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Review

Forecasting Western Corn Rootworm (*Diabrotica virgifera virgifera* LeConte) Density and Non-Chemical Control of Larvae: A Practical Review

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Abstract: The western corn rootworm (WCR) (*Diabrotica virgifera virgifera* LeConte; Chrysomelidae) is one of the most significant maize pests in Europe, with farmers spending a substantial amount (approximately 140 EUR) on its control. In the context of climate change, WCRs could pose an even greater threat to EU maize production, particularly as the European Union continues to withdraw an increasing number of effective yet environmentally harmful active agents. Biological control methods have now emerged to the forefront in creating sustainable agriculture. In this review, we carried out an extensive literature analysis on methods for forecasting WCRs and evaluated the practical applicability of the latest non-chemical control methods targeting its larvae. Effective forecasting is essential for successful pest management, enabling informed planning and the selection of the most suitable control methods. Several traditional predicting techniques remain in use today, but recent advancements have introduced modern electronic forecasting units combined with sensor-equipped pheromone and colour traps, as well as thermal sum calculations. Research has demonstrated that crop rotation is one of the most effective methods for controlling WCR larvae. Biological agents, such as entomopathogenic fungi (*Beauveria bossiana* and *Mettarrhyzum anasoplia*), entomopathogenic nematodes (*Heterorhabditis bacteriophora*), and botanical insecticides such as azadirachtin can significantly reduce larval populations and root damage, thereby maintaining infestation levels below the economic threshold. Genetically modified maize plants that produce specific toxins, along with conventional breeding efforts to increase root system regeneration, are also promising tools for the sustainable management of this pest. This review summarizes the solutions for prediction of western corn rootworm infestations and non-chemical control of its larvae. Accurate forecasting methods provide a clear picture of infestation levels in a given area, enabling precisely targeted control measures. In all cases, the control should be directed primarily against the larvae, thereby reducing root damage and reducing the size of the emerging imago population. This review demonstrates that biological control methods targeting larvae can be as effective as pesticides, supporting sustainable pest management.

Keywords: azadirachtin; entomopathogenic nematodes; *Heterorhabditis bacteriophora*; forecasting of western corn rootworm; seed dressing; western corn rootworm larvae; *Diabrotica virgifera virgifera*

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1. Introduction

The western corn rootworm (WCR) (*Diabrotica virgifera virgifera* LeConte 1868), an insect native to North America, is one of the most destructive pests of maize (*Zea mays* L.) in the temperate zones of Europe [1,2]. It first appeared on the continent in Belgrade in 1992, with larval damage detected the following year on 100,000 ha of maize grown in monoculture [3,4]. In Hungary, the first specimens were identified in 1995 in the Great Plain region, with economic damage first observed in 1998, and by 2001, the pest had spread

throughout the country. Today, it is found in many countries, including those north and east of Hungary, where it has been capable of causing significant economic damage [5].

The WCR has a high invasion potential, capable of migrating up to 50 km from infestation sites within a year [4]. It starts to cause economic damage five to seven years after its initial appearance [3]. The primary pests are the older larvae, which chew through the support roots of plants [6], thereby causing a characteristic lodging referred to as goose-necking in the literature [7,8]. During dry years, these roots cannot regenerate, and a windstorm or heavy rainfall can topple entire maize plants [9].

The adult individuals cause fertilisation problems by feeding on pistils and anthers [10]. They also reduce the photosynthetically active surface area by chewing through leaf veins [11] and act as vectors in spreading the bacterium *Pantoea ananatis*, which causes leaf spotting in plants [12]. The cost of controlling WCRs has risen proportionally with its spread. In Hungary, the price of maize seeds per hectare is approximately 150 EUR today. The cost of soil insecticides currently effective against western corn rootworm larvae is approximately 80 EUR per hectare, while the average price of preparations used against imago is 20 EUR per hectare, plus the application cost of 20–40 EUR per hectare. The combined cost of chemicals used against larvae and imagoes (application + material) is almost the same as the cost of hybrid maize seeds per hectare. In case maize is used as a preceding crop, it is necessary to control larval damage and prepare for controlling the imagoes as well [13]. Beyond agrotechnical methods, the most common control methods in Europe include soil disinfection with insecticides, seed dressing, and insecticidal crop treatments [14–16]. On the American continent, one highly significant tool for combating the pest is the cultivation of transgenic plants [17].

Several review articles have been published on the chemical control of WCR larvae and its effectiveness [18,19]. However, starting in 2018, the European Union initiated a large-scale withdrawal of active agents to protect the environment and human health. As a result, environmentally friendly biological solutions that pose no risk to human life and protect beneficial organisms have gained importance. In order to ensure successful pest control, proper forecasting is absolutely essential, as it allows for planning and selecting the most appropriate control method [20]. Nevertheless, the prediction methods focusing specifically on WCRs have not been systematically reviewed so far. The options for the biological control of insects of the genus *Diabrotica* were reviewed 26 years ago by Kuhlmann et al. [21], but a comprehensive review of the literature on this topic from subsequent periods is still lacking. Accordingly, the objectives of the present review article are (1) to summarise methods for forecasting WCRs and (2) to explore the studies published over the past 25 years on non-chemical control methods for WCRs that have been successfully integrated into actual practice. It is crucial to focus on controlling the larvae, since this can reduce the number of adult individuals, thereby preventing the damage caused by imagoes and the related significant yield losses.

For this review, existing peer-reviewed articles, books, and book chapters were searched using electronic databases such as ISI Web of Science, Scopus, Google Scholar, and Matarka (the latter for Hungarian articles). Studies published up to August 2024 were collected. For the topic of "forecasting", no start date was set; the keywords used were the English and scientific names of the WCR and "forecast", "prediction", "prognoses", and "detection". However, in the case of the "practical applicability of non-chemical control methods", only studies that had been published since 1999 were considered. Here, we combined the English and scientific names of the WCR with "larvae", together with the expressions "non-chemical control or management", "agrotechnical control", "crop rotation", "empathogenic fungi", "entomopathogenic nematodes", "plant extracts", "botanical insecticides", "gene technology", and "plant breeding". We selected only studies written in English or Hungarian, including those reporting practical applications.

2. Methods for Forecasting Western Corn Rootworm

Since the pest was first described in 1868, researchers have been concerned with how to predict the damage it might cause [22]. By observing the mature imagoes, it is possible to estimate the size of the population and larval pressure for the following year. Given that the eggs and larvae live in the soil, they are difficult to quantify [23]. Both direct and indirect methods are used to predict the presence of WCRs (Figure 1).

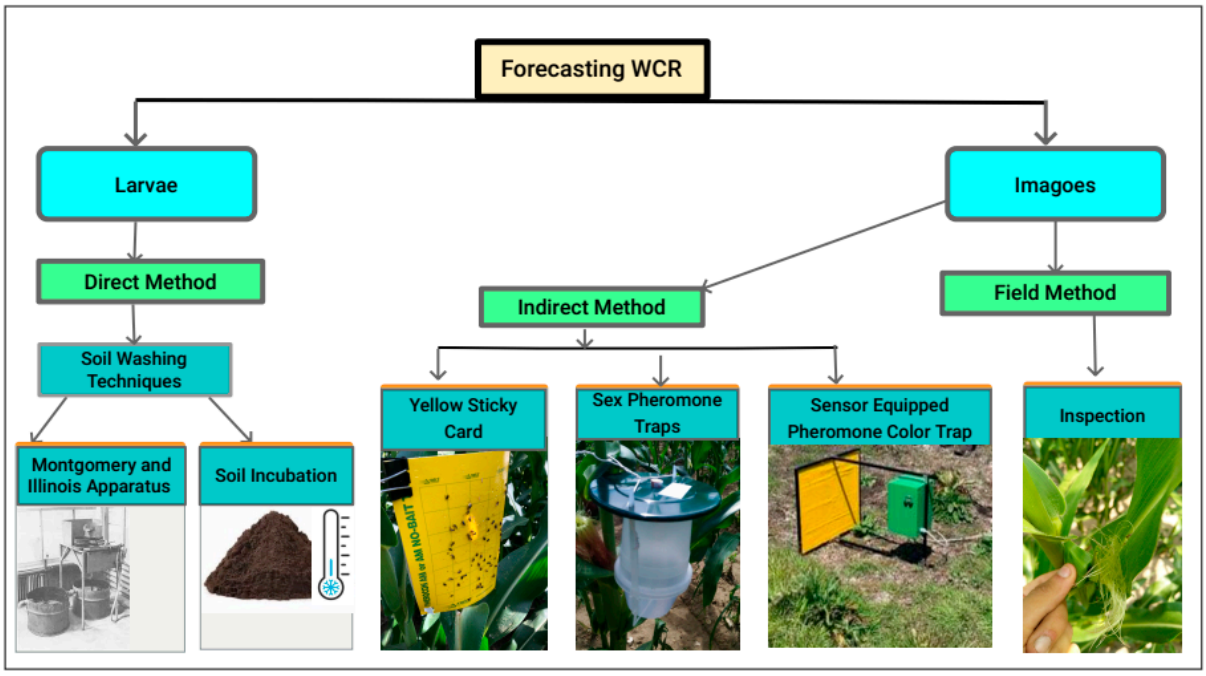


Figure 1. Overview of the methods for forecasting western corn rootworm (*Diabrotica virgifera virgifera*).

Direct prediction methods have two main approaches: rearing techniques and soil washing techniques [22]. Indirect methods infer the potential larval emergence of the following year [4]. Two methods are known for sampling during the dormant period. The soil washing method involves sedimentation to separate soil samples using water in order to determine the number of eggs present, the tools for which are the Montgomery and Illinois apparatus [22], respectively. The other method is soil incubation, where samples taken from maize fields are kept at a constant temperature until the eggs hatch [24].

Field sampling methods include determining larval counts and assessing the number of adult populations. During larval sampling, digging up plants allows for an accurate assessment of the number of larvae damaging the roots [7]. This method can precisely estimate the expected number of adult individuals and the potential yield loss using a damage estimation equation developed for WCRs (yield loss% = 0.001 + 0.765x, where x = number of larvae per plant) [7].

Several methods have been developed to determine the extent of root damage [25]. Today, the IOWA and modified IOWA scales are used for this assessment. In case of severe root damage, yield losses can reach up to 40% [25]. In addition to affecting yield quantity, larval damage also significantly impacts quality [26]. According to U.S. data, economic damage occurs when the level of damage reaches a value of 2.5 on the Iowa scale [27]. This value was later raised to 2.75 [28], then to 3 [29], and finally to 3.5 [30].

Adult counts can be monitored using various trapping methods, such as sex pheromone traps with fyke nets and sticky mechanisms, yellow sticky cards, and cucurbitacin traps. In central and eastern Europe, mainly yellow sticky traps have been used when WCR population levels exceeded a threshold. However, pheromone traps appeared to be most sensitive for early detection purposes, allowing also for tracking of flight activity during the summer and predicting the larval pressure for the following year [31–33].

The rapid advancement of information systems has enabled the development of electronic forecasting units combined with sensor-equipped pheromone and colour traps. Sensory devices can detect insects caught with traps attached to them, which requires precise programming of the electrical device to recognize certain species and distinguish between them. The catch data of traps attached to these devices can be saved, recorded, and even compared with the results of sensor apparatuses installed in other parts of the country. These devices have been determined to be 95.84% accurate in studies [34].

The development of WCR larvae and imagoes can also be tracked through thermal sum calculations. This method uses the minimum temperature required for pest development as a baseline. Once soil temperature reaches this threshold, the base temperature value is subtracted from the mean temperature, and these values are summed over subsequent days to assess the pest's biological status. The western corn rootworm developmental base temperature for overwintering eggs is 11 to 12 degrees Celsius. Approximately 165 degrees Celsius is required for larval development, while 400 degrees Celsius is required for the emergence of adult individuals. Male individuals appear four to six days earlier than females [35].

Field sampling can also be performed with the naked eye, using methods such as the plant inspection surveys described by Chiang [7], which involve counting and collecting adults on 1 to 10 plants or in tent isolators. It is important to note that imagoes exhibit intense flight activity in the early morning hours and just before sunset [36].

3. Practical Applications of Non-Chemical Methods for Controlling Western Corn Rootworm Larvae

The non-chemical control of WCR larvae can have agrotechnical, biological, gene technological, and plant-breeding-related approaches (Figure 2).

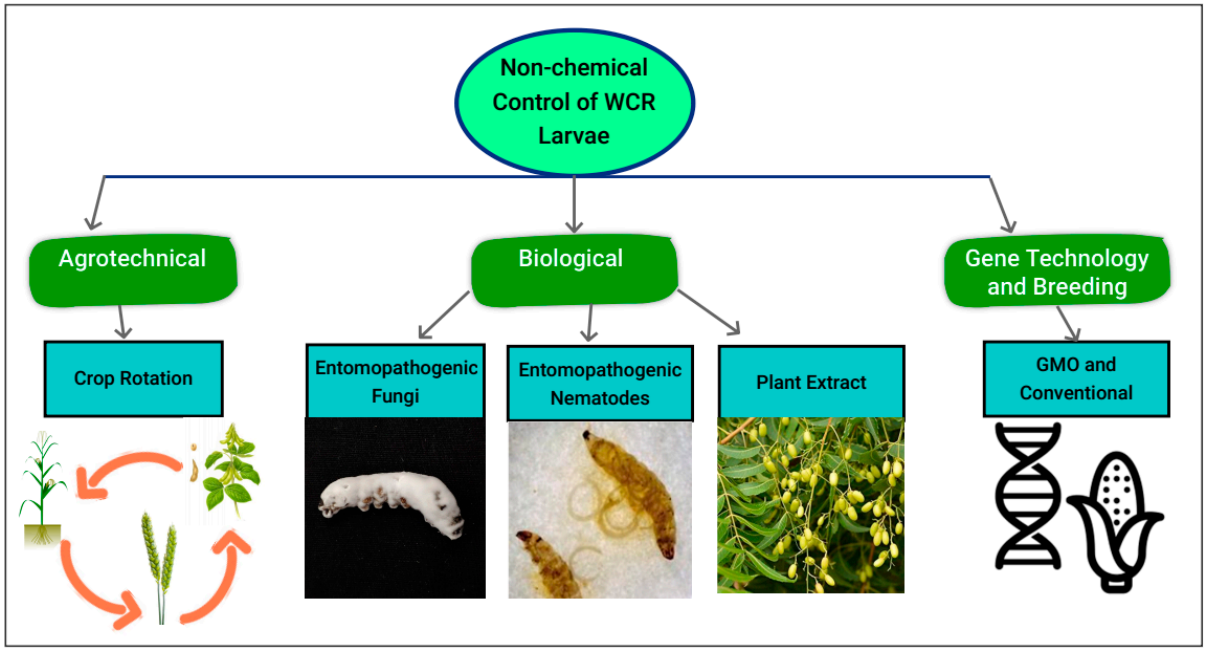


Figure 2. Overview of the methods for non-chemical control of western corn rootworm (*Diabrotica virgifera virgifera*) larvae.

3.1. Agrotechnical Control

Crop rotation is one of the most efficient control methods for reducing the damage caused by WCR larvae [1,16,37]. This method is particularly suitable because the WCR is a single-generation species, with females laying eggs in the soil of maize fields [38]. Therefore, environmentally friendly and cost-effective pest control can be achieved [39]. Numerous studies indicated that within Europe, e.g., in Austria, Hungary, and Italy, this

agrotechnical method can significantly reduce the spread intensity and the damage caused by the WCR [37,40–42]. In northeastern Italy, different crop rotation systems "(i.e., a structural one with one-time maize planting in a three-year rotation and a flexible one with continuous maize planting interrupted when beetle populations exceed the threshold)" were proven to be equally efficient for maintaining the WCR below economic threshold densities without the need for insecticides [43].

Szalai et al. [37] simulated the necessity of crop rotation in relation to WCR damage using a multifactorial model. Their study found that if crop rotation occurred on only 40% of maize fields, economic damage was inevitable in every case. A similar model study conducted in Austria examined the significance of crop rotation in controlling the WCR while considering various climate scenarios in the calculations. This research also supported the conclusion that reducing maize's share in crop rotation is one of the most effective methods for controlling this pest [42]. The applicability of these methods is significantly influenced by the size of the farm, the number of crop species cultivated, and economic considerations. To pursue more profitable crops, smaller farms tend to grow maize more frequently than larger farms. Because of these limiting factors, maize is often grown in monoculture, which favours the proliferation of the WCR. It is important to note that crop rotation alone does not necessarily prevent damage by this insect. A good example of this was the observation of economic damage to maize following a soybean preceding crop in the soybean–maize rotation system prevalent in some regions of North America as early as 1987. These strains are called "crop rotation-resistant" strains [44]. Recent studies on rotation-resistant strains revealed that the damage caused to soybean plants is due not to some attractant but simply to "rotation boredom", as the soybean–maize rotation is very popular in the United States [45]. Nevertheless, it can be stated that crop rotation remains the most important non-chemical control method wherever it is an economically viable solution.

3.2. Options for the Biological Control of WCR Larvae

3.2.1. Entomopathogenic Fungi (EPF)

The most effective EPFs used against soil-dwelling pests include *Beauveria* and *Metarhizium* species belonging to the *Ascomycota* phylum [46]. Notably, *Beauveria bassiana* (Bals.-Cri. Vuill, Clavicipitaceae, Hypocreales) and *Metarhizium anisopliae* (Metschnikoff, Clavicipitaceae, Hypocreales) are entomopathogenic fungi that are natural enemies of many soil-dwelling pests, including the larvae of the WCR [47,48]. Rudeen et al. [48] conducted experiments in field conditions with these two species in Iowa, United States, in areas with high WCR larval density. Their studies concluded that formulations with *Beauveria bassiana* and *Metarhizium anisopliae* had efficacies of 60% ± 6.3% and 55% ± 6.4%, respectively, against the WCR. In an experiment conducted in Austria in 2014 and 2015, the efficacy of a biological formulation containing the same two entomopathogenic fungi was compared with chemical control options against the larvae of the WCR. The results of the experiment showed no significant differences between the treatments, suggesting that the effectiveness of the biological formulations is comparable to that of chemical control [49].

Pilz et al. [50] conducted surveys in several European countries (Hungary, Austria, Romania, and Serbia) between 2005 and 2008 under field conditions. They compared the root systems of plants dug up with soil and examined the number of WCR larvae and pupae in the soil with the naturally occurring fungal species isolated from the soil. Their study revealed that 1.4% of WCR larvae, 0.2% of pupae, and 0.05% of adults emerging from the soil were naturally infected with fungi. Additionally, their research found that maize-growing soil in Hungary had a higher incidence of *Metarhizium anisopliae* infection than that in other countries.

A laboratory experiment conducted in Slovakia also confirmed the effectiveness of EPFs against the larvae of the WCR. In these tests, the fungi *Beauveria bassiana*, *Beauveria brongniartii* (Sacc), and *Metarhizium anisopliae* were used, and larvae were treated with an experimental material at a concentration of 2×10^7 conidia/mL. Fourteen days after

the experiment was set up, a mortality rate of 25.83% to 60.57% was observed among the pests [51].

3.2.2. Entomopathogenic Nematodes

Pilz et al. [50] discovered natural enemies of the WCR that are naturally soil-dwelling, namely, *Heterorhabditis* sp., *Steinernema* sp. (EPNs), and they were the first to report on these in Europe [52]. In addition to entomopathogenic fungi, various EPNs can form the basis of another effective biological control method in agricultural practice. Such species include *Steinernema glasseri* Steiner, *Steinernema arenarium* (Artyukhovsky), *Steinernema abassi* (Elawad, Ahmad, and Reid), *Steinernema bicornatum* (Tallosi, Peters, and Ehlers), *Steinernema feltiae* (Filipjev), *Steinernena carpocapsae* Weiser, and *Heterorhabditis bacteriophora* (Poinar), which are all capable of entering WCR larvae and killing their hosts by reproducing inside them [53–56]. Among the species studied, *H. bacteriophora* proved to be the most effective, causing 77% mortality among WCR larvae in lab conditions [52]. The nematodes act as vectors that introduce their co-living bacteria (*Photorhabdus luminescens*) into the pests, and the propagating bacteria kill the target species and liquefy their bodies, thereby providing food and opportunity to reproduce for the nematodes [53–56].

EPNs detect their prey by releasing volatile compounds [57,58]. They enter the target organism through the anal and oral orifices, as well as via the epidermal route using their mouth stylet. The invaded WCR larvae typically die within two to three days after being entered by the nematodes [52]. In the body of the dead larvae, the nematodes continue to reproduce and develop, and after 12 days, approximately 4000 new virulent nematode larvae (L3, juvenile infective) are released, and they seek out a new host organism [52]. To facilitate widespread use in plant protection, the industrial-scale propagation of nematodes has been made feasible, allowing them to reach and control maize WCR larvae even in areas where their natural fauna is limited. Based on manufacturing conditions and costs, *H. bacteriophora* is the most suitable entomopathogenic nematode for use in biological control, but studies have suggested that *S. arenarium* and *S. feltiae* are also promising candidates [52].

In Brazil, the results of Santos et al. [59] also provided evidence that nematodes of the *Steinernema* and *Heterorhabditis* genera showed the best larvicide effect for controlling *Diabrotica speciosa*. Further experiments showed 3% to 15% reduction in root damage on the Iowa scale and 14% to 54% reduction on the scale for damage to the various root stages as a result of using *Heterorhabditis bacteriophora* in lab and greenhouse conditions, respectively [60,61].

Toepfer et al. [60,61] conducted efficacy studies of *H. bacteriophore* entomopathogenic nematodes against WCR larvae in Hungary in field conditions between 2004 and 2010, using various application technologies (surface spraying and injection into the soil). Whereas soil injection involved applying 230,000 nematodes per square meter, surface spraying used 400,000 nematodes per square meter. The study concluded that these two methods of application reduced the level of root damage by at least 50%. The highest efficacy was observed when the nematodes were injected into the soil simultaneously with the sowing; the observed larvicide effect was up to 68%. In comparison with surface spraying, the advantages of injecting simultaneously with the sowing process included a lower water need and a lack of harmful UV exposure [62]. In a subsequent experiment, nematodes were injected simultaneously with sowing at a rate of 2 billion/hectare and using a water quantity of 200 L; the observed efficacy in terms of root chewing was 79% in this study [60,61].

In the years 2010–2011, six field experiments were conducted in Germany, Austria, and Hungary on different soil types using the nematode *H. bacteriophora*. The nematodes were applied in the planting rows either directly in a liquid form or as a granulate dissolved in water. It was concluded that the biological agent, regardless of its formulation, retained its vitality and persistence until the WCR larvae hatched [63]. In 2012, the mass production of *Heterorhabditis bacteriophora* was successfully achieved, and it was commercially released under the name Dianem [13].

Geisert et al. [64] conducted laboratory studies with various entomopathogenic strains against WCR larvae, where they infected L3 larvae with 60 to 120 nematodes (*H. bacteriophora*, *H. megidis*, *S. feltiae*, *S. carpocapsae*, *S. diaprepesi*, *S. riobrave*, *S. rarum*, *S. diaprepesi*). Their findings revealed that *H. bacteriophora* and *S. diaprepesi* were the most effective in terms of efficacy against WCR larvae, with mortality rates over 80%. In contrast, *Steinernema carpocapsae* proved to be significantly the least effective, causing only 30% larval mortality.

Field experiments conducted by Toepfer et al. [60,61] confirmed that *H. bacteriophora* nematodes, applied at sowing, showed a proportionately smaller decline in effectiveness over time compared with soil insecticides comprising chemical active ingredients (cypermethrin, chlorpyrifos, tefluthrin). This suggests that these nematodes are able to reproduce in the soil, thereby reducing the size of the subsequently emerging imago population [61].

These field studies concluded that *H. bacteriophora* applied at a rate of 2 billion per hectare showed appropriate efficacy in combating WCR larvae [65]. Another study suggested that the most outstanding results could be achieved when the ecological conditions were favourable for the nematodes (a soil temperature of at least 10 °C; moist soil conditions) [65]. The viability of nematodes is also influenced by the type of the soil. Research results showed that the vitality and larvicide effect of nematodes was better in sandy soils with looser texture than in heavy soils with high clay content [60]. An experiment conducted in Southern Hungary that compared biological (*H. bacteriophora*, *B. bassiana*, *M. anisopliae*) and chemical (clothianidin 107 g/ha, tefluthrin 15 kg/ha) insecticides found that chemical products reduced soil fauna to a greater extent and killed numerous useful organisms besides the target species [66]. Further favourable effects of using entomopathogenic nematodes include applicability in ecological farming, as well as a reduced risk of potentially developing resistance [13].

There are several options for field applications of entomopathogenic nematodes and entomopathogenic fungi. One such solution is to spray the soil with fungal propagation formulas or nematodes and then mix them into the soil. The disadvantage of this method is that the application involves two workflows, and the biological agents are exposed to abiotic effects on the soil surface until soil cultivation takes place. The other solution is the injection of biological agents directly into the soil within the course of sowing. However, this latter method needs a specialized application device [13].

3.2.3. Azadirachtin as Plant Extracts

Biologically derived pesticides such as azadirachtin are gaining increasing importance, as they are usually effective to combat pests and are not persistent in the environment, therefore exhibiting low toxicity to humans and beneficial organisms [67]. In view of the sustainability principles, these control methods have an important role in the practice of plant protection. The extract of the seeds of neem (*Azadirachta indica* A. Juss), a tree from Southeastern and South Asia that contains the active ingredient azadirachtin as a main component, meets the above criteria [68].

The neem tree, also known as the margosa tree or Indian lilac (*Azadirachta indica*), is an evergreen plant belonging to the Meliaceae family. Its genetic centre is in the subtropical regions of India, from where it spread to various countries in Asia and Africa [69]. A mature tree can produce 40 to 50 kg of fruit annually, which collectively contains approximately 5 kg of seeds, from which up to 50 g of pure azadirachtin active ingredient can be extracted. In addition to azadirachtin, which is the main active ingredient extracted from the seed oil of the tree, neem contains more than a hundred biologically active ingredients that greatly reduce the chance of developing resistance to its main active ingredient [70]. Azadirachtin has a broad of action, including feeding inhibition, repellent effects, and growth regulation, as it disrupts the production of the hormone ecdysone, thereby blocking the moulting cycle. In addition to the above, it affects reproduction by inhibiting egg laying and causes fertility disorders and sterility [71–75]. Like colchicine, azadirachtin may also interfere with mitosis, and it has a direct histopathological effect on the intestinal epithelial cells, muscles, and adipose tissues, which leads to restricted movements and reduced flying

activity [76]. Because of its systemic and translocating properties, this active ingredient provides a long-lasting action [77]. Numerous studies have been conducted showing that the active ingredient has already been successfully used against several foliar pests in the form of crop treatment; it proved to be equally effective against Hymenoptera [78], Lepidoptera [79–81], and Hemiptera [82–84].

Tóth et al. [34] conducted experiments in greenhouse with a granular formulation of the active ingredient azadirachtin against WCR larvae. Initially, they applied biological tests based on filter paper and artificial feeding under controlled laboratory conditions to test the ovicidal and larvicidal effects of this biological insecticide. Their studies revealed that the azadirachtin active ingredient did not exhibit ovicidal effects, but its larvicidal effect proved to be effective. After three days, they estimated the LD50 value for larval control to be approximately 22 µg azadirachtin/mL. In their second experiment, they set up potted plant trials under greenhouse conditions to test the active ingredient, aiming to determine the extent to which this new azadirachtin-containing granular product could reduce the root damage caused by maize WCR larvae. Their studies demonstrated that the recommended dosage of 27 g of active ingredient per hectare was insufficient to reduce larval counts. However, the tenfold standard dose (380 g active ingredient/ha) eliminated all larvae and fully protected maize roots.

Since 2020, research has been conducted in Hungary on the application of azadirachtin as a seed-dressing agent against WCRs in field conditions, with very promising results reported [5]. Experiments were conducted over two years (2020 and 2021) at multiple locations in Hungary (in the West Transdanubia and Great Plain regions). Two azadirachtin-containing products were tested using a seed-dressing technology against WCR larvae. The first product was Neemazal T/S (Trifolio-M GmBH, Germany), which is available on the European market and contains 1% (10 g/L) of the active ingredient azadirachtin A. The other was Neemazal F (5% azadirachtin A+B; 50 g/L), a more concentrated product authorised in India (Coromandel International Limited Bio Products Division Thyagavalli, India) and used in the study. According to the Safety Data Sheet of the latter, the 5% active ingredient content of Neemazal F consists of 80% azadirachtin A and 20% azadirachtin B. In addition to said main components, both products contain smaller amounts of other neem components characteristic of plant extracts, which could significantly reduce the development of resistance to the active ingredient in the future. The experiments confirmed that this active agent, when used in a seed-dressing technology, could effectively combat WCR larvae and mitigate the extent of the resulting root damage. These treatments reduced larval counts to nearly one-third compared with the untreated control plot, as well as lowering m. Iowa scale root damage values. In areas with lower larval density (West Transdanubia, m.Iowa: 3.23), seed-dressing concentrations ranging from 50% (0.022 mg/seed) to 100% (0.043 mg/seed) were the most effective, while in the Great Plain region, where larval density was higher (m.Iowa: 4.42), seed-dressing concentrations ranging from 125% (0.053 mg/seed) to 150% (0.065 mg/seed) proved to be the most effective [5].

3.3. Other Options: Gene Technology and Plant Breeding

Among transgenic plants, the most widespread are maize plants that produce Bt. toxins, which allow for simple control of the WCR [85,86]. The literature also mentions other GMO-based methods against the WCR, such as plants engineered to produce protein toxins from *Chromobacterium pisicinae* (Kämpfer, b-proteobacteria), which can achieve high mortality rates (50–75%) [87]. The advantage of these technologies is that they can easily reduce pest populations below the economic threshold. However, the use of such methods is banned in many places (e.g., in the EU), and it is likely that long-term use could lead to the selection of resistant populations [85,86]. To delay the selection of resistant WCR populations to present and future transgenic traits, more diversified management, including crop rotation and use of non-Bt. maize with soil-applied insecticide, should be implemented [88].

In the fight against WCR larvae, conventional breeding efforts are also underway, focusing on the rapid regenerative ability of the root system of the maize crop. Studies showed that nine maize hybrids bred in Croatia (Institute of Agriculture, Osijek) and two North American hybrids (Pioneer Hi-Bred Int. Inc.) could significantly regenerate from larval damage, thereby avoiding substantial economic losses [89]. Similar results were observed in a study conducted in the United States, where it was found that two maize hybrids, Mp708 and Tx601, showed resistance to WCR larvae [90].

4. Conclusions

Because of cultivation in monoculture, the WCR causes increasing economic damage to maize. Controlling this pest has become difficult in the European Union because of a growing number of withdrawn active ingredients (e.g., neonicotinoids, organophosphorus acids). The list of permitted insecticides has shrunk so much that we are now facing the risk of developing resistance to the currently marketed effective insecticides. In view of the above, there exists a need for novel and effective control methods that can be incorporated into integrated agricultural practices.

Successful control must be based on accurate and professional forecasting. Among the forecasting methods, predicting the adult WCR is the most informative approach, from which the extent of larval pressure can be anticipated. Rearing and soil-washing techniques require tough physical activities and do not offer precise estimation. Sticky yellow traps, one of the oldest methods, are still the most popular for predicting this pest; however, they are not selective in terms of insect attraction. A better choice for detection is the sex pheromone trap, which emits only the sex pheromone produced by WCR females and attracts only the male individuals of the species. Such traps are easier to assess, and changes in population dynamics become much easier to monitor, which is why farmers should use this forecasting method. New techniques are based on electronic forecasting units combined with sensor-equipped pheromone and colour traps, as well as thermal sum calculations.

Plenty of studies have been published regarding the practical applicability of non-chemical control methods on WCR larvae in the last 25 years. Among those studied, crop rotation, entomopathogenic fungi (EPF), entomopathogenic nematodes (EPN), and botanical insecticides stand out as highly significant methods for effectively combating WCR larvae. The economic aspect of non-chemical control methods is also an important issue. In general, compared with chemical soil disinfectant granules currently approved in the European Union, the cost of treatments with EPF or EPN is approximately double. Nevertheless, the cost of azadirachtin application with active ingredient dressing technology is only a third of the cost of chemicals. GMO and conventional breeding approaches were also developed to produce toxins in maize and to boost the regenerative ability of the crop's root system; however, growing GMO crops is not yet allowed in the European Union.

5. Future Directions

We believe that, considering today's technological developments, pest forecasting tools based on sensor technology will also soon become widespread. In addition, the accuracy and evaluation of these forecasts will be much simpler, as changes in the population dynamics of the pest will become digitally traceable and less exposed to physical work and will therefore become part of the lexical knowledge of farmers. Such devices today are in continuous development, and their acquisition is very expensive. However, it is believed that in the near future, they will be a breakthrough in the detection of pests.

We believe that the control methods presented in our review article are expected to become increasingly integrated into plant protection practices in the future, in line with sustainability principles. It can be predicted that in the European Union, azadirachtin, as a botanical insecticide, will be highly utilized among non-chemical methods in controlling WCR larvae. Since it can be used directly as seed dressing or granules, its application follows the protocols of chemical insecticides, and therefore, farmers are likely to prefer

them to nematodes and fungi. Since the latter two groups are living organisms, more attention needs to be paid to their storage, so their application is much more complicated and expensive.

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