CSIRO PUBLISHING

COMMUNICATION
https://doi.org/10.1071/CH22097

AUSTRALIAN JOURNAL OF CHEMISTRY

Floral constituents of the Australian tar tree, *Semecarpus australiensis*

Soo Jean Park[A,B,*], Jodie Cheesman[C], Donald N. S. Cameron[A,B], Stefano G. De Faveri[C] and Phillip W. Taylor[A,B]

For full list of author affiliations and declarations see end of paper

***Correspondence to:**
Soo Jean Park
Applied BioSciences, Macquarie University, Sydney, 2109, Australia
Email: soojean.park@mq.edu.au

Handling Editor:
Charlotte Williams

ABSTRACT

Floral constituents of the Australian tar tree, *Semecarpus australiensis*, distributed in Melanesia and Northern Australia, were extracted with solvent, and analyzed by gas chromatography-mass spectrometry. The main constituents were 16- and 18-carbon fatty acids and their ethyl esters. Amongst the 67 identified compounds, zingerone was detected in minute quantity, providing the chemical basis for previous observations of fruit fly attraction to the flowers. The present study is the first to report the chemical profile of tar tree flowers.

Keywords: fatty acids, floral volatiles, fruit fly, GC-MS, isoeugenol, native cashew, salicylates, Tephritidae.

Introduction

The flowers of many orchid species of the genus *Bulbophyllum* contain phenylpropanoids or phenylbutanoids that attract male *Bactrocera* and *Zeugodacus* fruit flies (Diptera, Tephritidae).[1] Their most common responses are to raspberry ketone and methyl eugenol, although many species do not respond to either of these compounds or respond only weakly.[2] More recent studies have found that some *Bactrocera* and *Zeugodacus* species respond to zingerone,[1a,1d,3] which occurs as a floral scent in *Bu. patens* and *Bu. baileyi*.[1a,1b,1d] In addition, isoeugenol and methyl isoeugenol have been found to attract some species that are non-responsive or weakly responsive to raspberry ketone and methyl eugenol.[3c,3d,4] Isoeugenol and methyl isoeugenol occur in the essential oils of various plant taxa, including different species of *Citrus, Clusia, Pimenta*, and *Etlingera*.[5] Reasons for the attraction of male fruit flies to phenylpropanoids and phenylbutanoids are incompletely understood,[6] and vary across compounds and fruit fly species. For example, males of some species of *Bactrocera* acquire methyl eugenol as a pheromone precursor[7] or as a metabolic enhancer to increase mating competitiveness.[8] In return, *Bulbophyllum* orchids benefit through pollination,[1a,1b,1f] hence the interaction between the orchids and fruit flies is mutually beneficial.

In an evolutionarily unrelated interaction, the flowers of *Passiflora maliformis* release zingerone from their filaments and attract *Bactrocera jarvisi*.[9] Earlier records report that *B. jarvisi* is also attracted to flowers of the tar tree, also known as native cashew, *Semecarpus australiensis*, as well as to *Bu. baileyi*.[10] Zingerone in *Bu. baileyi* is responsible for attraction of *B. jarvisi*.[10b]

The tar tree grows naturally in rainforests, including the northeast part of Queensland and Northern Territory in Australia, and across a wide range in Melanesia including Torres Strait Islands, New Guinea, New Britain, and Aru Islands.[11] The tar tree produces cream-colored flowers in spring. Staminate (male) flowers, about 1.5 mm long, are sessile, while pistillate (female) flowers, about 4 mm long, are on pedicels. To date, the chemistry of tar tree flowers is unknown. The present study aims to (1) characterize the compounds in the tar tree flowers and (2) confirm the chemical basis of previous observations on the attraction of *B. jarvisi* males. The present study describes constituents in the solvent extracts of both staminate and pistillate tar tree flowers analyzed by

Received: 27 April 2022
Accepted: 22 September 2022
Published: 23 November 2022

Cite this:
Park SJ *et al.* (2022)
Australian Journal of Chemistry
75(12), 945–952. doi:10.1071/CH22097

gas chromatography-mass spectrometry (GC-MS) and identifies the chemical basis for the attraction of fruit flies to these flowers.

Results and discussion

Extracts of tar tree flowers contained a diverse suite of 67 compounds, including 11 aliphatic acids, 13 aliphatic esters, 2 aliphatic alcohols, 2 monoterpenes, 4 aldehydes, 15 aromatic compounds, and 20 hydrocarbons (Table 1). The proportions of the classes of the detected compounds in pistillate and staminate flowers are illustrated in Fig. 1. The aliphatic esters had the highest proportions in both sexes (51.9% in pistillate flowers and 35.2% in staminate flowers), followed by the aliphatic acids (21.8% in pistillate and 21.3% in staminate flowers). While many hydrocarbons were detected, their proportions were small (2.7% in pistillate flowers and 5.4% in staminate flowers). The proportions of aromatic compounds were 4.7 and 4.5% in pistillate flowers and staminate flowers, respectively. The remainder consisted of monoterpenes (2.6% in pistillate flowers and

Table 1. Identified compounds in *Smecarpus australiensis*.

No	Identity	MM	RI	RI (Ref)	P, μg	P, RSD (n = 10)	S, μg	S, RSD (n = 10)
1	3-Methylbutanoic acid	102.13	831	834[12]	0.010	0.004	0.012	0.008
2	1,2,4-Trimethylcyclohexane	126.24	874	881[13]	0.008	0.003	0.012	0.004
3	1-Ethyl-4-methylcyclohexane	126.24	877	888[13]	0.004	0.002	0.009	0.002
4	*n*-Nonane	128.26	900	900	0.018	0.08	0.023	0.006
5	1-Ethyl-3-methylcyclohexane	126.24	912	931[A][14]	0.007	0.002	0.009	0.003
6	2-Heptenal	112.17	954	957[15]	0.153	0.074	0.059	0.021
7	Benzaldehyde	106.12	960	958[16]	0.265	0.051	0.340	0.043
8	*β*-Myrcene	136.23	991	993[17]	0.019	0.009	0.078	0.013
9	1-Decene	140.27	990		0.020	0.010	0.043	0.017
10	*n*-Decane	142.28	1000	1000	0.028	0.012	0.078	0.024
11	*D*-Limonene	136.23	1025	1028[18]	2.230	0.950	9.187	2.399
12	Benzyl alcohol	108.14	1028	1032[19]	0.119	0.021	0.123	0.005
13	Benzeneacetaldehyde	120.15	1036	1042[20]	0.010	0.005	0.018	0.008
14	3-Methylbenzaldehyde	120.15	1065	1086[21]	0.244	0.120	0.266	0.015
15	*n*-Heptanoic acid	130.18	1071	1078[22]	0.036	0.013	1.801	2.064
16	*n*-Nonanal	142.24	1104	1108[23]	0.302	0.086	0.319	0.058
17	Phenylethyl alcohol	122.16	1114	1113[24]	0.171	0.050	0.185	0.038
18	Benzyl nitrile	117.15	1134	1143[21]	0.009	0.004	0.010	0.006
19	2-Ethylhexyl acetate	172.26	1146	1144[25]	0.018	0.024	0.008	0.004
20	*n*-Octanoic acid	144.21	1173	1180[26]	0.187	0.060	0.243	0.048
21	Diethyl succinate	174.19	1178	1176[27]	0.313	0.039	0.342	0.012
22	Methyl salicylate	152.15	1191	1192[28]	1.233	0.466	1.078	0.255
23	1-Dodecene	168.32	1193		0.105	0.015	0.146	0.035
24	*n*-Dodecane	170.33	1200		0.008	0.002	0.018	0.003
25	*n*-Decanal	156.27	1204	1208[29]	0.021	0.009	0.045	0.024
26	Benzothiazole	135.19	1221	1228[30]	0.026	0.014	0.041	0.003
27	*n*-Nonanoic acid	158.24	1264	1265[31]	0.036	0.014	0.057	0.014
28	Ethyl salicylate	166.17	1268	1269[31]	0.664	0.171	0.7544	0.064
29	*n*-Tridecane	184.36	1300		0.217	0.467	1.011	0.832
30	4-Allyl-2-methoxyphenol (eugenol)	164.20	1356	1359[29]	0.025	0.018	0.018	0.005

(*Continued on next page*)

Table 1. (*Continued*)

No	Identity	MM	RI	RI (Ref)	P, µg	P, RSD (n = 10)	S, µg	S, RSD (n = 10)
31	*n*-Decanoic acid	172.26	1363	1370[21]	0.476	0.119	0.561	0.115
32	4-Hydroxy-3-methoxybenzaldehyde (vanillin)	152.15	1386		0.113	0.047	0.122	0.021
33	1-Tetradecene	196.37	1393		0.174	0.051	0.328	0.109
34	*n*-Tetradecane	198.38	1400		0.158	0.071	0.386	0.119
35	2-Methoxy-4-propenylphenol (*trans*-isoeugenol)	164.20	1445	1451[32]	0.297	0.101	0.269	0.091
36	Ethyl 4-ethoxybenzoate	194.23	1520	1522[33]	0.708	0.408	0.705	0.536
37	5-MeC15	226.44	1553		0.077	0.045	0.108	0.019
38	*n*-Dodecanoic acid (lauric acid)	200.32	1558	1558[34]	0.423	0.070	0.488	0.048
39	3-MeC15	226.44	1573	1570[35]	0.475	0.061	0.590	0.021
40	1-Hexadecene	224.43	1596		0.148	0.108	0.313	0.150
41	*n*-Hexadecane	226.44	1600		0.147	0.112	0.438	0.226
42	*n*-Tetradecanal	212.37	1611	1611[36]	0.025	0.015	0.039	0.023
43	Benzophenone	182.22	1624	1628[33]	0.336	0.044	0.438	0.077
44	4-(4-hydroxy-3-methoxyphenyl)-2-butanone (zingerone)	194.23	1646	1640[37]	0.015	0.006	0.043	0.018
45	Undecyl cyclopentane	224.42	1655	1656[35]	0.076	0.048	0.261	0.171
46	2-MeC16	240.47	1665	1665[21]	0.052	0.031	0.153	0.078
47	*n*-Heptadecane	240.47	1700		0.046	0.024	0.101	0.0254
48	*n*-Tetradecanoic acid (myristic acid)	228.37	1759	1758[38]	0.198	0.164	0.401	0.261
49	2-MeC17	254.49	1771	1771[35]	0.065	0.042	0.173	0.068
50	Ethyl tetradecanoate (ethyl myristate)	256.42	1792	1795[39]	0.627	0.395	0.812	0.441
51	*n*-Octadecane	254.49	1800		0.457	0.318	1.284	0.590
52	1-Hexadecanol	242.44	1862	1883[21]	0.122	0.109	0.416	0.212
53	Ethyl pentadecanoate	270.45	1893	1893[40]	0.196	0.103	0.247	0.121
54	(*Z*)-9-hexadecenoic acid (palmitoleic acid)	254.41	1938	1941[21]	2.739	1.093	3.218	0.616
55	*n*-Hexadecanoic acid (palmitic acid)	256.42	1969	1968[21]	7.187	1.492	9.936	4.796
56	Ethyl (*Z*)-9-hexadecenoate (ethyl palmitoleate)	282.46	1979	1976[41]	0.534	0.505	0.301	0.073
57	Ethyl (*E*)-9-hexadecenoate	282.46	1982	1978[42]	0.059	0.038	0.074	0.024
58	Ethyl hexadecanoate (ethyl palmitate)	284.48	1992	1996[21]	21.143	10.501	17.333	7.867
59	1-Octadecanol	270.49	2068	2063[43]	0.136	0.134	0.526	0.270
60	Ethyl heptadecanoate	298.50	2093	2089[44]	0.264	0.213	0.179	0.106
61	(*Z*,*Z*)-9,12-octadecadienoic acid (linoleic acid)	280.45	2134	2136[21]	0.848	0.319	0.421	0.107
62	(*Z*)-9-octadecenoic acid (oleic acid)	282.46	2142	2146[21]	7.322	4.418	5.113	1.610
63	Ethyl (*Z*,*Z*)-9,12-octadecadienoate (ethyl linoleate)	308.50	2161	2165[31]	8.868	5.331	5.900	2.209
64	Ethyl (*Z*,*Z*,*Z*)-9,12,15-Octadecatrienoate	306.48	2165	2153[45]	5.178	1.501	4.881	1.910
65	Ethyl (*Z*9)-octadecenoate (ethyl oleate)	310.51	2171	2172[41]	4.966	2.372	3.018	1.008
66	Ethyl (*E*9)-octadecenoate	310.51	2181	2174[42]	0.730	0.330	0.501	0.12
67	Ethyl octadecanoate	312.53	2192	2197[41]	1.863	1.152	1.341	0.502

MM, molar mass; RI, Kovats retention index; RI (Ref), RI from the literature that used a column with 5% diphenyl/95% dimethyl polysiloxane as the stationary phase.
[A]Obtained from 100% dimethylpolysiloxane column; P, pistillate (female); S, staminate (male), RSD, relative standard deviation.

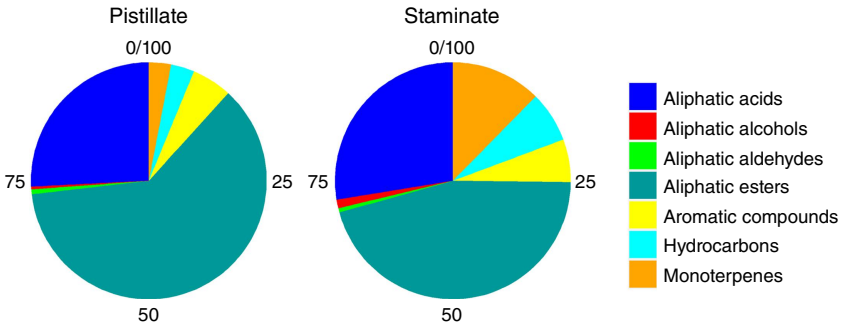

Fig. 1. Proportions of the compound classes in female and male flowers of *S. australiensis*.

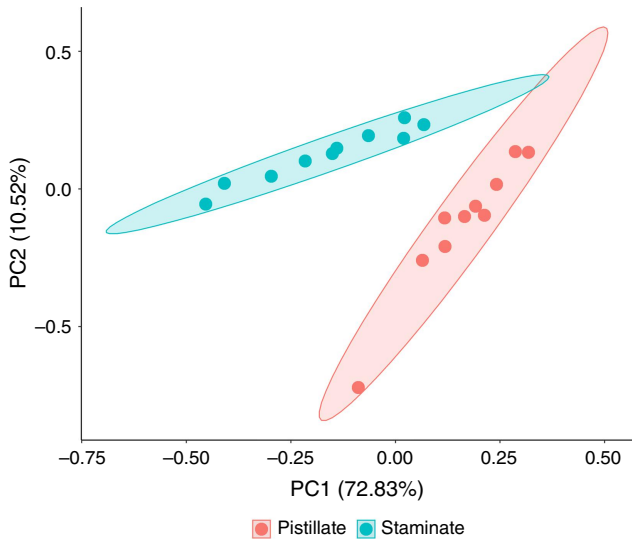

Fig. 2. Principal component analysis of the individual compounds in pistillate and staminate flowers; Biplot of the first and second principal components.

9.6% in staminate flowers), aliphatic aldehydes (0.6% in pistillate flowers and 0.4% in staminate flowers), and aliphatic alcohols (0.3% in pistillate flowers and 0.9% in staminate flowers). *D*-limonene is noticeably higher in staminate flowers than in pistillate flowers (Table 1). Principal component analysis identified notable differences in the compositions of pistillate and staminate flowers (Fig. 2). Further detailed investigation will be required to identify the functional or physiological role of such compositional differences in these flowers.

Flowers release a more diverse suite of volatiles at higher levels than other plant parts.[46] The floral volatiles are by-products of plant secondary metabolism, and some volatiles function to attract pollinators or as a defence against florivores and pathogens.[46] The fatty acids, 16- and 18-carbon species, i.e. C16:1 (palmitoleic acid), C16:0 (palmitic acid), C18:1 (oleic acid) and C18:2 (linoleic), and their ethyl esters are the predominant constituents in the tar tree flower samples. The fatty acids are important compounds in plants. For example, most cutin monomers are derived from the 16- and 18-carbon fatty acids to form the macromolecules that are the framework of the plant cuticles.[47] The 18-carbon unsaturated fatty acids can be nitrated to act as signalling mediators in the plant-defence system in oxidative stress situations.[48] It is also known that linoleic acid serves as a hydroperoxyl intermediate for the biosynthesis of green leaf volatiles.[49] The other detected compounds are also commonly found in plants. For example, *D*-limonene inhibits spore germination of the rice blast fungus (*Magnaporthe oryzae*) and is expressed at higher levels in response to the up-regulation of a terpene synthase gene when rice plants have the fungal infection.[50] Methyl salicylate is widespread as a herbivore-induced volatile to communicate herbivore attacts in plants. For example, barley exposed to deuterated methyl salicylate showed significant qualitative and quantitative changes in the chemical profile of the headspace.[51] Methyl salicylate is also known as a mobile signal to induce systemic acquired resistance against biotrophic pathogens in the tobacco plant.[52]

Ethyl and methyl salicylates are the major aromatic constituents in solvent extracts of the tar tree flowers, while phenylpropanoids and phenylbutanoids are present only in minute amounts. Although the amount of zingerone is minute (Table 1), the presence of this compound explains the attraction of *B. jarvisi*.[3a,53] The attraction of *B. jarvisi* is specific to zingerone and has been confirmed by systematic modification of zingerone and testing the synthesized analogs in the field.[54] In the study, >99% of flies captured by traps containing zingerone and its analogs were *B. jarvisi*. Hence, it is likely that zingerone is responsible for the observed interaction between the plant and fruit fly species. There appears to be no record of other fruit fly species being attracted to tar tree flowers. Along the tar tree natural distribution range, it is likely that different local fruit fly species are attracted to one or more of the phenylpropanoids and phenylbutanoids. For example, tar tree flowers contain *trans*-isoeugenol which is attractive to males of a New Caledonian fruit fly, *B. curvipennis*.[3d]

In summary, the present study addressed the chemical basis of the interaction between *B. jarvisi* and the native tar tree. Solvent extracts from tar tree flowers were dominated by unsaturated fatty acids and their ethyl esters. Amongst the 67 identified compounds, zingerone, a fruit fly attractant, was detected in minute amounts, providing a likely explanation for reported attraction of *B. jarvisi* fruit flies. Co-evolutionary interactions between plants and their insect

pollinators mediated by plant secondary metabolites are known.[55] The commonly used fruit fly attractants are plant secondary metabolites or derivatives, which have been deployed for decades to monitor and control horticultural pest fruit flies.[56] There are many unexplored plant species, but future investigations will greatly benefit from a targeted approach, for example, studies on species with previous observations and records will increase the chances of new attractant discovery and address the chemistry of a species of interest.

Experimental

Chemicals

3-Methylbutanoic acid, *n*-nonane, 2-heptenal, benzaldehyde, *β*-myrcene, 1-decene, *n*-decane, *D*-limonene, benzyl alcohol, 3-methylbenzaldehyde, *n*-heptanoic acid, *n*-nonanal, phenethyl alcohol, *n*-octanoic acid, methyl 2-hydroxybenzoate, 1-dodecene, *n*-dodecane, *n*-decanal, benzothiazole, *n*-nonanoic acid, ethyl 2-hydroxybenzoate, *n*-tridecane, 3-alyl-6-methoxyphenol, *n*-decanoic acid, 4-hydroxy-3-methoxybenzaldehyde, 1-tetradecene, *n*-tetradecane, 3-allyl-6-methoxyphenol, 1-tetradecene, tetradecane, (*E*)-2-methoxy-4-(1-propenyl)phenol, *n*-dodecanoic acid, *n*-hexadecane, *n*-tetradecanal, benzophenone, 4-(4-hydroxy-3-methoxyphenyl)-2-butanone, *n*-heptadecane, *n*-tetradecanoic acid, ethyl tetradecanoate, *n*-octadecane, 1-hexadecanol, ethyl pentadecanoate, (*Z*)-9-hexadecenoic acid, *n*-hexadecanoic acid, ethyl (*Z*)-9-hexadecenoate, ethyl (*E*)-9-hexadecenoate, ethyl hexadecanoate, (*Z*,*Z*)-9,12-octadecadienoic acid, ethyl (*Z*,*Z*)-9,12-octadecadienoate, ethyl (Z,*Z*,*Z*)-9,12,15-octadecatrienoate, 1-octadecanol, (*Z*,*Z*)-9,12-octadecadienoic acid, (*Z*)-9-octadecenloic acid, ethyl (*Z*)-9-octadecenoate, and ethyl octadecanoate were purchased from Sigma-Aldrich. 3-Ethylhexyl acetate was purchased from Tokyo Chemical Industry. All compounds were analytical grade with 98% purity or higher.

Collection of flowers

The flowers (Queensland Herbarium voucher number: AQ952605) were collected from male and female tar trees located in Silver Crescent Park, Palm Cove, Queensland, Australia (−16.758420, 154.669235) in October 2018. Branches with staminte or pistillate flowers were cut from the trees. Ten flowers of each sex were separately cut into fine pieces in a ceramic bowl using microscissors. The fine flower pieces were transferred to a 2.0 mL clear vial containing 1.0 mL of absolute ethanol (six replicates for each sex). The samples were transported to Macquarie University, Australia, and stored at 4°C for 2–3 weeks until extracted.

Extraction of flowers

Milli-Q water (1.0 mL) (Millipore) was added to each sample vial, vortexed for 30 s, and then ultrasonicated for 30 min. The aqueous extract was transferred to a 10 mL separating funnel. The aqueous phase was extracted with the organic solvents (3 × 2.0 mL, 10% ethyl acetate in hexane). The organic layers were combined, washed with Milli-Q water (6.0 mL), and dried over Na_2SO_4. The solvents were evaporated using a Rotary evaporator (Buchi), and the residue was re-dissolved in 200 μL of 10% ethyl acetate (v/v) in hexane. The extracted samples were stored at −30°C until analyzed. An aliquot (5 μL) of the three internal standards, 1-octanol, methyl *n*-dodecanoate, and methyl *n*-hexadecanoate, were incorporated to give 2.6, 2.5, and 2.6 μg/mL, respectively.

Gas chromatography-mass spectrometry (GC-MS) analysis

GC-MS analysis was performed on a Shimadzu GCMS TQ8040 spectrometer equipped with a split/splitless injector, fused silica capillary column (SH-Rtx-5MS, 30 m × 0.25 mm I.D. × 0.25 μm film thickness) with cross-bond 5% diphenyl/95% dimethyl polysiloxane as the stationary phase and integrated mass spectrometry (MS). Helium gas (BOC, North Ryde, NSW, Australia) (99.999%) was used as a carrier gas with a constant flow of 1.5 mL/min. An aliquot of 1 μL of a sample was injected in splitless mode, and the injection port temperature was 270°C. The initial column temperature was set at 60°C and held for 4 min, increased to 220°C at a rate of 2°C/min, then increased to 320°C at a rate of 30°C/min and held for 3 min. The interface and ion source box temperatures were set at 250 and 200°C, respectively. The ionization method was electron impact at a voltage of 70 eV. The spectra were obtained over a mass range of *m/z* 41–600. The data were processed by Shimadzu GCMS Post-run software. The retention indices were obtained by analyzing a run of a standard mixture of C_8 to C_{40} alkane with a sample. For identifications, mass spectra were compared with the NIST library (NIST17-1, NIST17-2, NIST17s) to identify related molecules. Fragmentation patterns and retention indices published in the literature were used to determine candidate molecules. The identity of a candidate molecule was confirmed by comparing retention time and fragmentations of the authentic molecule. The solvents used, including absolute ethanol, hexane, and ethyl acetate, were routinely analyzed by GC-MS to identify any impurities. The percentage of each compound was calculated from the quantification or semi-quantification of the constituents.

Standard solutions of five known concentrations of the individual compounds were prepared to quantify individual compounds. These contained three internal standards with the same concentration used in the extracted samples, in 5 mL volumetric flasks. The standard solutions were analyzed by GC-MS along with the flower samples. The standard curves of the authentic samples were generated by linear regression of peak area ratios of a compound to the internal standard against concentration ratios of a compound to that of the internal standard. Equations obtained by linear regression

were used to calculate the concentrations of the compounds in a sample, and the amount of a compound per flower was subsequently estimated by taking account of the final sample volume of a sample prepared from ten flowers.

Several compounds that are not commercially available were estimated by the use of a surrogate.[57] The response factor of 1,2,4-trimethyl cyclohexane was used to estimate the quantities of 1-ethyl-4-methylcyclohexane and 1-ethyl-3-methylcyclohexane. The response factor of benzaldehyde was used to estimate the quantity of benzeneacetaldehyde. The response factor of methyl hexadecanoate was used to estimate the quantities of 3-methyl pentadecane, undecyl cyclopentane, 2-methyl hexadecane, 2-methyl heptadecane. The response factor of ethyl (*Z*)-9-hexadecenoate was used to estimate the quantity of ethyl (*E*)-9-hexadecenoate. The response factor of ethyl (*Z*)-9-octadecenoate was used to estimate the quantity of ethyl (*E*)-9-octadecenoate.

Data analysis

Principal component analysis was carried out to compare the compositions of pistillate and staminate flowers.

Supplementary material

Supplementary material is available online.

References

[1] (a) Tan K-H, Nishida R. Mutual reproductive benefits between a wild orchid, *Bulbophyllum patens*, and *Bactrocera* fruit flies via a floral synomone. *J Chem Ecol* 2000; 26(2): 533–46. doi:10.1023/A:1005477926244
(b) Tan K-H, Nishida R. Synomone or kairomone? – *Bulbophyllum apertum* flower releases raspberry ketone to attract Bactrocera fruit flies. *J Chem Ecol* 2005; 31(3): 497–507. doi:10.1007/s10886-005-2023-8
(c) Tan K-H, Nishida R, Toong Y-C. Floral synomone of a wild orchid, *Bulbophyllum cheiri*, lures Bactrocera fruit flies for pollination. *J Chem Ecol* 2002; 28(6): 1161–72. doi:10.1023/A:1016277500007
(d) Tan KH, Nishida R. Zingerone in the floral synomone of *Bulbophyllum baile*yi (Orchidaceae) attracts Bactrocera fruit flies during pollination. *Biochem Syst Ecol* 2007; 35(6): 334–41. doi:10.1016/j.bse.2007.01.013
(e) Tan KH, Nishida R. editors. Pollination of bactrocerophilous *Bulbophyllum* orchids. Proceedings of the 20th World Orchid Conference. Singapore: Singarpore Botanic Gardens; 2011.
(f) Tan KH, Tan LT, Nishida R. Floral phenylpropanoid cocktail and architecture of *Bul*bophyllum vinaceum orchid in attracting fruit flies for pollination. *J Chem Ecol* 2006; 32(11): 2429–41. doi:10.1007/s10886-006-9154-4
(g) Katte T, Tan KH, Su Z-H, Ono H, Nishida R. Floral fragrances in two closely related fruit fly orchids, *Bulbophyllum hortorum* and *B. macranthoides* (Orchidaceae): assortments of phenylbutanoids to attract tephritid fruit fly males. *Appl Entomol Zool* 2020; 55(1): 55–64. doi:10.1007/s13355-019-00653-x

[2] (a) Drew RAI. The response of fruit fly species (Diptera: Tephritidae) in the South Pacific area to male attractants. *Aust J Entomol* 1974; 13(4): 267–70. doi:10.1111/j.1440-6055.1974.tb02206.x
(b) Fletcher BS. The biology of dacine fruit flies. *Annu Rev Entomol* 1987; 32(1): 115–44. doi:10.1146/annurev.en.32.010187.000555

[3] (a) Fay HAC. A highly effective and selective male lure for *Bactrocera jarvisi* (Tryon) (Diptera: Tephritidae). *Aust J Entomol* 2012; 51(3): 189–97. doi:10.1111/j.1440-6055.2011.00847.x
(b) Nakahira M, Ono H, Wee SL, Tan KH, Nishida R. Floral synomone diversification of *Bulbophyllum* sibling species (Orchidaceae) in attracting fruit fly pollinators. *Biochem Syst Ecol* 2018; 81: 86–95. doi:10.1016/j.bse.2018.10.002
(c) Royer JE, Agovaua S, Bokosou J, Kurika K, Mararuai A, Mayer DG, *et al.* Responses of fruit flies (Diptera: Tephritidae) to new attractants in Papua New Guinea. *Aust Entomol* 2018; 57(1): 40–9. doi:10.1111/aen.12269
(d) Royer JE, Mille C, Cazeres S, Brinon J, Mayer DG. Isoeugenol, a more attractive male lure for the cue-lure-responsive pest fruit fly *Bactrocera curvipennis* (Diptera: Tephritidae: Dacinae), and new records of species responding to zingerone in New Caledonia. *J Econ Entomol* 2019; 112(3): 1502–7. doi:10.1093/jee/toz034
(e) Royer JE, Tan KH, Mayer DG. Comparative trap catches of male *Bactrocera*, *Dacus*, and *Zeugodacus* Fruit Flies (Diptera: Tephritidae) with four floral phenylbutanoid lures (anisyl aetone, cue-lure, raspberry ketone, and zingerone) in Queensland, Australia. *Environ Entomol* 2020; 49(4): 815–22. doi:10.1093/ee/nvaa056

[4] (a) Royer JE. Responses of fruit flies (Tephritidae: Dacinae) to novel male attractants in north Queensland, Australia, and improved lures for some pest species. *Aust Entomol* 2015; 54(4): 411–26. doi:10.1111/aen.12141
(b) Royer JE, Khan M, Mayer DG. Methyl-isoeugenol, a highly attractive male lure for the cucurbit flower pest *Zeugodacus diversus* (Coquillett) (syn. *Bactrocera diversa*) (Diptera: Tephritidae: Dacinae). *J Econ Entomol* 2018; 111(3): 1197–201. doi:10.1093/jee/toy068

[5] (a) Paula JAM, Ferri PH, Bara MTF, Tresvenzol LMF, Sá FAS, Paula JR. Infraspecific chemical variability in the essential oils of *Pimenta pseudocaryophyllus* (Gomes) L.R. Landrum (Myrtaceae). *Biochem Syst Ecol* 2011; 39(4): 643–50. doi:10.1016/j.bse.2011.05.013
(b) Wong KC, Sivasothy Y, Boey PL, Osman H, Sulaiman B. Essential oils of *Etlingera elatior* (Jack) R. M. Smith and *Etlingera littoralis* (Koenig) Giseke. *J Essent Oil Res* 2010; 22(5): 461–6. doi:10.1080/10412905.2010.9700372
(c) de L, Nogueira PC, Bittrich V, Shepherd GJ, Lopes AV, Marsaioli AJ. The ecological and taxonomic importance of flower volatiles of *Clusia species* (Guttiferae). *Phytochemistry* 2001; 56(5): 443–52. doi:10.1016/S0031-9422(00)00213-2
(d) Choi H-S, Sawamura M. Composition of the essential oil of *Citrus tamurana* Hort. ex Tanaka (Hyuganatsu). *J Agric Food Chem* 2000; 48(10): 4868–73. doi:10.1021/jf000651e

[6] Raghu S. Functional significance of phytochemical lures to dacine fruit flies (Diptera: Tephritidae): an ecological and evolutionary synthesis. *Bull Entomol Res* 2004; 94(5): 385–99. doi:10.1079/BER2004313

[7] Nishida R, Tan KH, Serit M, Lajis NH, Sukari AM, Takahashi S, *et al.* Accumulation of phenylpropanoids in the rectal glands of males of the Oriental fruit fly, *Dacus dorsalis*. *Experientia* 1988; 44(6): 534–6. doi:10.1007/BF01958941

[8] (a) Wee S-L, Tan K-H, Nishida R. Pharmacophagy of methyl eugenol by males enhances sexual selection of *Bactrocera carambolae*. *J Chem Ecol* 2007; 33(6): 1272–82. doi:10.1007/s10886-007-9295-0
(b) Kumaran N, Prentis PJ, Mangalam KP, Schutze MK, Clarke AR. Sexual selection in true fruit flies (Diptera: Tephritidae): transcriptome and experimental evidences for phytochemicals increasing male competitive ability. *Mol Ecol* 2014; 23(18): 4645–57. doi:10.1111/mec.12880

[9] Park SJ, De Faveri SG, Cheesman J, Hanssen BL, Cameron DNS, Jamie IM, *et al.* Zingerone in the flower of *Passiflora maliformis* attracts an Australian fruit fly, *Bactrocera jarvisi* (Tryon). *Molecules* 2020; 25(12): 2877. doi:10.3390/molecules25122877

[10] (a) May AWS. Queensland host records for the Dacinae (fam. Trypetidae). *Queens J Agric Sci* 1953; 10: 36–79
(b) May AWS. Queensland host records for the Dacinae (fam. Trypetidae). Second supplementary lists. *Queens J Agric Sci* 1960; 17: 195–200.

[11] Zich FA, Hyland BPM, Whiffin T, Kerrigan RA. Australian tropical rainforest plants, Edition 8. 2020. Available at https://apps.lucidcentral.org/rainforest/

[12] Huang B, Qin L, Chu Q, Zhang Q, Gao L, Zheng H. Comparison of headspace SPME with hydrodistillation and SFE for analysis of the volatile components of the roots of *Valeriana officinalis* var. *latifolia*. *Chromatographia* 2008; 69(5): 489. doi:10.1365/s10337-008-0921-y
[13] Bramston-Cook R. Kovats indices for C2-C13 hydrocarbons and selected oxygenated/halocarbons with 100% dimethylpolysiloxane columns. 2013. Available at http://lotusinstruments.com/wp/wp-content/uploads/List-of-Kovats-Indices-for-C2-C13-Hydrocarbons.pdf
[14] Ohnishi S, Shibamoto T. Volatile compounds from heated beef fat and beef fat with glycine. *J Agric Food Chem* 1984; 32(5): 987–92. doi:10.1021/jf00125a008
[15] Xie J, Sun B, Zheng F, Wang S. Volatile flavor constituents in roasted pork of Mini-pig. *Food Chem* 2008; 109(3): 506–14. doi:10.1016/j.foodchem.2007.12.074
[16] Qiao Y, Xie B, Zhang Y, Zhang Y, Fan G, Yao XL, *et al.* Characterization of aroma active compounds in fruit juice and peel oil of jinchen sweet orange fruit (*Citrus sinensis* (L.) Osbeck) by GC-MS and GC-O. *Molecules* 2008; 13(6): 1333–44. doi:10.3390/molecules13061333
[17] Leela NK, Vipin TM, Shafeekh KM, Priyanka V, Rema J. Chemical composition of essential oils from aerial parts of *Cinnamomum malabatrum* (Burman f.) Bercht & Presl. *Flavour Fragr J* 2009; 24(1): 13–6. doi:10.1002/ffj.1910
[18] Angioni A, Barra A, Coroneo V, Dessi S, Cabras P. Chemical composition, seasonal variability, and antifungal activity of *Lavandula stoechas* L. ssp. stoechas essential oils from stem/leaves and flowers. *J Agric Food Chem* 2006; 54(12): 4364–70. doi:10.1021/jf0603329
[19] Mebazaa R, Mahmoudi A, Fouchet M, Santos MD, Kamissoko F, Nafti A, *et al.* Characterisation of volatile compounds in Tunisian fenugreek seeds. *Food Chem* 2009; 115(4): 1326–36. doi:10.1016/j.foodchem.2009.01.066
[20] Cole RA, Haber WA, Setzer WN. Chemical composition of essential oils of seven species of *Eugenia* from Monteverde, Costa Rica. *Biochem Syst Ecol* 2007; 35(12): 877–86. doi:10.1016/j.bse.2007.06.013
[21] Radulović N, Blagojević P, Palić R. Comparative study of the leaf volatiles of *Arctostaphylos uva-ursi* (L.) Spreng. and *Vaccinium vitis-idaea* L. (Ericaceae). *Molecules* 2010; 15(9): 6168–85. doi:10.3390/molecules15096168
[22] Javidnia K, Miri R, Kamalinejad M, Khazraii H. Chemical composition of the volatile oil of aerial parts of *Valeriana sisymbriifolia Vahl.* grown in Iran. *Flavour Fragr J* 2006; 21(3): 516–8. doi:10.1002/ffj.1660
[23] Fan G, Lu W, Yao X, Zhang Y, Wang K, Pan S. Effect of fermentation on free and bound volatile compounds of orange juice. *Flavour Fragr J* 2009; 24(5): 219–25. doi:10.1002/ffj.1931
[24] Pérez RA, Navarro T, de Lorenzo C. HS–SPME analysis of the volatile compounds from spices as a source of flavour in 'Campo Real' table olive preparations. *Flavour Fragr J* 2007; 22(4): 265–73. doi:10.1002/ffj.1791
[25] Ramsey JD, Flanagan RJ. Detection and identification of volatile organic compounds in blood by headspace gas chromatography as an aid to the diagnosis of solvent abuse. *J Chromatogr A* 1982; 240(2): 423–44. doi:10.1016/S0021-9673(00)99622-5
[26] Pino JA, Mesa J, Muñoz Y, Martí MP, Marbot R. Volatile components from mango (*Mangifera indica* L.) cultivars. *J Agric Food Chem* 2005; 53(6): 2213–23. doi:10.1021/jf0402633
[27] Fan W, Qian MC. Characterization of aroma compounds of Chinese "Wuliangye" and "Jiannanchun" liquors by aroma extract dilution analysis. *J Agric Food Chem* 2006; 54(7): 2695–704. doi:10.1021/jf052635t
[28] Edris AE, Chizzola R, Franz C. Isolation and characterization of the volatile aroma compounds from the concrete headspace and the absolute of *Jasminum sambac* (L.) Ait. (Oleaceae) flowers grown in Egypt. *Eur Food Res Technol* 2007; 226(3): 621–6. doi:10.1007/s00217-007-0623-y
[29] Alissandrakis E, Tarantilis PA, Harizanis PC, Polissiou M. Comparison of the volatile composition in thyme honeys from several origins in Greece. *J Agric Food Chem* 2007; 55(20): 8152–7. doi:10.1021/jf071442y
[30] Fan W, Qian MC. Identification of aroma compounds in Chinese 'Yanghe Daqu' liquor by normal phase chromatography fractionation followed by gas chromatography[sol]olfactometry. *Flavour Fragr J* 2006; 21(2): 333–42. doi:10.1002/ffj.1621
[31] Tava A, Pecetti L, Ricci M, Pagnotta MA, Russi L. Volatile compounds from leaves and flowers of *Bituminaria bituminosa* (L.) Stirt. (Fabaceae) from Italy. *Flavour Fragr J* 2007; 22(5): 363–70. doi:10.1002/ffj.1806
[32] Fernández de Simón B, Esteruelas E, Muñoz ÁM, Cadahía E, Sanz M. Volatile compounds in acacia, chestnut, cherry, ash, and oak woods, with a view to their use in cooperage. *J Agric Food Chem* 2009; 57(8): 3217–27. doi:10.1021/jf803463h
[33] Rohloff J, Bones AM. Volatile profiling of *Arabidopsis thaliana* – Putative olfactory compounds in plant communication. *Phytochemistry* 2005; 66(16): 1941–55. doi:10.1016/j.phytochem.2005.06.021
[34] Miyazawa M, Nishiguchi T, Yamafuji C. Volatile components of the leaves of *Brassica rapa* L. var. *perviridis* Bailey. *Flavour Fragr J* 2005; 20(2): 158–60. doi:10.1002/ffj.1335
[35] Kenig F, Simons D-JH, Crich D, Cowen JP, Ventura GT, Rehbein-Khalily T. Structure and distribution of branched aliphatic alkanes with quaternary carbon atoms in Cenomanian and Turonian black shales of Pasquia Hills (Saskatchewan, Canada). *Org Geochem* 2005; 36(1): 117–38. doi:10.1016/j.orggeochem.2004.06.014
[36] Palic R, Stojanovic G, Alagic S, Nikolic M, Lepojevic Z. Chemical composition and antimicrobial activity of the essential oil and CO_2 extracts of the oriental tobacco, Prilep. *Flavour Fragr J* 2002; 17(5): 323–6. doi:10.1002/ffj.1084
[37] Garneau FX, Bouhajib M, Collin GJ, Gagnon M, ApSimon JW. The glycosidically bound volatile compounds of *Picea mariana* (Mill.) B.S.P. *J Essent Oil Res* 1994; 6(1): 43–6. doi:10.1080/10412905.1994.9698323
[38] Formisano C, Senatore F, Bruno M, Bellone G. Chemical composition and antimicrobial activity of the essential oil of *Phlomis ferruginea Ten.* (Lamiaceae) growing wild in Southern Italy. *Flavour Fragr J* 2006; 21(5): 848–51. doi:10.1002/ffj.1740
[39] Ledauphin J, Guichard H, Saint-Clair J-F, Picoche B, Barillier D. Chemical and sensorial aroma characterization of freshly distilled calvados. 2. identification of volatile compounds and key odorants. *J Agric Food Chem* 2003; 51(2): 433–42. doi:10.1021/jf020373e
[40] Kim J-S, Chung HY. GC-MS analysis of the volatile components in dried boxthorn (*Lycium chinensis*) fruit. *J Korean Soc Appl Biol Chem* 2009; 52(5): 516–24. doi:10.3839/jksabc.2009.088
[41] Povolo M, Pelizzola V, Ravera D, Contarini G. Significance of the nonvolatile minor compounds of the neutral lipid fraction as markers of the origin of dairy products. *J Agric Food Chem* 2009; 57(16): 7387–94. doi:10.1021/jf8040353
[42] Andriamaharavo NR. Retention Data. NIST Mass Spectrometry Data Center; 2014.
[43] Marongiu B, Piras A, Porcedda S, Tuveri E, Maxia A. Isolation of *Seseli bocconi* Guss., subsp. *praecox* Gamisans (Apiaceae) volatile oil by supercritical carbon dioxide extraction. *Nat Prod Res* 2006; 20(9): 820–6. doi:10.1080/14786410500364684
[44] Woerdenbag HJ, Windono T, Bos R, Riswan S, Quax WJ. Composition of the essential oils of *Kaempferia rotunda* L. and *Kaempferia angustifolia* Roscoe rhizomes from Indonesia. *Flavour Fragr J* 2004; 19(2): 145–8. doi:10.1002/ffj.1284
[45] Tzakou O, Said A, Farag A, Rashed K. Volatile constituents of *Ailanthus excelsa* Roxb. *Flavour Fragr J* 2006; 21(6): 899–901. doi:10.1002/ffj.1739
[46] Muhlemann JK, Klempien A, Dudareva N. Floral volatiles: from biosynthesis to function. *Plant Cell Environ* 2014; 37(8): 1936–49. doi:10.1111/pce.12314
[47] Fich EA, Segerson NA, Rose JKC. The plant polyester cutin: biosynthesis, structure, and biological roles. *Annu Rev Plant Biol* 2016; 67: 207–33. doi:10.1146/annurev-arplant-043015-111929
[48] (a) Mata-Pérez C, Sánchez-Calvo B, Padilla MN, Begara-Morales JC, Valderrama R, Corpas FJ, *et al.* Nitro-fatty acids in plant signaling: New key mediators of nitric oxide metabolism. *Redox Biol* 2017; 11: 554–61. doi:10.1016/j.redox.2017.01.002
(b) He M, Ding N-Z. Plant unsaturated fatty acids: multiple roles in stress response. *Front Plant Sci* 2020; 11: 562785. doi:10.3389/fpls.2020.562785

[49] Hammerbacher A, Coutinho TA, Gershenzon J. Roles of plant volatiles in defence against microbial pathogens and microbial exploitation of volatiles. *Plant Cell Environ* 2019; 42(10): 2827–43. doi:10.1111/pce.13602
[50] Chen X, Chen H, Yuan JS, Köllner TG, Chen Y, Guo Y, *et al.* The rice terpene synthase gene OsTPS19 functions as an (*S*)-limonene synthase in planta, and its overexpression leads to enhanced resistance to the blast fungus *Magnaporthe oryzae*. *Plant Biotechnol J* 2018; 16(10): 1778–87. doi:10.1111/pbi.12914
[51] Ninkovic V, Glinwood R, Ünlü AG, Ganji S, Unelius CR. Effects of methyl salicylate on host plant acceptance and feeding by the aphid *Rhopalosiphum padi*. *Front Plant Sci* 2021; 12: 710268. doi:10.3389/fpls.2021.710268
[52] Park S-W, Kaimoyo E, Kumar D, Mosher S, Klessig DF. Methyl salicylate Is a critical mobile signal for plant systemic acquired resistance. *Science* 2007; 318(5847): 113–6. doi:10.1126/science.1147113
[53] Wee S-L, Peek T, Clarke AR. The responsiveness of *Bactrocera jarvisi* (Diptera: Tephritidae) to two naturally occurring phenylbutaonids, zingerone and raspberry ketone. *J Insect Physiol* 2018; 109: 41–6. doi:10.1016/j.jinsphys.2018.06.004
[54] Hanssen BL, Park SJ, Royer JE, Jamie JF, Taylor PW, Jamie IM. Systematic Modification of Zingerone Reveals Structural Requirements for Attraction of Jarvis's Fruit Fly. *Sci Rep* 2019; 9(1): 19332. doi:10.1038/s41598-019-55778-4
[55] Stevenson PC, Nicolson SW, Wright GA. Plant secondary metabolites in nectar: impacts on pollinators and ecological functions. *Funct Ecol* 2017; 31(1): 65–75. doi:10.1111/1365-2435.12761
[56] Vargas RI, Shelly TE, Leblanc L, Piñero JC. Chapter Twenty-Three - Recent Advances in Methyl Eugenol and Cue-Lure Technologies for Fruit Fly Detection, Monitoring, and Control in Hawaii. In: Litwack G, editor. Vitam Horm. Vol. 83. Academic Press; 2010. pp. 575–95.
[57] Jenke D, Odufu A. Utilization of internal standard response factors to estimate the concentration of organic compounds leached from pharmaceutical packaging systems and application of such estimated concentrations to safety assessment. *J Chromatogr Sci* 2012; 50(3): 206–12. doi:10.1093/chromsci/bmr048

Data availability. Not applicable.

Conflicts of interest. The authors declare no conflicts of interest.

Declaration of funding. This research was supported by the Australian Research Council Industrial Transformation Training Centre (ITTC) for Fruit Fly Biosecurity Innovation (Project IC50100026), funded by the Australian Government.

Acknowledgements. The authors thank Prof Darren Crayn and Mr Stuart Worboys for helping the identification of the Australian tar trees and locating them.

Author affiliations
[A]Applied BioSciences, Macquarie University, Sydney, 2109, Australia.
[B]Australian Research Council Centre for Fruit Fly Biosecurity Innovation, Macquarie University, North Ryde, NSW 2109, Australia.
[C]Horticulture and Forestry Science, Queensland Department of Agriculture and Fisheries, Mareeba, Qld, Australia.