MINI-REVIEW

Plant Signaling & Behavior 6:3, 369-371; March 2011; ©2011 Landes Bioscience

Roles of (*Z*)-3-hexenol in plant-insect interactions

Jianing Wei and Le Kang*

State Key Laboratory of Integrated Management of Pest Insects and Rodents; Institute of Zoology; Chinese Academy of Sciences; Beijing, China

Key words: green leaf volatiles, (*Z*)-3-hexenol, direct defense, indirect defense, priming

Green leaf C6-volatiles are among the most important herbivore-induced plant volatiles (HIPVs). They play important roles in mediating the behavior of herbivores and their natural enemies, and in triggering the plant-plant communication to prevent further attacks. Recently, wound-induced ubiquitous (*Z*)-3-hexenol, a C6-alcohol synthesized in the lipoxygenase/HPL pathway, was proved to be the most important info chemical for the herbivore repellence/attraction and natural enemy attraction in tritrophic interactions, as well as for the induction of gene expression in neighboring unattacked plants. In spite of the conflict functions of (*Z*)-3-hexenol in direct and indirect plant defenses, its positive roles in the indirect defense and the priming effect are consistent. Therefore, this compound can be used to develop novel insect pest control strategies.

To date, nearly 2000 volatile compounds have been identified in plant species from over 90 families.[1] These compounds are released from plant organs above or below the ground, and some are induced by biotic activities. Herbivore feeding stimulates the plants to release green leaf volatiles (GLVs), terpenoids, nitrogen-containing nitriles and oximes, methyl salicylate, etc. Production of these volatiles by plants involves at least three biosynthetic pathways: the fatty acid/lipoxygenase pathway for green leaf volatiles, the isoprenoid pathway for terpenoids, and the shikimic acid pathway for methyl salicylate.[2] Herbivore-damaged plants emit some of the most common GLVs and terpenoids that play important roles in mediating the behaviors of herbivores and their natural enemies, as well as in triggering the plant-plant communication.[1] Recently, functional studies on green leaf C6-volatiles have received wide attention and made exciting progresses. Especially, accumulating evidences on the C6-volatile (Z)-3-hexenol support its role in mediating indirect defense responses of plant.

Biosynthesis of (*Z*)-3-hexenol

Many inducible defense responses are activated by oxylipins, which are the oxygenated derivatives of fatty acids generated via the oxylipin branch pathways.[1] Based on the current available knowledge, most healthy plant species do not release GLVs such as C6-aldehydes, -alcohols or -esters, but large amounts of GLVs are emitted when plants are wounded.[3-5] In addition, release of these C6-volatiles occurs not only locally at the wounding sites, but also systemically in the distal leaves.[4,6] Through the oxylipin pathway that is parallel to the biosynthetic pathway for JAs, plant C6-volatiles are synthesized using 13-hydroperoxides of linoleic or linolenic acid (LA) as substrate and using fatty acid hydroperoxide lyase (HPL) as catalytic enzyme.[7,8] In the lipoxygenase/HPL pathway, the plant first produces C6-aldehydes, which are then catalyzed by alcohol dehydrogenase to form the corresponding C_6-alcohols, e.g. (*Z*)-3-hexenol.[8] Studies of mutant tomato plants and transgenic Nicotiana plants showed that JA-deficient plants (*spr2*) and HPL-deficient plants (as-*hpl*) released significantly reduced level of C6-alcohols than wild-type counterparts, suggesting that these enzymes are key regulators for the hexenol production in oxylipin pathway.[9-11] Therefore, the biosynthesis and release of (*Z*)-3-hexenol are closely related to HPL pathway, which is one of the two major competing oxylipin-pathway branches that produce stress-inducible compounds.[12]

Response of Herbivores to (*Z*)-3-hexenol

Herbivore- or wound-induced (*Z*)-3-hexenol can directly affect the physiology and behavior of herbivores through its positive (attractive) and negative (repelling and deterring) properties. Electrophysiological studies revealed that most herbivorous insects exhibited distinct electroantennogram (EAG) responses to (*Z*)-3-hexenol.[13] Compared with the weak EAG responses of two Inner Mongolia grasshopper species to hexanol, strong EAG responses were induced by C6-alcohols [(*Z*)-2-hexenol, (*E*)-2-hexen-ol and (Z)-3-hexenol], aldehyde [(*E*)-2-hexenal], ester [(Z)-3-hexenyl acetate] and C7-alcohol (1-hepatanol).[14] In addition, (*Z*)-3-hexenol stimulated similar EAG responses in the first instar nymphs and the adults of a grasshopper species *Melanoplus sanguinipes* to other C6-alcohols, aldehydes and esters.[15,16] A agromyzid fly species *Liliomyza sativae* displayed a higher EAG response to C6-alcohols than to the alcohols with longer or shorter chains, and it also showed higher EAG responses to synthetic C6-alcohols (hexanol, (*Z*)-3-hexenol and (*E*)-2-hexenol) than to C6-aldehyde (hexenal) and -acetate [(*E*)-3-hexenyl acetate]; however, the geometric isomers of 3-hexenol led to similar EAG amplitude of leafminers.[17] Despite these advances in electrophysiological studies, behavioral bioassays in laboratories and in fields are essential for exploring the roles of (*Z*)-3-hexenol in the host selection of herbivores. It has been suggested for long that (*Z*)-3-hexenol can significantly reduce the preference and performance of aphids.[18] In

*Correspondence to: Le Kang; Email: lkang@ioz.ac.cn
Submitted: 12/10/10; Accepted: 12/10/10
DOI: 10.4161/psb.6.3.14452

contrast, the adult emerald ash borer, *Agrilus planipennis*, was more attractive to the (*Z*)-3-hexenol containing purple prism traps than to the unbaited controls; supplement of other C6-volatiles such as (*Z*)-3-hexenal or (*Z*)-3-hexenyl acetate in binary or ternary combinations, did not show any synergistic effect.[19] In another study, the mixture of C6-volatiles ((*Z*)-3-hexenol and (*Z*)-3-hexenyl acetate) and benzaldehyde in a natural ratio, but not individual compound, attracted more female fruit moth, *Cydia molesta*.[20] Furthermore, the preference analysis of adult flies or moths for JA-deficient and wild-type (WT) plants revealed that C6-volatiles, including (*Z*)-3-hexenol, stimulate the behavioral preferences of these herbivores for host plants.[10,11] In addition to adult, larval herbivores also respond to (*Z*)-3-hexenol. The neonate larvae of Asian corn borer *Ostrinia furnacalis* showed repellent behavior to the synthetic (*Z*)-3-hexenol,[21] while the Lepidopteran herbivore's larvae *Manduca sexta* preferred wild-type (WT) *Nicotiana attenuate* plants over GLV-deficient mutants, suggesting that C6-volatiles are able to elicit the feeding of these insects.[22] Although it is difficult to conclude whether (*Z*)-3-hexenol is an attractant or a repellent, accumulating evidences suggested that (*Z*)-3-hexenol is, at least in part, important plant-derived infochemical that modulates the behavior of herbivorous insects, and the release of (*Z*)-3-hexenol should be the defensive responses of the plants.

Response of Natural Enemies to (*Z*)-3-hexenol

C6-alcohols have long been involved in indirect plant defense in tritrophic interactions. Extensive evidences imply that nearly all the natural enemies can perceive and positively respond to these volatile compounds. A pioneer study revealed that *Manduca sexta* larvae-induced release of (Z)-3-hexenol, linalool and cis-a-bergamotene functioned as indirect defense by attracting predators, which resulted in increasing herbivore mortality in the field.[23] Subsequently, with the combined techniques of chemical analysis (GC-MS), electrophysiological detection (GC-EAD) and behavioral bioassays, several studies showed that (*Z*)-3-hexenol is not only the most common inducible chemical in 10 plant species from seven families when they were wounded by herbivore insects or physical damage, but also the most important infochemical to attract parasitic wasps compared with other inducible chemicals and their blends.[5,24] Moreover, these results proved that the reduced release of (Z)-3-hexenol in JA-deficient tomato mutant (*spr2*) led to decreased level of attractiveness and parasitism by leafminer's parasitoid compared with WT plants, confirming the distinct role of (Z)-3-hexenol in the host finding of parasitoids.[11] A recent study demonstrated that the oral secretion of larval *Manduca sexta* is able to rapidly convert the Z-isomers of hexenal, hexenol and hexenyl acetate to the corresponding E-isomers, and the (*Z*)/(*E*) ratio changes increase the foraging success of the generalist hemipteran predator *Geocoris* spp. in the field.[25] These studies clearly indicated the innate responses of natural enemies to C6-volatiles. In fact, the odor learning by natural enemies is also an important mechanism for host location. The synchronization of biological rhythms in the tritrophic interactions of Lima bean, leafminer and parasitoid showed that the parasitoid emergence coincided with (Z)-3-hexen-ol emission, and the naive wasps displayed strong preference for this chemical. In contrast, the oviposition-experienced wasps preferred both (*Z*)-3-hexenol- and terpene-dominated volatiles released in the peak time of leafminer larvae feeding under the photoperiod of light:dark regimes, which optimized their parasitizations.[26] The results suggested that the feeding of leafminer, the defense chemical biosynthesis of plant, and the activities of parasitoid in this system can achieve rhythmic synchronization under natural light-dark photoperiod, but not under constant lightness or darkness.

(*Z*)-3-hexenol in Plant-plant Communication

In addition to the defensive role in tritrophic interaction, airborne (*Z*)-3-hexenol from wounded plants is also proposed to trigger the pre-defense responses of neighboring unattacked plants; this phenomenon is called plant-plant communication or priming effect of volatiles. A novel study showed that after mechanical damage and caterpillar regurgitation, corn seedlings pre-exposed to synthetic (Z)-3-hexen-ol produced significantly higher level of JA and volatile sesquiterpenes than the seedlings without previous exposure.[27] Another study demonstrated that simultaneous exposure to (Z)-3-hexenol and ethylene increased the herbivore-induced volatile emissions in intact maize plants.[28] Recently, our group performed a transcriptional analysis of *Arabidopsis thaliana* defensive response to priming volatiles, tand the results showed that (Z)-3-hexenol can activate the gene expression in the defense related pathways of Arabidopsis (Zhang S, Wei J and Kang L, unpublished data). However, the exact role of (Z)-3-hexenol in priming process requires further investigation.

Perspective

Here, we reviewed the current knowledge about the defensive roles of (Z)-3-hexenol in tritrophic interactions and its priming effects in plant-plant communication. Benefit from the recent progress on plant-insect interactions using advanced chemical analysis and plant materials genetically modified in single trait of signaling pathways,[9-11,25] the functions of airborne (Z)-3-hexenol in plant defenses and communications become more and more clear. However, due to the limited number of available model systems (Arabidopsis, Nicotiana and Lima bean plants) and the relatively simple interactions among plants, herbivores and natural enemies, the exact mechanisms of volatile production, the olfactory responses of herbivores and natural enemies and the signal transduction and underlying molecular mechanisms of boarder communities and more complex systems still remain elusive. Therefore, other model systems need to be developed to understand the exact functions of plant-derived ubiquitous compounds in plant-insect interactions. In addition, a conflict in (Z)-3-hexenol activity is expected because it is potentially used by both herbivores and natural enemies to locate the host and host prey in multitrophic interactions under natural conditions. However, we believe that the major role of this compound is to mediate indirect defense and to prime neighboring plants. Thus, (Z)-3-hexenol can be used to develop insect pest control strategies. Further investigation on its quantitative and qualitative administration under natural condition is needed to elucidate the underlying mechanisms and to develop novel control strategies.

Acknowledgements

The research was supported by the Chinese Academy of Sciences (Project No: KSCX2-EW-N-05-03 and KSCX2-YW-N-045) and the National Nature Science Foundation of China (30921063 and 30970434).

References

1. Dudareva N, Negre F, Nagegowda DA, Orlova I. Plant volatiles: Recent advances and future perspectives. Crit Rev Plant Sci 2006; 25:417-40.
2. Paré PW, Tumlinson JH. De novo biosynthesis of volatiles induced by insect herbivory in cotton plants. Plant Physiol 1997; 114:1161-7.
3. Dicke M, Gols R, Ludeking D, Posthumus MA. Jasmonic acid and herbivory differentially induce carnivore-attracting plant volatiles in lima bean plants. J Chem Ecol 1999; 25:1907-22.
4. Thaler JS, Farag MA, Pare PW, Dicke M. Jasmonate-deficient plants have reduced direct and indirect defenses against herbivores. Ecol Lett 2002; 5:764-74.
5. Wei J, Wang L, Zhu J, Zhang S, Nandi OI, Kang L. Plants attract parasitic wasps to defend themselves against insect pests by releasing hexenol. PLoS ONE 2007; 2:852.
6. Kessler A, Baldwin IT. Plant responses to insect herbivory: The emerging molecular analysis. Annu Rev Plant Biol 2002; 53:299-328.
7. Browse J. Jasmonate: An oxylipin signal with many roles in plants. Vitam Horm 2005; 72:431-56.
8. Matsui K. Green leaf volatiles: hydroperoxide lyase pathway of oxylipin metabolism. Curr Opin Plant Biol 2006; 9:274-80.
9. Kessler A, Halitschke R, Baldwin IT. Silencing the jasmonate cascade: Induced plant defenses and insect populations. Science 2004; 305:665-8.
10. Halitschke R, Stenberg JA, Kessler D, Kessler A, Baldwin IT. Shared signals—'alarm calls' from plants increase apparency to herbivores and their enemies in nature. Ecol Lett 2008; 11:24-34.
11. Wei J, Wang L, Zhao J, Li C, Ge F, Kang L. Ecological trade-offs between jasmonic acid-dependent direct and indirect plant defenses in tritrophic interactions. New Phytol 2011; 189:557-67.
12. Feussner I, Wasternack C. The lipoxygenase pathway. Annu Rev Plant Biol 2002; 53:275-97.
13. Bruce TJA, Wadhams LJ, Woodcock CM. Insect host location: a volatile situation. Trends Plant Sci 2005; 10:269-74.
14. Chen HH, Zhao YX, Kang L. Comparison of the olfactory sensitivity of two sympatric steppe grasshopper species (Orthoptera : Acrididae) to plant volatile compounds. Sci China C Life Sci 2004; 47:115-23.
15. Kang L, Charlton R, Hopkins TL. olfactory sensitivity of the grasshopper, *Melanoplus sanguinipes*, to plant volatiles and their blends. Entomol Sinica 1995; 2:136-44.
16. Kang L, Hopkins TL. Behavioral and olfactory responses of grasshopper hatchlings, *Melanoplus sanguinipes*, to plant odours and volatile compounds. Chin Sci Bull 2004; 49:136-41.
17. Zhao YX, Kang L. Role of plant volatiles in host plant location of the leafminer, *Liriomyza sativae* (Diptera: Agromyzidae). Physiol Entomol 2002; 27:103-11.
18. Hildebrand DF, Brown GC, Jackson DM, Hamiltonkemp TR. Effects of Some leaf-emitted volatile compounds on aphid population increase. J Chem Ecol 1993; 19:1875-87.
19. Grant GG, Ryall KL, Lyons DB, Abou-Zaid MM. Differential response of male and female emerald ash borers (Col., Buprestidae) to (Z)-3-hexenol and manuka oil. J Appl Entomol 2010; 134:26-33.
20. Natale D, Mattiacci L, Hern A, Pasqualini E, Dorn S. Response of female *Cydia molesta* (Lepidoptera : Tortricidae) to plant derived volatiles. B Entomol Res 2003; 93:335-42.
21. Huang CH, Yan FM, Byers JA, Wang RJ, Xu CR. Volatiles induced by the larvae of the Asian corn borer (*Ostrinia furnacalis*) in maize plants affect behavior of conspecific larvae and female adults. Insect Sci 2009; 16:311-20.
22. Halitschke R, Ziegler J, Keinanen M, Baldwin IT. Silencing of hydroperoxide lyase and allene oxide synthase reveals substrate and defense signaling crosstalk in *Nicotiana attenuata*. Plant J 2004; 40:35-46.
23. Kang L, Chen B, Wei JN, Liu TX. Roles of thermal adaptation and chemical ecology in Liriomyza distribution and control. Annu Rev Entomol 2009; 54:127-45.
24. Kessler A, Baldwin IT. Defensive function of herbivore-induced plant volatile emissions in nature. Science 2001; 291:2141-4.
25. Allmann S, Baldwin IT. Insects betray themselves in nature to predators by rapid isomerization of green leaf volatiles. Science 2010; 329:1075-8.
26. Zhang SF, Wei JN, Guo XJ, Liu TX, Kang L. Functional synchronization of biological rhythms in a tritrophic system. PloS One 2010; 5:11064.
27. Engelberth J, Alborn HT, Schmelz EA, Tumlinson JH. Airborne signals prime plants against insect herbivore attack. Proc Natl Acad Sci USA 2004; 101:1781-5.
28. Ruther J, Kleier S. Plant-plant signaling: Ethylene synergizes volatile emission in *Zea mays* induced by exposure to (Z)-3-hexen-1-ol. J Chem Ecol 2005; 31:2217-22.