The Response of Alate Diuraphis noxia (Kurdjumov) (Hemiptera: Aphididae) to Volatile Substances from Four Non-Host Plant Extracts Under Laboratory Conditions

Article *in* African Entomology · December 2014

DOI: 10.4001/003.022.0425

J.M. Richter
Plant Science, Entomology

Godfried Prinsloo
Agricultural Research Council, South Africa

The response of alate *Diuraphis noxia* (Kurdjumov) (Hemiptera: Aphididae) to volatile substances from four non-host plant extracts under laboratory conditions

J.M. Richter[1,3], G.J. Prinsloo[2*] & T.C. de K. Van Der Linde[3]

[1]*Department of Agriculture and Rural Development, Private Bag X01, Glen, 9360 South Africa*
[2]*Agricultural Research Council-Small Grain Institute, Private Bag X29, Bethlehem, 9700 South Africa*
[3]*Department of Zoology and Entomology, University of the Free State, P.O. Box 339, Bloemfontein, 9300 South Africa*

Volatile compounds present in essential plant oils are known to influence insect behaviour. They are generally of low molecular weight, highly volatile, have a unique mode of action and are sometimes of low toxicity to non-target organisms. One example of a volatile compound extracted from a plant is methyl salicylate which is being used to reduce aphid infestation on barley in some European countries. The use of such volatile compounds was therefore considered as an alternate control option to be included in the control programme of *Diuraphis noxia* (Kurd.) after plant resistance-breaking biotypes started to develop in South Africa. The aim of this study was to test the response of alate *D. noxia* to plant extracts from four plant species in a four-arm olfactometer. Aqueous and light mineral oil extracts of *Artemisia afra* (Jacq.), *Datura stramonium* (L.), *Tagetes minuta* (L.) and *Tulbaghia violacea* (Harv.), which grow naturally in some dryland wheat production regions of South Africa, were tested. These plants were chosen based on possible insect repelling properties known to occur in other species of the same genera. Aphids were strongly repelled by the oil extract of *T. violacea* and the aqueous extract of *A. afra*. The oil extract of *A. afra*, both *T. minuta* extracts and *T. violacea* aqueous extract were less repellent but still elicited an exceptional repelling response. Aphids were not repelled by *D. stramonium* extracts. The effectiveness of these basic extracts in repelling *D. noxia* should, however, be tested under field conditions before they can be recommended as a control option.

Key words: Russian wheat aphid, plant extracts, olfactometer tests, alternate control, aphid repellent.

INTRODUCTION

Plant resistance is a cornerstone of integrated management of the Russian wheat aphid (*Diuraphis noxia*) (Kurd.) (Homoptera: Aphididae) (Prinsloo *et al*. 2007). Biotypes capable of infesting resistant cultivars in South Africa were, however, recorded during 2005 (Jankielsohn 2012; Tolmay *et al*. 2007). This aphid, which is the most important wheat pest in the dryland wheat production areas of South Africa, therefore creates a new challenge to wheat production in the Free State Province of South Africa (Jankielsohn 2012). The utilization of broad spectrum insecticides that are harmful to natural enemies increased and novel alternatives should be added to the programme to reduce the use of these insecticides. Novel management strategies that could be used include insect repellents, attractants and antifeedants (Mann *et al*. 2012). Such strategies are even more important for emerging farmers who do not always have sufficient resources for chemical control interventions (Marasas *et al*. 1997).

Certain essential plant oils contain chemical compounds that affect insect behaviour. They are generally of low molecular weight, highly volatile, have a unique mode of action and can be of low toxicity to non-target organisms (Mann *et al*. 2012). If a strategy could be followed to reduce aphid infestation below economic or insecticide application thresholds, these compounds will exert much weaker selection for development of resistance by the pest compared to insecticides (Prinsloo *et al*. 2007). Since *D. noxia* infests wheat plants when they are small, the number of tillers, awns, spikes and seeds that are fixed during these plant growth stages are all affected. A logical control strategy would be to reduce early migration of *D. noxia* into the wheat crop.

It is known that methyl salicylate is released by several plant species upon damage by insects and

*Author for correspondence. E-mail: prinsloogj@arc.agric.za

it is also known as and inducer of plant defence against pathogens (Shulaev *et al*. 1997). Exposing cereal plants to this essential oil in the laboratory led to reduced aphid acceptance, and its application in the field reduced populations of the aphids *Rhopalosiphum padi* (L.) (Homoptera: Aphididae) and *Sitobion avenae* (F.) (Homoptera: Aphididae) in European cereals by up to 50 % (Pettersson *et al*. 1994; Glinwood & Pettersson 2000; Ninkovic *et al*. 2003). This aphid behaviour-modifying semiochemical, was also found to repel *D. noxia* in olfactometric tests in South Africa, but varying results were found during field trials indicating an interaction between semiochemicals and plant resistance (Prinsloo *et al*. 2007).

There are several plant species growing naturally in wheat production areas of the Free State Province that are also known as traditional medicinal plants (Van Wyk *et al*. 2000) which may have insect repelling properties. They are not known as host plants of *D. noxia* and it is hypothesized that some of these plant species could be used to repel migrating alate (winged) *D. noxia* from the wheat crop at an early stage. Four of these species, namely *Artemisia afra* (Jacq. ex Willd.) (Asteraceae), *Datura stramonium* (L.) (Solonaceae), *Tagetes minuta* (L.) (Asteraceae) and *Tulbaghia violacea* (Harv.) (Amaryllidaceae) are present in the wheat production regions (pers. obs). Possible insect repelling properties are known from other species in the same genus (Bruce *et al*. 2002; Isman 2006; Zehnder *et al*. 2007; Halbert *et al*. 2008; Işik & Görűr 2009; Van Wyk *et al*. 2000).

The objective of the study was therefore to investigate the behaviour-modifying effect of volatile substances emitted by plant extracts prepared from the above-mentioned four plant species.

MATERIAL AND METHODS

Plant material

Green leaves and stems of *A. afra*, *D. stramonium*, *T. minuta* and *T. violacea* were collected before flowering from plants growing naturally in the vicinity of Bethlehem, Free State Province (28°14′S 28°18′E), South Africa.

Plant extracts

Aqueous and light mineral oil (Citrole 100®) extracts were prepared from each plant species by finely chopping plant material and infusing 10 g with either 30 ml of boiling water or 30 ml of light mineral oil. The mixtures were covered with Parafilm and after 24 h at room temperature the extracts were filtered through glass wool. Extracts were stored in amber glass at 4 °C until needed.

Aphids

The Russian wheat aphid colony originated from collections made in cultivated wheat fields at ARC-Small Grain Institute, Bethlehem (28°06′S 28°18′E), South Africa. The aphid colony consisted of the original RWASA1 biotype and was started 10 years ago and is regularly supplemented by new aphids. Aphids were reared in cages (45 × 45 × 60 cm) in a greenhouse on the susceptible wheat cultivar, Betta, at 20 ± 2 °C and natural light conditions. Colony growth was allowed until alate formation. Alates which moved from the plants and gathered at the roof of the cage were collected and used in olfactometer trials.

Olfactometry

A four-arm olfactometer (Pettersson 1993) was used to test the response of aphids as described by Prinsloo *et al*. (2007). The tests were performed in a dark room with light provided by double fluorescent lamp tubes (20W/640S Cool White, Philips) suspended 50 cm directly above the olfactometer.

A single alate aphid was introduced into the olfactometer and observed for 10 min, during which the number of times an aphid entered and the time spent inside a treated and control arm zone were recorded using the computer programme OLFA (Olfa: Exeter Software New York, U.S.A.). Olfactometers and odour sources with charcoal filters were replaced after every five replications until 50 aphids were tested for each plant extract. The mean duration per entry (seconds) was calculated as: number of entries/total time spent in the arm. In addition the percentage difference between the control and treated arms the entry number and the entry duration was calculated for each replicate. This figure, indicates the magnitude by which the number of entries and time spent in the control arm differs from the treatment arm, and expresses the repellent response of the insect towards the volatile substance. For example, a substantial percentage difference between entries made into the control and treated arms meant that the aphid prefers the control arm and therefore is strongly repelled, while a small percentage indicates that the aphid is weakly repelled by the extract. If the difference resulted in a negative percentage it meant that the aphid preferred the treated arms above the control arms and was attracted to the extract.

Data analysis

Initially the mean number of entries into the arm zones and the mean duration of an entry between the treated and control arms were compared, using matched, paired *t*-tests at 5 % test level (Snedecor & Cochran 1967). To determine the extract with the highest repellence potential a one-way ANOVA and Fisher's protected least significant difference (LSD) test at 5 % level were then used to test between percentage differences in repellence by the respective plant species extracts (Genstat 14, VSN International 2013).

RESULTS

Separate extracts

Volatiles from the different plant extracts elicited an obvious response in alate *D. noxia*. These aphids made significantly fewer entries into olfactometer arms containing volatiles from both extract types of *A. afra, T. minuta* and *T. violacea* (Fig. 1) (*T. violacea* aqueous extract: $t = –4.8$, $P < 0.001$; *T. violacea* oil extract: $t = –4.8$, $P < 0.001$; *T. minuta* aqueous extract: $t = –21.1$, $P < 0.001$; *T. minuta* oil extract: $t = –10.7$, $P < 0.001$; *A. afra* aqueous extract: $t = –13.96$, $P < 0.001$; *A. afra* oil extract: $t = –12.68$, $P < 0.001$). The aphids made significantly fewer entries into the treatment arms of *D. stamonium* aqueous extract ($t = –3.69$, $P < 0.001$) but did not respond significantly to the oil extract of the same plant ($t = 0.44$, $P = 0.661$) (Fig. 1).

The mean duration of entries made into the treated olfactometer arms was also significantly shorter for *A. afra, T. minuta* and *T. violacea* (*T. violacea* aqueous extract: $t = –4.7$, $P < 0.001$; *T. violacea* oil extract: $t = –5.64$, $P < 0.001$; *T. minuta* aqueous extract: $t = –8.42$, $P < 0.001$; *T. minuta* oil extract: $t = –5.51$, $P < 0.001$; *A. afra* aqueous extract: $t = –6.92$, $P < 0.001$; *A. afra* oil extract: $t = –7.59$, $P < 0.001$) (Fig. 2). Because this calculated value is an indication of not only how many times the aphids entered the arms, but also the time spent in the arm, the mean duration of an entry is a better indication of the response of the aphids towards the volatiles present in the arms. The mean duration of an entry made into the treated arms of *D. stramonium* did not differ significantly from the duration in the control arms (aqueous extract: $t = –0.24$, $P = 0.814$; oil extract $t = –0.22$, $P = 0.825$) (Fig. 2). This shows that although the number of

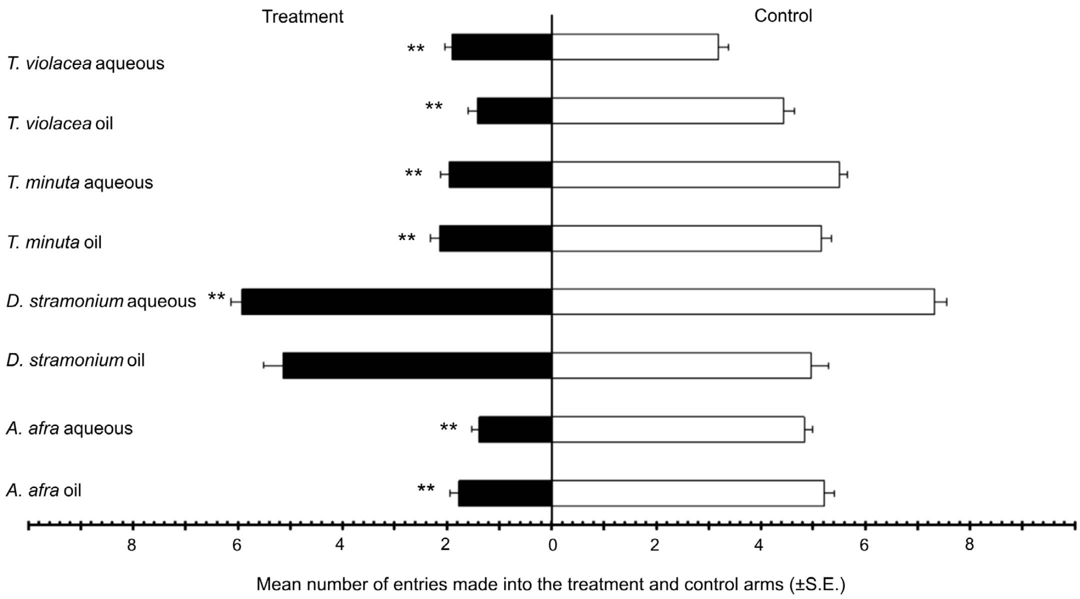

Fig. 1. Mean number of entries made by alate *Diuraphis noxia* into different olfactometer arms containing volatiles released by different extract types of four different plants (black bars) or volatiles from the control solutions. Black and white bars are compared with each other within an extract and bars followed by ** are significantly different from the white bars (paired *t*-test, $P < 0.001$).

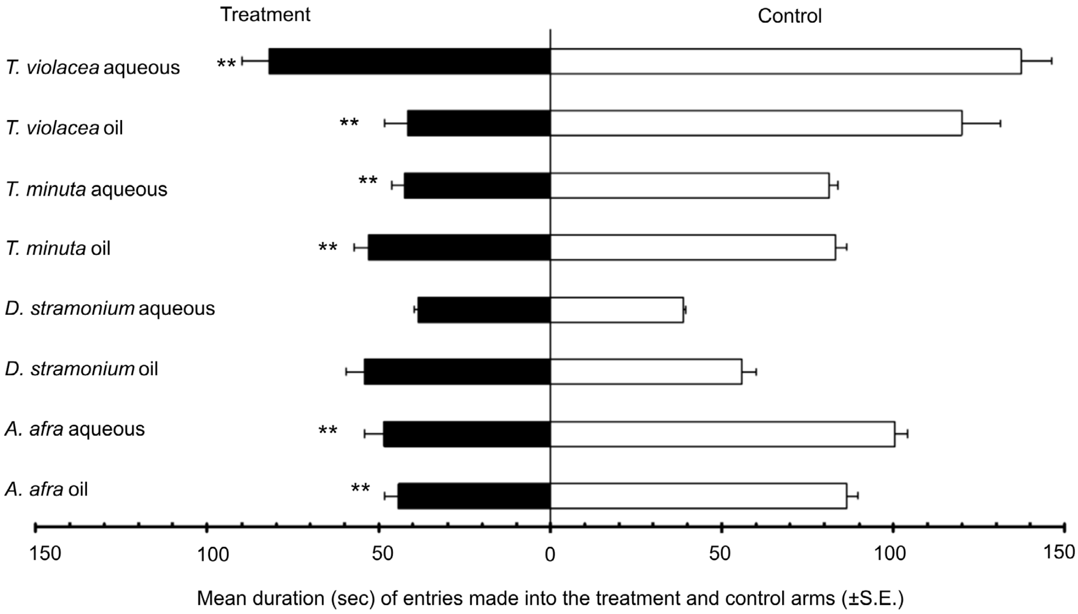

Fig. 2. Mean duration (s) of entries made by alate *D. noxia* into different olfactometer arms containing volatiles released by different extract types of four different plants (black bars) or volatiles from the control solutions. Black and white bars are compared with each other within an extract and bars followed by ** are significantly different from the white bars (paired *t*-test, $P < 0.001$; $n = 50$/extract).

entries indicates that the aqueous extract was repellent (Fig. 1), the duration of the entries in the different arms did not differ and aphids were therefore not repelled by the volatiles from these extracts.

Comparison of extracts

Based on the percentage differences between number of entries it was found that alate *D. noxia* were highly repelled by the *A. afra* aqueous and *T. violacea* oil extracts (Fig. 3) ($F = 27.52$, d.f = 7, 399, $P < 0.001$). This percentage difference was also not significantly lower in the *A. afra* oil and *T. minuta* aqueous extracts and therefore this four extracts all seem to be potentially effective repellents. The *T. violacea* and *D. stramonium* aqueous extracts were the least repellent, while the *D. stramonium* oil extract slightly attracted the aphids.

The percentage difference in duration of an entry indicates the overall response of the aphids was highly repelled by the *T. violacea* oil extract followed by *A. afra* aqueous extract (Fig. 4) ($F = 13.31$, d.f. = 7, 399, $P < 0.001$). The percentage difference in entries compared to the percentage difference in entry duration reveals that the oil extract of *A. afra* and aqueous extracts of *T. minuta* and *T. violacea* elicited a weaker repellent response (Fig. 4). The aphids did not respond well to both *D. stramonium* extracts, and these two could be rendered as non-repellent.

DISCUSSION

Plant resistance-breaking biotypes of *D. noxia* are a new challenge in the control of this pest, specifically in areas where emerging farmers with limited resources are operating. The release of odour-masking substances into the air by non-host plant species is considered to provide some protection to the associated crop host plants (Finch & Collier 2000). The authors demonstrated that host plant selection by the cabbage root fly was disrupted when its host plants were surrounded by a range of different plants, including weeds, peas, rye grass and clover (Finch & Collier 2000).

Alate *D. noxia* responded differently to the different plant extracts which may indicate differences in the solubility and volatility of the different compounds present in the plants. Two basic extraction types were utilized to render it user-friendly for the farmers. Volatile compounds such as 1,8-cineole, α- and β-thujone, borneol and camphor

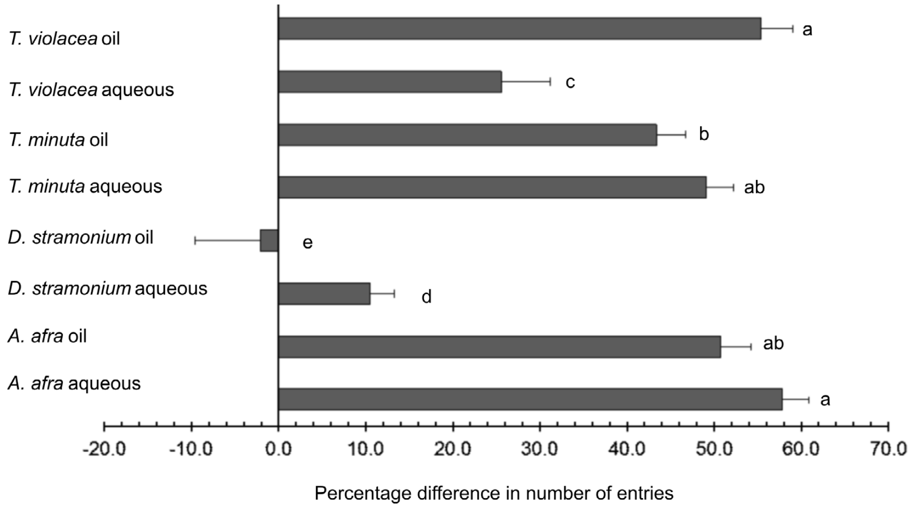

Fig. 3. Percentage differences between the number of entries made by alate *Diuraphis noxia* into the treated and control olfactometer arms. Bars followed by different letters indicate significant differences between extracts ($P < 0.001$).

are known from *A. afra*, while for *T. violacea* two unnamed volatile sulphur compounds cause the characteristic garlic smell (Van Wyk *et al*. 2000). Zhou *et al*. (2013) showed that intercropping with common garlic and the use of volatile compounds from garlic repelled *S. avenae* from wheat fields, which renders the identification of specific reactive compounds from *T. violacea* and *A. afra* important for testing against *D. noxia*. Although intercropping with these plants would not be a practical solution for emerging farmers, plant extracts and specific volatile compounds from these plants, could be used to mask volatile substance coming from the host plants and in that way keep aphids away from the crop. Therefore the response of *D. noxia* to each of these compounds should be investigated.

Although all studied plants grow naturally in the Free State Province, only *A. afra* and *T. violacea* are perennial and green material is available throughout the year. In the coldest parts of the Free State, *A. afra* plants may die back during winter, but the

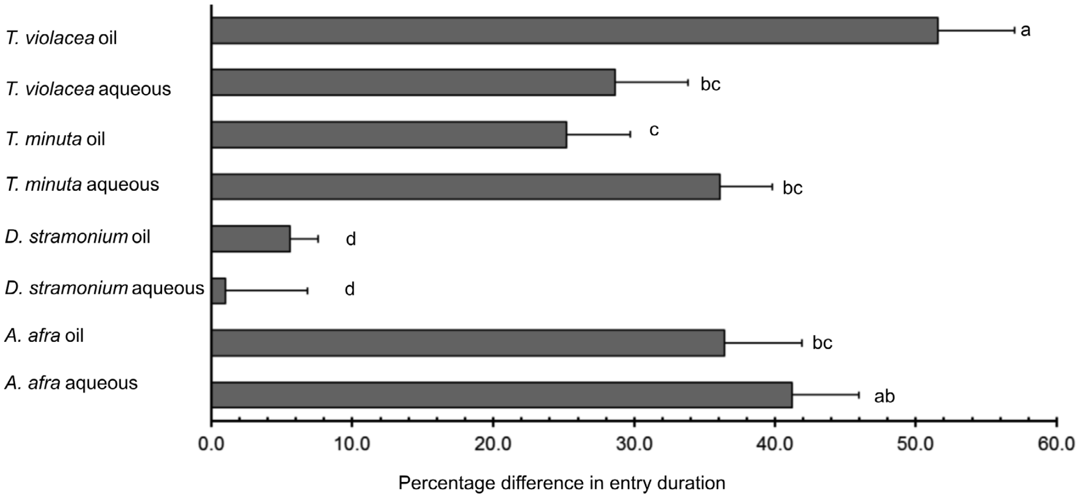

Fig. 4. Percentage differences between the mean duration of entries made by alate *Diuraphis noxia* into the treated and control olfactometer arms. Bars followed by different letters indicate significant differences between extracts ($P < 0.001$).

regrowth occurs rapidly afterwards (Van Wyk *et al*. 2000). *Tagetes minuta*, which also shows some potential as an aphid repellent is an annual, and green plant material is not available in winter. Plant material of several medicinal plants are usually collected and dried so that it could be stored for later use (Van Wyk *et al*. 2000). This method should also be investigated for *T. minuta* with plants collected in the previous summer. Since *D. noxia* is colonizing the wheat crop early in the season, plant extracts to mask odour from wheat plants should be prepared and applied soon after plant emergence (Aalbersberg *et al*. 1989). The identification of the key reactive compounds from these plant extracts could be helpful in the formulation of aphid repellent products that could be used shortly after planting when aphids are migrating into the fields.

The current study demonstrated that crude extracts from three of the tested plants have the potential to repel alate *D. noxia*. It is evident that *D. noxia* was stronger repelled by the *T. violacea* oil and *A. afra* aqueous extracts and that these could be recommended for use by farmers. In the second instance, the oil extract of *A. afra*, both *T. minuta* extracts and *T. violacea* aqueous extracts also elicited strong repellent responses and could also be used. The response to *T. violacea* oil extract was lower and would possibly not be as efficient as the others, while aphids did not respond to both extracts of *D. stramonium*. Although these substances show the potential to be used to minimize the migration of alate *D. noxia* into wheat fields they should be tested for efficacy, phytotoxicity and mammalian toxicity under field conditions before recommendations could be made to farmers on the application thereof.

ACKNOWLEDGEMENTS

We would like to thank P. Radebe and J. Kilian for assistance and the ARC-Small Grain Institute for using their facilities during the study.

REFERENCES

AALBERSBERG, Y.K., HEWITT, P.H. & VAN DER WESTHUIZEN, M.C. 1989. Characteristics of the population build-up of the Russian wheat aphid *Diuraphis noxia* and the effect on wheat yield in the eastern Orange Free State. *Annals of Applied Biology* **114**: 231–242.

BRUCE, T.J., CORK, A., HALL, D.R. & DUNKELBLUM, E. 2002. Laboratory and field evaluation of floral odours from African marigold, *Tagetes erecta*, and sweet pea, *Lathyrus odoratus*, as kairomones for the cotton bollworm *Helicoverpa armigera. International Organization of Biological Control Bulletin* **25**: 1–9.

FINCH, S. & COLLIER, R.H. 2000. Host-plant selection by insects: a theory based on 'appropriate/inappropriate landings' by pest insects of cruciferous plants. *Entomologia Experimentalis et Applicata* **96**: 91–102.

GLINWOOD, R.T. & PETTERSSON, J. 2000. Change in response of *Rhopalosiphum padi* spring migrants to the repellent winter host component methyl salicylate. *Entomologia Experimentalis et Applicata* **94**: 325–330.

HALBERT, S.E., CORSISNI, D., WIEBE, M. & VAUGHN, S.F. 2008. Plant-derived compounds and extracts potential as aphid repellents. *Annals of Applied Biology* **154**: 303–307.

IŞIK, M. & GÖRŰR, G. 2009. Aphidicidial activity of seven essential oils against the cabbage aphid, *Breviocoryne brassicae* L. (Hemiptera: Aphididae). *Munis Entomology and Zoology* **4**: 424–431.

ISMAN, M.B. 2006. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. *Annual Review of Entomology* **51**: 45–66.

JANKIELSOHN, A. 2012. Distribution of Russian wheat aphid (Hemiptera: Aphididae) biotypes in South Africa and Lesotho. *Journal of Economic Entomology* **104**: 1736–1741.

MANN, R.S., TIWARI, S., SMOOT, J.M., ROUSEFF, R.L. & STELINSKI, L.L. 2012. Repellency and toxicity of plant-based essential oils and their constituents against *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae). *Jounal of Applied Entomology* **136**: 87–96.

MARASAS, C., ANANDAJAYASEKERAM, P., TOLMAY, V., MARTELLA, D., PURCHASE, J. & PRINSLOO, G. 1997. *Socio-economic Impact of the Russian Wheat Aphid Control Research Program*. Southern African Centre for Cooperation in Agricultural and Natural Resources and Training, Gaborone, Botswana.

NINKOVIC, V., AHMED, E., GLINWOOD, R. & PETTERSSON, J. 2003. Effects of two types of semiochemicals on population development of the bird cherry oat aphid *Rhopalosiphum padi* in a barley crop. *Agricultural and Forest Entomology* **5**: 27–33.

PETTERSSON, J. 1993. Odour affecting autumn migration of *Rhopalosiphum padi* (L.) (Hemiptera: Homoptera). *Annals of Applied Biology* **122**: 417–425.

PETTERSSON, J., PICKETT, J.A., PYE, B.J., QUIROZ, A., SMART, L.E., WADHAMS, L.J. & WOODCOCK, C.M. 1994. Winter host component reduces colonisation by bird cherry-oat aphid, *Rhopalosiphum padi* (L.) (Homoptera, Aphididae), and other aphids in cereal fields. *Journal of Chemical Ecology* **20**: 2565–2574.

PRINSLOO, G.J., NINKOVIC, V., VAN DER LINDE, T.C., VAN DER WESTHUIZEN, A.J., PETTERSSON, J. & GLINWOOD, R. 2007. Test of semiochemicals and a resistant wheat variety for Russian wheat aphid management in South Africa. *Journal of Applied Entomology* **131**: 637–644.

SHULAEV, V., SILVERMAN, P. & RASKIN, I. 1997. Airborne signalling by methyl salicylate in plant pathogen resistance. *Nature* **385**: 718–721.

SNEDECOR, G.W. & COCHRAN, W.G. 1967. *Statistical Methods.* 6th Edn. Iowa State University Press, Ames, Iowa, U.S.A.

TOLMAY, V.L., LINDEQUE, R.C. & PRINSLOO, G.J. 2007. Preliminary evidence of a resistance-breaking biotype of the Russian wheat aphid, *Diuraphis noxia* (Kurdjumov) (Homoptera: Aphididae), in South Africa. *African Entomol*ogy **15**: 228–230.

VAN WYK, B.E., VAN OUTSHOORN, B. & GERICKE, N. 2000. *Medicinal Plants of South Africa*. Briza Publications, Pretoria, South Africa.

ZEHNDER, G., GURR, G.M., KÜHNE, S., WADE, M.R., WRATTEN, S.D. & WYSS, E. 2007. Arthropod pest management in organic crops. *Annual Review of Entomology* **52**: 57–80.

ZHOU, H.B., CHEN, J.L., LIU, Y., FRANCIS, F., HAUBRUGE, E., BRAGARD, C., SUN, J. & CHENG, D.F. 2013. Influence of garlic intercropping or active emitted volatiles in releasers on aphid and related beneficial in wheat fields in China. *Journal of Integrative Agriculture* **12**: 467–473.

Accepted 18 June 2014