New Pyridine Alkaloids from Rove Beetles of the Genus *Stenus* (Coleoptera: Staphylinidae)

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Three new pyridine alkaloids were detected in the pygidial glands of some *Stenus* species. The chemotaxonomic significance of the occurrence of these alkaloids and stenusine in different *Stenus* species is discussed. The antimicrobial properties of (*Z*)- and (*E*)-3-(2-methyl-1-butenyl)-pyridine and the deterrent activities of stenusine and norstenusine were investigated.

Key words: Rove Beetles, *Stenus*, Stenusine, Pyridine Alkaloids

Introduction

The rove beetle genus *Stenus* Latreille belongs to the most species-rich genera of the animal kingdom. Up to now, 2377 taxa including 8 fossil species are known worldwide (Puthz, 2008).

Since the beginning of the last century, *Stenus* beetles have intrigued scientists due to their extraordinary method of locomotion on water surfaces (Piffard, 1901). Upon falling on water they excrete a secretion from their pygidial glands that lowers the surface tension of water and propels the beetle forward very quickly. In the 1970's, this secretion was investigated by Schildknecht *et al.* (1976) using the common species *S. comma*. They found the alkaloid stenusine [*N*-ethyl-3-(2-methylbutyl)-piperidine, **4**] (Fig. 1) to be the active ingredient in the secretion responsible for this movement on water surfaces, also called skimming.

In the present paper we describe the detection of three new pyridine alkaloids, **1**–**3**, in *Stenus* species other than *S. comma* (Fig. 1). The chemotaxonomic significance of the occurrence of the alkaloids **1**–**4** in different *Stenus* species is discussed and the antimicrobial properties of **1** and **2** and the deterrent activities of **4** and **5** are investigated.

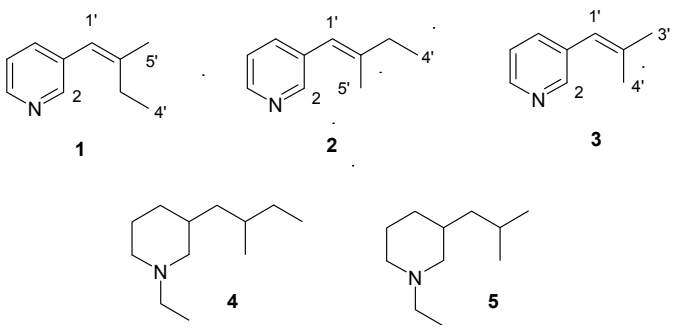

Fig. 1. Chemical structures of (*Z*)- (**1**), (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**), 3-(1-isobutenyl)-pyridine (**3**), stenusine (**4**), and norstenusine (**5**).

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Results and Discussion

Detection, structure elucidation, and synthesis of the pyridine alkaloids

Most previous studies on the chemical composition of the pygidial gland secretion of the rove beetle genus *Stenus* have been carried out using *S. comma*. We analyzed the pygidial gland content of a broader range of species of the subgenus *Hypostenus* (8) and *Metastenus* (2) by means of GC-MS.

All investigated species, except for *S. solutus* and *S. cicindeloides*, showed stenusine (**4**) as their major compound. *S. solutus* and *S. cicindeloides* contained another major compound showing a very similar EI mass spectrum (m/z = 147 [M^+], m/z = 146, m/z = 132 and m/z = 117) like actinidine, but they eluted at two different retention times (t_R = 9.21 min and t_R = 9.38 min). In order to unambiguously assign the retention time of actinidine we used an extract of valerian (*Valeriana officinalis*), a natural source of actinidine, as a reference. Actinidine from valerian showed the typical mass spectrum, but eluted at a third retention time (t_R = 10.00 min). As actinidine had been reported from other rove beetles (Huth and Dettner, 1990), *e.g.* from *Philontus laminatus*, we additionally used an extract of the abdominal glands of *P. laminatus* as a reference, which confirmed the retention time of actinidine from *Valeriana officinalis* (Fig. 2). As a result, we concluded that the substances from the investigated *Stenus* species are not actinidine and might be so far unknown in insects or in animals in general.

Because the unknown substances occurred amongst others in *S. solutus*, we dissected pygidial glands of 80 beetles of this species in order to collect sufficient material for structure elucidation. The crude sample was analyzed by LC-ESIMS and NMR spectroscopy at 500 MHz using a cryogenically cooled probe. Even though the NMR sample was a mixture of all gland contents, we achieved ^{1}H and 2D NMR spectra (^{1}H-^{1}H COSY, HSQC, HMBC), which suggested the substance to be (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) (Fig. 1). The ESIHR mass spectrum of **2** exhibited a $[M+H]^+$ peak at m/z 148.1118, consistent with a molecular formula $C_{10}H_{13}N$. The ^{1}H NMR data showed 4 signals (H-2 δ 8.39, H-4 δ 7.71, H-5 δ 7.39, H-6 δ 8.33) characteristic for an in position 3 substituted pyridine. The ^{1}H-^{1}H COSY correlations H-1' δ 6.27/3H-5' δ 1.87, H-1'/2H-3' δ 2.25 and 2H-3'/3H-4' δ 1.15 were in agreement with the 2-methyl-1-butenyl side chain. The ^{13}C chemical shifts of C-1' δ 120.9, C-3' δ 34.5, C-5' δ 18.0 of the 2-methyl-1-butenyl group were compared with the calculated values (ACD/CNMR: ChemSketch Window) δ 119.1, 33.4, 16.1 and led to the assumption of the (*E*)-configuration. For the (*Z*)-isomer δ 119.1, 28.8, 19.9 were calculated.

The (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) were synthesized by Wittig olefination from readily available starting materials over two steps (Fig. 3). 2-Bromobutane was converted into the phosphonium salt (Bestmann and Kratzer, 1963) in 68% yield. Subsequent Wittig reaction with nicotinaldehyde under salt-free conditions gave the desired compounds **1** and **2** in 43% yield as a 1.7:1 mixture, which could be separated by MPLC. In accordance with the literature (Brückner, 1996), we obtained predominantly the (*Z*)-isomer.

The (*Z*)- and (*E*)-configuration was exactly assigned by 1D NOE difference measurements of the synthetic compounds. Irradiation on the 5'-methyl group at δ 1.92 of the (*Z*)-isomer **1** indicated clearly an NOE with H-1' δ 6.23. On the other hand, the 5'-methyl group at δ 1.87 of **2** showed NOEs between 5'-CH_3/H-4 δ 7.71 and 5'-CH_3/H-2 δ 8.39 (Fig. 4).

Comparison of the retention times and mass spectra obtained with a mixture of extracts of *S. similis*, *S. solutus*, and *Philonthus laminatus* (Fig. 2) with those of synthetic (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) proved the major compounds in *S. similis* to be **1** (t_R = 9.21 min) and in *S. solutus* to be **2** (t_R = 9.38 min).

Compound **3** with a molecular ion peak in the EI mass spectrum at m/z 133 could be detected by GC-MS in the pygidial glands of *S. solutus*, *S. cicindeloides*, *S. binotatus* and *S. pubescens*. The [M^+] peaks of (*Z*)-3-(2-methyl-1-butenyl)-pyridine (**1**) and **3** differ by 14 mu.

The structure of **3** could be 3-(1-isobutenyl)-pyridine, which was synthesized in the same way as **1** and **2**. 2-Iodopropane reacted with triphenylphosphane to the phosphonium salt in 68% yield. Wittig reaction of the phosphonium salt with nicotinaldehyde under salt-free conditions gave 3-(1-isobutenyl)-pyridine (**3**) (82%). As expected the ^{1}H and ^{13}C NMR data of **3** differ only in the side chain from those of **1** and **2**. The methyl groups 3' and 4' were observed in the ^{1}H NMR spectrum at δ 1.89 and δ 1.81 and in the ^{13}C NMR

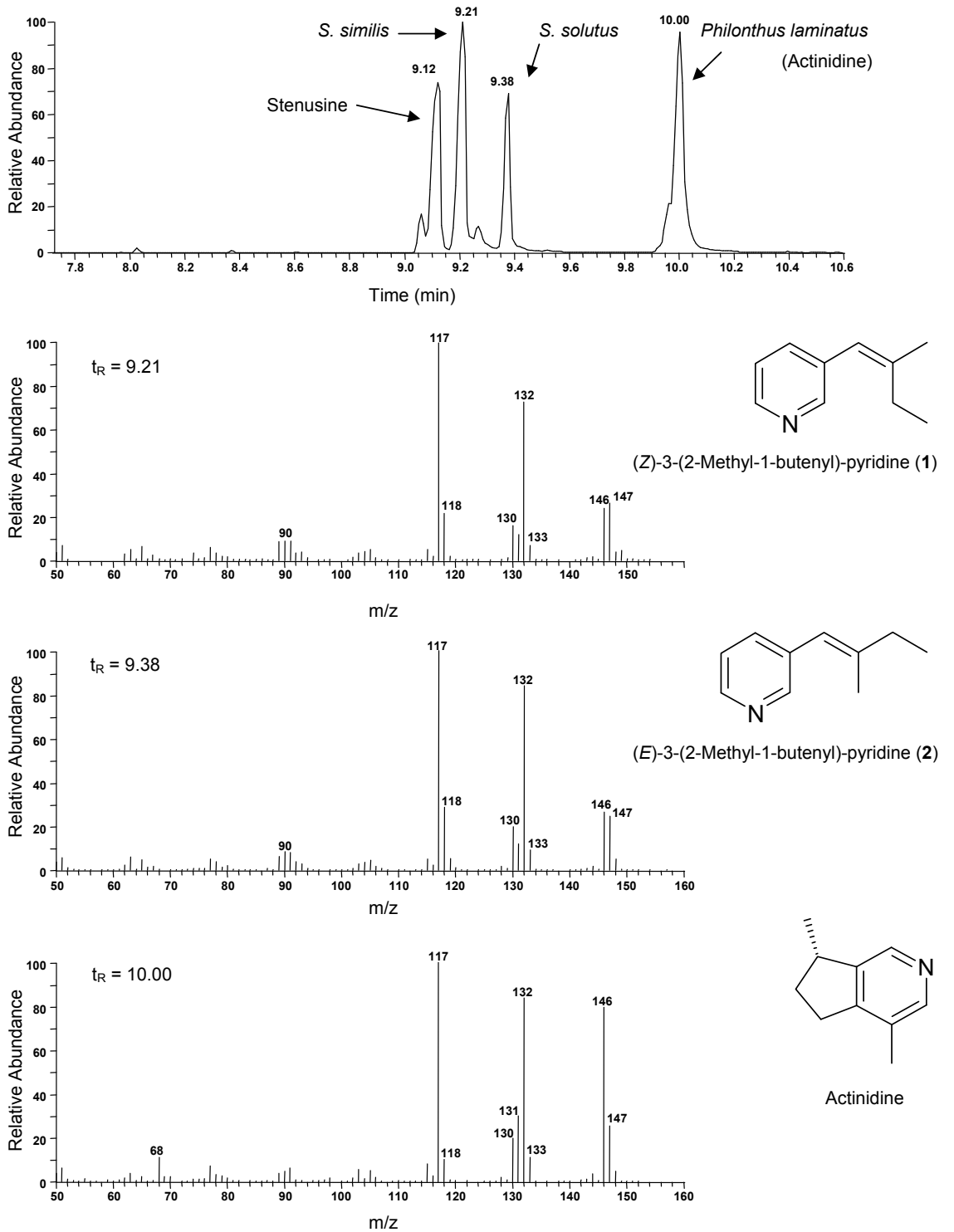

Fig. 2. Partial total-ion chromatogram of the mixed sample of *S. similis*, *S. solutus* and *Philonthus laminatus* and the EI mass spectra of (*Z*)- (**1**), (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) and actinidine.

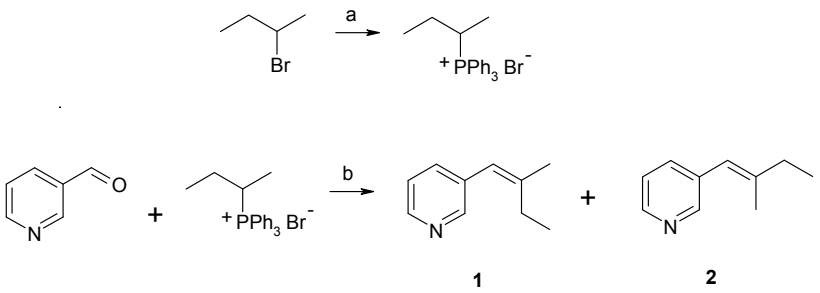

Fig. 3. Synthesis of (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**). (a) PPh_3, neat, 120 °C, 24 h, 68% (based on PPh_3); (b) NaHMDS, benzene, 2 h, room temperature, 43%.

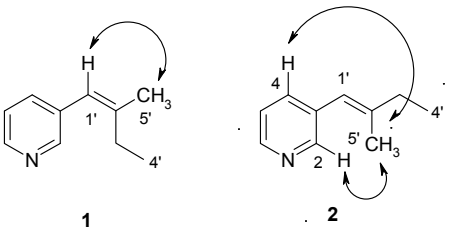

Fig. 4. 1D NOE correlations of (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**).

spectrum at δ 27.2 and δ 19.6. The retention times (t_R = 8.41 min) and mass spectra of compound **3** from the four *Stenus* species and of the synthesized sample were identical. This proved the occurrence of the alkaloid 3-(1-isobutenyl)-pyridine (**3**) (Fig. 1) in the pygidial glands of *S. solutus*, *S. cicindeloides*, *S. binotatus* and *S. pubescens*.

Chemotaxonomic significance

(*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) represent new natural products which are mainly, but not exclusively, abundant in the subgenus *Hypostenus*. The two investigated *Hypostenus* species *S. fulvicornis* und *S. latifrons* which do not contain the compounds represent exceptions. The pygidial glands of *S. binotatus* and *S. pubescens*, both from the subgenus *Metastenus*, contain both isomers **1** and **2**, and 3-(1-isobutenyl)-pyridine (**3**) (Table I). *S. cicindeloides* was reported to be able to skim on water surfaces (Billard and Bruyant, 1905) even though it hardly contains any stenusine (**4**); one possibility is that 3-(2-methyl-1-butenyl)-pyridine and 3-(1-isobutenyl)-pyridine replace stenusine as spreading alkaloid in this species. In a simple experiment one drop of stenusine (**4**), a 1.7:1 mixture of (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) or 3-(1-isobutenyl)-pyridine (**3**) was given at a water surface on which some small particles of foam polystyrene were swimming. The spreading velocity of **4** was the highest followed by the mixture of **1** and **2**. Compound **3** exhibited the lowest spreading velocity.

The close relationships of the chemical structures of the new alkaloids **1**, **2** and **3** with stenusine (**4**) and norstenusine (**5**) become evident from Fig. 1. Stenusine (**4**) and norstenusine (**5**) are the *N*-ethyl-octahydro products of the new alkaloids **1**, **2** and **3**.

In an earlier study we showed that the ratio of the four stereoisomers of the alkaloid stenusine (**4**) differs with species (Lusebrink *et al.*, 2007). The stereoisomeric ratio [(2'*R*,3*R*)-, (2'*S*,3*R*)-, (2'*S*,3*S*)- and (2'*R*,3*S*)-stenusine] of the German (GER) and Canadian (CDN) *S. comma* (subgenus *Stenus*) differs enormously: GER 17%:37%:43%:3% vs. CDN 0%:0%:55%:45%. Furthermore, the monoterpene isopiperitenol reported for the German *S. comma* (Schildknecht, 1970), was missing in the Canadian specimens, but was always abundant in the German *S. comma*.

Table I. *Stenus* species in which *Z/E*-isomers of 3-(2-methyl-1-butenyl)-pyridine (**1**, **2**), 3-(1-isobutenyl)-pyridine (**3**), and stenusine (**4**) were detected by GC-MS.

Compound	Subgenus									
	Metastenus	*Metastenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*	*Hypostenus*
	Species									
	S. binotatus Ljungh 1804	*S. pubescens* Stephens 1833	*S. fulvicornis* Stephens 1833	*S. latifrons* Erichson 1839	*S. fornicatus* Stephens 1833	*S. pinguis* Casey 1884 CDN	*S. tarsalis* Ljungh 1804	*S. similis* Herbst 1784	*S. solutus* Erichson 1840	*S. cicindeloides* Schaller 1783
1	+	+	–	–	+	+	++	++	+	+
2	++	++	–	–	+	++	+	+	++	+
3	+	+	–	–	–	–	–	–	+	+
4	++	++	++	++	++	++	++	++	+	+

+ Abundant; ++ abundant in higher amounts.
CDN species collected in Canada.

This indicates that the German and the Canadian *S. comma* show wide differences in the pygidial gland secretions. Morphological and molecular studies could potentially explain the differences in their secondary metabolites. A total inventory of the pygidial gland content, including stereoisomeric ratios of stenusine (**4**), could be promising for a comprehensive chemotaxonomic study.

Biological function

It is well known, that *Stenus* beetles impregnate their body surface with their pygidial gland secretion (Betz, 1999). This behaviour is called secretion grooming and suggests that the secretion involves substances which prevent the beetle from infection by epibiontic microorganisms like bacteria and fungi. The pygidial gland constituents stenusine (**4**) and norstenusine (**5**) have antimicrobial activity. According to our study (Lusebrink *et al.*, 2008), agar diffusion tests with a 57:43 (v/v) blend of (*Z*)- (**1**) and (*E*)-3-(2-methyl-1-butenyl)-pyridine (**2**) showed that the blend of **1** and **2** has higher antimicrobial properties at a content of 800 μg/disc than stenusine on in-

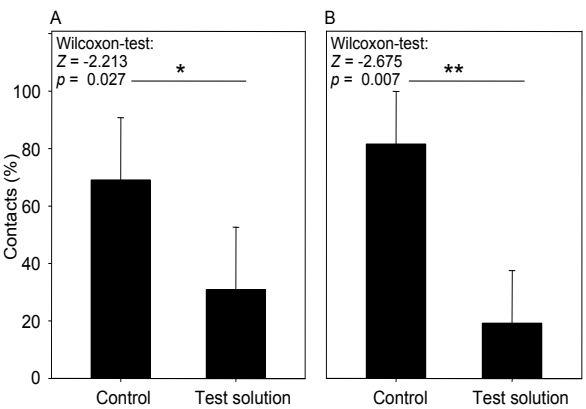

Fig. 5. Deterrencies of (A) stenusine (50 μg/ml milk) and (B) norstenusine (30 μg/ml milk) against *Leptothorax platythorax* ants. Both alkaloids were emulsified with UHT-milk and tested against UHT-milk controls. Bioassay A was conducted with a highly active ant colony, whereas test B was performed several weeks later, when the same colony exhibited a lower activity. Therefore A and B can not be directly compared. Each bioassay consisted of ten replicates. The bar diagram shows the percentile distribution of contacts with the control and test solution, including standard deviation bars. *, Significant difference ($p < 0.05$); **, high significant difference ($p < 0.01$).

sect pathogenic bacteria (Gram-positive: *Bacillus sphaericus*; Gram-negative: *E. coli* and *Serratia entomophila*). Moreover it was obvious, that this blend was considerably more active than the pure (*Z*)-isomer **1**. Thus a mixture of both isomers of 3-(2-methyl-1-butenyl)-pyridine contributes to the defence against epibiontic microorganisms of *S. solutus* and *S. cicindeloides* which only contain minor amounts of the antibacterial stenusine (**4**).

Both **4** and norstenusine (**5**) exhibit distinct deterrent activities against *Leptothorax platythorax* ants in feeding experiments. According to Schoonhoven *et al.* (2005) a chemical deterrent inhibits feeding when it is present in a place where insects would normally feed. During a binary choice test, ants were able to choose between test solutions in milk and milk controls (both solutions contained minor amounts of saccharose), and *L. platythorax* ants preferred milk over a test solution of milk with 50 μg/ml **4**. Thus **4** showed high deterrent activities (Fig. 5A). This bioassay was performed with a colony of highly active *L. platythorax* ants that showed lower activity when tested 5 weeks later. Therefore the experiment with norstenusine (**5**) can not be directly compared with the stenusine (**4**) bioassay. However, in this current experiment **5** also exhibited significant deterrency against the ants even at a concentration of **5** in milk as low as 30 μg/ml (Fig. 5B). Because most *Stenus* species emit pygidial gland secretion after molestations, these results show that the secretion also represents a true defensive secretion which is directed against enemies such as predatory arthropods like ants or spiders.

Experimental

General

GC-MS: Finnigan MAT GCQ instrument equipped with a BPX5 column (SGE, length 25 m, diameter 0.22 mm). Oven temperature program: 80 °C for 2 min, raised with 4 °C/min to 125 °C and then with 30 °C/min to 275 °C. LC-ESIMS: Finnigan MAT TSQ instrument equipped with a Ultrasep RP-18E column (length 100 mm, diameter 1 mm, particle size 5 μm). Gradient system starting from H_2O/MeCN 9:1 (each of them containing 0.2% HOAc) to 1:9 within 15 min; flow rate, 50 μl/min. MPLC: Büchi B688 pump and Büchi B687 gradient former. NMR: Bruker Avance DRX 500 and Bruker AC-300 instruments.

Collection and determination of Stenus beetles, and extraction of their pygidial glands

S. pinguis was collected near Meanook (Alberta, Canada) and the Canadian *S. comma* in Red Lodge Provincal Park (Alberta, Canada). All other species were collected in the surroundings of Bayreuth, Germany. The species collected in Germany were determined using the determination key of Lohse (1964). The Canadian species were identified by Dr. Volker Puthz (Schlitz, Germany), a *Stenus* specialist, who has described about 50% of all *Stenus* species.

The beetles (all adults) were killed as well as stored in a freezer at a temperature of –20 °C. For each sample, 10 glands of the same species were dissected and transferred into a conically ending vial with 25 μl EtOAc SupraSolv for GC (Merck). Each sample was homogenized by sonication for 5 min. Glands of 80 *S. solutus* beetles were dissected and extracted.

Bioassays

A deterrency bioassay was performed with a laboratory colony of *Leptothorax platythorax* (gift from Prof. Dr. J. Steidle, University of Hohenheim, Germany). Experiments were conducted in a controlled climate chamber with constant humidity (40%) at 25 °C and 16 h light:8 h dark. The behaviour of the ants was studied within a formicarium (46 cm × 31 cm, height 13 cm) lined with cement. The colonies were fed with saccharose- and honey-water and dead insects. Since **4** and **5** are not water-soluble, both alkaloids were emulsified with ultra-high temperature-processed (UHT)-milk (100 ml contained: 1.5 g fat, 3.4 g protein, 4.8 g carbohydrates and 10 g saccharose). During the binary choice the ant colony had 10 min to choose between the test solutions and pure milk controls. From experiment to experiment the positions of test solutions and controls were randomly changed. When the ants visited a droplet and fed at the milk solutions for at least 3 s (after the first mandible contact), the solution was considered as non-deterrent.

Each bioassay comprised of ten replicates and was statistically evaluated with the Wilcoxon-test for paired samples. Autoreplications could not be prevented.

Preparation and spectroscopic data of the alkaloids

(Z)- (**1**) *and (E)-3-(2-methyl-1-butenyl)-pyridine* (**2**): Triphenylphosphane (13.12 g, 50 mmol) and 2-bromobutane (10.96 ml ≙13.7 g, 100 mmol) were heated for 24 h at 120 °C in a sealed tube. The obtained colourless crystals were finely powdered and washed three times with dry THF and dried under vacuum (13332 Pa) for 2 h yielding 13.58 g (34 mmol, 68%) of 2-butyltriphenylphosphonium bromide, m.p. 225 °C. To a rapidly stirred suspension of 2-butyltriphenylphosphonium bromide (1.00 g, 2.5 mmol) in dry benzene (10 ml) was added at room temperature NaHMDS (1.25 ml, 2 M in THF, 2.5 mmol). After 30 s nicotinaldehyde (0.3 ml, 3.2 mmol) was dropped to the deep red suspension and stirring was continued for 2 h. The resulting pale yellow slurry was filtered and the solvent was evaporated. The residue was purified by flash chromatography on silica gel (*n*-hexane/EtOAc 3:1 v/v, R_f 0.4) yielding a 1.7:1 mixture of **1** and **2** (158 mg, 1.07 mmol, 43%) as a colourless oil. The mixture of **1** and **2** could be separated by MPLC on silica gel MN Nucleodur 100, particle size 12 μm (*n*-hexane/EtOAc 19:1 v/v).

(Z)-3-(2-Methyl-1-butenyl)-pyridine (**1**): ^{1}H NMR (300 MHz, MeOH-d_4): δ = 8.35 (1H, d, $J_{2,4}$ = 2.4 Hz, H-2), 8.34 (1H, dd, $J_{5,6}$ = 4.8 Hz, $J_{4,6}$ = 1.5 Hz, H-6), 7.65 (1H, ddd, $J_{4,5}$ = 7.8 Hz, $J_{4,6}$ = 1.5 Hz, $J_{2,4}$ = 2.4 Hz, H-4), 7.38 (1H, ddd, $J_{4,5}$ = 7.8 Hz, $J_{5,6}$ = 4.8 Hz, $J_{2,5}$ = 0.6 Hz, H-5), 6.23 (1H, br. s, H-1'), 2.25 (2H, dq, $J_{3',4'}$ = 7.5 Hz, $J_{1',3'}$ = 0.6 Hz, 2H-3'), 1.92 (3H, d, $J_{1',5'}$ = 1.5 Hz, 3H-5'), 1.10 (3H, t, $J_{3',4'}$ = 7.5 Hz, 3H-4'). – ^{13}C NMR (75 MHz, MeOH-d_4): δ = 149.9 (C-2), 147.4 (C-6), 145.8 (C-2'), 137.9 (C-4), 136.3 (C-3), 125.1 (C-5), 122.0 (C-1'), 26.7 (C-3'), 23.9 (C-5'), 13.3 (C-4'). – GC-MS: t_R = 9.21 min. – EI: *m*/*z* (%) = 147 (25) [M$^+$], 146 (24), 132 (71), 130 (15), 118 (22), 117 (100). – ESI-HRMS: 148.1118 [$C_{10}H_{14}N^+$] (calcd. 148.1121).

(E)-3-(2-Methyl-1-butenyl)-pyridine (**2**): ^{1}H NMR (300 MHz, MeOH-d_4): δ = 8.39 (1H, d, $J_{2,4}$ = 2.4 Hz, H-2), 8.33 (1 H, dd, $J_{5,6}$ = 4.8 Hz, $J_{4,6}$ = 1.5 Hz, H-6), 7.71 (1H, ddd, $J_{4,5}$ = 7.8 Hz, $J_{4,6}$ = 1.5 Hz, $J_{2,4}$ = 2.4 Hz, H-4), 7.39 (1H, ddd, $J_{4,5}$ = 7.8 Hz, $J_{5,6}$ = 4.8 Hz, $J_{2,5}$ = 0.6 Hz, H-5), 6.27 (1H, br. s, H-1'), 2.25 (2H, dq, $J_{3',4'}$ = 7.5 Hz, $J_{1',3'}$ = 1.2 Hz, 2H-3'), 1.87 (3H, d, $J_{1',5'}$ = 1.5 Hz, 3H-5'), 1.15 (3H, t, $J_{3',4'}$ = 7.5 Hz, 3H-4'). – ^{13}C NMR (75 MHz, MeOH-d_4): δ = 150.3 (C-2), 147.2 (C-6), 145.6 (C-2'), 138.2 (C-4), 136.6 (C-3), 125.0 (C-5), 120.9 (C-1'), 34.5 (C-3'), 18.0 (C-5'), 13.2 (C-4'). – GC-MS: t_R = 9.38 min. – EI: *m*/*z* (%) = 147 (24) [M$^+$], 146 (25), 132 (82), 130 (18), 118 (28), 117 (100). – ESI-HRMS: 148.1118 [$C_{10}H_{14}N^+$] (calcd. 148.1121).

3-(1-Isobutenyl)-pyridine (**3**): Triphenylphosphane (14.43 g, 55 mmol) and 2-iodopropane (5.00 ml, 50 mmol) in toluene (12 ml) were heated for 72 h under reflux. The formed colourless crystals were separated, washed with cold toluene and dried under vacuum (13332 Pa) for 2 h giving 14.68 g (34 mmol, 68%) of 2-propyltriphenylphosphonium iodide, m.p. 193 °C. To a stirred suspension of 2-propyltriphenylphosphonium iodide (9.08 g, 21 mmol) in dry THF (100 ml) was dropped NaHMDS (10.5 ml, 2 M in THF, 21 mmol). After 20 min nicotinaldehyde (1.95 ml, 20.7 mmol) was added during 5 min to the deep red solution. The stirring was continued for 14 h, the solvent evaporated, and the residue distributed between H_2O and CH_2Cl_2. The aqueous phase was extracted two times with CH_2Cl_2. Silica gel (30 g) was added to the combined organic phases and the solvent was removed. This silica gel was given to the top of a silica gel column. Elution with *n*-hexane/EtOAc 3:1 v/v yielded after evaporation of the solvent **3** (2.25 g, 16.89 mmol, 82%) as colourless oil.

^{1}H NMR (300 MHz, MeOH-d_4): δ = 8.37 (1H, d, $J_{2,4}$ = 2.1 Hz, H-2), 8.33 (1 H, dd, $J_{5,6}$ = 4.8 Hz, $J_{4,6}$ = 1.5 Hz, H-6), 7.64 (1H, ddd, $J_{4,5}$ = 7.9 Hz, $J_{4,6}$ = 1.5 Hz, $J_{2,4}$ = 2.1 Hz, H-4), 7.34 (1H, ddd, $J_{4,5}$ = 7.9 Hz, $J_{5,6}$ = 4.8 Hz, $J_{2,5}$ = 0.7 Hz, H-5), 6.22 (1H, br. s, H-1'), 1.89 (3H, d, $J_{1',3'}$ = 1.6 Hz, 3H-3'), 1.81 (3H, d, $J_{1',4'}$ = 1.6 Hz, 3H-4'). – ^{13}C NMR (75 MHz, MeOH-d_4): δ = 150.3 (C-2), 147.3 (C-6), 140.0 (C-2'), 137.9 (C-4), 136.3 (C-3), 124.9 (C-5), 122.4 (C-1'), 27.2 (C-3'), 19.6 (C-4'). – GC-MS: t_R = 8.41 min. – EI: *m*/*z* (%) = 133 (100) [M$^+$], 132 (68), 119 (85), 117 (63), 91 (57). – ESIHRMS: 133.0886 [$C_9H_{11}N^+$] (calcd. 133.08915).

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