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# Primary screening for the effect of orange oil against pathogens and pests on tomato and cucumber

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## Abstract

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Considering the positive and especially negative sides of the use of chemical plant protection products, attention is increasingly being directed towards finding, and using products of natural origin that have qualities that affect harmful microflora and microfauna. The study conducted tested the effectiveness of different concentrations of orange oil against *Phytophthora infestans*, *Alternaria solani* and *Aphis gossypii*. A variety of reactions of the tested pathogens and aphid to the tested products were established. Against *Phytophthora infestans*, the best results on the 7th and 9th day of reporting (over 50% efficacy) were observed in the three concentrations of the products Oo1 and Oo2. The average effectiveness for the entire reporting period was highest for Oo1 – 1% – 63.76%. Against *Alternaria solani*, it was found that the initial effect of the tested products was very good, but a gradual decrease in the effect was observed in the following days. On average for the reporting period, the highest efficiency was registered with product Oo1 – 1% (47.91%). The results obtained in the experiments against *Aphis gossypii* show that the relatively best efficiency of the four tested variants is the product Oo1 in a concentration of 1% – 56.02% on the 5th day after the third treatment. It proves that with the increase in the concentration of the tested products, the sensitivity of the tested pathogens and enemies also increases. Best results against *Phytophthora infestans*, *Alternaria solani* and *Aphis gossypii* show product Oo1 in highest test concentration 1%.

*Keywords:* screening; orange oil; pathogens; aphid

## Introduction

Plant essential oils (PEOs) are gaining attention as eco-friendly alternatives to synthetic pesticides. They are biodegradable, target-specific, and safe for humans and other life forms. PEOs have been widely explored for their biopesticidal properties, including bioherbicides, bioinsecticides, and biofungicides (Gupta et al., 2023). They have shown efficacy against a wide range of pests and in numerous crop systems (Isman, 2020).

However, there are challenges in using PEOs as pesticides due to their high volatility and production costs (Gupta et al., 2023). Encapsulation techniques, such as emulsification and ionic gelation, are being explored to improve their stability and allow for controlled release (Ayllón-Gutiérrez et al., 2024).

Plant essential oils offer a promising source of naturally derived antimicrobials. Recently, these oils and extracts from various plants have gained substantial popularity and scientific interest. The increasing consumer demand for natural preservatives is driven by safety concerns regarding chemical additives (Tabassum and Vidyasagar, 2013). Plant essential oil-based pesticides are effective against various pests and some pathogenic fungi. They can be used as fumigants, granules, or sprays, leading to lethal toxicity, repellence, or deterring oviposition in insects (Isman and Machial, 2006).

Potato blight (*Phytophthora infestans*) is one of the primary diseases responsible for significant economic losses among potato and tomato growers due to decreased crop quality and yield. Globally, annual losses in potato production are estimated at $6 billion. Depending on the severity of the epidemic, this disease can devastate 40 to 70% of the potato harvest (Runno-Paurson et al., 2019). With the strong socio-political pressure to limit the use of certain pesticides in the EU by 2030, cultivating the 5th most produced crop in the EU may become increasingly challenging. Therefore, it is vital to provide farmers with a variety of effective and reasonable pesticide alternatives. Currently, botanical biopesticides (plant extracts) are viewed as one strategy to achieve the Green Deal goals. These biopesticides are generally considered low risk compared to synthetic ones (Acheuk et al., 2022). Moreover, in addition to directly affecting pathogens, they can also act as plant resistance inducers (Burketova et al., 2015).

Early blight of tomato, caused by *Alternaria solani*, can lead to significant losses in tomato production. The disease can cause yield losses ranging from 20% to 40% in seedling establishment and up 50% to 86% in fruit yield (Parvin et al., 2021). The severity of the disease depends on various factors, including environmental conditions, the susceptibility of the tomato variety, and the presence of the pathogen. Effective management strategies, such as crop rotation, resistant varieties, and the use of fungicides, can help mitigate these losses. Additionally, botanical exudates like neem oil have shown promising results in reducing disease severity and increasing yield (Soni and Arsia, 2021).

The cotton aphid, *Aphis gossypii* Glover (*Hemiptera*: *Aphididae*), is a pest with a broad range of host plants, including cotton, potato, and melons (Li et al., 2022). It causes harm through direct feeding on foliage and stems and the transmission of plant viruses (Zhao et al., 2013). Wingless aphids exhibit faster development compared to their winged counterparts and possess adaptations that enhance their fecundity, making them more challenging to control (Braendle et al., 2006). Insecticides remain the primary method for managing *A. gossypii* in many agricultural settings. However, the extensive and frequent use of insecticides has led to *A. gossypii* populations developing resistance to all major insecticide groups (Herron et al., 2001).

Orange oil has been found to exhibit antimicrobial properties against a range of pathogens, including bacteria and fungi. It disrupts the cell membranes of pathogens, inhibiting their growth and spread (Obidi et al., 2013). Some essential oils act as repellents and antifeedants, deterring pests from feeding on plants. This helps in reducing pest damage and protecting crops (Ayllón-Gutiérrez et al., 2024).

The use of essential oils in plant protection aligns with the principles of sustainable agriculture. Being biodegradable and non-toxic to humans, pets, and beneficial insects, essential oils offer an eco-friendly solution that minimizes the environmental impact. They also reduce the risk of pesticide resistance development among pathogens, which is a growing concern with conventional pesticides (Ghalem, 2016; Mohd et al., 2022).

Advancements in encapsulation techniques have enhanced the stability and efficacy of essential oils in pest and disease management. Encapsulation methods such as emulsification, ionic gelation, and complex coacervation protect essential oils from oxidation and volatilization, allowing for controlled release and prolonged activity. This innovative approach ensures that the oil remains effective over an extended period, reducing the need for frequent applications (Mohd et al., 2022).

The "Green Revolution" resulted in a significant boost in crop productivity. However, it also brought significant ecological impacts due to the extensive use of synthetic agrochemicals, prompting concerns about agricultural sustainability. The widespread application of synthetic pesticides has caused environmental degradation, pest resistance, and potential risks to various non-target species, including plants, animals, and humans (Gupta et al., 2023).

The aim of this scientific work is to explore тhe effectiveness of orange oil against pathogens and aphids on tomatoes and cucumbers.

## Material and Methods

The effect of orange oil (Oo) 4 products (Oo1, Oo2, Oo3, Oo4) in different concentrations (0.6%; 0.8%, 1%), was tested to *Phytophthora infestans*, *Alternaria solani* and *Aphis gossypii*. Orange oil formulation:

- Oo1 – 80% orange oil;
- Oo2 – 50% orange oil + 30% paraffin oil;
- Oo3 – 30% orange oil + 50% paraffin oil;
- Oo4 – 10% orange oil + 70% paraffin oil.

Control is the use of pathogens (no add products in Petri dishes) and insects, and etalon standard Limocid (0.8%) for cotton aphid. The experiments were conducted at laboratory and unheated steel-glass greenhouse at the Maritsa Vegetable Crops Research Institute – Plovdiv (MVCRI).

The isolates utilized in this study, originate from the phytopathological collection, housed within the Phytopathology laboratory at MVCRI – Plovdiv. Thornberry's method was applied to test the four products against the pathogens (Vasileva, 2023).

The diameter of the colony was recorded on the 3rd, 5th, 7th, and 9th days. The results were analyzed using the modified Abbott' (1925) formula.

Treatments against cotton aphids (*Aphis gossypii*) were carried out three times at 7-day intervals. Phenophase of the crop at the start of treatment: 5 – 6 true leaves of cucumber variety – Tony $F_1$. The effectiveness (%) was reported according to the formula of Henderson and Tilton (1955) against cotton aphid (*Aphis gossypii*).

Throughout the investigation, we employed software tools, including the 'MS Excel Analysis ToolPak Add-Ins' (https://support.office.com) and 'R-3.1.3,' in conjunction with 'RStudio-0.98' and the installed package 'agricolae 1.2-2.

## Results and Discussion

As a result of the research carried out with the tested products against pathogens and insects, significant differences in terms of effectiveness were found (Table 1, 2 and 3).

Crop protection primarily depends on synthetic pesticides for pest control. However, their ongoing use is facing increasing scrutiny due to negative impacts, including long-term environmental persistence that leads to water and soil contamination, and residue retention in food, which poses significant risks to human and animal health. Within integrated pest management frameworks, biopesticides offer a viable alternative for crop protection, providing eco-friendly and safe solutions for humans and non-target organisms. Essential oils, consisting of complex mixtures of low-molecular-weight, highly volatile compounds, have emerged as promising candidates for plant-derived bioinsecticides that meet sustainable biological standards (Devrnja et al., 2022). Many essential oils and their bioactive compounds have attracted interest as botanical pesticides because of their high effectiveness, varied modes of action, and favorable safety profiles for mammals and non-target organisms (Chaudhari et al., 2021).

The tested products against the pathogen *Phytophthora infestans* showed a different effect on the 3rd day of reporting. The concentration of 0.6% on the 3rd day shows the strongest limitation of mycelial growth in product Oo1 – 46.67% and in Oo2 – 40.00%. The effectiveness of the products at a dose of 0.6% is most clearly visible on the 7th day of the reading 67.11% – Oo1. Tendency to unsatisfactory results is reported at the low concentration of 0.6% for all tested products. The activity of the products against the pathogen was significantly better on subsequent days than the reading with increasing concentration. The best results on the 7th and 9th day of the reporting (over 50% efficacy) were observed, in the three concentrations of the products Oo1 and Oo2. The average efficacy for the period was the highest for Oo1 – 1% – 63.76%. The highest values of variation (166.35%) were recorded for Oo2 – 1%. The standard deviation was in the range from 6.34 to 11.17 (Table 1).

The results for *Alternaria solani* show a better initial effect, expressed in a significant limitation in the growth of the mycelium. The effectiveness on the 3rd day is in the range of 57.58% – 33.33%. The best results on this day are registered with productOo1 (1%, 0.8%, 0.6%). In the following days, a gradual decrease in the effect of the tested products is observed. On average for the entire reporting period, the highest effectiveness is registered with the product Oo1 – 1% (47.91%), and the lowest is with Oo4 – 0.6% (12.32%). The variation is most pronounced with Oo3 – 1% (Table 2).

The results obtained in the experiments show that the product Oo1 in a concentration of 1% – 56.02% on the 5th

**Table 1. Effectiveness of products Oo 1, 2, 3, 4 against *Phytophthora infestans***

| *Phytophthora infestans* | | | | | For all days | | |
|---|---|---|---|---|---|---|---|
| Product/Concentration | 3 -day | 5-day | 7-day | 9-day | Average | VAR | SD |
| Oo1- 0.6% | 46.67 | 54.35 | 67.11 | 58.27 | 56.60 | 72.31 | 7.36 |
| Oo1- 0.8% | 46.67 | 60.87 | 71.05 | 62.12 | 60.18 | 101.64 | 8.73 |
| Oo1- 1% | 53.33 | 63.04 | 73.68 | 65.00 | 63.76 | 69.75 | 7.23 |
| Oo2 -0.6% | 40.00 | 43.48 | 55.26 | 39.62 | 44.59 | 53.62 | 6.34 |
| Oo2- 0.8% | 40.00 | 47.83 | 60.53 | 35.38 | 45.94 | 121.08 | 9.53 |
| Oo2 -1% | 43.33 | 54.35 | 65.79 | 36.35 | 49.96 | 166.35 | 11.17 |
| Oo3- 0.6% | 20.00 | 28.26 | 47.37 | 24.81 | 30.11 | 143.88 | 10.39 |
| Oo3- 0.8% | 26.67 | 32.61 | 52.63 | 28.65 | 35.14 | 142.05 | 10.32 |
| Oo3 -1% | 33.33 | 39.13 | 55.26 | 29.62 | 39.34 | 128.03 | 9.80 |
| Oo4- 0.6% | 13.33 | 21.74 | 34.21 | 34.62 | 25.98 | 106.79 | 8.95 |
| Oo4- 0.8% | 13.33 | 26.09 | 36.84 | 38.46 | 28.68 | 134.85 | 10.06 |
| Oo4 -1% | 20.00 | 30.43 | 44.74 | 36.15 | 32.83 | 107.75 | 8.99 |

*Oo – orange oil; VAR – coefficient of variation; SD – Standard deviations

*Source*: Authors' own elaboration

**Table 2. Effectiveness of products Oo 1, 2, 3, 4 against *Alternaria solani***

| *Alternaria solani* | | | | | For all days | | |
|---|---|---|---|---|---|---|---|
| Product/Concentration | 3-day | 5-day | 7-day | 9-day | Average | VAR | SD |
| Oo1- 0.6% | 54.55 | 36.84 | 37.50 | 34.92 | 40.95 | 83.37 | 7.91 |
| Oo1- 0.8% | 57.58 | 44.74 | 41.73 | 37.69 | 45.44 | 73.90 | 7.44 |
| Oo1- 1% | 57.58 | 48.68 | 46.15 | 39.23 | 47.91 | 57.51 | 6.57 |
| Oo2 -0.6% | 40.91 | 34.21 | 32.69 | 30.77 | 34.65 | 19.43 | 3.82 |
| Oo2- 0.8% | 41.82 | 39.47 | 35.77 | 33.23 | 37.57 | 14.58 | 3.31 |
| Oo2 -1% | 45.45 | 43.42 | 35.58 | 33.08 | 39.38 | 35.77 | 5.18 |
| Oo3- 0.6% | 36.36 | 25.53 | 14.81 | 7.23 | 20.98 | 161.46 | 11.00 |
| Oo3- 0.8% | 42.42 | 35.79 | 20.38 | 8.62 | 26.80 | 232.18 | 13.20 |
| Oo3 -1% | 43.94 | 38.68 | 22.12 | 10.00 | 28.69 | 241.61 | 13.46 |
| Oo4- 0.6% | 33.33 | 5.26 | 5.77 | 4.92 | 12.32 | 196.31 | 12.13 |
| Oo4- 0.8% | 34.85 | 7.11 | 7.69 | 6.92 | 14.14 | 190.69 | 11.96 |
| Oo4 -1% | 36.36 | 13.16 | 11.54 | 9.23 | 17.57 | 159.48 | 10.94 |

*Oo – orange oil; VAR – coefficient of variation; SD – Standard deviations
*Source*: Authors' own elaboration

day after the third treatment has the relatively best efficacy of the four tested variants, with a maximum efficacy value reported for the standard Limocid of 0.8% – 67.25% on the 3rd day after the third treatment. In the remaining variants, the values of the maximum efficacy reported vary from 42.77% to 47.13% against cotton aphid. On average for the entire period, the highest efficacy is in the third treatment with Oo1 – 1% – 50.50%. It is established that the weakest effect of the products is in the first treatment. The variation is most pronounced in Oo3 – 1% (20.51%) compared to the standard Limocid (62.37%) (Table 3).

Research on innovative, sustainable, eco-friendly pesticides is a major global focus, aimed at reducing synthetic inputs in agriculture, protecting biodiversity, and ensuring food safety for consumers. Botanical substances, particularly essential oils, are among the most promising natural pesticides. In this context, essential oils (EOs) could play a crucial role in developing eco-friendly pesticides. One study described and evaluated a citrus peel-based nano-formulation of EO, and its biological activity against A. gossypii in both laboratory and field settings. The phytotoxicity towards plants was also assessed. The developed nano-insecticide demonstrated effective aphicidal activity in both laboratory and field trials, even at moderate EO concentrations (Laudani et al., 2022).

There is an urgent need to develop sustainable and environmentally friendly pesticide formulations due to the numerous adverse effects of synthetic pesticides on human health and the environment. Essential oils (EOs), which are volatile oils produced as secondary metabolites in medicinal plants, show activity against pests, insects, and pathogenic fungi. The chemical composition of EOs is influenced by factors such as plant species or cultivar, geographical origin, environmental conditions, agricultural practices, and extraction methods. Increasing research highlights the herbicidal, insecticidal, acaricidal, nematicidal, and antimicrobial properties of EOs, confirming their efficacy and suitability as

**Table 3. Effectiveness of products Oo 1, 2, 3, 4 against cotton aphid (*Aphis gossypii*) in cucumbers variety Toni $F_1$ grown in greenhouses**

| Variant | Concentration, % | Effectiveness, %/ Days after treatment | | | | Common | | |
|---|---|---|---|---|---|---|---|---|
| | | 1 | 3 | 5 | 7 | Average | VAR | SD |
| 1- treatment | | | | | | | | |
| Oo 1 | 0.6 | 33.87 | 38.91 | 37.85 | 34.83 | 36.37 | 4.32 | 2.40 |
| | 0.8 | 34.39 | 39.39 | 41.92 | 35.17 | 37.72 | 9.49 | 3.56 |
| | 1 | 35.95 | 44.15 | 43.97 | 37.06 | 40.28 | 14.43 | 4.39 |
| 2- treatment | | | | | | | | |
| Oo1 | 0.6 | 36.02 | 42.16 | 38.50 | 35.79 | 38.12 | 6.58 | 2.96 |
| | 0.8 | 36.82 | 43.00 | 43.88 | 36.09 | 39.95 | 12.36 | 4.06 |
| | 1 | 38.25 | 47.70 | 44.94 | 39.87 | 42.69 | 14.46 | 4.39 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 3- treatment | | | | | | | | |
| Oo1 | 0.6 | 38.65 | 43.20 | 42.40 | 36.61 | 40.22 | 7.28 | 3.12 |
| | 0.8 | 43.78 | 44.97 | 45.74 | 39.19 | 43.42 | 6.45 | 2.93 |
| | 1 | 46.64 | 53.22 | 56.02 | 46.10 | 50.50 | 18.03 | 4.90 |
| 1- treatment | | | | | | | | |
| Oo2 | 0.6 | 34.02 | 34.60 | 38.60 | 33.89 | 35.28 | 3.75 | 2.24 |
| | 0.8 | 34.13 | 38.50 | 41.54 | 34.76 | 37.23 | 8.97 | 3.46 |
| | 1 | 34.21 | 39.22 | 42.58 | 34.00 | 37.50 | 12.96 | 4.16 |
| 2- treatment | | | | | | | | |
| Oo2 | 0.6 | 34.12 | 40.44 | 41.37 | 35.01 | 37.74 | 10.26 | 3.70 |
| | 0.8 | 36.30 | 42.78 | 42.04 | 36.03 | 39.29 | 9.83 | 3.62 |
| | 1 | 37.02 | 44.37 | 43.31 | 38.48 | 40.80 | 9.68 | 3.59 |
| 3- treatment | | | | | | | | |
| Oo2 | 0.6 | 37.03 | 42.77 | 41.40 | 35.00 | 39.05 | 9.96 | 3.64 |
| | 0.8 | 39.16 | 41.62 | 47.04 | 38.82 | 41.66 | 10.82 | 3.80 |
| | 1 | 38.68 | 47.13 | 43.64 | 40.10 | 42.39 | 10.76 | 3.79 |
| 1- treatment | | | | | | | | |
| Oo3 | 0.6 | 33.36 | 33.77 | 37.56 | 32.62 | 34.33 | 3.65 | 2.21 |
| | 0.8 | 33.93 | 38.31 | 41.39 | 34.11 | 36.94 | 9.69 | 3.59 |
| | 1 | 34.41 | 38.67 | 42.15 | 34.15 | 37.35 | 10.92 | 3.82 |
| 2- treatment | | | | | | | | |
| Oo3 | 0.6 | 33.41 | 38.81 | 37.87 | 34.34 | 36.11 | 5.20 | 2.63 |
| | 0.8 | 34.07 | 40.52 | 38.23 | 35.61 | 37.11 | 6.09 | 2.85 |
| | 1 | 35.39 | 43.41 | 43.37 | 38.03 | 40.05 | 12.03 | 4.00 |
| 3- treatment | | | | | | | | |
| Oo3 | 0.6 | 37.02 | 38.36 | 37.93 | 35.02 | 37.08 | 1.65 | 1.48 |
| | 0.8 | 38.28 | 40.73 | 46.05 | 38.10 | 40.79 | 10.30 | 3.71 |
| | 1 | 36.62 | 46.06 | 46.81 | 38.33 | 41.96 | 20.51 | 5.23 |
| 1- treatment | | | | | | | | |
| Oo4 | 0.6 | 33.23 | 33.51 | 36.55 | 31.44 | 33.68 | 3.37 | 2.12 |
| | 0.8 | 33.94 | 33.49 | 36.84 | 34.05 | 34.58 | 1.75 | 1.53 |
| | 1 | 33.61 | 38.67 | 42.15 | 34.15 | 37.15 | 12.21 | 4.03 |
| 2- treatment | | | | | | | | |
| Oo4 | 0.6 | 33.71 | 36.24 | 37.23 | 33.66 | 35.21 | 2.45 | 1.81 |
| | 0.8 | 34.85 | 43.26 | 43.30 | 38.54 | 39.99 | 12.54 | 4.09 |
| | 1 | 34.44 | 42.48 | 43.37 | 37.16 | 39.36 | 13.72 | 4.28 |
| 3- treatment | | | | | | | | |
| Oo4 | 0.6 | 38.36 | 37.04 | 36.63 | 33.77 | 36.45 | 2.80 | 1.93 |
| | 0.8 | 36.93 | 45.83 | 44.10 | 38.79 | 41.41 | 13.43 | 4.23 |
| | 1 | 36.62 | 46.06 | 46.81 | 38.33 | 41.96 | 20.51 | 5.23 |
| 1- treatment | | | | | | | | |
| Limocid | 0.8 | 47.90 | 56.48 | 64.17 | 59.86 | 57.10 | 35.66 | 6.90 |
| 2- treatment | | | | | | | | |
| Limocid | 0.8 | 48.54 | 66.05 | 65.24 | 51.45 | 57.82 | 62.37 | 9.12 |
| 3- treatment | | | | | | | | |
| Limocid | 0.8 | 51.09 | 67.25 | 52.80 | 51.99 | 55.78 | 44.20 | 7.68 |

*Oo – orange oil; VAR – coefficient of variation; SD – Standard deviations

*Source*: Authors' own elaboration

sustainable and eco-friendly biopesticides (Assadpour et al., 2024).

Commercial synthetic pesticides have been demonstrated to be harmful to humans, animals, non-target plants, nearby organisms, and the environment. Additionally, there is concern about pests developing resistance to these chemicals. Several factors limit the widespread use of biopesticides, including cost, production difficulties, mild effectiveness, and insufficient formulations. Regulatory issues in numerous countries also impede the adoption of biopesticides. Despite these challenges, biopesticides show promise due to their eco-friendly properties, unique chemical compositions, and diverse modes of action (Khursheed et al., 2022).

Essential oils can be applied in various forms, including sprays, soil drenches, and seed treatments. Their versatility allows for integration into different stages of crop production, from pre-planting to post-harvest. By incorporating essential oils into integrated pest management (IPM) programs, farmers can achieve effective disease control, while promoting a healthier and more resilient agricultural system (Jamshidi et al., 2023; Ghalem, 2016).

## Conclusion

As a result of the primary studies conducted to establish the effect of orange oil against *Phytophthora infestans*, *Alternaria solani* and *Aphis gossypii*, a very good initial effect was found. It is reported that the average effectiveness for observation period against *Phytophthora infestans* and *Alternaria solani* is highest at Oo1 (concentration1%), 63.76% and 47.91%, respectively. The data obtained show a better effect of the tested products against *Phytophthora infestans* compared to *Alternaria solani*. Against the *Aphis gossypii*, the highest effectiveness is at the third treatment with Oo1 (concentration 1%) – 50.50%.

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