

LETTER

Volatile communication between plants that affects herbivory: a meta-analysis

Richard Karban,^{1*} Louie H. Yang,¹
and Kyle F. Edwards²

Abstract

Volatile communication between plants causing enhanced defence has been controversial. Early studies were not replicated, and influential reviews questioned the validity of the phenomenon. We collected 48 well-replicated studies and found overall support for the hypothesis that resistance increased for individuals with damaged neighbours. Laboratory or greenhouse studies and those conducted on agricultural crops showed stronger induced resistance than field studies on undomesticated species, presumably because other variation had been reduced. A cumulative analysis revealed that early, non-replicated studies were more variable and showed less evidence for communication. Effects of habitat and plant growth form were undetectable. In most cases, the mechanisms of resistance and alternative hypotheses were not considered. There was no indication that some response variables were more likely to produce large effects. These results indicate that plants of diverse taxonomic affinities and ecological conditions become more resistant to herbivores when exposed to volatiles from damaged neighbours.

Keywords

Eavesdropping, induced resistance, plant behaviour, plant signalling, volatiles.

Ecology Letters (2014) 17: 44–52

INTRODUCTION

When some plants are attacked by herbivores, they release chemical cues that cause other individuals to change their traits and become more resistant to herbivory. Communication between plants was first observed and reported more than 30 years ago (Rhoades 1983; Baldwin & Schultz 1983) and the number of reported cases has grown rapidly in the recent past (Heil & Karban 2010). We consider a process to be plant volatile communication if it involves signalling by a plant that causes a response in the same or a different individual that receives the cue. Emission or display of a cue is plastic and the response of the receiver is conditional on receiving the cue. We require that emitting the cue could potentially benefit the emitter, although this has proven difficult or impossible to establish (Karbon 2008). For an interaction to be considered communication, the responder must have the choice of responding to the cue or not, a requirement which excludes allelopathy (Schenk & Seabloom 2010). We make no assumptions about the intended target of the cues; many plants use volatile cues to co-ordinate their own defences against herbivores when one branch is attacked and other branches on the same individual respond by increasing defences (Karbon *et al.* 2006; Frost *et al.* 2007; Heil & Silva Bueno 2007; Rodriguez-Saona *et al.* 2009). There is no agreed upon definition of communication; our use of the term is broader than most and includes phenomena that some authors prefer to call eavesdropping or signalling.

Early reports of plant communication met with great interest from scientists and the popular press. David Rhoades

observed that caterpillars placed on willow trees near damaged neighbours grew less well than those placed on trees near undamaged neighbours (Rhoades 1983). He hypothesised that the reduction in performance was caused by airborne communication from the damaged trees that increased resistance in neighbours. However, he was unable to repeat his initial results and his experimental design lacked true replication (D. Rhoades, pers. comm., Fowler & Lawton 1985). In addition, the poor performance of caterpillars on trees in close proximity to infested neighbours could have been caused by the introduction of insect pathogens rather than by communication between plants. Early experiments conducted on plants in growth chambers reported that plants exposed to volatiles coming from chambers containing feeding herbivores became more resistant, but the experimental designs used in these studies also lacked true replication (Baldwin & Schultz 1983; Bruin *et al.* 1992).

Following an influential review by Simon Fowler and John Lawton, most ecologists decided that communication between plants was a phenomenon that had been considered and debunked and that the phenomenon did not occur in nature (Fowler & Lawton 1985; Dicke & Bruin 2001). In addition to the limitations associated with experimental design of the early studies, communication between plants that benefited neighbours did not make sense to many ecologists. Early descriptions of this phenomenon were referred to as ‘talking trees’ by both the popular press and some scientists in the field. Natural selection would not be expected to favour the emission of cues that provided neighbouring competitors with information about herbivores. Coinciding with the resurgence

¹Department of Entomology, University of California, Davis, CA, 95616, USA

²Kellogg Biological Station, Michigan State University, 3700 E. Gull Lake Dr., Hickory Corners, MI, 49060, USA

*Correspondence: E-mail: rkarban@ucdavis.edu

in scientific interest in communication has been a resolution of this apparent contradiction. 'Talking trees' is a particularly poor jargon for these phenomena since 'listening trees' may more accurately reflect the process and should be more in keeping with how evolution might work. It is well accepted that plants respond to environmental cues including light and nutrients to make allocation decisions about the plastic growth and abscission of shoots and roots (Ballare 1999). It is easy to imagine selection favouring individuals that adjust their allocation decisions and their defences based on diverse sources of reliable information that they acquire from their environments, including eavesdropping on the wounding responses of neighbouring plants.

This ecological phenomenon almost got lost because it was first over-sensationalised by the popular press and then it became taboo for scientists to consider. D. Rhoades (pers. comm.) was unable to secure funding to continue his work in this field and he left science. Interest in communication between plants revived after the publication of several well-documented and more carefully controlled cases in 2000 (Arimura *et al.* 2000; Birkett *et al.* 2000; Dolch & Tscharrntke 2000; Karban *et al.* 2000) along with more sympathetic reviews (e.g. Dicke & Bruin 2001). Over the past few years, communication between plants has been more widely accepted, and a recent review listed 14 studies (Heil & Karban 2010). However, this literature has recently expanded, and not all of the studies have found evidence for communication that affects herbivory (Fig. 2).

In some cases, communication between individuals may be co-opting a process that originally evolved to allow plants to co-ordinate their own systemic responses. For many plants, vascular connections among plant modules (branches, roots, etc.) are greatly limited, and systemic signals cannot spread throughout the vasculature of an individual (Waisel *et al.* 1972; Orians 2005). For individuals of these species, volatile communication may represent a more effective and rapid means of signalling among different organs within a single individual (Farmer 2001; Karban *et al.* 2006; Heil & Adame-Alvarez 2010). Furthermore, the relatively short distances over which volatile signals are effective increases the likelihood that signals will only provide information to other tissues on the same individual as the emitter. Many plants reproduce vegetatively or with limited dispersal so that individuals that are spatially close neighbours are more likely to share genes. Plant populations that are viscous in this way can more easily evolve traits that benefit responders since emitters are more likely to benefit by increasing the inclusive fitness of closely related neighbours (Hamilton 1964). In addition, if individuals are able to recognise kin and respond differently to cues from kin compared to strangers, then cooperative behaviours such as communication are more likely to evolve. Recent empirical results supported this hypothesis; communication was more effective among closely related sagebrush individuals than among distantly related individuals (Karbon *et al.* 2013).

Many of the volatile chemicals that plants emit when they have been damaged by herbivores produce a diversity of ecological consequences, any of which could help to explain their selective advantage (Penuelas & Llusia 2004). For example, volatile emissions protect plants from heat and oxidative

stresses (Loreto & Velikova 2001; Behnke *et al.* 2007). The volatiles released by herbivory are often directly repellent to herbivores (Bernasconi *et al.* 1998; De Moraes *et al.* 2001) and allelopathic to plant competitors (Karbon 2007; Inderjit *et al.* 2009). These same volatiles attract the predators and parasites of herbivores that can decrease levels of damage inflicted by herbivory (Dicke & Sabelis 1988; Turlings *et al.* 1990; Thaler 1999; Kessler & Baldwin 2001). However, there is still surprisingly little convincing evidence that predators responding to volatile cues released by damaged plants actually increase the fitness of those plants under natural conditions (Allison & Hare 2009; Kessler & Heil 2011). Theoretical concerns that plant communication contradicts evolutionary common sense have largely been addressed in recent years.

We have conducted a meta-analysis of the published and unpublished studies available to us that have considered communication between plants that affect their interactions with herbivores. We have addressed these specific questions: (1) Does volatile plant communication increase resistance of plants to herbivory, on average? (2) Which response variables are affected by volatile plant communication? (3) Under what ecological conditions is volatile plant communication observed? (4) Is there a publication bias in reporting plant communication involving volatile cues?

THE DATA SET

The data set was compiled by conducting keyword searches in the ISI Web of Science up to June 2012, by collecting studies from recent reviews, and by contacting colleagues who are actively working in the this field. We included 48 studies in the meta-analysis that met the following criteria: (1) plants were subjected to at least two treatments – exposure to a volatile cue and a control; (2) the authors provided means, some measure of variance, and sample sizes for each treatment group; (3) there were at least two independent replicates of each treatment group; (4) the study tested for an effect of these treatments on herbivores or plant damage caused by herbivory. We did not require that the treatments be randomly assigned so that we included 'natural experiments' in which the authors observed differences among groups but could be less confident about cause and effect relationships. When studies were pseudoreplicated, we reduced the number of replicates reported by the authors but still included the study in the meta-analysis, albeit weighted less heavily. However, studies were excluded if they had only one true replicate per treatment. We did not include studies documenting responses to herbivore-induced plant volatiles involving predators and parasites or indirect plant responses that benefited these higher trophic levels, as this subject has been reviewed frequently elsewhere (e.g. Allison & Hare 2009; Kessler & Heil 2011). Studies of associational resistance in which a volatile cue was not likely to be induced by experimental treatments were not included. There were many cases in which multiple experiments conducted at different sites, at different times, or involving closely related response variables were available. In these cases, we included only a single pair of means for the two treatments. We reported mean values over time and space when these data were presented by the authors or could be

easily calculated. However, publications involving different emitter species, different receiver species, or different and unrelated response variables were considered different ‘studies’ and were included in the analysis.

Our final data set consisted of 48 studies published (and unpublished) between 1983 and 2012. Our literature search ended in June 2012. Our data set included 33 plant species in 15 different families.

META-ANALYSIS

We performed a meta-analysis using the log response ratio to quantify effect size across the experiments in our data set. The log response ratio is defined as $\log(x_T/x_C)$, where x_T is the treatment mean for plants exposed to volatiles and x_C is the control mean. To incorporate the fact that uncertainty in effect size varies across experiments, we calculated the sample variance for the log response ratio after Hedges *et al.* (1999). There are several potential sources of variation in effect size in the data set: sampling error, quantified using the sample variance; variation among species, some of which were used in multiple experiments (‘Species’); variation among studies, some of which included experiments on multiple species (‘Study’); variation due to phylogenetic relatedness, which was approximated using taxonomic family as a predictor (‘Family’); variation due to the predictors of interest which we described below; and variation due to other unknown causes (‘Other’). Sampling error was accounted for by specifying *a priori* sample variances for each experiment, which effectively weighed effect sizes by their variance. Other between-experiment heterogeneity was accounted for with an experiment-level random effect and effects of Species, Family and Study were all accounted for using additional random effects. Thus, our meta-analysis was structured as a multilevel random effects model (Gelman & Hill 2006) that accounted for multiple sources of random variation while also testing for hypothesised predictors of effect size. Preliminary analyses indicated that Species and Family did not explain significant variation in effect size, and these were excluded from further analyses in order to reduce the number of parameters in the model.

We were interested in exploring how multiple potential predictors may affect the responses of plants to volatile cues: laboratory vs. field experiments, damage type, plant growth form, habitat characteristics, conspecific vs. heterospecific cues, and the type of response that was measured. However, this resulted in a large number of potential predictors relative to the number of experiments in the data set, and several of these predictors were partially collinear with each other. For example, most of the studies that used artificial damage to induce the emission of cues (32 of 35 observations) were conducted in the field, while most of the studies that used insect damage were conducted in the laboratory (nine of 13 observations). Consistent with these issues of predictor collinearity, a model that included all predictors found that no single predictor explained significant variation in effect size, once the other predictors had been accounted for (see Table S1). Therefore, we chose to test each of the predictors using ‘univariate’ models in which only the focal predictor was included, along with the sampling error and random effects

terms described above (Other and Study). Thus, a key caveat of our results is that the effects of these partially collinear predictors cannot be fully distinguished by our results. Nonetheless, we can describe which factors were associated with variation in effect sizes, and our results suggest future experiments that can better disentangle these patterns. In these univariate models, we excluded those studies for which we could not calculate a log response ratio. Studies with continuous independent factors, studies without replication, and studies that did not specify the type of damage to plants were all excluded from these further analyses.

The models were fit using the Bayesian mixed models package MCMC glmm (Hadfield 2010), in R version 2.15.2 (R Development Core Team 2012). MCMC chains were sampled 10^6 times, saving every 100th sample to eliminate autocorrelation. For binary predictors (e.g. laboratory vs. field), we report 95% highest posterior density (HPD) credible intervals for the difference in effect size between the two categories. For categorical predictors with more than two levels, we quantified the variation explained by this factor using the standard deviation across factor levels (Gelman & Hill 2006), and we reported the 95% HPD credible interval for this standard deviation.

RESULTS AND DISCUSSION

Volatile communication between plants is common

Of the 48 observations in this data set, 39 of them showed evidence for volatile-induced plant resistance to herbivory, eight showed evidence for induced susceptibility, and one study showed an effect size of zero. Overall, our MCMC analysis indicated significant evidence for volatile-induced plant resistance to herbivory (Fig. 1, posterior mean log response ratio effect size = -0.31 , 95% CI = -0.44 to -0.18 , pMCMC < 0.001). An analysis that controlled for non-independence of multiple observations described in a single citation found the same result (posterior mean log response ratio effect size = -0.44 , 95% CI = -0.63 to -0.25 , pMCMC < 0.001). Plant species and plant family were also included as random factors in additional models but we did not find that some plant taxa were more likely to respond to volatile cues of herbivory than others.

Cumulative meta-analysis allows researchers to determine whether the mean effect size changes over time (Leimu & Koricheva 2004). For example, a cumulative meta-analysis of studies of induced plant responses to herbivory revealed that early studies reported large effect sizes but as the paradigm of induced resistance became better accepted by the field, mean effect size decreased and more studies reported finding no significant differences caused by induction (Nykanen & Koricheva 2004). A historical view of the data in this meta-analysis showed that evidence for a general pattern of volatile-induced plant resistance to herbivory accumulated gradually in the literature from 1983 to the present (Fig. 1). Observations from several early studies were not properly replicated but were influential and controversial (Rhoades 1983; Baldwin & Schultz 1983; Myers & Williams 1984). While some early studies suggested induced susceptibility (Rhoades 1983), the

Rhoades 1983; *Salix sitchensis*
 Rhoades 1983; *Salix sitchensis*
 Rhoades 1983; *Salix sitchensis*
 Baldwin and Schultz 1983; *Acer saccharum*
 Baldwin and Schultz 1983; *Populus euroamericana*
 Williams & Myers 1984; *Alnus rubra*
 Williams & Myers 1984; *Alnus rubra*
 Myers & Williams 1984; *Alnus rubra*

Bruin et al. 1992; *Gossypium hirsutum*
 Hildebrand et al. 1993; *Solanum lycopersicum*
 Hildebrand et al. 1993; *Nicotiana tabacum*
 Dolch and Tschardtke 2000; *Alnus glutinosa*
 Dolch and Tschardtke 2000; *Alnus glutinosa*
 Tschardtke et al. 2001; *Alnus glutinosa*
 Tschardtke et al. 2001; *Alnus glutinosa*
 Karban et al. 2003; *Nicotiana attenuata*
 Karban et al. 2003; *Nicotiana attenuata*
 Karban et al. 2003; *Nicotiana attenuata*
 Engelberth et al. 2004; *Zea mays*
 Engelberth et al. 2004; *Zea mays*
 Karban et al. 2004; *Lomatium dissectum*
 Karban et al. 2004; *Lupinus polyphyllus*
 Karban et al. 2004; *Valeriana californica*
 Kessler et al. 2006; *Nicotiana attenuata*
 Heil and Silva Bueno 2007; *Phaseolus lunatus*
 Heil and Silva Bueno 2007; *Phaseolus lunatus*
 Ton et al. 2007; *Zea mays*
 Karban 2007; *Wyethia mollis*
 Frost et al. 2008; *Populus deltoides x nigra*
 Shiojiri and Karban 2008a; *Artemisia tridentata*
 Shiojiri and Karban 2008b; *Artemisia cana*
 Shiojiri and Karban 2008b; *Artemisia douglasiana*
 Rodriguez-Saona et al. 2009; *Vaccinium corymbosum*
 Peng et al. 2010; *Brassica oleracea*
 Peng et al. 2010; *Brassica oleracea*
 Ramadan et al. 2011; *Zea mays*
 Pearse et al. 2012; *Achyrrachaena mollis*
 Pearse et al. 2012; *Achyrrachaena mollis*
 Pearse et al. 2012; *Lupinus nanus*
 Pearse et al. 2012; *Lupinus nanus*
 Pearse et al. 2012; *Sinapis arvensis*
 Pearse et al. 2012; *Sinapis arvensis*
 Pearse et al. 2013; *Salix exigua*
 Pearse et al. 2013; *Salix lemmonii*
 Savchenko et al. 2013; *Arabidopsis thaliana*
 Ishizaki, Shiojiri, Karban unpublished; *Wyethia mollis*
 Karban et al unpublished; *Artemisia tridentata*
 Shiojiri unpublished; *Solidago canadensis*
 Shiojiri unpublished; *Cardiocrinum cordatum*
 Shiojiri unpublished; *Pachysandra terminalis*
 Shiojiri unpublished; *Anthriscus aemula*
 Shiojiri unpublished; *Viola odorata*
 Shiojiri unpublished; *Taraxacum officinale*
 Shiojiri unpublished; *Lathyrus japonica*
 Shiojiri unpublished; *Rosa rugosa*
 Shiojiri unpublished; *Artemisia indica*

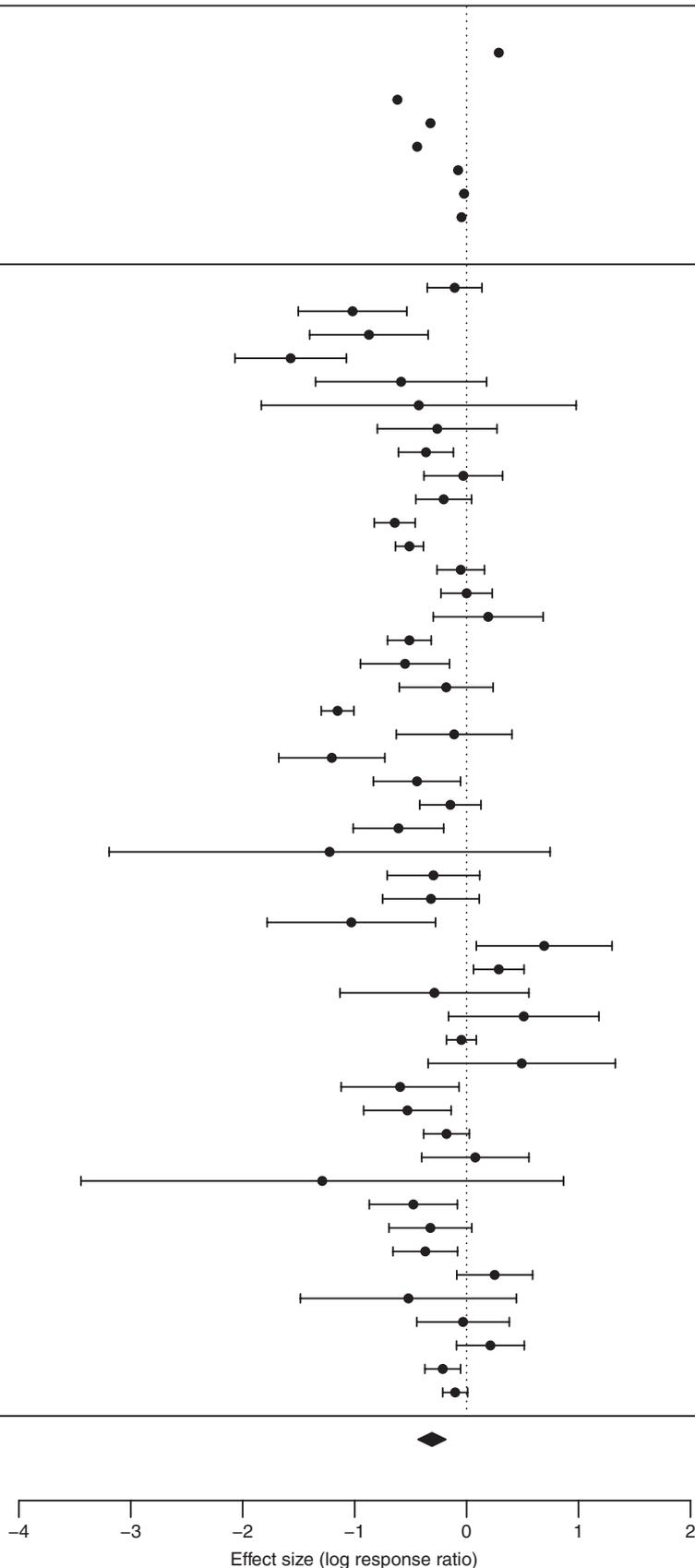


Figure 1 Log response ratios for studies of volatile communication affecting resistance. Negative effect sizes (log response ratios) indicate induced resistance, whereas positive values indicate induced susceptibility. Horizontal error bars indicate 95% credible intervals; studies lacking error bars were not replicated and were not included in the meta-analysis although they were influential in the development of the field. References for these studies are listed in the Supporting Information.

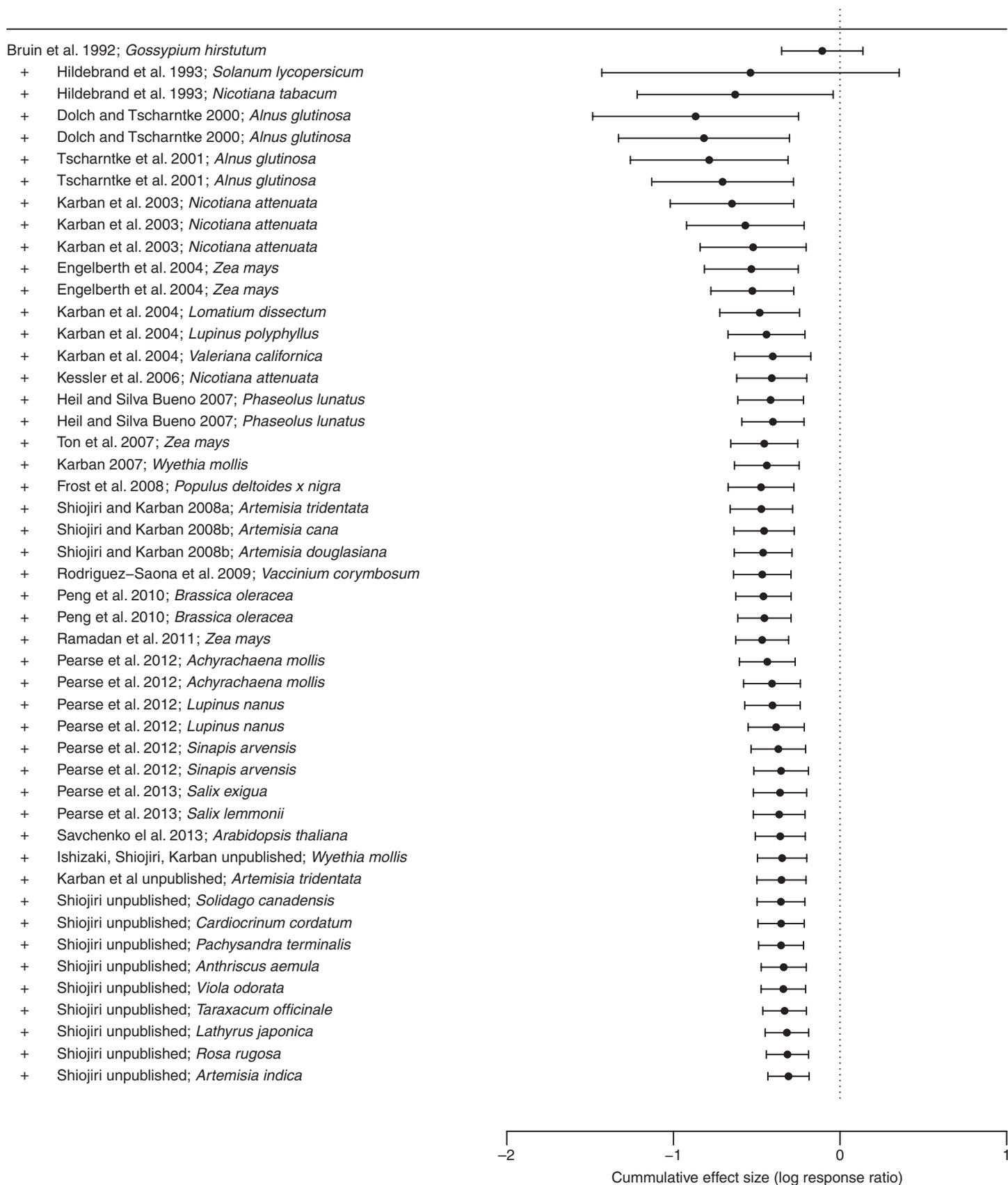


Figure 2 Cumulative effect size (log response ratios) for all studies that adds cumulative effect sizes chronologically. Error bars show 95% credible intervals for studies with replication. Negative effect sizes with error bars that do not include 0 indicate significant effects of induced resistance.

overall weight of subsequent studies showed a significant general pattern of volatile-induced plant resistance (Fig. 2). Early critics of this field correctly pointed out that the first studies were pseudo-replicated (Fowler & Lawton 1985). They speculated that the evidence for communication affecting induced resistance could have been caused by lack of true replication. However, a meta-analysis that included only studies with true replication yielded stronger support for the overall pattern of volatile communication producing induced resistance than one that also included the early pseudo-replicated studies (Fig. 1, analyses not presented).

These results (Fig. 1) indicate that plant communication cannot be regarded as an aberrant phenomenon that is observed only occasionally under unusual circumstances. When ecologists have looked for this phenomenon, they have found evidence supporting its existence. Some of these researchers started their experiments expecting to find such evidence; others were extremely sceptical. This meta-analysis includes far more studies than previous reviews. However, communication that affects herbivory is not ubiquitous. Some studies failed to find it and others found that exposure to volatile cues from damaged neighbours made plants more susceptible to their herbivores, rather than more resistant (positive effects in Fig. 1). Induced susceptibility may result from 'inappropriate' plant responses when tradeoffs exist in resistance to several different challenges or from herbivores successfully manipulating plants responses (e.g. gall makers). Further examination of these examples should provide valuable insights into control of plant signalling.

What response variables were measured?

It is useful to clarify exactly what this significant result indicates. Most of the studies included in the meta-analysis measured plant damage as a response to volatile cues. This response variable is both relatively easily to measure and includes a variety of steps – herbivores must locate the host plant, find it acceptable, choose to consume it, and so on. In only a few of the studies included in the meta-analysis are actual plant characteristics known that ultimately influence plant resistance and even in the best studied cases, our knowledge of the mechanisms of resistance is incomplete. For example, tomato plants that were exposed to cues from experimentally wounded sagebrush neighbours increased their accumulations of proteinase inhibitors, chemicals known to interfere with herbivore digestion (Farmer & Ryan 1990). However, accumulation of proteinase inhibitors is only one part of a much more complex induced response, and the internal chemical environment of the tomato leaf can largely inactivate proteinase inhibitors (Duffey & Felton 1989). For many of the plants included in this meta-analysis, we lack evidence implicating any particular defensive mechanism. In summary, we know that plants receiving volatile cues experience reduced levels of damage from herbivores, but we do not understand the mechanisms leading to this enhanced resistance.

Most (83%) of the studies included in the meta-analysis reported that herbivory was reduced for plants that were exposed to volatile cues. In some cases, chemicals implicated in plant defence have been measured, but rarely have the

researchers evaluated alternative hypotheses. For example, volatiles emitted by experimentally damaged plants can directly repel herbivores rather than, or in addition to, causing changes in resistance levels of neighbours (Bernasconi *et al.* 1998; De Moraes *et al.* 2001). In contrast, the volatiles emitted by experimentally clipped sagebrush reduced damage experienced by neighbours but had no direct repellent effects on grasshoppers, the herbivores responsible for a lot of this damage (Karban & Baxter 2001). However, direct repellence has not been examined in most experiments as an alternative mechanism.

What conditions favour volatile communication between plants?

In addition to asking whether this phenomenon occurs, we were interested in understanding the conditions that may have favoured plants that respond to volatile cues. In order to address hypotheses associating resistance mediated by volatiles with various conditions, we conducted univariate analyses within the meta-analysis.

We predicted that if variation due to other sources makes it difficult to detect relatively small effects due to communication, studies done under uniform environmental conditions or those involving genetically uniform plants may be more likely to show significant effects. This line of reasoning led to the prediction that studies conducted in the laboratory will show stronger effects of communication than those conducted in the field. Similarly, we predicted that studies of genetically more homogeneous agricultural crops will show stronger effects of communication than those conducted on genetically diverse natural plants. Laboratory studies generally tended to report stronger evidence of volatile-induced plant resistance to herbivory than field studies did (Fig. 3, posterior mean effect = -0.37 , 95% CI = -0.73 to 0.01 , pMCMC = 0.05). Interpretation of this result may be confounded because most of the studies in the laboratory used real insects to damage plants (9 of 13), while most of studies in the field used artificial damage (32 of 35). Studies using actual insects to damage plants produced stronger effects than those that used artificial damage (Fig. 3, posterior mean effect = -0.27 , 95% CI -0.56 to 0.03 , pMCMC = 0.07). Similarly, agricultural systems showed a non-significant tendency towards stronger induced resistance than natural systems (Fig. 3, posterior mean effect = -0.25 , 95% CI = -0.66 to 0.13 , pMCMC = 0.21).

Systemic induced resistance among tissues of individuals can be strongly constrained by the vascular architecture ('plumbing') of the plant (Viswanathan & Thaler 2004; Orians 2005). Plant species vary considerably in the extent and importance of these vascular constraints (sectoriality). Woody species that are more constrained have greater xylem vessel size and lower vessel density than more integrated species (Zanne *et al.* 2006). Species that had higher tolerance to drought and wind were more constrained while those that had higher tolerance to flooding and shade were less constrained by vascular architecture. As a result, woody species from arid environments were predicted to be less able to rely on vascular signalling and more likely to require volatile communication to co-ordinate systemic responses (Waisel *et al.* 1972; Zanne *et al.* 2006). We found little support for these predic-

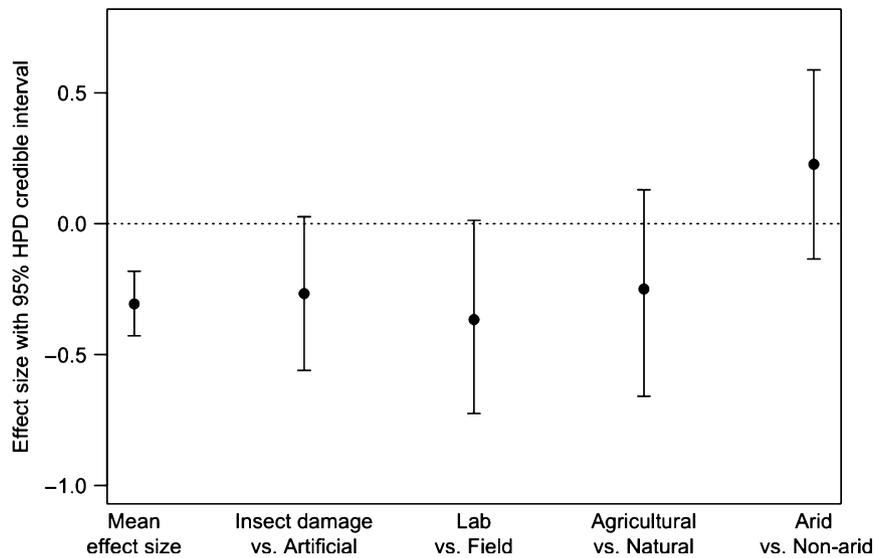


Figure 3 Univariate tests of various factors that influence effect sizes (log response ratios) of communication. Negative values indicate induced resistance associated with the first of the paired factors, positive values indicate induced susceptibility, and error bars are 95% highest posterior density (HPD) credible intervals.

tions based on habitat or plant growth form. Plants from dry habitats tended to show more induced susceptibility than those from more mesic habitats (Fig. 3, posterior mean effect = 0.22, 95% CI = -0.13 to 0.59, pMCMC = 0.22). Although this effect was not significant, it was in the opposite direction from our prediction about the distribution of volatile communication. We detected no trend in the likelihood of woody plants showing volatile-induced resistance compared to non-woody plants (posterior mean effect = -0.10, 95% CI = -0.33 to 0.44, pMCMC = 0.41). Similarly, perennial plants were no more likely to show evidence of volatile-induced resistance than annuals (posterior mean effect = 0.09, 95% CI = -0.31 to 0.49, pMCMC = 0.65).

There have been some suggestions that communication between plants may have evolved to allow plants to coordinate their own systemic responses and that the signalling system allowing for within-plant communication has been co-opted for between-plant signalling (Karbon *et al.* 2006; Frost *et al.* 2007; Heil & Adame-Alvarez 2010). If this has been the case, we predicted that volatile cues would be more effective between conspecifics than between heterospecific plants because conspecifics are more similar genetically. However, we found no evidence that effects of volatile signalling were stronger between conspecifics than between heterospecifics (posterior mean effect = -0.04, 95% CI = -0.44 to 0.33, pMCMC = 0.83). One possible explanation for this surprising result is that some of the volatiles that plants use to communicate are highly conserved and many species respond to the same cues.

As mentioned above, this meta-analysis includes studies that measure effects on several kinds of response variables: performance of insect herbivores as well as plant responses, particularly the percentage of leaves that have been damaged. The effect of volatile cues on induced resistance was not significantly different when plant measurements or insect measure-

ments were used to assess induced responses (posterior mean effect = 0.23, 95% CI = -0.18 to 0.60, pMCMC = 0.24). In fact, considering the great variety of response variables that were included in this analysis, it is surprising that any clear signal was visible through all of the noise.

Is there publication bias?

Researchers conducting meta-analyses often attempt to determine whether the results that are reported in the literature present an unbiased sample of the population of results or whether investigators chose to publish only those results that were significant or supported their preconceived hypotheses

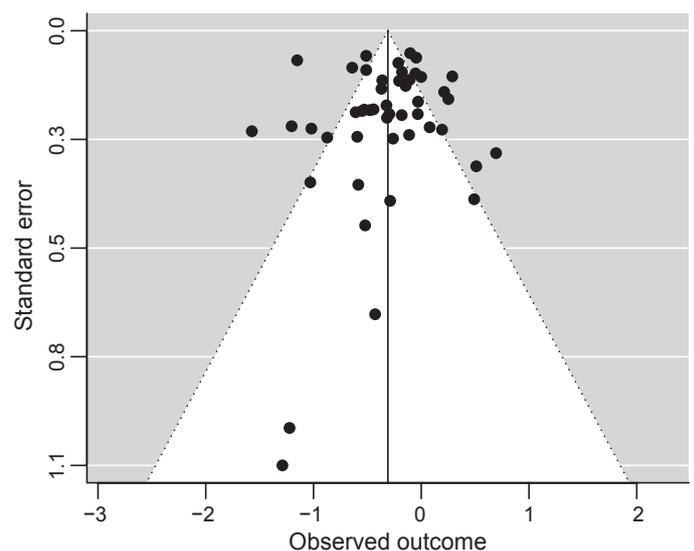


Figure 4 A funnel plot showing the distribution of effect sizes and standard errors.

about nature. One way to evaluate this 'file-drawer problem' and other reporting biases is to use funnel plots that compare effect size and SE (Palmer 1999). An unbiased set of observations should be symmetrical around the true effect size, which was the case for the data set reported here (Fig. 4). However, the absence of points in the lower right of the funnel plot suggests the possibility that studies with small sample sizes and large SE that failed to find induced resistance or found induced susceptibility may have been under-reported.

CONCLUSIONS

Many studies involving diverse plants reported evidence of volatile communication resulting in increased resistance to herbivore attack, indicating that this is a widespread natural phenomenon. Unlike early studies of this phenomenon, the studies considered in this review were well-replicated with independent sampling units.

Many of these studies did not identify mechanisms involved, even whether a plant response was responsible for the effects. Alternative hypotheses, such as direct repellency of herbivores by volatiles, were often not ruled out and would be worth considering in future studies. Determining the plant responses involved, particularly the volatile cues that were responsible will be well worth future effort. As expected, conditions that minimised background variation, particularly laboratory studies and studies of genetically homogeneous crop species, were more likely to detect significant effects of volatile cues on induced resistance. Future studies are needed to separate effects due to experimental conditions (laboratory vs. field) from those caused by different inducing agents (herbivores vs. artificial damage). Surprisingly, woody plants from arid habitats were no more likely to show evidence of volatile communication than plants of other growth forms or habitats. Negative results are difficult to interpret and more studies are required to evaluate these tentative conclusions. If plants commonly use volatile cues to regulate their defences, agriculturalists may be able to manipulate plant resistance to pests using these cues. Future studies should explore the possibility of using volatile cues in production agriculture as a means of regulating defences of crops.

Volatile communication between plants is an interesting ecological finding because it suggests that plants share information with other nearby individuals. Hence, plants are less isolated and independent than previously assumed. Many plants perceive volatile cues in their environments and respond to those cues by changing their defences against herbivores.

ACKNOWLEDGEMENTS

We thank Kaori Shiojiri and Satomi Ishizaki for sharing unpublished results with us and Mikaela Huntzinger, Ian Pearse and anonymous referees for improving the manuscript.

AUTHORSHIP

RK organised the study and wrote the manuscript, LHY and KFE conducted statistical analyses, prepared the figures and edited the manuscript.

REFERENCES

- Allison, J.D. & Hare, J.D. (2009). Learned and naive natural enemy responses and the interpretation of volatile organic compounds as cues or signals. *New Phytol.*, 184, 768–782.
- Arimura, G., Ozawa, R., Shimoda, T., Nishioka, T., Boland, W. & Takabayashi, J. (2000). Herbivory-induced volatiles elicit defence genes in lima bean leaves. *Nature*, 406, 512–513.
- Baldwin, I.T. & Schultz, J.C. (1983). Rapid changes in tree leaf chemistry induced by damage: evidence for communication between plants. *Science*, 221, 277–279.
- Ballare, C.L. (1999). Keeping up with the neighbours: phytochrome sensing and other signalling mechanisms. *Trends Plant Sci.*, 4, 97–102.
- Behnke, K., Ehrling, B., Teuber, M., Louis, S., Hansch, R., Polle, A. *et al.* (2007). Transgenic, non-isoprene emitting poplars don't like it hot. *Plant J.*, 51, 485–499.
- Bernasconi, M.L., Turlings, T.C.J., Ambrosetti, L., Bassetti, P. & Dorn, S. (1998). Herbivore-induced emissions of maize volatiles repel the corn leaf aphid, *Rhopalosiphum maidis*. *Entomol. Exp. Appl.*, 87, 133–142.
- Birkett, M.A., Campbell, C.A.M., Chamberlin, K., Guereri, E., Hick, A.J., Martin, J.L. *et al.* (2000). New roles for cis-jasmone as an insect semiochemical and in plant defense. *Proc. Natl Acad. Sci.*, 97, 9329–9334.
- Bruin, J., Dicke, M. & Sabelis, M.W. (1992). Plants are better protected against spider mites after exposure to volatiles from infested conspecifics. *Experientia*, 48, 525–529.
- De Moraes, C.M., Mescher, M.C. & Tumlinson, J.H. (2001). Caterpillar-induced nocturnal plant volatiles repel conspecific females. *Nature*, 410, 577–580.
- Dicke, M. & Bruin, J. (2001). Chemical information transfer between plants: back to the future. *Biochem. Syst. Ecol.*, 29, 981–994.
- Dicke, M. & Sabelis, M.W. (1988). How plants obtain predatory mites as bodyguards. *Neth. J. Zool.*, 38, 148–165.
- Dolch, R. & Tschirntke, T. (2000). Defoliation of alders (*Alnus glutinosa*) affects herbivory by leaf beetles on undamaged neighbors. *Oecologia*, 125, 504–511.
- Duffey, S.S. & Felton, G.W. (1989). Plant enzymes in resistance to insects. In: *Biocatalysis in Agricultural Biotechnology*. (eds Whitaker, J.R., Sonnet, P.E.). American Chemical Society, Toronto, pp. 289–313.
- Farmer, E.E. (2001). Surface-to-air signals. *Nature*, 411, 854–856.
- Farmer, E.E. & Ryan, C.A. (1990). Interplant communication: airborne methyl jasmonate induces synthesis of proteinase inhibitors. *Proc. Natl Acad. Sci.*, 87, 7713–7716.
- Fowler, S.V. & Lawton, J.H. (1985). Rapidly induced defenses and talking trees - the devil's advocate position. *Am. Nat.*, 126, 181–195.
- Frost, C.J., Appel, H.M., Carlson, J.E., De Moraes, C.M., Mescher, M.C. & Schultz, J.C. (2007). Within-plant signalling via volatiles overcomes vascular constraints on systemic signalling and primes responses against herbivores. *Ecol. Lett.*, 10, 490–498.
- Gelman, A. & Hill, J. (2006). *Data Analysis Using Regression and Multilevel/Hierarchical Models*. Cambridge University Press, Cambridge, UK.
- Hadfield, J.D. (2010). MCMC methods for multi-response generalized linear mixed models: the MCMCglmm R package. *Journal of Statistical Software* 33, 1–22.
- Hamilton, W.D. (1964). The genetical evolution of social behaviour. *J. Theor. Biol.*, 7, 1–52.
- Hedges, L.V., Gurevitch, J. & Curtis, P.S. (1999). The meta-analysis of response ratios in experimental ecology. *Ecology*, 80, 1150–1156.
- Heil, M. & Adame-Alvarez, R.M. (2010). Short signalling distances make plant communication a soliloquy. *Biol. Lett.*, 6, 843–845.
- Heil, M. & Karban, R. (2010). Explaining evolution of plant communication by airborne signals. *Trends Ecol. Evol.*, 25, 137–144.
- Heil, M. & Silva Bueno, J.C. (2007). Within-plant signaling by volatiles leads to induction and priming of an indirect plant defense in nature. *Proc. Natl Acad. Sci.*, 104, 5467–5472.
- Inderjit, von Dahl, C.C. & Baldwin, I.T. (2009). Use of silenced plants in allelopathy bioassays: a novel approach. *Planta*, 229, 569–575.

- Karban, R. (2007). Experimental clipping of sagebrush inhibits seed germination of neighbors. *Ecol. Lett.*, 10, 791–797.
- Karban, R. (2008). Plant behaviour and communication. *Ecol. Lett.*, 11, 727–739.
- Karban, R. & Baxter, K.J. (2001). Induced resistance in wild tobacco with clipped sagebrush neighbors: the role of herbivore behavior. *Insect Behav.*, 14, 147–156.
- Karban, R., Baldwin, I.T., Baxter, K.J., Laue, G. & Felton, G.W. (2000). Communication between plants: induced resistance in wild tobacco plants following clipping of neighboring sagebrush. *Oecologia*, 125, 66–71.
- Karban, R., Shiojiri, K., Huntzinger, M. & McCall, A.C. (2006). Damage-induced resistance in sagebrush: volatiles are key to intra- and interplant communication. *Ecology*, 87, 922–930.
- Karban, R., Shiojiri, K., Ishizaki, S., Wetzel, W.C. & Evans, R.Y. (2013). Kin recognition affects plant communication and defence. *Proc. Biol. Sci.*, 280, 20123062. DOI: 10.1098/rspb.2012.3062.
- Kessler, A. & Baldwin, I.T. (2001). Defensive function of herbivore-induced plant volatile emissions in nature. *Science*, 291, 2141–2144.
- Kessler, A. & Heil, M. (2011). The multiple faces of indirect defenses and their agents of natural selection. *Funct. Ecol.*, 25, 348–357.
- Leimu, R. & Koricheva, J. (2004). Cumulative meta-analysis: a new tool for detection of temporal trends and publication bias in ecology. *Proc. Biol. Sci.*, 271, 1961–1966.
- Loreto, F. & Velikova, V. (2001). Isoprene produced by leaves protects the photosynthetic apparatus against ozone damage, quenches ozone products, and reduces lipid peroxidation of cellular membranes. *Plant Physiol.*, 127, 1781–1787.
- Myers, J.H. & Williams, K.S. (1984). Does tent caterpillar attack reduce the food quality for red alder foliage? *Oecologia*, 62, 74–79.
- Nykanen, H. & Koricheva, J. (2004). Damage-induced changes in woody plants and their effects on insect herbivore performance: a meta-analysis. *Oikos*, 104, 247–268.
- Orians, C. (2005). Herbivores, vascular pathways, and systemic induction: facts and artifacts. *J. Chem. Ecol.*, 31, 2231–2242.
- Palmer, A.R. (1999). Detecting publication bias in meta-analyses: a case study of fluctuating asymmetry and sexual selection. *Am. Nat.*, 154, 220–233.
- Penuelas, J. & Llusia, J. (2004). Plant VOC emissions: making use of the unavoidable. *Trends Ecol. Evol.*, 19, 402–404.
- Rhoades, D.F. (1983). Responses of alder and willow to attack by tent caterpillars and webworms: evidence for phenomonal sensitivity of willows. In: *Plant Resistance to Insects, Symposium Series 208*. (ed Hedin, P.A.). American Chemical Society, Washington D. C., pp. 55–68.
- Rodriguez-Saona, C.R., Rodriguez-Saona, L.E. & Frost, C.J. (2009). Herbivore-induced volatiles in the perennial shrub, *Vaccinium corymbosum*, and their role in inter-branch signaling. *J. Chem. Ecol.*, 35, 163–175.
- Schenk, H.J. & Seabloom, E.W. (2010). Evolutionary ecology of plant signals and toxins: a conceptual framework. In: *Plant Communication from an Ecological Perspective*. (eds Baluska, F., Ninkovic, V.). Springer-Verlag, Berlin, pp. 1–19.
- Team, R.D.C. (2012). *A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria.
- Thaler, J.S. (1999). Jasmonate-inducible plant defences cause increased parasitism of herbivores. *Nature*, 399, 686–688.
- Turlings, T.C.J., Tumlinson, J.H. & Lewis, W.J. (1990). Exploitation of herbivore-induced plant odors by host-seeking parasitic wasps. *Science*, 250, 1251–1253.
- Viswanathan, D.V. & Thaler, J.S. (2004). Plant vascular architecture and within-plant spatial patterns in resource quality following herbivory. *J. Chem. Ecol.*, 30, 531–543.
- Waisel, Y., Liphshitz, N. & Kuller, Z. (1972). Patterns of water movement in trees and shrubs. *Ecology*, 53, 520–523.
- Zanne, A.E., Sweeney, K., Sharma, M. & Orians, C.M. (2006). Patterns and consequences of differential vascular sectoriality in 18 temperate tree and shrub species. *Funct. Ecol.*, 20, 200–206.

SUPPORTING INFORMATION

Additional Supporting Information may be downloaded via the online version of this article at Wiley Online Library (www.ecologyletters.com).

Editor, Rebecca Irwin

Manuscript received 25 July 2013

First decision made 2 September 2013

Manuscript accepted 3 October 2013