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EFFECT OF FORMULATED PLANT OILS APPLIED ALONE AND IN COMBINATIONS ON WHEAT LEAF RUST DISEASE AND GRAIN YIELD

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ABSTRACT

Antifungal activity of certain plant oils and some of their combinations on wheat leaf rust, *Puccinia recondita* f.sp. *tritici* was evaluated under field conditions. All treatments of the tested plant oils, applied alone or in pairs, showed significant reduction in leaf rust severity of wheat cultivar, Giza 160. Of the formulated essential oils applied singly, caraway oil and peppermint oil were the most effective at the rate of 0.8% in reducing leaf rust severity which ranged from 76.3-86.0% in the plots treated with caraway oil and 72.9-81.5% in plots treated with peppermint oil, respectively. The mixtures of caraway + jojopa oils, caraway + pepper tree oils and caraway + peppermint oils were the best combinations in controlling leaf rust disease which gave reduction in leaf rust severity ranged between 81.4% and 100% during the two growing seasons. Grain yield (ton/acre) was also increased by the application of plant oil combinations revealing an increase in grain yield ranged from 27.0-38.0% of caraway + jojopa oil combination, 25.7-37.5% of caraway + pepper tree oil combination and 25.9-34.8% of caraway + peppermint oil combination which, subsequently, led to a relative increase in 100-kernel weight. There were no significant differences were shown between Sumi-8 fungicide and the three oil mixtures concerning fungicidal field efficacy and improvement of grain yield. It was recommended that natural oils used and their combinations might be considered as an effective and safe method for the management program of wheat rust.

INTRODUCTION

Essential oils isolated from plants which consist of acyclic and monocyclic monoterpenes, have been shown to be effective agents against phytopathogenic fungi and bacteria (Miah *et al.*, 1990; Kang, 1992; Naigre *et al.*, 1996; Eldoksch and Abd El-Moity, 1997 and Hassanien and Eldoksch, 1997). Ypema and Gold (1999 indicated that biorational fungicides, those based on natural products and synthesized analogues of naturally occurring biochemicals, are more acceptable than conventional fungicides because of an assumed reputation for being environmentally safe and less hazardous to humans and other non-target organisms.

Leaf rust disease of wheat caused by the fungus, *Puccinia recondita* f. sp. *tritici* develops on leaves causing considerable losses in yield and grain quality (Abdel-Hak *et al.*, 1980 and Shafik *et al.*, 1992). Control of rust diseases is usually carried out using resistant varieties and application of synthetic fungicides (Hofle *et al.*, 1995 and Kolmer, 1995). Successful experiments have been made to manage wheat leaf rust disease using

natural products and biocontriol agents (Sajid *et al.*, 1995 and Eldoksch *et al.*, 2001). Several compound mixtures have been widely used in controlling economic pests including phytopathogenic fungi (Kubo and Tanignchi, 1988 and Gravanis *et al.*, 1998). The main purpose of mixing pesticidal compounds in one formulation or mixing more than one formulation at the moment of application is to increase biological activity and toxicity against target organisms infesting plant crops (Hewlett, 1960 and 1969; El-Sebae *et al.*, 1964, O'Brein, 1967 and Kang *et al.*, 1992).

The present research is devoted to study the antifungal activity of certain formulated essential oils when applied alone or in combinations against wheat leaf rust, *Puccinia recondita* f. sp. *tritici* under field conditions. The effect of these natural oils and their mixtures in comparison with Sumi-8 fungicide on wheat grain yield and 100-kernel weight was also evaluated.

MATERIALS AND METHODS

Plant materials used

Five oily plant extracts and their combinations were used for antifungal activity studies against wheat leaf rust fungus. The tested plant materials selected for this study are listed in Table (1) and includes Latin and Family names, part used and important constituents.

Table (1): Plant materials used and important constituents of extracted oils.

Plant material	Latin name	Family	Extracted part	Important constituents
Pepper tree	*Schinus terebinthifolius*	Anacardiaceae	Leaves	Terpenes; gallic acid (Lemk, 1992)
Caraway	*Carum carvi* L.	Umbelliferae	Fruits	Carvone; Limonene (Bouwmeester *et al.*, 1998)
Peppermint	*Mentha piperita* L.	Labiatae	Leaves	Menthol (Trease and Evans, 1985)
Parsley	*Petroselinum sativum*	Umbelliferae	Seeds	Apiole (Trease and Evans, 1985)
Jojopa	*Simmondsia chinensis*	Simmondsiaceae	Jojopa bean	Resins; Erucic acid (EPA, 1999)

The scientific names of different plant samples were confirmed by Department of Aromatic & Medicinal Plants, ARC, Alexandria. Jojopa oil was provided by a certified plant oil company. The fungicide Sumi-8, 5% EC (active ingredient, Diniconazole), Sumitomo Chemical Company, Japan, was used as a reference for comparison.

Preparation of formulated essential oils and their (1:1) mixtures

Samples (100-150 g) of dried plant materials were ground into fine powder and soaked for three days in 80% ethanol, filtered and concentrated by using rotary evaporator and then it was partitioned with petroleum ether (40-60). The petroleum ether extract was filtered, concentrated by evaporation to obtain the essential oil. Method of preparing emulsions of the extracted plant oils was followed (Eldoksch and Abd El-Moity, 1997) to

emulsify the oil material in water to be used for greenhouse and field application. Control treatments were treated only with water plus emulsifier. Mixtures of formulated oils were applied in pairs at (1:1) ratio.

Field experiments

Field trials on wheat plants were carried out at Alexandria University Experimental Station during 2000/2001 and 2001/2002 growing seasons, to evaluate the efficacy of some extracted plant oils applied singly or in pairs to control wheat leaf rust, *Paccinia recondita tritici* as well as their potential effects on grain yield and 100-kernel weight. The experiment was conducted in a randomized complete block design consisting of 11 treatments including the control and each treatment was replicated three times. The field experiment was divided into plots each 1/400 acre. Wheat cultivar Giza 160 was sown in November 22nd and 26th of the two successive growing seasons. Plots were surrounded by rust spreader border sown with a mixture of highly susceptible varieties to rust infection. Seeding rate was 60 kg/acre. All other agricultural practices were applied as recommended for wheat production. At the booting stage (Large, 1954), artificial rust inoculation was carried out according to Tarvet and Cassel (1951). The extracted plant oils and their combinations were applied one week after rust inoculation using a Solo back sprayer (mist blower) at the rate of 500 ml/plot (200 L/acre). Rust severity was determined on flag leaves of 10 plants randomly selected from each plot at the full rust development stage according to the modified Cobb's scale (Peterson *et al.*, 1948). At the harvest stage, grain yield as ton/acre on whole plot basis and weight (g) of 100 kernels were recorded. Treatment effectiveness was calculated according to the following equation:

$$\text{Treatment effectiveness } \% = \frac{B - A}{B} \times 100$$

where: A = rust severity in treatment.
B = rust severity in control.

The obtained results were statistically analyzed according to Snedecor and Cochran (1971).

RESULTS AND DISCUSSION

Data of the effect of extracted oils or their combinations on reducing wheat leaf rust during two successive growing seasons 2000/2001 and 2001/2002 are presented in Tables (2 and 3). The data showed that all treatments applied alone or in pairs significantly reduced rust disease severity on wheat cultivar Giza 160 throughout the two growing seasons. In 2000/2001 season, Table (2), results showed that, when oils applied singly, caraway oil and peppermint oil exhibited a significant inhibitory effect on leaf rust *Puccinia recondita* f. sp. *tritici* with 86.0 and 81.5% reduction in leaf rust severity, respectively, followed by pepper tree oil (74.4%) and then parsley oil

(69.0%). Mixture of caraway oil + pepper tree oil was the best combination against leaf rust disease and gave a complete protection revealing 100% reduction in rust disease incidence. Mixture of caraway oil + jojopa oil came next in its efficacy with 94.6% reduction in leaf rust severity without significant differences between their effects on rust disease, followed by caraway + peppermint, peppermint + parsley and then caraway + parsley with 84.2%, 76.8% and 75.2% reduction in leaf rust severity, respectively. Sumi-8 fungicide showed a high antifungal activity against leaf rust fungus giving about 95.7% reduction in leaf rust severity compared with the untreated control and the other treatments. Data also indicated that there was no significant differences in reducing leaf rust severity in plots treated with Sumi-8 fungicide and plots treated with the three oil mixtures of caraway + pepper tree oil, caraway + jojopa oil and caraway + peppermint oil. Results also indicated that grain yield (ton/acre) was significantly increased by the application of tested oils and their combinations. The formulated mixtures of caraway + pepper tree oils, caraway + jojopa oils and caraway + peppermint oils were the best treatments in increasing the grain yield and quality giving about 25.7%, 27.0% and 25.9% increase in grain yield, respectively. This subsequently led to a relative increase in 100 kernel weight (g) over the check by 8.1%, 9.5% and 5.0%, respectively.

Table (2): Effect of extracted oils applied alone or in pairs (1:1) on leaf rust, grain yield and 100-kernel weight of wheat cultivar Giza 160 in 2000/2001 season.

plant oils and mixtures	Application rate (ml/L)	Rust severity (%)	Relative reduction (%)	Grain yield (ton/acre)	Relative increase (%)	100-kernel weight (g)	Relative increase (%)
Pepper tree oil	7	16.2	74.4	3.88	26.3	4.42	5.4
Caraway oil	8	10.4	86.0	3.76	23.9	4.37	4.3
Peppermint oil	8	11.7	81.5	3.84	25.5	4.40	5.0
Parsley oil	8	19.6	69.0	3.36	14.9	4.32	3.2
Caraway + Peppermint	4 + 4	10.0	84.2	3.86	25.9	4.40	5.0
Caraway + Parsley	4 + 4	15.7	75.2	3.63	21.2	4.38	4.6
Caraway + Pepper tree	4 + 3.5	0.0	100	3.85	25.7	4.55	8.1
Peppermint + Parsley	4 + 4	14.7	76.8	3.49	18.0	4.36	4.1
Caraway + Jojopa	4 + 4	3.4	94.6	3.92	27.0	4.62	9.5
Fungicide (Sumi-8)	0.35	2.7	95.7	3.84	25.5	4.41	5.2
Control	-	63.3	0.0	2.86	0.0	4.18	0.0
$L.S.D_{0.05}$	-	14.6	-	0.59	-	0.17	-

These results are in accordance with those reported by Sajid *et al.*(1995), who found that neem oil extracted from *Azadirachta indica* completely inhibited germination of *Puccinia recondite* urediospores in the lab, but in the field neem oil at 4% checked leaf rust on wheat after four applications. They also found a significant improve in grain yield and quality. Also, Eldoksch *et al.*(2001) found that field application of jojopa oil, eucalyptus oil and caraway oil at 1% (10 ml/L) showed effective control against leaf rust disease on wheat which led to an improvement in grain yield and quality. In 2001/02 growing season, results indicated mostly the same trend as in 2000/01 growing season. All formulated oils applied singly exhibited reasonable control of rust disease on wheat crop with percentage reduction of rust severity which ranged between 78.8% in case of pepper tree

oil and 66.9% in case of parsley oil. The grain yield was also increased relative to the untreated control by about 31.7% of caraway oil formulation and 16.8% of parsley oil formulation Of the oil mixtures applied under field conditions, caraway + jojopa oil combination (1:1) was the best in reducing leaf rust disease with 91.7% reduction in rust severity followed by caraway plus pepper tree (90.6%), caraway plus peppermint (81.4%), peppermint plus parsley (79.4%) and then caraway plus parsley (76.8%). Results also indicated that grain yield (ton/acre) was significantly improved by the application of certain oil mixtures. The plots treated with caraway / jojopa oil mixture was the best in this respect giving about 38.0% increase in grain yield (3.68 ton/acre) and this subsequently led to an increase in 100 kernel weight (g) revealing about 10.7% increase relative to untreated control plots. Mixture of caraway + pepper tree oil came next in its efficacy causing increase in grain yield by about 37.5% (3.65 ton/acre), and the rest of applied mixtures gave also a significant grain yield increase ranged between 34.8% (3.50 ton/acre) in case of caraway + peppermint oil mixture and 20% (2.85 ton/acre) in case of peppermint + parsley oil mixture.

The data also revealed that there is an improvement in grain quality showing an increase of the 100-kernel weight (g) over the check as shown in Table (3) which indicated that the three mixtures of caraway oil with each of jojopa oil, pepper tree oil and peppermint oil were the best in this respect. On the other hand, Sumi-8 fungicide exhibited effective control against leaf rust disease causing 87.9% reduction in leaf rust severity and an increase in grain yield by about 33.7% (3.44 ton/acre) and also an increase in 100-kernel weight by 9.7%.

Table (3): Effect of extracted oils applied alone or in pairs (1:1) on leaf rust, grain yield and 100-kernel weight of wheat cultivar Giza 160 in 2001/2002 season.

plant oils and mixtures	Application rate (ml/L)	Rust severity (%)	Relative reduction (%)	Grain yield (ton/acre)	Relative increase (%)	100-kernel weight (g)	Relative increase (%)
Pepper tree oil	7	11.7	78.8	3.21	29.0	4.36	8.5
Caraway oil	8	13.1	76.3	3.34	31.7	4.38	8.9
Peppermint oil	8	15.0	72.9	2.99	23.7	4.45	10.3
Parsley oil	8	18.3	66.9	2.74	16.8	4.37	8.7
Caraway + Peppermint	4 + 4	10.3	81.4	3.50	34.8	4.44	10.1
Caraway + Parsley	4 + 4	12.8	76.8	3.20	28.7	4.31	7.4
Caraway + Pepper tree	4 + 3.5	5.2	90.6	3.65	37.5	4.45	10.3
Peppermint + Parsley	4 + 4	11.4	79.4	2.85	20.0	4.33	7.8
Caraway + Jojopa	4 + 4	4.6	91.7	3.68	38.0	4.47	10.7
Fungicide (Sumi-8)	0.35	6.7	87.9	3.44	33.7	4.42	9.7
Control	-	55.3	0.0	2.28	0.0	3.99	0.0
$L.S.D_{0.05}$	-	11.5	-	0.34	-	0.18	-

It is of interest to mention that certain plant oils when mixed at (1:1) ratio and applied in pairs under field conditions exhibited synergistic effect which led to a higher reduction in leaf rust severity than obtained when these oils were applied singly as shown in Tables (2 and 3). Increasing fungicidal activity in mixtures of compounds was reported by Gravanis *et al.*(1998), who found that a combination of garlic extract and flutriafol exhibited high protection of tomato plants infected with five pathogens. EPA (United States

Environmental Protection Agency, 1999) indicated that jojopa oil acts as a pesticide by forming a physical barrier between an insect pest or a phytopathogenic fungus and the leaf surface. On the other hand, caraway oil has proved to have fungicidal and bactericidal activity (Eldoksch and Abdel-Moity, 1997 and Hassanien and Eldoksch, 1997) and also exhibited an effect on wheat leaf rust disease (Eldoksch *et al.*, 2001). The combination of the two oils (caraway / jojopa oil mixture) showed a higher effect on leaf rust disease of wheat (Tables 2 and 3). This fungicidal activity enhancement may be due to the synergistic effect resulted in the joint action of the two oils in the combination. Increasing the fungicidal activity in tested oil mixtures is in accordance with that reported by Hewlett (1960), El-Sebae *et al.*(1964), O'Brein (1967) and Gravanis *et al.*(1998), who indicated that when certain compounds are jointly applied to living organisms, the potency of the combination might be greater than that expected from the potencies of the components if applied alone since synergism and potentiation might occur.

It could be concluded that the use of certain natural oils when applied in mixtures such as caraway/jojopa, caraway/pepper tree and caraway/peppermint oil mixtures could be accepted as an environmentally friendly green fungicides for managing wheat leaf rust disease and improve the grain yield and quality.

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التقييم الحقلى لتأثير بعض الزيوت النباتية المجهزة عندما تطبق منفردة وفى خلائط على صدأ أوراق القمح وتحسين محصول الحبوب

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تم دراسة التأثير المثبط ضد فطر صدأ أوراق القمح لبعض الزيوت النباتية المجهزة للتطبيق الحقلى وذلك عندما تطبق منفردة أو فى خلائط بنسبة (١ : ١) خلال موسمين متتاليين لمحصول القمح (٢٠٠٠/٢٠٠١ ، ٢٠٠١/٢٠٠٢).

ولقد إتضح من النتائج أن كل الزيوت النباتية المجهزة والمطبقة بمفردها أو فى أزواج قد سببت خفضا معنويا فى شدة الإصابة بصدأ أوراق القمح المتسبب عن الفطر بأكسينياريكونديتا تريتيسای ومن الزيوت النباتية المطبقة بمفردها وجد أن تجهيزة كل من زيت الكراوية وزيت النعناع الفلفلى قد أظهرا التأثير الواقى الأفضل عند التطبيق بمعدل ٠,٨% مسببا إنخفاض فى شدة صدأ الأوراق خلال موسمى النمو بمدى يتراوح مابين ٧٦,٣ ، ٨٦,٠% وذلك فى الأحواض المعاملة بتجهيزة زيت الكراوية وبإنخفاض يتراوح مابين ٧٢,٩% ، ٨١,٥% فى الأحواض المعاملة بتجهيزة زيت النعناع الفلفلى ثم تبعهما فى التأثير تجهيزة زيت شجر الفلفل بإنخفاض مابين ٧٤,٤ ، ٧٨,٨%

ولقد تبين من النتائج أن الخلائط المجهزة لمستخلص زيت الكراوية مع كل من زيت الجوجوبا، زيت شجر الفلفل وزيت النعناع الفلفلى قد أظهروا المكافحة والوقاية الأفضل ضد مرض صدأ أوراق القمح مع حدوث إنخفاض فى شدة الإصابة تتراوح مابين ٨١,٤% ، ١٠٠% خلال موسمى النمو لمحصول القمح.

ولقد إتضح من النتائج أيضا أن محصول الحبوب (طن/فدان) قد تحسن بتطبيق هذه الخلائط المجهزة مع حدوث زيادة فى محصول الحبوب تراوح مابين ٢٧ - ٣٨% فى الأحواض المرشوشة بالمخلوط (كراوية - جوجوبا) ومابين ٢٥,٧ - ٣٧,٥% فى الأحواض المرشوشة بالمخلوط الزيتى (كراوية + شجر الفلفل) ومابين ٢٥,٩ إلى ٣٤,٨% فى حالة الأحواض المرشوشة بالمخلوط الزيتى (كراوية + نعناع فلفلى)

ولقد أوضحت النتائج أيضا أن الخلائط الثلاثة قد أحدثت زيادة معنوية فى وزن ١٠٠ حبة قمح، ولم توجد أى فروق معنوية بين إستخدام المبيد الفطرى سومى-8 والخلائط الزيتية الثلاثة الواعدة من حيث الكفاءة الحقلية كمبيدات فطرية أو التأثير على زيادة محصول الحبوب.

وتوصى الدراسة بإمكانية إستخدام هذه الخلائط الزيتية المجهزة للرش الحقلى الوقائى فى برامج مكافحة أمراض صدأ القمح والتى قد تساعد فى تدعيم الإتجاه إلى تطبيق ونشر الزراعة العضوية.