Commentary PCE

The emerging importance of microbial volatile organic compounds

Birgit Piechulla[1] & Jörg Degenhardt[2]

[1]University of Rostock, Institute for Biological Sciences, Albert-Einstein-Str. 3, D-18059 Rostock, Germany

[2]Martin Luther University Halle, Institute for Pharmacy, Hoher Weg 8, D-06120 Halle, Germany

Correspondence B. Piechulla, email: birgit.piechulla@uni-rostock.de

Volatiles are ubiquitously present on earth due to their physical and chemical properties (high vapour pressure, low boiling point, low molecular mass). It is obvious that volatiles are constituents of the atmosphere, but they also appear in porous material, e.g. plants, rocks, soil pores, and are dissolved in aqueous media. Volatiles originate from both anthropogenic and biogenic sources. While animal and plant volatile emissions have been comprehensively studied in the past, volatiles of microorganisms (i.e. bacteria and fungi) have been mostly neglected (Fig. 1). Only recently has the wealth of microbial volatile organic compounds (mVOCs) been discovered. Approximately 1000 mVOCs released by about 400 bacteria and fungi are described in the literature to date. These volatiles are summarized in the database ‚mVOC' which is publicly accessible (http://bioinformatics.charite.de/mvoc/, Lemfack et al. 2013). Considering that only about 10,000 microbial species are currently described, although at least a million species are expected to exist on earth, the VOC profiles of a surprisingly small number of microorganisms have been investigated so far. The more microorganisms we investigate in the future, the more volatiles with novel structures are likely to be discovered as this new research field expands (von Reuss et al. 2010).

Besides the elucidation of their chemical structures, unraveling the biological functions of mVOCs will be one of the major tasks in the future. Exposure of plants to mVOCs containing the methyl esters of the plant hormones salicylic acid and

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: doi/10.1111/pce.12254

jasmonic acid, and the phytohormone ethylene can alter gene expression in plants (Ping and Boland, 2004). Most likely, these mVOCs interfere with the plant's regulatory pathways of the respective hormones. The growth-promoting activity of several *Bacillus* strains was attributed to the volatiles acetoin and 2,3–butanediol, Exposure of *Arabidopsis* mutants with deficiencies in hormonal regulatory pathways suggested that these volatiles interfere with the cytokinin and ethylene signaling pathways. Since these pathways form a tightly interwoven network, the growth, defense reactions, and systemic induced resistance of the plant are affected (Ping and Boland, 2004). About ten years ago, Ryu et al. (2003) showed that 2,3–butanediol was a key compound conferring plant resistance. Recently, the precursor acetoin was also demonstrated to trigger induced systemic resistance against *Pseudomonas syringae* in *Arabidopsis thaliana* (Rudrappa et al. 2010). Other bitrophic mVOC based interactions in which bacteria or fungi affect plant growth, the movement and survival of nematodes, and inter-species interactions between bacteria and fungi in the soil, result in morphological and phenotypical alterations of the receiving organism (reviewed in Effmert et al. 2012). There are also instances of mVOCs being closely associated with insect feeding behaviors (Davis et al. 2013). Volatiles of *Staphylococcus aureus* attract the Mexican fruit fly *Anastrepha ludens* (Robacker and Flath 1995), and the predatory hoverfly is attracted by bacteria of honeydew pea aphids (Leroy et al. 2011).

An additional piece of evidence for the complex mVOC functional network is presented in this issue of Plant, Cell & Environment by D'Alessandro et al. (2013). They found that volatile 2,3–butanediol released by endophytic bacteria like *Enterobacter aerogenes* does not only increase pathogen resistance of maize, but also decreases its resistance against the herbivorous larvae of *Spodoptera littoralis*. The altered state of the plant can also affect the interaction with the parasitic wasp *Cotesia marginiventris*, a predator of the *S. littoralis* larvae (D'Alessandro et al., 2013).

These investigations illustrate that microbial volatiles play important biological roles in multitrophic interactions. They have to be considered in ecological experiments and ecosystem analysis, since microorganisms are ubiquitous in the biosphere and sterility as such does not exist in nature (Fig.1). As our knowledge of intra- and interorganismal mVOC-based interactions gradually increases, volatile perception, signal transduction and phenotypical responses in the receiver organisms need to be investigated and elucidated in detail. To better understand the regulatory networks in the plant, Wenke et al. (2012) analyzed molecular and biochemical alterations in *A. thaliana* co-cultivated with rhizobacteria. They observed the activation of defense genes along with WRKY transcription factors that mediate these responses. Additional experiments also indicated the presence of WRKY-independent pathways that are activated by mVOC exposure. These results are not astonishing, since some bacteria have the capability to emit complex mixtures of compounds (Kai et al. 2010). It will be a challenge to identify the respective bioactive mVOCs, determine their biologically relevant concentrations, and assess whether they act as individual compounds or in mixtures. Since mVOC synthesis is a dynamic process coupled to the metabolic activity of the microorganism, the morphological, phenotypic and molecular reactions of the plants can be the result of contrasting actions of mVOCs, e.g. 2,3-butanediol promotes plant growth while dimethyldisulfate inhibits growth.

Most likely, we have presently discovered only the ‚tip of the iceberg' of mVOCs and volatile-mediated interactions in bi- or multitrophic networks of organisms that are found aboveground and belowground (Fig. 1). The central task for the future will be the elucidation of a plethora of bacterial and fungal VOCs and their biological and ecological roles. This will include far-reaching spatiotemporal dynamics as well as environmental perturbations. Therefore, this research on mVOCs may lead to the discovery and development of novel agricultural tools.

References

D'Alessandro M., Erb M., Ton J., Brandenburg A., Karlen D., Zopfi J. & Turlings TCJ. (2013) Volatiles produced by soil-borne endophytic bacteria increase plant pathogen resistance and affect tritrophic interactions, *Plant Cell Environment*, in press.

Davis T.S., Crippen T.L., Hofstetter R.W. & Tomberlin J.K. (2013) Microbial volatile emissions as insect semiochemicals. *Journal of Chemical Ecology* **39**, 687-806.

Effmert U., Kalderás J., Warnke R. & Piechulla B. (2012) Volatile mediated interactions between bacteria and fungi in the soil. *Journal of Chemical Ecololgy* **38**, 665-703.

Kai M., Crespo E., Cristescu S.M., Harren F.J.M., Francke W. & Piechulla B. (2010) Serratia odorifera: analysis of volatile emission and biological impact of volatile compounds on Arabidopsis thaliana. *Appl Microbiol Biotechnol* **88**, 965–976.

Lemfack M.C., Nickel J., Dunkel M., Preissner R. & Piechulla B. (2013) mVOC: a database of microbial volatiles. *Nucleic Acid Research*, in press

Leroy P.D., Sabri A., Heuskin S., Tronart P., Lognay G., Verheggen F.J., Francis F., Bostaux Y., Felton G.W. & Haubruge E. (2011) Microorganisms from aphid honeydew attract and enhance the effivacy of natural enemies. *Nat Commun* **2**, 348.

Ping L. & Boland W. (2004) Signals from the underground: bacterial volatiles promote growth in Arabidopsis. *Trends in Plant Science* **9**, 263-266.

Robacker D.C. & Flath R.A.(1995) Attractans from *Staphylococcus aureus* cultures for mexican fruit fly, *Anastrepha ludens*. *Journal of Chemical Ecology* **21**, 1861-1874.

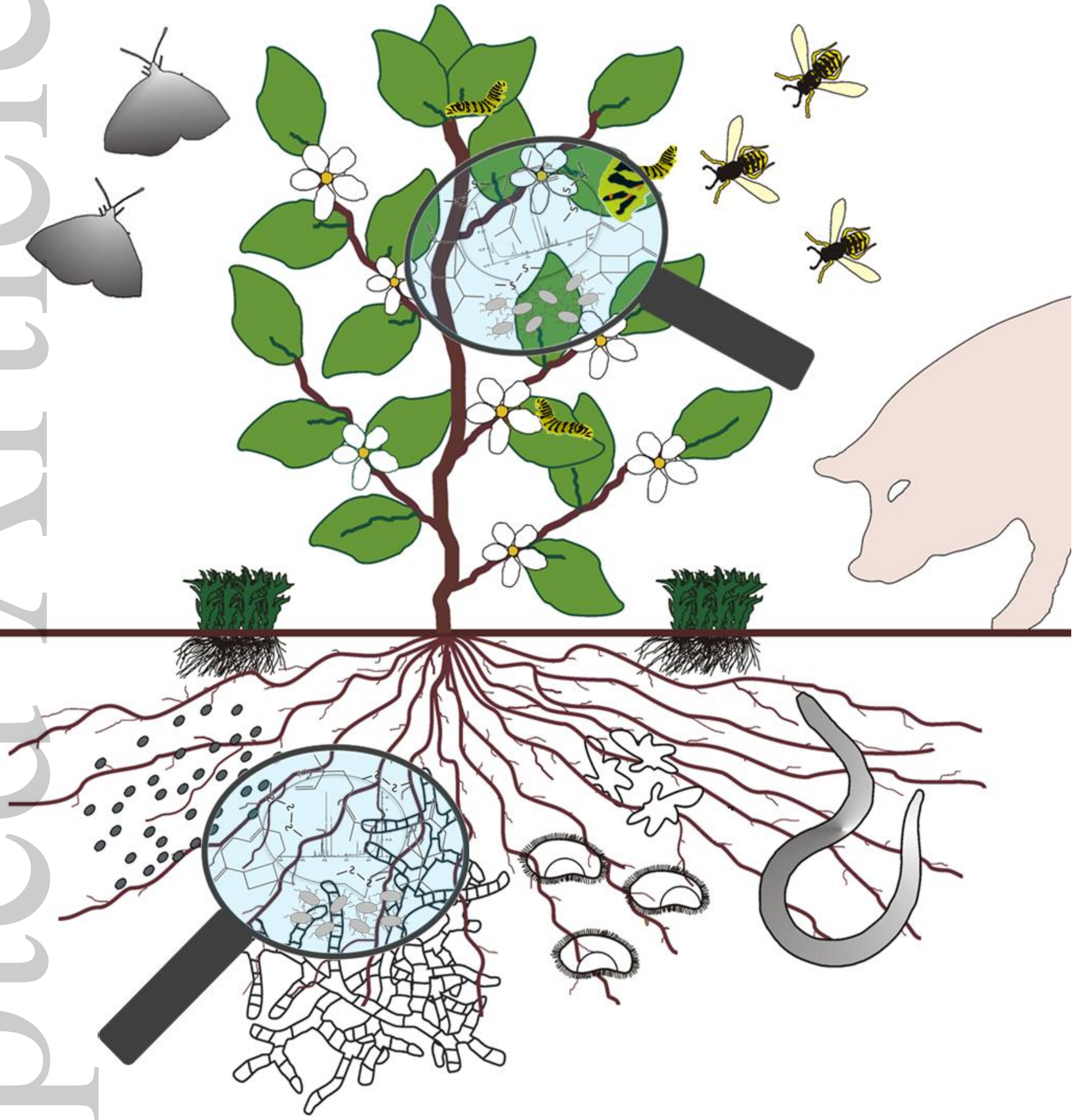
Ryu C.M., Farag M.A., Hu C.H. Reddy M.S., Wie H.X., Pare P.W. & Kloepper J.W. (2003) Bacterial volatiles promote growth in *Arabidopsis*. *Proceedings of National Academy of Science U.S.A* **100**, 4927–4932.

von Reuss S.H., Kai M., Piechulla B. & Francke W. (2010) Octamethylbicyclo[3.2.1]octadienes from *Serratia odorífera. Angewandte Chemie* **122**, 2053-2054.

Wenke K., Wanke D., Kilian J., Berendzen K., Harter K. & Piechulla B. (2012) Volatiles of two growth-inhibiting rhizobacteria commonly engage AtWRKY18 function. *Plant J* **70**, 445-459.

Figure legend

Figure 1: Schematic presentation of various organisms (belowground: plant root, bacteria, fungi, ciliates, amoeba, nematodes; aboveground: vertebrates, (e.g. wild boar), plant leaves and flowers, invertebrates (e.g. caterpillars, butterflies, bees and flies)) involved in bi- and multitrophic volatile-based interactions. Magnifying glasses indicate the universal presence of bacteria and fungi and their potential for mVOC production (drawn by Marco Kai and Uta Effmert).



pce_12254_f1