Nematicidal Potentialities of Alginate Controlled-Release Formulations of Aldicarb and Cadusafos on The Root-Knot Nematode *Meloidogyne incognita* Infecting Tomato

Hamdy R. Soltan,[1] A. S. Marei and A.Mousa[2]

ABSTRACT

Two separate pot experiments was conducted to evaluate the nematicidal activity of the alginate controlled release (CR) formulations of aldicarb (FA_3, FA_6 and FA_9) and cadusafos (FC_3, FC_6 and FC_9) versus the corresponding granule formulations against the root-knot nematode *Meloidogyne incognita* infecting tomato under green house conditions. The results indicated that all the tested formulations significantly reduced the number of galls, egg masses, and 2nd juveniles in the soil as compared with the untreated pots (control). On the other hand, the CR formulations of aldicarb and cadusafos could be reduced the galls, egg masses, and 2nd juveniles in soil significantly compared with their granular formulations. The percentage reduction in nematode galls for all the CR formulations of aldicarb and cadusafos were ranged from (95.1% to 96.04%) and (87.1% to 88.03%) for aldicarb and cadusafos, respectively. Whereas, the lowest percentage of reduction of the number of tomato galls was recorded with aldicarb and cadusafos granule formulation (61.3% and 66.71%), respectively. Also, the maximum reduction of the number of 2nd juveniles in the soil was achieved by the FA_3 (86.73%) and FC_3 (89.08%), while for its corresponding granule was (59.05% and 46.23%). It could be noted that the nematicidal efficacy of the CR formulations against *M. incognita* reached to approximately 1.77 folds by FA_3 and 1.93 folds by FC_3 comparable to the commercial formulations. The possible effects of different aldicarb formulations residues on the chemical constituents of tomato fruits were determined and recorded at the end of the experiment (72 days). The results obtained indicating that all the aldicarb and cadusafos formulations had significantly decreased of the β-carotene, lycopene, and ascorbic acid content of tomato fruits comparable to the untreated pots. No significant differences were found between all the aldicarb formulations and the control on the percentage of the total soluble solid (T.S.S.) whereas there were significantly increased in the percentage of T.S.S. of tomato plant treated with the CR cadusafos formulations. The FA_3 and FC_3 could be applied on tomato plant to control nematode with a minimum side effect on the quality control parameters such as lycopene, T.S.S. and ascorbic acid content of tomato juice compared with the commercial formulations.

Key word: controlled release formulations, root-knot nematode, plant chemical constituent.

INTRODUCTION

Root-knot nematodes (*Meloidogyne* spp.) are among the most economically damaging genera of plant parasitic nematodes on horticultural and field crops, causing an estimated US$100 billion loss globally on an annual basis (Oka *et al.*, 2000). *Meloidogyne incognita* is one of the most harmful root-knot nematode species, which infects a wide range of vegetable crops in Egypt (Ibrahim *et al.*, 2000). Root-knot nematodes infection of tomatoes limits fruit production by 30% (Anwar and McKenry, 2010). Among various control measures, use of chemicals has been considered as an effective strategy for control of plant parasitic nematodes when other methods like cultural practices, resistant varieties and bio-control agents are unable to protect crops from these pests (Randhawa *et al.*, 2001; Sakhuja and Jain, 2001). Currently various fumigants and non-fumigants are available in the market against these pests (Rich *et al.*, 2004). Granular non-fumigant nematicides are more easily applied and safer for farmers compared with fumigants (Lamberti *et al.*, 2000). Among the most extensively used non-fumigant granular are aldicarb and cadusafos which are carbamate and organophosphate based nematicides. Although chemical nematicides are effective, easy to apply and show rapid effects, they have begun withdrawn from the market owing to concerns about public health and environmental safety (Rich *et al.*, 2004). Controlled-release formulations technology offers the potential to reduce the environmental losses of pesticides and hence may be increased their efficacy, reduce their side effect to non-target organisms and reduce their harmful effect on growth and nutrient contents of crops. Biodegradable and synthetic matrices for carbofuran formulations were assessed against *M. incognita* infecting tomato by Radwan *et al.* (2006). Alginate gels are biodegradable and pesticides can be easily incorporated into the matrix using an aqueous

[1]Department of Applied Entomology, Faculty of Agriculture, Alexandria University, Egypt
[2]Agriculture Research Centre Sabahia Research Station, Alexandria Central Pesticide Research Lab
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system at ambient temperature (Soltan, 1991; Soltan 1996 and Soltan *et.al.,* 2014). For all the reasons mentioned above, alginate gels were selected as matrices for preparation of new controlled-release formulations of aldicarb and cadusafos nematicides. The objective of this research was to investigate if by using aldicarb or/and cadusafos in the form alginate controlled-release formulations, its efficacy vis-à-vis *M. incognita* control asspcoated tomato plants could be improved by evaluating the nematicidal efficacy expressed as the percent reduction of total number of galls, egg masses and 2nd juveniles nematode in the soil. The chemical constituents of tomato fruits were determined that may be affected by the aldicarb and cadusafos released from the different formulations

MATERIALS AND METHODS

Preparation on controlled – release formulations of aldicarb and cadusafos:-

Different types of alginate controlled-release formulations (CRF.s) for each nematicide (aldicarb and/or cadusafos) were prepared in the present study according to the method described by Soltan *et.al.* 2014. Among the CRF's prepared, FA_3, FA_6 and FA_9, for aldicarb and FC_3, FC_6 and FC_9, for cadusafos were tested. Commercial formulation (15G) for aldicarb and 10G for cadusafose was provided by Agrochem Company, Egypt, and FMC Co., U.S.A, respectively.

Nematicidal activity of aldicarb and cadusafos formulations against *Meloidogyne incognita* on tomato seedlings

Plastic pots (20 cm in diameter and 25 cm deep) were filled with 3Kg sand soil. Two tomato seedling (*Lycopersicon esculentum* L. var. strain B), three weeks old, were transplanted. Recommended practices for fertilization and irrigation were followed and the pots were left for one week before treatments in order to acclimate the tomato seedling under the greenhouse condition. After planting, treatments were arranged in a randomized complete block design with 6 replications for each treatment in addition to the control. Pots were all inoculated with the suspension of 5000 2nd juveniles of *M. incognita* per pot. Aldicarb formulations (FA_3, FA_6, FA_9 and 15G) and cadusafos formulations (FC_3, FC_6, FC_9, and 10G), 3 days latter were applied on the surface layer of the soil at the recommended rate (1.5mg and 2.4mg a.i. /kg soil) for aldicarb and cadusafos, respectively. The efficiency of the formulations tested was judged, seven weeks after nematicide formulations treatment, by determining the number of galls per root system, egg masses and 2nd juveniles' nematode in the soil. The number of gall per root system of plant was accounted visually. The number of egg masses per root system was counted after staining with phloxin B (prepared by dissolving 0.12 g phloxin B per liter of water). The galled roots were placed in this solution for 15-20 minutes. The roots were gently rinsed in tap water. The egg masses were stained red and counted directly. On the other hand, the 2nd juveniles per 250 g soil were extracted according to Baermann plate technique (Goodey, 1963). The soil samples were suspended by tap water at least three times, and then screened through 200, 325 mesh sieve, respectively. The juveniles were washed and decanted from 325 mesh sieve to dish equipped with a tissue paper, and were left for 24 hours before counting under microscope.

Impact of aldicarb and cadusafos formulations on some internal quality parameters of tomato fruits:

Internal quality parameters of treated and untreated tomato fruits were measured at the end of experiment such as β-carotene, lycopene, ascorbic acid, and total soluble solid (T.S.S.). The method described by AOAC (1965) was followed to extract β-carotene and lycopene. The β Carotene color was measured at 450 nm against blank according to the method described by Umiel and Gabelman (1971) and lycopene measured at 503 nm according to the method described by Rangama(1977). Ascorbic acid was measured according to the method of AOAC (1984), Extraction and determination of total soluble solid (T.S.S.) in tomato fruits according to the method reported by Pearson (1979).

Statistical analysis:

Data were analyzed by analysis of variance and mean comparison by least significant difference (L.S.D.) measured at the probability level of 0.05 using SAS statistical software.

RESULTS AND DISCUSSION

Nematicidal activity of aldicarb and cadusafos formulations against *Meloidogyne incognita* on tomato plants:

The data in Tables (1 and 2) indicated that all the aldicarb formulations significantly reduced the number of root galling and the larval number of *M. incognita* as compared with the untreated pots. No significant differences among alginate controlled release formulations were found in the number of galls which were ranged from 87.1% to 95.1%. The granular formulation (15G) was the lowest effective formulation (61.3%) in this concern. The numbers of galls /root system recorded were 11.17, 18.33, and 29.5 for FA_3, FA_6, and FA_9, respectively. Whereas, for aldicarb 15G was 88.1 and control was 227.83.

Table 1. Effect of different aldicarb formulations on *Meloidogyne incognita* infecting tomato under greenhouse conditions

Treatment	*Mean number of galls/root system	Mean number of egg masses /root system	Mean number of 2nd juveniles /250 g soil
Control	227.8±37 c	32400±4.51 d	668.17±1.37 d
FA_3	11.3±7.8 a	7185±4.0 a	86.6±0.8 a
FA_6	18.3±6.64 a	10410.7±4.51 ab	126.53±0.94 a
FA_9	29.5±4.98 a	12663.7±6.67 b	207.3±0.94 b
15G	88.1±23.04 b	17136.7±3.8 c	273.61±1.0 c

*Each value is a mean ±SD of six replicates

Values followed by the same letter are not significantly different at $p \geq 0.05$.

Table 2. % Reduction in nematode parameters as affected by different aldicarb formulations

Treatment	Effectiveness on galls (%)	Effectiveness on egg masses (%)	Effectiveness on 2nd juveniles/ 250g soil (%)
Control	-	-	-
FA_3	95.1	77.82	86.73
FA_6	91.95	67.87	80.76
FA_9	87.1	60.95	68.87
15G	61.3	47.11	59.05

It should be cited that the CR formulations may be extended the nematicidal efficacy of aldicarb on tomato plant than its granular formulation. It can be concluded that there were negative correlation between the release rate of aldicarb formulations and the mean total number of galls/root of tomato seedlings, which in turn lead to increasing in tomato yield. In addition, the CRFs highly reduced the mean total number of egg masses comparable to the granular formulation. The number of egg masses reduction expressed as percentage of the total number of egg masses were ranged from 60.95% to 77.82% for CRFs and 47.11% for the granular formulation. Also, the controlled release formulations were the most effective formulations in the percent reduction on juveniles into galls and were ranged from 61.7% to 83.96%. Whereas the granular formulation considered the lowest effective one in this respects (44.9%). Deleon, (1984) reported that aldicarb decreased the population of *M. incognita*, by more than 57%. There was no significant difference on the mean of total egg asses into galls between FA_3 and FA_6 as well as between FA_6 and FA_9. Concerning to the number of 2nd juveniles 250g soil, the data showed that FA_3 and FA_6 were the most effective in reduction of the number of 2nd juveniles. Generally, it can be concluded that the highest of nematicidal efficacy was caused by the controlled-released formulations FA_3 (86.73%), FA_6 (80.76%), FA_9 (68.87%)] whereas, the granular formulation gave the lowest effect (59.05%) according to the total number of 2nd juveniles / 250 gm soil. These results are in agreement with those obtained by El-Shoura *et al.*, (1992). They found that the controlled-release formulations of carbofuran were the most effective treatment in reducing the *M. javanica* infecting tomato plant. Also, Parlaza et al., (1979) found that aldicarb caused a marked reduction of *Meloidogyne spp.* population on carrot.

Data presented in Tables (3 and 4) indicated that cadusafos controlled-release formulations significantly reduced the number of root galls and 2nd juveniles in the soil than that of its commercial granule formulation. Radwan *et al.* (2012) reported that cadusafos granule was the least effective compound tested and caused 77.51 and 86.63% reduction in root galling and J_2 population in the soil, respectively.

Among the CR formulations, no significant deference in the number of galls and 2nd juveniles in the soil between FC_3 and FC_6. The percent reductions in nematode galls for all the controlled-release formulations were ranged from 88.03 to 96.04%. On the other hand, the lowest percent reduction in mean total number of galls was recorded associated tomato seedlings treated with the commercial formulation (66.71%) comparable to untreated treatment. Ibrahim (1994) found that the cadusafos applied at the rate (15-45 mg/pot) was totally suppressed the root-knot nematode on tomato plant.

Once again, according to the number of egg masses the result cited that the formulations tested were suppressed the mean total number comparable to the control and with a wide range of efficacy based on the formulation types.

Table 3. Effect of different cadusafos formulations on *Meloidogyne incognita* infecting tomato under greenhouse conditions

Treatment	*Mean number of galls/root system	Mean number of egg masses /root system	Mean number of 2nd juveniles /250 g soil ±S.D.
Control	227.8±37 d	32400±4.51 d	668.17±1.37 d
FC_3	9±4.81 a	6626.7±3.51 a	72.94±0.7 a
FC_6	16.2±4.16 a	9800±4.58 ab	104.5±0.77 a
FC_9	27.2±4.9 b	11176.7±3.51 b	184.25±1.34 b
10G	75.8±18.5 c	15263.3±2.1 c	239±0.92 c

*Each value is a mean ±SD of six replicates

Values followed by the same letter are not significantly different at $p \geq 0.05$.

Table 4. % Reduction in nematode parameters as effected by different cadusafos formulations

Treatment	Effectiveness on galls (%)	Effectiveness on egg masses (%)	Effectiveness on 2nd juveniles/ 250g soil (%)
Control	-	-	-
FC_3	96.04	79.55	89.08
FC_6	92.86	69.75	84.36
FC_9	88.03	65.5	72.42
10G	66.71	52.89	46.23

Generally, the CR formulations were more effective in reducing the mean total number of eggs than the granular formulation. In addition, the results indicated that the percent reduction in number of eggs into galls were ranged from 65.5% to 79.55% for CR formulations whereas, the percentage was 52.89% for commercial formulation.

The same trend was obtained concerning the mean total number of all juveniles into galls. The higher reduction was obtained from the roots treated with the CR formulations, mainly FC_3 (88.7%) and the lowest reduction was obtained from pots treated with the granule formulation (50.41%) compared to untreated pots. However, the efficacy of cadusafos formulations tested could be arranged descending as follow: FC_3-FC_6>FC_9>10G according to all parameters used to evaluate the efficacy of cadusafos formulations. These data was supported by those data obtained during the present course of study that reported the highest release rate, residue levels in soil as well as residue levels in the root zone of tomato plant was from the pots treated with FC_3 while, the lowest from the granular formulation. Also, it should be noted that the decreasing in the mean total number of eggs and the mean total number of all juveniles into gall, indicated that the alginate incorporated cadusafos might be improved its toxicity behavior from non-persistence nematicide to persistent one and/or improving the penetration of cadusafos into nematode galls.

Impact of aldicarb and cadusafos formulations on some internal quality parameters of tomato fruits:

Pesticide residues may cause no visible injury to the plants on which they are used. Some of the less apparent injurious effects of pesticides on plants are interference with biochemical and physiological processes in it. Any of this interference may temporary or permanently retard the growth of the plant and then crop production (yield), may lower its nutrition value, may even prevent its use as food, and may affect its quality characterization (Hance, 1981). The quality of plant and its validity for edible use by consumer depends on the contaminations with pesticide residues. This role of pesticides in altering the nutrient levels in the plants should be considered when using chemical available as potential pesticide. The effect of the aldicarb formulations tested on the quality parameters in tomato to juice was recorded and the data were presented in Table (5). The results indicated that all the aldicarb formulations had significantly decreased of the β-carotene, lycopene and ascorbic acid content comparable to the untreated pots in tomato fruits. No significant difference was found among all the aldicarb formulations and the control on the percentage of the total soluble solid (T.S.S.).

Also, no significant differences were found among the controlled-release formulations (CRFs) on the β-Carotene content, and ascorbic acid content whereas, there were significant difference between FA_9 and both FA_3 and FA_6 on the lycopene content.

Table 5. Effecte of Aldicarb Formulations Residue on some Quality Parameters of Tomato Fruits

Treatment	β-Carotene Content (ppm)	Lycopene Content (ppm)	Ascorbic acid Content (ppm)	Total soluble solid (T.S.S.) (%)
Control	57.64±15.2 b	4.09±0.33 b	44.8±3.5 b	7.06±0.11 a
FA_3	36.36 ± 7.7 b	3.46 ± 0.31 b	31.5 ± 2.0 b	7.13 ± 0.11 a
FA_6	30.96 ± 10.7 b	3.55 ± 0.22 c	33.83 ± 4.04 b	7.13 ± 0.11 a
FA_9	41.24 ± 12 a	3.78 ± 0.35 a	33.25 ± 3.5 a	7.2 ± 0.2 a
15G	48.66 ± 18.8	3.32 ± 0.26	22.16 ± 2.0	7.15 ± 0.2

*Each value is a mean ±SD of six replicates.
Values followed by the same letter are not significantly different at p≥0.05.

On the other hand, the commercial formulation (15G) had the highest effect on the all quality control parameters, comparable to the CR formulations. These results are in disagreement with those reported by Baojaji and Mohajam (1978) who found that aldicarb had significant effect on each lycopene content and ascorbic acid content, but is in agreement with the results of the aldicarb in reducing β-Carotene content. From the above-mentioned results, it should be cited that the injury caused by the nematicide could be minimized by alteration the formulation type such as using the alginate controlled release formulation. Also among the alginate controlled release formulation, the FA_9 could be applied on tomato plant to control nematode with a minimum side effect on the quality control parameter such as the percentage of total soluble solids and ascorbic acid content of tomato juice. These parameters are very important in tomato storage period, tomato mature stage, nutrition value, palatability and the processing of different product from tomato juice.

These parameters are very important in tomato storage period, tomato mature stage, nutrition value, palatability and the process carried out of produce different products from tomato juice.

The effects of cadusafos released from the different formulations (CRFs and 10 G), which were uptaken from soil by tomato plant and translocated and reached to tomato fruits presented in Table (6). The statistical analysis of these data clearly indicated that there were significant differences in the reducing of β-Carotene, lycopene, and ascorbic acid content between the tomato treated with cadusafos formulations and untreated one. In opposite, there were significant increases in the percentage of T.S.S. for the tomato plant treated with the CR cadusafos formulations as compared with the granular formulation as well as with the control. Among the CRFs, no significant differences were found in β-carotene content and total soluble solid. In addition, no significant difference was found in the ascorbic acid content between FC_3 and FC_6. Also, there was significant difference in the ascorbic acid content between FC_9 and the other tested formulations. On the other hand, the commercial formulation (10G) of cadusafos had the lowest value in total soluble solid and lycopene content comparable to the CR formulations. There was a highly significant difference between CF_9 and the other formulations in the ascorbic acid content. From all the results mentioned previously, we could conclude that the effect of cadusafos on the quality parameters tested were based upon the release rate of cadusafos which in turn based upon the formulation types. It is well established that certain organophosphorus compounds influence the chemical composition of the plant fruits to which they are applied. The insecticide, dimethoate did not influence T.S.S. and sugars of peppers. (Shahin *et al.*, 1989).

Table 6. Effecte of Cadusafos Formulations Residue on some Quality Parameters of Tomato Fruits

Treatment	β-Carotene Content (ppm)	Lycopene Content (ppm)	Ascorbic acid Content (ppm)	Total soluble solid (T.S.S.) (%)
Control	57.64±15.2 b	4.09±0.33 a	44.8±3.5 a	7.06±0.11 b
FC_3	26.95 ± 13.7 b	3.47 ± 0.16 b	33.25 ± 3.5 b	7.33 ± 0.11 b
FC_6	38.7 ± 6.1 b	3.66 ± 0.25 b	35 ± 3.5 b	7.36 ± 0.11 b
FC_9	37.65 ± 2.4 a	3.64 ± 0.31 a	39.08 ± 7.0 a	7.35 ± 0.2 a
10G	49.76 ± 5.7	3.35 ± 0.12	33.25 ± 3.5	7.1 ± 0.2

*Each value is a mean ±SD of six replicates.
Values followed by the same letter are not significantly different at p≥0.05.

Profenofos residues increased T.S.S. and acidity, but decreased the glucose, protein and ascorbic acid content of tomatoes (Ismail *et al.*, 1993). Radwan *et al.*, (1995) reported that pirimiphos-methyl residues appeared to have significant effect on the total soluble sugars and ascorbic acid content of tomato fruits and broad bean seeds. Consequently to obtain good results in pest chemical control in modern agriculture, the environmental factors, e.g. contamination of the treated edible crops with cadusafos residue and also the possible effect on the quality and nutritional values should be carefully considered to avoid problems which may arise as results of side effect of cadusafos.

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الملخص العربي

الكفاءة الإبادية لمبيدي التيميك والكاديسفوس المجهزان في صورة حبيبات الاليجنيت المتحكم فى تحررهما ضد نيماتودا تعقد الجذور على نبات الطماطم تحت ظروف البيوت المحمية

حمدي سلطان وعبدالسلام مرعى واشرف موسى

تم تقييم النشاط الابادى لمبيدى الألديكارب (FA_3، FA_6 وFA_9) والكاديسفوس (FC_3، FC_6 وFC_9) المنسابان من تجهيزات الاليجنيت المتحكم فى تحرر المبيدان ضد نباتات تيماتودا تعقد الجذور على نباتات الطماطم المنزرعة فى اصص تحت ظروف البيوت المحمية. أشارت النتائج إلى أن جميع التجهيزات التى تم أختبارها ادت إلى خفض كبير في متوسط عدد العقد الجذرية وكتل البيض وكذلك يرقات النيماتودا فى التربة مقارنة مع الكونترول الغير معامل. من ناحية اخرى كانت تجهيزات الاليجنيت لكلا المبيدان اكثر فعالية نحو خفض متوسط عدد العقد وكتل البيض وكذلك يرقات النيماتودا فى التربة مقارنة بالتجهيزة التجارية المحببة. فقد أدت تجهيزات الاليجنيت لمبيد الألديكارب والكاديسفوس الى خفض متوسط عدد العقد الى (٩٥,١٪ -٩٦,٠٤٪) و(٨٧,١٪ - ٨٨,٠٣٪) لمبيد الألديكارب والكاديسفوس على التوالى. من ناحية أخرى، تم تسجيل أقل نسبة خفض العدد الإجمالي العقد بواسطة تجهيزات الألديكارب والكاديسفوس المحبب التجارية (٦١,٣٪ و٦٦,٧١٪) على التوالى. فى حين ان الحد الاقص لخفض عدد العقد على جذور نباتات الطماطم تم الحصول عليها عند تطبيق كلا من FA_3 (٨٦,٧٣٪) وFC_3 (٨٩,٠٨٪) بينما باقى تجهيزات الاليجنيت لكلا المبيدان تراوحت مابين ٥٩,٠٥٪ و٤٦,٢٣٪. ويجدر الاشارة ان الفعالية الابادية لتجهيزة FA_3 ضد نيماتودا تعقد الجذور كانت تقريبا ١,٧٧ ضعف التجهيزة التجارية بينما كانت تجهيزة FC_3 حوالى ١,٩٣ ضعف التجهيزة التجارية.

أظهرت النتائج المتحصل عليها لتجهيزات مبيدى الالديكارب والكاديسفوس الى حدوث انخفاض معنوى للبيتا كاروتين وصبغة اللايكوبين وحامض الاسكوربيك مقارنة بنباتات الطماطم الغير معاملة. كما لايوجد فروق معنوية فيما بين جميع تجهيزات مبيد الالديكارب والنبات الغير معاملة فى قيم المواد الصلبة الكلية الذائبة بيتما كانت هناك زيادة فى النسبة المئوية لتلك المواد عند معاملة نباتات الطماطم بمبيد الكاديسفوس المجهز فى صورة متحكم فى انسياب المبيد. ويمكن تطبيق اى من تجهيزات FA_3 وFC_3 على نبات الطماطم لمكافحة نيماتودا تعقد الجذور بأقل تأثير ضار على بعض المكونات الكيمائية لثمار الطماطم مثل صبغة اللايكوبين وحامض الاسكوربيك والمواد الصلبة الكلية الذائب مقارنة بالتجهيزات التجارية.