JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Volatile Constituents of Fermented Sugar Baits and Their Attraction to Lepidopteran Species

A. M. El-Sayed,* V. J. Heppelthwaite, L. M. Manning, A. R. Gibb, and D. M. Suckling

HortResearch, Canterbury Research Centre, Lincoln, 8152, New Zealand

The volatile compounds emanating from four fermented sugar baits, palm sugar, golden cane syrup, port wine, and molasses, were isolated by headspace sampling and analyzed by gas chromatography−mass spectrometry. Three classes of compounds including esters, alcohols, and aromatic compounds were identified in the headspace of the four fermented sugar baits. There was a high degree of qualitative similarity between the headspace contents of the four fermented sugar baits, although quantitatively they varied considerably. Ethyl acetate, 3-methylbutanol, ethyl hexanoate, 2-phenylethanol, ethyl octanoate, ethyl (*E*)-4-decenoate, ethyl decanoate, and ethyl dodecanoate were the major compounds identified in the headspace of the four fermented sugar baits. The efficacy of the four fermented sugar baits was investigated in field trapping experiments. Fermented palm sugar and golden cane syrup were superior in attracting significant numbers of moths as compared to port wine and molasses. Fermented molasses was the least attractive among the four baits. Over 90% of the insects caught were noctuids with *Graphania mutans* and *Tmetolophota* spp. being the main noctuids captured (over 55%) in the four fermented sugar baits. Male and female *G. mutans* were equally attracted to the four sugar baits. A number of tortricid species were also trapped.

KEYWORDS: Fermentation; sugar baits; trapping; moths; esters; headspace; molasses; port wine; palm sugar; golden cane syrup

INTRODUCTION

Fermented sugar baits are well-known as attractants for many insect species (*1*−*5*). Early in the twentieth century, they were used for monitoring and detection of economically important lepidopteran pests (*4*, *6*, *7*). Noctuids, geometrids, tortricids, and pyralids are the major lepidopteran groups that are attracted to these types of baits (*1*−*3*, *5*). However, since the discovery of the first sex pheromone in 1959 (*8*) and the further identification of the sex pheromones of over 2500 lepidopteran species (*9*), the use as well as the research in the area of fermented sugar baits have significantly declined. While nearly all moth sex pheromones are species specific and attract only males, fermented sugar baits are generic and attract both males and females. Therefore, fermenting sugar baits provide an advantage over sex pheromones, because they can be used for targeting a wide range of insect pests. However, there are several disadvantages associated with fermented sugar baits, including the attraction of nontarget insects, mainly Hymenoptera, and generally lower attractiveness than pheromones. Also, they are inconvenient to use as monitoring tools, because the wet bait traps are heavy, maintenance is time consuming, and aging significantly affects attractiveness.

* To whom correspondence should be addressed. E-mail: AEl-Sayed@hortresearch.co.nz.

There are a significant number of sex attractants available to monitor male activity of many important insect pests. However, few attractants are commercially available to monitor female activity. Therefore, optimization of the fermented sugar baits, especially with regard to handling and preparation, would provide a new tool to monitor female activity of a range of economically important insect pests. Identification of the volatile compounds present in the headspace of the fermented sugar baits would represent an essential step toward transformation of these wet sugar baits into the more convenient dry baits that are easier to handle and prepare. Also, this might allow for the transformation of these baits from generalist to more specialist attractants, targeting only specific insect pests and thus minimizing the catch of nontarget species.

Numerous publications on the chemistry of the headspace of various fermented food products are available. The compounds identified are mainly alcohols, esters, carboxylic acids, ketones, and aldehydes (*10*−*14*). However, few studies have attempted to analyze the headspace of fermented sugar baits in correlation with insect attraction (*3*). In the first step toward isolation and identification of the active compounds in the headspace of fermented sugar baits, we have (i) investigated the efficacy of various fermented sugar baits for attracting insect pests and (ii) isolated and analyzed the headspace of fermented sugar baits using headspace sampling followed by gas chromatography−mass spectrometry (GC-MS) analysis.

10.1021/jf048521j CCC: $30.25 © 2005 American Chemical Society
Published on Web 01/25/2005

MATERIALS AND METHODS

Preparation of Fermented Sugar Baits. The fermented sugar baits prepared in this study included (i) port wine (Ormond, Rich Ruby, New Zealand); (ii) stock food grade molasses (Thick Black Strap, New Zealand); (iii) golden syrup (cane sugar and water, Chelsea Golden Syrup, New Zealand); and (iv) palm sugar (JHC, Thailand). The starter solution consisted of 1.65 g of brewer's yeast, *Saccharomyces cerevisiae* (Lavin 71B, New Zealand) mixed with 150 mL of water and 5 mL of regular white sugar and fermented overnight at 22 °C, before being added to each of the sugar bait solutions. A 150 mL aliquot of the starter solution was added to 1350 mL of the attractant solution and maintained for 3 days at 22 °C before being put out in the field. Three concentrations of each bait were prepared, i.e., 5 (75 mL of each bait plus 1.275 L of water), 10 (150 mL of each bait plus 1.2 L of water), and 20% (300 mL of each bait plus 1.05 L of water).

Headspace Collection System. A 500 mL aliquot of 6 day old fermented bait was placed in a 0.5 L bottle. The bottle was then confined in a 4 L glass cylinder that was tightly closed using ground glass fittings and a clamp. A charcoal-filtered air stream was pulled over the fermented sugar bait, and the headspace was collected on a trap containing 50 mg of Tenax-GR 35/60 (Alltech Associates Inc.) in a 15 mm × 5 mm glass tube. Before use, Tenax traps were thermally conditioned at 200 °C under a stream of nitrogen. The airflow in the headspace collection system was 2 L min^{-1}, and each collection session lasted for 1.2 h. Before use, the charcoal filter used to clean the incoming air was thermally activated using a lab oven at 200 °C. Immediately after volatile collection, each Tenax trap was extracted with 5 × 200 μL aliquots of hexane (Analar, BDH Laboratory Supplies, Poole, England), and 500 ng of benzyl acetate (Sigma Aldrich, United States, 98% chemical purity) was added as an internal standard. Sample volumes were reduced to 100 μL at ambient temperature under a stream of argon. Samples were sealed and stored at −80 °C.

GC-MS. The concentrated extracts of headspace from fermented sugar baits were analyzed using a Saturn 2200 GC-MS (Varian Inc., Walnut Creek, CA). The GC-MS system was equipped with a 30 m × 0.25 mm i.d. × 0.25 μm, VF5-MS capillary column (Varian Inc., CA). The oven was programmed from 40 (held for 2 min) to 240 °C at 4 °C min^{-1}. Samples were injected with the GC injector in the splitless mode at a temperature of 220 °C. The spectra were recorded at an ionization voltage of 70 eV over a mass range m/z of 20−499. The transfer line and the trap were held at 250 and 180 °C, respectively. Tentative structural assignments were made by comparing their mass spectra with the MS library (NIST 98), as well as by comparison with Kovats retention indices (*29*) published in the literature.

Field Trapping Experiments. The field trial was conducted at Lincoln Grange Orchard, Lincoln, New Zealand, in mature apple trees starting in December 2003. Green plastic 2 L Unitraps (International Pheromone Systems Ltd, Cheshire, United Kingdom) were filled with 500 mL of trapping solution. Each trap had an aluminum wire mesh held in place above the level of the liquid to keep moths from falling into the attractant solution. The mesh was held up by a wire frame 65 mm high. Traps were washed in a 2% detergent solution (Decon Laboratories Ltd., Sussex) and rinsed in clean water before use. Each trap had an insect kill strip (active ingredient 2,2-dichloroethenyl dimethyl phosphate, DDVP) fixed with a Velcro sticky dot two-thirds up from the bottom of the trap. A data logger (Gemini Data Loggers Limited, West Sussex) was placed in the orchard to record temperature information for the length of the trial period. Three replicates of each solution were arranged in a randomized complete design in the apple orchard. This was achieved by establishing three trap lines at about 20 m intervals in the orchard consisting of 14 "trapping stations" representing all concentrations and compositions of the four sweet baits, at about 20 m intervals in each trap line. Blank traps containing only water were used as the controls. Moths were removed from the traps three times a week and frozen in glass tubes for later identification. The field trial was run for a 3 month period from December 15, 2003 to February 23, 2004.

Statistics. The variance of mean captures obtained with each sugar bait were stabilized using the arcsine square root transformation of *x* and the significance of treatment effect tested using analysis of variance. Significantly different treatment means were identified using Fisher's protected least significant difference test (*15*). The Shannon−Wiener diversity index was calculated as an indicator of species richness for moths caught in each of the different sugar baits (*16*).

Table 1. Volatiles Compound Isolated from the Fermented Sugar Baits by Headspace Sampling

		ng/h			
compound[a]	Kovats[b]	golden syrup	palm sugar	port wine	molasses
ethyl acetate	608	2.31	1.58		1.67
3-methylbutanol	735	3.78	3.24	7.01	10.90
3-methylbutyl acetate	877	0.64	0.83	3.79	1.25
ethyl hexanoate	1001	9.24	4.80	1.11	4.14
ethyl heptanoate	1108	0.16	0.39		0.21
2-phenylethanol	1122	5.11	3.49	1.55	1.54
isophorone	1135		0.22		
ethyl octanoate	1205	116.57	74.81	12.33	52.45
ethyl (*Z*)-9-octenoate	1200		0.78		0.09
ethyl nonanoate	1297	1.42	1.59	0.18	0.99
butyl octanoate	1345	1.06	0.77	0.05	
ethyl (*E*)-4-decenoate	1388	38.84	30.93	0.78	4.86
ethyl decanoate	1397	254.39	82.42	3.32	61.74
3-methylbutyl octanoate	1444	4.10	2.56	0.12	1.35
ethyl dodecanoate	1593	50.77	17.20	0.40	6.87
isopropyl tetradecanoate	1059		1.06		
2-ethylhexyl 4-methoxy-cinnamate	1154		0.45		
3-methylbutyl hexanoate	1262	5.89			
2-phenylethyl acetate	1265	0.53			
ethyl undecanoate	1494	0.41			
ethyl hex-3-enoate	1003			4.44	
diethyl butanedioate	1191			0.61	
ethyl oct-3-enoate	1209			0.02	
2-phenylethyl dodecanoate	1631			0.08	
ethyl hexa-2,4-dienoate	1111			1.33	
4-ethylphenol	1178				0.23
methyl salicylate	1206				5.26
4-ethenyl-1,2-dimethoxy-benzene	1368				1.94
propyl dodecanoate	1626				0.46
2,3-dihydrobenzofuran	1226				4.98
1-ethenyl-4-methoxybenzene	1235				0.51
4-ethenyl-2-methoxyphenol	1313				1.09
ethyl tetradecanoate	1734		0.45		0.42
nonan-2-ol	1114				0.44
linalool	1112				0.39
1,2,3,4-tetrahydro-1,1,6-trimethylnaphthalene	1266				0.47
decyl acetate	1220				0.35
1-(1H-pyrrol-2-yl)-ethanone	1072				0.28
2-methylpropyl acetate	767			0.14	
benzyl alcohol	1043				0.38

[a] Compound identified by mass spectra correlation and Kovats index. [b] Kovats index on a DB-5 column.

RESULTS

Headspace Analysis. Chemical analysis of the headspace of the four fermented sugar baits is given in **Table 1**. It should be noted that quantification of the compounds was relative because the response of all compounds was considered the same as the internal standard and the identification was tentative because it was based on comparison with mass spectral data and Kovats indices published in the literature. Three prominent classes of compounds, esters, alcohols, and aromatic compounds, were identified in the headspace of the four sugar baits. Esters were the most prominent chemical class with numbers of carbons ranging from 2 to 15. Volatile compounds produced as a result of yeast anaerobic metabolism, in which the monosaccharide sugars will eventually break down and form esters with acids,

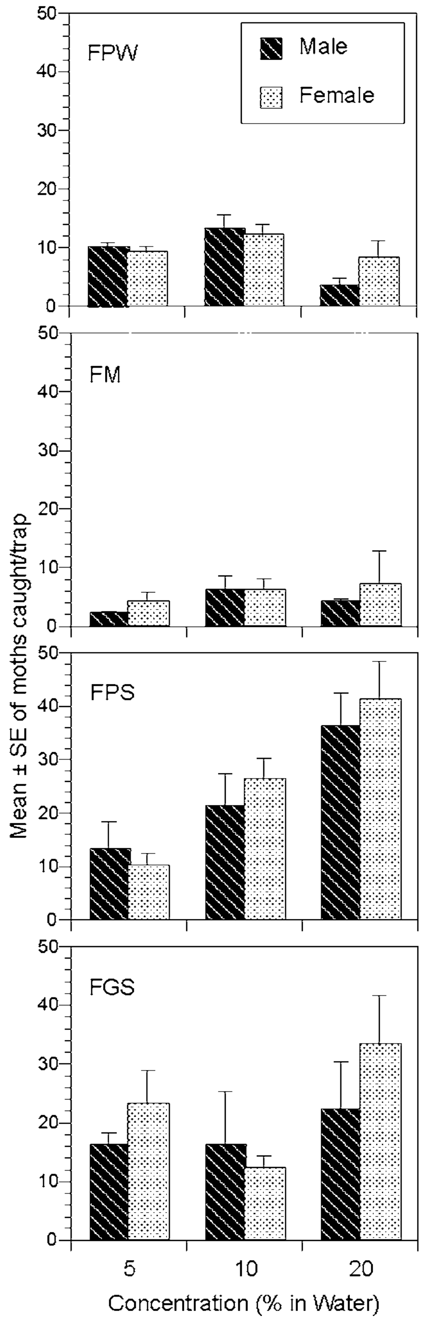

Figure 1. Trapping of male and female *G. mutans* in bucket traps baited with various concentration of fermented sugar baits. PW, fermented port wine; FM, fermented molasses; PS, fermented palm sugar; and GS, fermented golden cane syrup. Error bar indicates standard error.

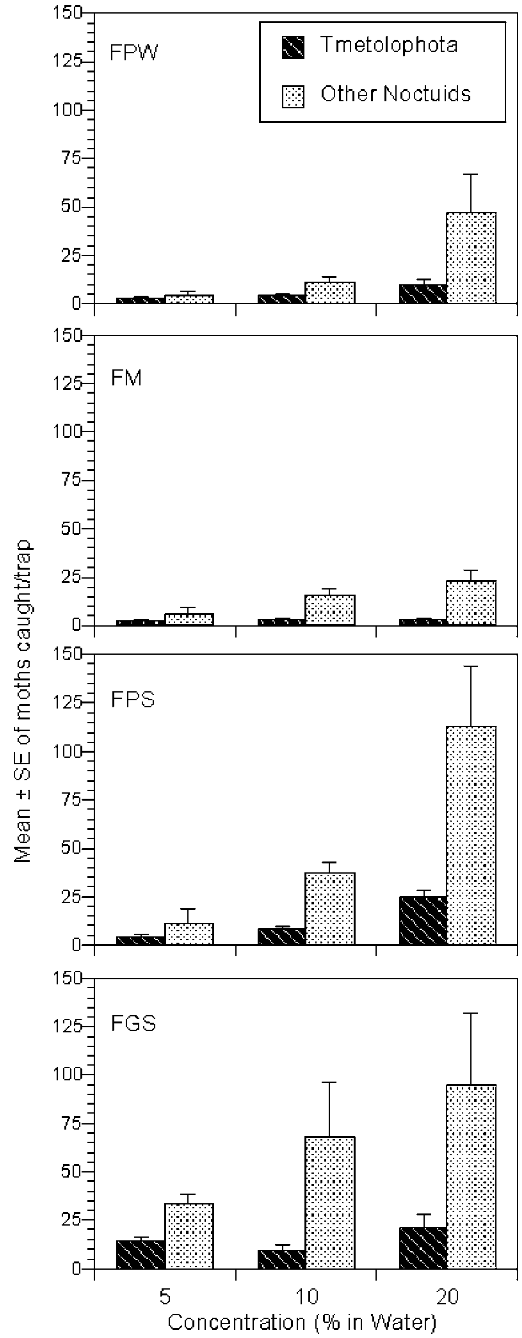

Figure 2. Trapping of *Tmetolophota* spp. and other noctuids in bucket traps baited with various concentrations of sugar baits. PW, fermented port wine; FM, fermented molasses; PS, fermented palm sugar; and GS, fermented golden cane syrup. Error bar indicates standard error.

ethers with alcohols, and acetals with aldehydes and ketones, may in turn serve as substrates for further fermentation. Among the four sugar baits, molasses yielded the highest number of volatile compounds, and it was rich with aromatic compounds. Eight volatiles [including ethyl acetate, 3-methylbutanol, 2-phenylethanol, ethyl hexanoate, ethyl octanoate, ethyl (*E*)-4-decenoate, ethyl decanoate, and ethyl dodecanoate] were the major compounds identified in the headspace of the four fermented sugar baits. Fermented palm sugar and golden syrup release considerably more esters [i.e., ethyl hexanoate, ethyl octanoate, ethyl (*E*)-4-decenoate, and ethyl decanoate] than port wine and molasses (**Table 1**). Several esters including ethyl hex-3-enoate, diethyl butanedioate, ethyl oct-3-enoate, 2-phenylethyl dodecanoate, and ethyl hexa-2,4-dienoate were identified only in the headspace of the fermented port wine. Other esters including 2-phenylethyl acetate, propyl dodecanoate, ethyl tetradecanoate, and decyl acetate were identified only in the headspace of the fermented molasses.

Fermented Sugar Baits Efficacy vs Concentration. The efficacy of the fermented sugar baits varied significantly with changing concentration of the media (**Figures 1−3**). For instance, catches with the fermented port wine and molasses were less affected by changing concentration as compared to fermented palm sugar and golden syrup. Increasing the concentration of the fermented port wine from 5 to 20% did not affect the number of male or female *Graphania mutans* and tortricids caught ($p > 0.2$) (**Figures 1** and **3**), while the number of other noctuids including *Tmetolophota* spp. and other non noctuids species significantly increased ($p < 0.01$) (**Figures**

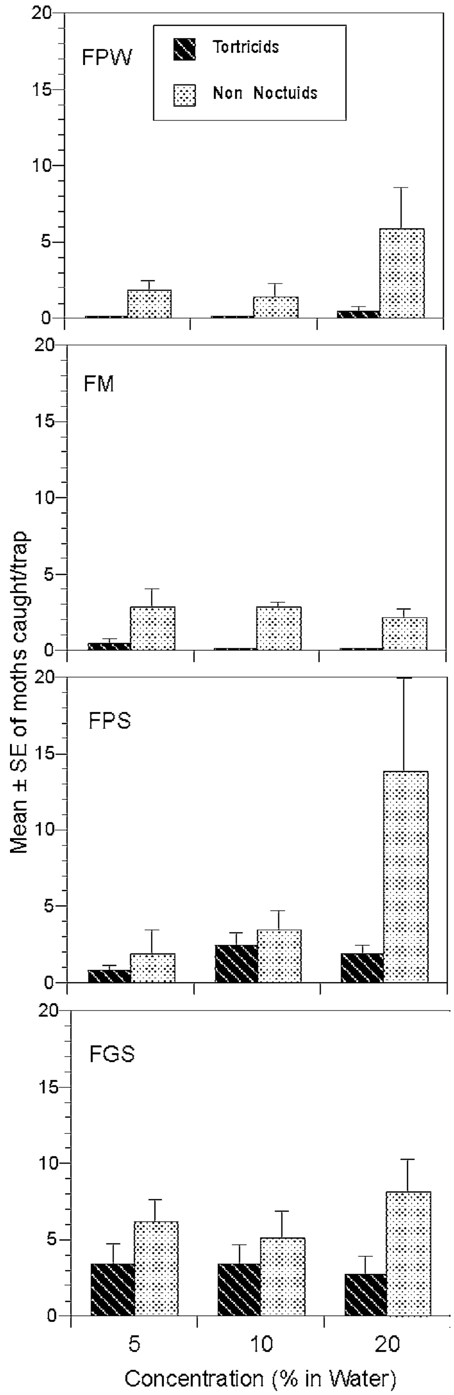

Figure 3. Trapping of tortricids and nonnoctuids in bucket traps baited with various concentrations of fermented sugar baits. PW, fermented port wine; FM, fermented molasses; PS, fermented palm sugar; and GS, fermented golden cane syrup. Error bar indicates standard error.

2 and **3**). With fermented molasses, increasing the concentration had no affect on the number of male or female *G. mutans*, *Tmetolophota* spp., and tortricids caught ($p > 0.3$) (**Figures 1−3**), while the number of other noctuids significantly increased ($p < 0.05$) (**Figure 2**). Increasing the concentration of the fermented palm sugar caused linear and significant increases in the number of moths caught except for tortricids ($p < 0.01$) (**Figures 1−3**). With fermented golden syrup, there was no significant increase in the number of male or female *G. mutans*, *Tmetolophota* spp., non noctuids, and tortricids caught ($p > 0.1$) (**Figures 1−3**). The number of other noctuids significantly increased as the concentration of the fermented golden syrup increased ($p > 0.05$) (**Figure 2**).

Fermented Sugar Baits Efficacy vs Composition. Efficacy of the fermented sweet baits varied significantly with composition of the media used ($p < 0.01$) (**Figures 1−3**). Fermented palm sugar and golden syrup were the most attractive baits among the four media tested ($p < 0.01$). Fermented port wine showed a moderate degree of attraction, while fermented molasses was the least attractive bait. The ratio of male to female *G. mutans* caught with various sugar baits was similar (**Figure 3**). Although the catches varied with baits composition, the diversity measure for species richness caught in each of the sugar baits, as indicated by Shannon−Wiener index, was similar (1.33 for port wine, 1.29 for golden syrup, 1.24 for molasses, and 1.27 for palm sugar).

DISCUSSION

Chemical analysis of the four different fermented baits revealed that most of the compounds present in the headspace were esters (**Table 1**). In a recent study, ethyl hexanoate, ethyl decanoate, ethyl dodecanoate, and 3-methylbutanol were identified in the headspace of Brazilian sugar cane spirit (*12*, *17*). Esters formed as a result of sugar decomposition during the fermentation process are likely to undergo further decomposition and produce alcohols and acetic acid. Other minor classes of compounds were identified including ketones, alcohols, and aromatic compounds. In this study, baits were allowed 3 days for fermentation before being taken to the field for 1 week. Headspace analysis was conducted within this period; therefore, the attractiveness of the bait is strongly correlated with the compounds identified in the headspace. It is striking that many of the compounds identified in the headspace of the fermented sugar baits are known as insect attractants or pheromone compounds (*9*). For example, 3-methylbutanol is known as an attractant to many noctuid species (*4*, *18−21*). Ethyl acetate has been identified as the male produced pheromone in the Mediterranean fruit fly, *Ceratitis capitata* (*22*). Ethyl hexanoate has been identified as the sex pheromone of the Deathshead cockroach, *Blaberus craniifer* (*23*). 2-Phenylethanol has been identified as the male sex pheromone for the Flounced chestnut, *Agrochola helvola* (*24*). Ethyl octanoate is an attractant for the Greater wax moth, *Galleria mellonella* (*25*). Methyl salicylate has been identified as an attractant for many species including the Western flower thrips, *Frankliniella occidentalis* (*26*). 2-Ethylphenol has been identified as an alarm pheromone of the Four spot roach, *Eublaberus distanti* (*27*).

Although the four fermented sugar baits have a very similar headspace profile, the quantities of the compounds identified varied considerably among baits. In the field trapping experiments, there were significant differences in the attractive efficacy between baits. Molasses and port wine were the least attractive, while palm sugar and golden syrup were the most attractive. On the basis of the headspace analysis, both palm sugar and golden syrup release more esters as compared to port wine and molasses. Therefore, it is likely that these esters account for the greater trapping efficacy of palm sugar and golden syrup as compared to port wine and molasses. In addition, the headspace of molasses was rich with aromatic compounds. Aromatic compounds are not commonly found to be kairomone or pheromone compounds for Lepidopteran species (*9*), and it is likely that these compounds might antagonize the attraction to molasses, thus reducing its efficacy. In a previous study, palm sugar was found to be more attractive for many lepidopteran species when compared to molasses (*2*). For most of the species caught, there was a direct correlation between the concentration of the media and the number of moths caught. Similarly, increasing the concentration of the palm sugar from 5 to 20% resulted in a significant increase in the number of *Mocis latipes* caught (*2*). Preliminary GC-MS analysis of the headspace from various concentrations of each bait indicates that increasing the concentration results in a significant increase in the quantity of the compounds, rather than any qualitative change in the headspace (El-Sayed et al., unpublished data).

Over 90% of the species caught in the fermented sugar baits were noctuids; similarly, noctuids were the major species caught in earlier experiments utilizing fermented sugar baits (*1*, *2*). *G. mutans* was the main species caught in the four fermented baits in our trials, while in North America, Tobacco budworm moth, *Heliothis virescens*, *M. latipes*, and the corn earworm, *Heliothis zea*, were the main noctuids species caught in fermented sugar baits (*1*, *2*, *6*). Because of the large number of moths caught in this study, we have determined the sex ratio for only *G. mutans* where an equal number of males and females were caught in each of the four fermented sugar baits. Similarly, the sex ratio for other noctuids (i.e., *H. virescens* and *M. latipes*) caught in fermented sugar baits was one to one (*1*, *2*). The catches of both male and female moths clearly indicate that attraction to these types of baits is mainly a feeding response. *G. mutans* is a native pest in New Zealand and of economical importance mainly in apple orchards. The sex pheromone has been identified for this species (*28*) and is used to monitor male activity. However, catches of both males and females in the fermented sugar baits open the opportunity for developing an attractant that targets both males and females of this pest. Initial field tests using 3-methylbutanol and acetic acid caught male and female *G. mutans* but in small numbers as compared to the number caught using fermented sugar baits traps (El-Sayed et al., unpublished data). Therefore, it is likely that GC-EAD and behavioral experiments will be required for identification of further active compounds present in the headspace of the fermented sugar baits that might enhance the efficacy of the dry lure, e.g., rubber septa or polyethylene vials releasing the attractive blend of volatiles.

ACKNOWLEDGMENT

Thanks to three anonymous reviewers for helpful comments on an earlier draft of the manuscript.

LITERATURE CITED

(1) Landolt, P. J.; Mitchell, E. R. Attraction of tobacco budworm moths (Lepidoptera: Noctuidae) to Jaggery, a palm sugar extract. *Florida Entomol*. **1997**, *80*, 402−407.

(2) Landolt, P. J. Attraction of *Mocis latipes* (Lepidoptera: Noctuidae) to sweet baits in traps. *Florida Entomol*. **1995**, *78*, 523−530.

(3) Utrio, P.; Eriksson, K. Volatile fermentation products as attractants for Macrolepidoptera. *Ann. Zool. Fenn*. **1977**, *14*, 98−104.

(4) Eyer, J. R.; Medler, J. T. Attractiveness to codling moth of substances related to those elaborated by heterofermentative bacteria in baits. *J. Econ. Entomol*. **1940**, *33*, 933−940.

(5) Suckling, D. M.; Thomas, W. P.; Burnip, G. M.; Robson, A. Monitoring lepidopterous pests at two Canterbury orchards. *Proc. 43rd N. Z. Weed Pest Control Conf*. **1990**, 322−327.

(6) Ditman, L. P.; Cory, E. N. The response of corn earworm moths to various sugar solutions. *J. Econ. Entomol*. **1933**, *26*, 109−115.

(7) Götz, B. Der sexualduftstoff als bekämpfungsmittel gegen die traubenwickler im *Freiland. Weinrebe*. **1941**, *4*, 75−89.

(8) Butenandt, von A.; Beckmann, R.; Stamm, D.; Hecker, E. Über den sexual-lockstoff des seidenspinners *Bombyx mori*. Reindarstellung und konstitution. *Z. Naturforsch*. **1959**, *14b*, 283−284.

(9) El-Sayed, A. M. The Pherobase: Database of insect pheromones and semiochemicals. http://www.pherobase.com, 2004.

(10) Patel, S.; Shibamoto, T. Effect of different strains of *Saccaromyces cerevisiae* on production of volatiles in Napa Gamay wine and Petite Sirah wine. *J. Agric. Food Chem*. **2002**, *50*, 5649−5653.

(11) Fu, S.-G.; Yoon, Y.; Bazemore, R. Aroma-active components in fermented Bamboo shoots. *J. Agric. Food Chem*. **2002**, *50*, 549−554.

(12) Nonato, E. A.; Carazza, F.; Silva, F. C.; Carvalho, C. R.; de Cardeal, Z. A headspace solid-phase microextraction method for the determination of some secondary compounds of Brazilian sugar cane spirits by gas chromatography. *J. Agric. Food Chem*. **2001**, *49*, 3533−3539.

(13) DeMilo, A. B.; Lee, C.-J.; Moreno, D. S.; Martinez, A. J. Identification of volatiles derived from *Citrobacter freundii* fermentation of a Trypticase Soy broth. *J. Agric. Food Chem*. **1996**, *44*, 607−612.

(14) Lee, C.-J.; DeMilo, A. B.; Moreno, D. S.; Mangan, R. L. Identification of the volatile components of E802 Mazoferm steepwater, a condensed fermented corn extractive highly attractive to the Mexican fruit fly (Diptera: Tephritidae). *J. Agric. Food Chem*. **1997**, *45*, 2327−2331.

(15) SAS Institute Inc. *Statview*; SAS Institute Inc.: Cary, NC, 1998.

(16) Zar, J. H. *Biostatistical Analysis*, 3rd ed.; Prentice Hall: New Jersey, 1996.

(17) Carneiro, I.; Nobrega, C. The analysis of volatile compounds from Brazilian sugar cane spirit by dynamic headspace concentration and gas chromatography−mass spectrometry. *Cienc. Tecnol. Aliment.* **2003**, *23*, 210−216.

(18) Landolt, P. J.; Higbee, B. S. Both sexes of the true armyworm (Lepidoptera: Noctuidae) trapped with the feeding attractant composed of acetic acid and 3-methyl-1-butanol. *Florida Entomol*. **2002**, *85*, 182−185.

(19) Landolt, P. J.; Alfaro, J. F. Trapping *Lacanobia subjuncta*, *Xestia c-nigrum*, and *Mamestra configurata* (Lepidoptera: Noctuidae) with acetic acid and 3-methyl-1-butanol in controlled release dispensers. *Environ. Entomol*. **2001**, *30*, 656−662.

(20) Landolt, P. J. New chemical attractants for trapping *Lacanobia subjuncta*, *Mamestra configurata*, and *Xestia c-nigrum*, (Lepidoptera: Noctuidae). *J. Econ. Entomol*. **2000**, *93*, 101−106.

(21) Landolt, P. J.; Hammond, P. C. Species composition of moths captured in traps baited with acetic acid and 3-methyl-1-butanol. *J. Lepidop. Soc*. **2001**, *55*, 53−58.

(22) Jang, E. B.; Light, D. M.; Binder, R. G.; Flath, R. A.; Carvalho, L. A. Attraction of female Mediterranean fruit flies to the five major components of male-produced pheromone in a laboratory flight tunnel. *J. Chem. Ecol*. **1994**, *20*, 9−20.

(23) Brossut, R.; Dubois, P.; Rigaud, J. Aggregation in *Blaberus craniifer*: Isolation and identification of the pheromone. *J. Insect Physiol*. **1974**, *20*, 529−543.

(24) Bestmann, H. J.; Vostrowsky, O.; Platz, H. Pheromone XII. Male sex pheromones of noctuids. (in Germany). *Experientia* **1977**, *33*, 874−875.

(25) Turker, L.; Togan, I.; Ergezen, S.; Ozer, M. Novel attractants of *Galleria mellonella* L (Lepidoptera Pyralidae Galleriinae). *Apidologie* **1993**, *24*, 425−430.

(26) Chermenskaya, T. D.; Burov, V. N.; Maniar, S. P.; Pow, E. M.; Roditakis, N.; Selytskaya, O. G.; Shamshev, I. V.; Wadhams, L. J.; Woodcock, C. M. Behavioral responses of western flower thrips, *Frankliniella occidentalis* (Pergande), to volatiles from three aromatic plants. *Insect Sci. Appl*. **2001**, *21*, 67−72.

(27) Brossut, R. Allomonal secretions in cockroaches. *J. Chem. Ecol*. **1983**, *9*, 143−158.

(28) Frérot, B.; Foster, S. P. Sex pheromone evidence for two distinct taxa within Graphania mutans (Walker). *J. Chem. Ecol*. **1991**, *17*, 2077−2093.
(29) Robards, K.; Haddad, P. R.; Jackson, P. E. *Principles and Practice of Modern Chromatographic Methods*; Academic Press: New York, 1994.

Received for review September 5, 2004. Revised manuscript received November 29, 2004. Accepted December 3, 2004. This study was funded by The Foundation for Research, Science & Technology (FRST), New Zealand.

JF048521J