pine essential oil     | 5 × 10³    | 4.60 ± 0.00⁹    | 3.44 ± 0.18⁹    | 4.25 ± 0.12⁹ | 4.29 ± 0.19⁹      |
| 7   |                        | 10 × 10³   | 2.31 ± 0.27⁹    | 1.44 ± 0.21⁹    | 1.43 ± 0.12⁹ | 3.13 ± 0.21⁹      |
| 8   |                        | 15 × 10³   | 0.66 ± 0.08⁹    | 0.60 ± 0.06⁹    | 0.38 ± 0.22⁹ | 2.38 ± 0.31⁹      |
| 9   |                        | 20 × 10³   | 0.49 ± 0.17⁹    | 0.48 ± 0.13⁹    | 0.30 ± 0.00⁹ | 1.45 ± 0.29⁹      |
| LSD (0.05) |                |            | 0.14            | 0.16            | 0.13        | 0.26              |

a–f: different letters indicate statistically different mean LSD (p = 0.05) = value from the last line.
essential oil was used resulted in the fact that the values of root length, shoot length, and vigor index were equal to zero (Table 5).

Soaking the seeds in both distilled water and essential oil solutions contributed to a statistically significant decrease in the infection of seedlings and the surface of

Table 2: Effect of essential oils on the germination energy and germination capacity of maize seeds

| No. | Treatment                        | Dose (ppm) | Germination energy (%) | Germination capacity (%) |
|-----|----------------------------------|------------|------------------------|--------------------------|
| 1   | Control                          | —          | 66.7 ± 12.2<sup>ab</sup> | 86.3 ± 5.7<sup>a</sup>  |
| 2   | Seeds soaked in distilled water   | —          | 65.3 ± 15.1<sup>ab</sup> | 83.7 ± 12.0<sup>ab</sup> |
| 3   | Clove essential oil               | 5 × 10<sup>3</sup> | 71.7 ± 3.5<sup>a</sup> | 75.3 ± 7.5<sup>bc</sup> |
| 4   |                                  | 10 × 10<sup>3</sup> | 5.3 ± 4.6<sup>c</sup> | 5.3 ± 4.6<sup>d</sup>  |
| 5   |                                  | 15 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>d</sup>  |
| 6   |                                  | 20 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>d</sup>  |
| 7   | Pine essential oil                | 5 × 10<sup>3</sup> | 68.0 ± 4.0<sup>ab</sup> | 68.0 ± 4.0<sup>c</sup> |
| 8   |                                  | 10 × 10<sup>3</sup> | 64.7 ± 5.0<sup>ab</sup> | 78.3 ± 5.7<sup>abc</sup> |
| 9   |                                  | 15 × 10<sup>3</sup> | 64.0 ± 4.0<sup>ab</sup> | 78.7 ± 12.2<sup>abc</sup> |
| 10  |                                  | 20 × 10<sup>3</sup> | 54.7 ± 15.1<sup>c</sup> | 73.3 ± 4.6<sup>bc</sup> |

LSD (0.05) | 14.62 | 10.92 |

<sup>a–d</sup>: different letters indicate statistically different mean LSD (p = 0.05) = value from the last line.

Table 3: Effect of essential oils on the length of roots and shoots of seedlings and the vigor index of maize seedlings

| No. | Treatment                        | Dose (ppm) | Root length (cm) | Shoot length (cm) | Vigor index |
|-----|----------------------------------|------------|------------------|-------------------|-------------|
| 1   | Control                          | —          | 1.86 ± 1.33<sup>a</sup> | 1.52 ± 0.96<sup>a</sup> | 130.67 ± 80.28<sup>a</sup> |
| 2   | Seeds soaked in distilled water   | —          | 1.60 ± 1.41<sup>ab</sup> | 1.43 ± 0.85<sup>a</sup> | 122.05 ± 76.64<sup>a</sup> |
| 3   | Clove essential oil               | 5 × 10<sup>3</sup> | 1.05 ± 0.81<sup>ab</sup> | 1.18 ± 0.84<sup>ab</sup> | 89.97 ± 65.94<sup>ab</sup> |
| 4   |                                  | 10 × 10<sup>3</sup> | 0.00 ± 0.00<sup>d</sup> | 0.02 ± 0.14<sup>c</sup> | 0.16 ± 1.14<sup>c</sup>  |
| 5   |                                  | 15 × 10<sup>3</sup> | 0.00 ± 0.00<sup>d</sup> | 0.00 ± 0.00<sup>d</sup> | 0.00 ± 0.00<sup>c</sup>  |
| 6   |                                  | 20 × 10<sup>3</sup> | 0.00 ± 0.00<sup>d</sup> | 0.00 ± 0.00<sup>d</sup> | 0.00 ± 0.00<sup>c</sup>  |
| 7   | Pine essential oil                | 5 × 10<sup>3</sup> | 0.74 ± 0.77<sup>c</sup> | 0.91 ± 0.81<sup>b</sup> | 61.35 ± 53.86<sup>b</sup> |
| 8   |                                  | 10 × 10<sup>3</sup> | 1.09 ± 0.88<sup>ab</sup> | 1.52 ± 1.02<sup>a</sup> | 120.44 ± 83.25<sup>a</sup> |
| 9   |                                  | 15 × 10<sup>3</sup> | 1.63 ± 1.27<sup>ab</sup> | 1.57 ± 1.05<sup>a</sup> | 126.22 ± 88.70<sup>a</sup> |
| 10  |                                  | 20 × 10<sup>3</sup> | 1.44 ± 1.41<sup>ab</sup> | 1.31 ± 1.11<sup>ab</sup> | 97.01 ± 83.24<sup>ab</sup> |

LSD (0.05) | 0.624 | 0.445 | 48.836 |

<sup>a–d</sup>: different letters indicate statistically different mean LSD (p = 0.05) = value from the last line.

Table 4: Effect of essential oils on the germination energy and germination capacity of winter wheat seeds

| No. | Treatment                        | Dose (ppm) | Germination energy (%) | Germination capacity (%) |
|-----|----------------------------------|------------|------------------------|--------------------------|
| 1   | Control                          | —          | 97.3 ± 2.3<sup>ab</sup> | 97.3 ± 2.3<sup>ab</sup> |
| 2   | Seeds soaked in distilled water   | —          | 97.3 ± 2.3<sup>ab</sup> | 97.3 ± 2.3<sup>ab</sup> |
| 3   | Clove essential oil               | 5 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>c</sup>  |
| 4   |                                  | 10 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>c</sup>  |
| 5   |                                  | 15 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>c</sup>  |
| 6   |                                  | 20 × 10<sup>3</sup> | 0.0 ± 0.0<sup>c</sup> | 0.0 ± 0.0<sup>c</sup>  |
| 7   | Pine essential oil                | 5 × 10<sup>3</sup> | 96.0 ± 4.0<sup>b</sup> | 96.0 ± 4.0<sup>b</sup> |
| 8   |                                  | 10 × 10<sup>3</sup> | 96.0 ± 4.0<sup>b</sup> | 97.3 ± 2.3<sup>ab</sup> |
| 9   |                                  | 15 × 10<sup>3</sup> | 97.3 ± 2.3<sup>ab</sup> | 97.3 ± 2.3<sup>ab</sup> |
| 10  |                                  | 20 × 10<sup>3</sup> | 100.0 ± 0.0<sup>a</sup> | 100.0 ± 0.0<sup>a</sup> |

LSD (0.05) | 3.47 | 3.07 |

<sup>a–c</sup>: different letters indicate statistically different mean LSD (p = 0.05) = value from the last line.
Table 5: Effect of essential oils on the length of roots and shoots of seedlings and the vigor index of winter wheat seedlings

| No. | Treatment               | Dose (ppm) | Root length (cm) | Shoot length (cm) | Vigor index |
|-----|-------------------------|------------|------------------|-------------------|-------------|
| 1   | Control                 | —          | 1.18 ± 0.53bc    | 1.85 ± 0.58bc    | 180.03 ± 56.76bc |
| 2   | Seeds soaked in distilled water | —         | 2.47 ± 1.44a     | 2.44 ± 1.06a     | 238.05 ± 104.83a |
| 3   | Clove essential oil     | 5 × 10³   | 0.00 ± 0.00d     | 0.00 ± 0.00d     | 0.00 ± 0.00d  |
| 4   |                         | 10 × 10³  | 0.00 ± 0.00d     | 0.00 ± 0.00d     | 0.00 ± 0.00d  |
| 5   |                         | 15 × 10³  | 0.00 ± 0.00d     | 0.00 ± 0.00d     | 0.00 ± 0.00d  |
| 6   |                         | 20 × 10³  | 0.00 ± 0.00d     | 0.00 ± 0.00d     | 0.00 ± 0.00d  |
| 7   | Pine essential oil      | 5 × 10³   | 0.81 ± 0.50c     | 1.54 ± 0.60c     | 148.42 ± 59.99c |
| 8   |                         | 10 × 10³  | 0.88 ± 0.39c     | 1.53 ± 0.52c     | 149.25 ± 50.66c |
| 9   |                         | 15 × 10³  | 1.37 ± 0.73bc    | 1.96 ± 0.82bc    | 190.24 ± 77.50bc |
| 10  |                         | 20 × 10³  | 1.69 ± 0.69b     | 2.11 ± 0.63ab    | 211.07 ± 63.07ab |
|     | LSD (0.05)              | —          | 0.742            | 0.474            | 48.046       |

a–d: different letters indicate statistically different mean LSD \((p = 0.05)\) = value from the last line.

| No. | Treatment               | Dose (ppm) | Maize | Winter wheat |
|-----|-------------------------|------------|-------|-------------|
| 1   | Control                 | —          | 30.7a | 26.7a       |
| 2   | Seeds soaked in distilled water | —         | 14.0b | 18.7b       |
| 3   | Clove essential oil     | 5 × 10³   | 13.0b | 0.0d        |
| 4   |                         | 10 × 10³  | 6.7bcd | 0.0d       |
| 5   |                         | 15 × 10³  | 0.0d  | 0.0d        |
| 6   |                         | 20 × 10³  | 0.0d  | 0.0d        |
| 7   | Pine essential oil      | 5 × 10³   | 9.3bc | 6.7c        |
| 8   |                         | 10 × 10³  | 8.0bc | 2.7d        |
| 9   |                         | 15 × 10³  | 4.0cd | 2.7d        |
| 10  |                         | 20 × 10³  | 8.0bc | 2.7d        |
|     | LSD (0.05)              | —          | 7.84  | 3.89        |

a–d: different letters indicate statistically different mean LSD \((p = 0.05)\) = value from the last line.

In organic farming where the use of chemical plant protection products is not allowed, a significant reduction in the occurrence of pathogens with the use of biological agents is already an important factor improving the health of crops. Combining biological control with other methods can allow obtaining sufficiently higher yields of good quality [56]. Research developed by other scientists also indicates that the use of essential oils can significantly reduce the development of pathogens of the Fusarium genus [57]. Clove essential oil significantly contributed to the inhibition of the development of the studied analyzed pathogens. In the case of chemical plant protection products, there are many studies about their effects on crops [58,59]. However, research is needed to assess the phytotoxicity of substances of natural origin in relation to agricultural crops.

Soaking seeds in distilled water contributed to a decrease in the infection of seedlings. In the case of pathogens present on the surface of the grains, rinsing the seed material may reduce the infestation of plants by fungal pathogens. Information available in the literature also indicates that soaking the seeds in water may have only a minor contribution in reducing seed infestation by pathogens of the genus Fusarium [60]. In the search for new, safe methods of plant protection that reduce the amount of chemicals released into the environment, the possibility of reducing the occurrence of fungal diseases by soaking seeds in solutions containing plant extracts was investigated [61]. The addition of essential oils to distilled water limited infestation in the roll experiment.

Soaking cereal seeds in essential oil solutions may inhibit germination [62]. In the conducted experiment, soaking of winter wheat and maize seeds in solutions of clove oil contributed to a significant, in most cases, complete inhibition of germination. Other results available in corn and winter wheat seeds. Clove essential oil contributed more to the reduction of infestation than pine essential oil (Table 6). In the case of winter wheat, the use of higher concentrations of pine essential oil reduced infection on a statistical level equal to the use of clove essential oil.

4 Discussion

The use of pine essential oil did not contribute to the complete inhibition of pathogen development. In plant protection, however, it is important to limit the occurrence of diseases below the threshold of economic harmfulness.

Table 6: Effect of essential oils on the infection by Fusarium of seeds and seedlings of maize and winter wheat

| No. | Treatment               | Dose (ppm) | Maize (cm) | Winter wheat (cm) |
|-----|-------------------------|------------|------------|-------------------|
| 1   | Control                 | —          | 30.7a      | 26.7a             |
| 2   | Seeds soaked in distilled water | —         | 14.0b      | 18.7b             |
| 3   | Clove essential oil     | 5 × 10³   | 13.0b      | 0.0d              |
| 4   |                         | 10 × 10³  | 6.7bcd     | 0.0d              |
| 5   |                         | 15 × 10³  | 0.0d       | 0.0d              |
| 6   |                         | 20 × 10³  | 0.0d       | 0.0d              |
| 7   | Pine essential oil      | 5 × 10³   | 9.3bc      | 6.7c              |
| 8   |                         | 10 × 10³  | 8.0bc      | 2.7d              |
| 9   |                         | 15 × 10³  | 4.0cd      | 2.7d              |
| 10  |                         | 20 × 10³  | 8.0bc      | 2.7d              |
|     | LSD (0.05)              | —          | 7.84       | 3.89              |

a–d: different letters indicate statistically different mean LSD \((p = 0.05)\) = value from the last line.
the literature also indicate that soaking wheat grains in a solution of clove oil can significantly reduce their germination [63]. The use of pine oil, depending on the concentration, had a lesser effect on germination or had no statistically significant effect on this parameter.

Seed dressing may contribute to limiting the length of the roots and shoots of crop seedlings [64]. Studies carried out by other scientists indicate that pine essential oil contributes less to the inhibition of root and seedling shoot growth than the selected other essential oils in the context of some plant species [65]. The infestation of cereal grains by pathogens of the Fusarium genus alone contributes to the inhibition of germination [66,67]. It can also lead to disturbances in the growth of seedlings and roots [68–70]. However, when essential oils are used, germination may be completely inhibited or seedling growth may be significantly inhibited.

It is worth paying attention to the properties of essential oils, such as biodegradability, easy availability, and low toxicity to organisms that are not the target of the treatment [71]. They are indicated as a possible substitute for chemicals for organic farming [72,73]. In addition, essential oils contain many ingredients that have different mechanisms of action. This is important in the context of counteracting resistance [74]. However, solutions that will reduce their volatility and increase bioavailability should be sought [75].

The introduction of biological plant protection products in the market, which could be used on a large scale, is one of the requirements for achieving the assumptions of the agricultural policy, which involves limiting the amount of chemical preparations released into the environment. However, attention is drawn to the need to introduce clearer regulations in the European Union regarding biological plant protection products [76,77].

5 Conclusion

The use of essential oils can significantly reduce the development of pathogens of the Fusarium genus. Individual essential oils show different fungicidal activity. Clove essential oil inhibited fungal growth to a greater extent than pine essential oil. The tested substances can significantly affect the germination parameters of the seeds of cultivated plants. In future, essential oils may be used as fungicides, but methods should be found to reduce their impact on crop development.

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