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Proceeding Paper

Innovative Functional Plastic Films as Cover for Stonewool Grow Blocks and Their Effect on Tomato Hydroponic Cultivation †

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Abstract: Alkaloid compounds derived from tomato cultivation wastes (leaves, stems, etc.) are proposed as natural pesticides in this study. The tomato waste derived alkaloids (tomatidine and tomatine) were encapsulated in LDPE films through an extrusion process, in order to develop polymeric films with a pesticide action as covers for the stonewool grow blocks used in hydroponic tomato cultivation. A reduced attraction of mealybugs and the presence of their larvae in the leaves in relation to the control culture, as well as improved productivity, was observed, with the total marketable production of the tested plants being higher.

Keywords: alkaloids; antimicrobial properties; natural pesticides; polymeric films; application in the field

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1. Introduction

Pesticides protect crops from pest infestations and yield reduction. However, pesticides pose potential risks to food safety, the environment, and all living things since they can emigrate from treated fields to air, other land, and water bodies. On 12 May 2021, the European Commission adopted the EU Action Plan: "Towards a Zero Pollution for Air, Water and Soil", a key deliverable of the European Green Deal, with the aim of minimizing the use and risk of chemical pesticides by 50% by 2030 [1] https://ec.europa.eu/environment/pdf/zero-pollution-action-plan/results-public-consultation_en.pdf (accessed on 20 November 2021). Extensive use of chemical pesticides can lead to chemical pollution of the soil and the water used for irrigation [2–4], and the farmers are exposed to high chemical hazards [5]. Moreover, the accumulation of pesticides' residues in food products can cause serious health problems to consumers, especially in the case of foods that are mainly consumed fresh, such as tomatoes [2]. In this context, the presented study proposed the use of alkaloid compounds (tomatidine and tomatine) derived from tomato cultivation waste (leaves, stems, etc.) as natural pesticides. Plants, naturally, have their own protective mechanism against pathogen microbes [6,7]. In Solanaceae family plants, alkaloids protect them from insect attacks while offering fungal and nematode resistance. Based on previous studies, tomatine has reported fungicidal properties and is mainly located in the stems and leaves of the tomato plant [7]. The alkaloids in Solanaceae family plants protect the plant from insect attacks and offer fungal and nematode resistance [8].

2. Materials and Methods

2.1. Materials

Residues of tomato cultivation (leaves, stems, and green and red tomato fruit) were provided by AGRITEX Energy S.A., and low-density polyethylene pellets were provided by ACHAIKA PASTICS S.A. Ammonium hydroxide (NH_4OH) and hydrochloric acid (HCl) were purchased from Sigma-Aldrich. Ethanol and acetic acid were of analytical grade; methanol was of HPLC grade and was purchased from Thermo Fisher Scientific, (Cleveland, OH, USA).

2.2. Methods

2.2.1. Extraction of Alkaloids

Ultrasound-assisted extraction (UAE) (XO-SM50 Ultrasonic Microwave Reaction System, China) was used for the receipt of the tomato residues' alkaloid content. An ethanol–acetic acid (95:5 v/v) mixture was used as the extraction solvent, and the solvent/tomato residues ratio by weight was 20:1. The UAE operating conditions were: 25 kHz frequency at 450 Watt and temperature 25 °C. After the extraction a centrifugation step took place at 3000 rpm for 10 min, and the supernatant was collected and stored in −80 °C (Ultra-Low Temperature Freezer (Model MDF-U3386S; Panasonic Corporation, Osaka Prefecture, Japan). The deep-frozen extracts were freeze-dried in Leybold-Heraeus GT 2A (Koln, Germany) for 8 h in high vacuum (3 mbar).

2.2.2. Alkaloids Detection

The choice of the best extracts for use as natural pesticides was made after the detection of tomatine and tomatidine with High Pressure Liquid Chromatography (HPLC) with a Photodiode Array Detector (PDA) (Shimadzu LC-2030C, Kyoto, Japan). For this study, freeze-dried extracts of the leaves, stem, and green and red tomatoes were dissolved in 0.2 N HCl, and the alkaloids were precipitated with 5% NH_4OH until a pH = 10 was reached. Subsequently, the mixture was centrifuged at 3000 rpm for 20 min at 4 °C, and the supernatant was rejected. The washing process was repeated twice. Any residual supernatant was removed by evaporation at 45 °C using a rotary evaporator (Buchi R200 rotavapor), and the collected precipitate was redissolved in methanol. Prior to HPLC, the samples were filtered through a 0.45 µm PTFE syringe filter. The column used was Phenomenex Luna® 5 µm C18 (2) 100 Å, LC Column 250 × 4.6 mm; the mobile phase was methanol (isocratic flow) with a flow rate of 1 mL/min, and the injection volume was 10 µL. The absorbance was detected at 200 nm. All samples were studied in triplicate. The contents of the alkaloids were calculated using calibration curves of standard tomatine and tomatidine samples. The calibration curve was quite linear over the concentration range from 0.02 to 1 $mg \cdot mL^{-1}$ (r = 0.9987), and the limit of detection was 0.01 $mg \cdot mL^{-1}$.

2.2.3. Polymeric Film Development

Based on the HPLC results, the extracts with a higher alkaloid content were used as bioactive agents for encapsulation in LDPE. For this purpose, two extrusion processes took place. Firstly, a twin-screw extruder (Prism EuroLab 16 mm) was used for incorporation of the selected bioactive extracts into the LDPE. The screw speed was 50 rpm, and the extrusion temperature was 180–190 °C. The concentration of the bioactive agent in the extrusion product was 20% w/w. This product was pelletized with a Brabender Pelletizer and was incorporated in a blown film extruder. More specifically, pellets with the encapsulated alkaloids were mixed with pure LDPE pellets for the production of the final polymeric film. The screw speed was 50 rpm, and the extrusion temperature was 170–175 °C. The content of the encapsulated alkaloids in the final polymeric film was 1% w/w.

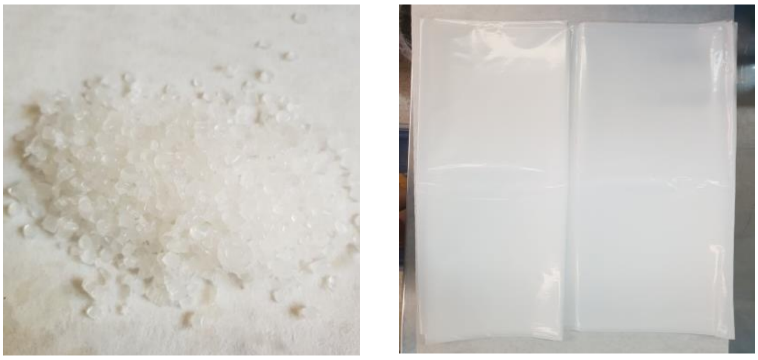
2.2.4. Application in the Field

The experiments took place in tomato hydroponic cultivation greenhouses of AGRITEX ENERGY S.A. in Imathia, Greece. The cultivation period of reference started in July

2020 and ended in January 2021. The total area of application was 294.4 m^2 and was divided equally into two parts: the first was the control tested area where the tomato cultivation took place in commercial Grodan stonewool, and the second area was the experimental application of the innovative polymeric film with encapsulated alkaloids as a cover for the stonewool. The stonewool dimensions in both cases were 120 × 20 × 7.5 cm. The studied cultivation was Brioso tomato cultivar. Evaluation of the cultivation performance was based on the following parameters: a) total plant yield (kg/m^2), b. marketable yield of plants (kg/m^2), c. non-marketable yield of plants (kg/m^2), d. average catches of mealybugs in chrome-adhesive traps (5 yellow traps per operation), and e. phytosanitary level, in all of the plants of the experimental piece, both visually and with a stereoscope.

3. Results

The measurement with HPLC showed that leaves, followed by stem extract, had the highest amount of α-tomatine, with a much lower concentration in the green tomato fruit extract, which is in agreement with the literature about tomatine [7,9]. Respectively, the tomatidine was mainly detected in the extracts of the leaves and the green fruits, thus inthe stem extract the alkaloids content was low. As for the red tomato extract, it was not detected because of its degradation during the fruit's ripening [10,11]. Based on these results, a mixture of leaves, stems, and green tomato fruit extracts was used as the bioactive agent for encapsulation with the extrusion method. The pellets produced from the twin-screw extrusion (Figure 1a) were incorporated in a blown film extruder, and the produced film (Figure 1b) was used as cover for the stonewool grow blocks during hydroponic tomato cultivation (Figure 2, Table 1).



(**a**) (**b**)

Figure 1. Presentation of (**a**) the pellets of the encapsulated tomato leaves–stem extract in LDPE and (**b**) the final polymeric film with 1% bioactive compounds.

Figure 2. Application of the polymeric films with the encapsulated bioactive agents on tomato hydroponic crops cultivation.

Table 1. Phytosanitary level per experimental piece sample per month.

Sample/Month	Aug-20	Sep-20	Oct-20	Nov-20	Dec-20	Jan-21
Control	Good *	Presence of mealybug larvae (average: 8 ± 2/leaf)	Presence of mealybug larvae (average: 5 ± 2/leaf)	Presence of mealybug larvae (average: 2 ± 1/leaf)	2 plants infected by botrytis	Good
Test	Good	Presence of mealybug larvae (average: 6 ± 2/leaf)	Presence of mealybug larvae (average: 3 ± 1/leaf)	Good	3 plants infected by botrytis	Good

* Good phytosanitary level: no presence of mealybug larvae; absence of infected plants.

In Figures 3–6, the results from the application of the innovative films in the tomato hydroponic crops are presented in comparison with the same crops covered with commercial stonewool grow blocks.

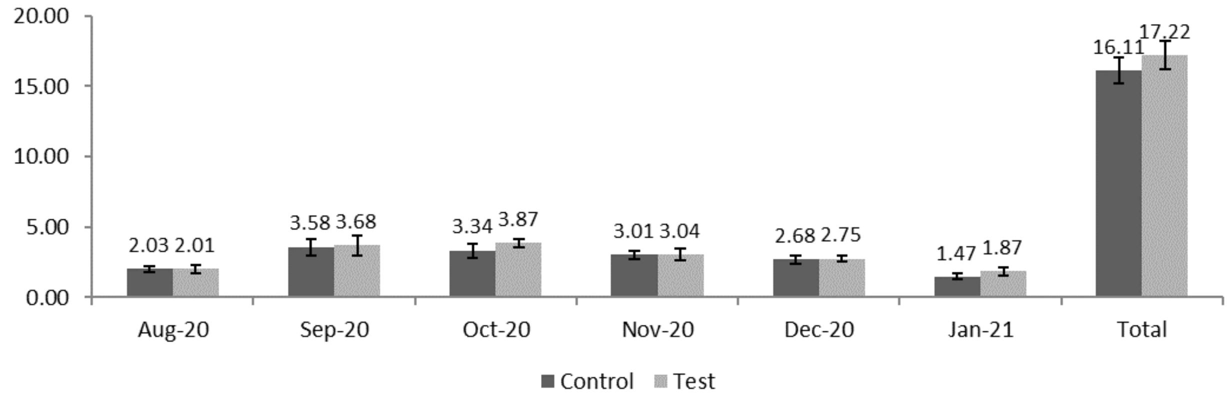


Figure 3. Total yield (kg/m^2) per experimental piece per month.

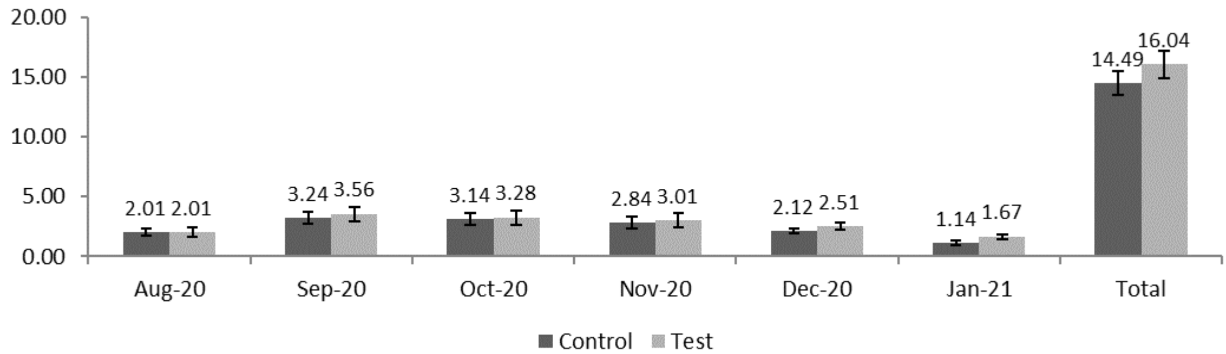


Figure 4. Marketable yield (kg/m^2) per experimental piece per month.

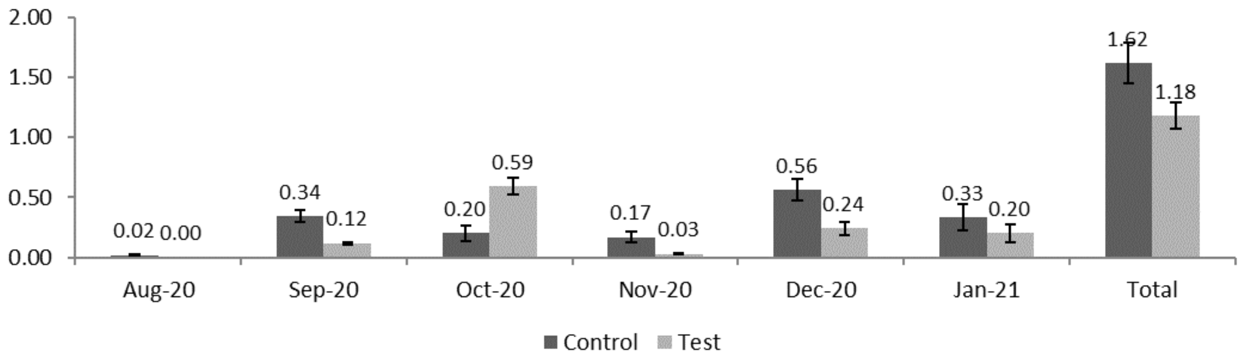


Figure 5. Non marketable yield (kg/m^2) per experimental piece per month.

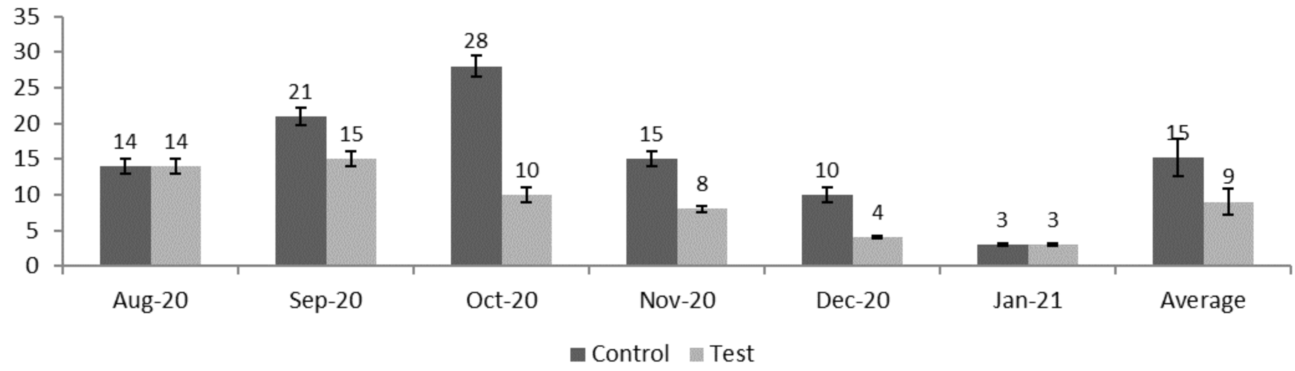


Figure 6. Average catches of mealybugs in chrome-adhesive traps per experimental piece per month.

4. Discussion

As can be observed in Figures 3–6, the test samples gave better results than the controls. More specifically, the test plants proved to be more productive than the control ones with higher total marketable yields. Moreover, a reduced attraction of mealybugs and the presence of larvae on the leaves was observed in the control plants; despite the fact that: (a) at the beginning of the experiments, the number of prosperous mealybugs counted both in the control and the test colored sticky traps was the same (14 prosperous/5 traps); (b) the plant protection operations that followed (biological and chemical) in both the control and the test were common; and (c) climatic conditions and hydro-lubrication were common.

5. Conclusions

Based on the results of the presented study, it is observed that the innovative polymeric films with encapsulated alkaloids derived from tomato cultivation by-products as a cover for the stonewool show significant activity towards the pesticide control of the Brioso tomato cultivation. This study served as a model for the practical application of the circular economy and zero pollution crop protection.

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Conflicts of Interest: The authors declare no conflict of interest.

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