

## DAMAGE-INDUCED RESISTANCE IN SAGEBRUSH: VOLATILES ARE KEY TO INTRA- AND INTERPLANT COMMUNICATION

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**Abstract.** Airborne communication between individuals, called “eavesdropping” in this paper, can cause plants to become more resistant to herbivores when a neighbor has been experimentally clipped. The ecological relevance of this result has been in question, since individuals may be too far apart for this interaction to affect many plants in natural populations. We investigated induced resistance to herbivory in sagebrush, *Artemisia tridentata*, caused by experimental clipping of the focal plant and its neighbors. We found no evidence for systemic induced resistance when one branch was clipped and another branch on the same plant was assayed for naturally occurring damage. In this experiment, air contact and plant age were not controlled. Previous work indicated that sagebrush received less damage when a neighboring upwind plant within 15 cm had been experimentally clipped. Here we found that pairs of sagebrush plants that were up to 60 cm apart were influenced by experimental clipping of a neighbor. Furthermore, we observed that most individuals had conspecific neighbors that were much closer than 60 cm. Air contact was essential for communication; treatments that reduced airflow between neighboring individuals, either because of wind direction or bagging, prevented induced resistance. Airflow was also necessary for systemic induced resistance among branches within an individual. Reports from the literature indicated that sagebrush is highly sectorial, as are many desert shrubs. Branches within a sagebrush plant do not freely exchange material via vascular connections and apparently cannot rely on an internal signaling pathway for coordinating induction of resistance to herbivores. Instead, they may use external, volatile cues. This hypothesis provides a proximal explanation for why sagebrush does not demonstrate systemic induced resistance without directed airflow, and why airborne communication between branches induces resistance.

**Key words:** *Artemisia tridentata*; communication; eavesdropping; herbivory; induced resistance; plant signal; sagebrush; sectoriality; volatile methyl jasmonate.

### INTRODUCTION

Communication between plants is a phenomenon of undetermined ecological importance. Laboratory results have convincingly demonstrated that markers of resistance to herbivores increased for focal plants in sealed jars sharing air contact with experimentally induced neighboring plants (e.g., Farmer and Ryan 1990, Arimura et al. 2000). However, the relevance of these laboratory findings to naturally occurring plant defenses remained unclear. Recent field experiments with two natural systems involving multiple species combinations indicate that plants sometimes become more resistant when growing near other individuals that have been damaged. Alder trees that were near attacked conspecific neighbors became more resistant to their primary herbivore, the alder leaf beetle (Dolch and Tscharrntke 2000, Tscharrntke et al. 2001). The importance and

ecological consequences of this interaction must still be determined.

Wild tobacco plants that were near experimentally clipped sagebrush suffered less leaf loss than tobacco near unclipped sagebrush. Experiments blocking air contact and soil contact indicated eavesdropping that required an airborne cue (Karban et al. 2000). Clipped sagebrush was found to release relatively large quantities of volatile methyl jasmonate that peaked four hours after wounding, although it is still unclear if methyl jasmonate is the active cue (Preston et al. 2004). Downwind neighbors became more resistant within the first five days of exposure to volatiles and remained resistant for the remainder of the season (Karban 2001). Over five seasons, tobacco plants near clipped sagebrush neighbors suffered 16–48% less leaf loss to herbivores than controls with unclipped neighbors (Karban and Maron 2002). This reduction in herbivory translated into increases in fruit production of 3–528% for tobacco plants very near clipped sagebrush neighbors (Karban and Maron 2002). However, when distances between sagebrush and tobacco were increased beyond 15 cm,

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eavesdropping between these two species was not detectable, suggesting that eavesdropping may have limited ecological relevance (Karban et al. 2003). Thus, the fitness increases were realized by few tobacco plants in nature, since most individuals did not grow in close enough proximity to sagebrush and those that did were adversely affected by competition (Karban and Maron 2002).

Both tobacco and sagebrush individuals are most likely to have conspecifics as nearest neighbors. Wild tobacco plants showed no evidence of communication among conspecific individuals in the field (Karban et al. 2003). Neither oxidative enzyme activity (a chemical marker of induced resistance) nor levels of leaf damage were affected by experimentally clipping neighboring tobacco plants. In contrast, sagebrush individuals near clipped sagebrush neighbors experienced approximately half the leaf loss to herbivores over the summer as plants with unclipped sagebrush neighbors (Karban et al. 2004). This effect was detected 14 days after clipping and persisted over the entire season. Most of this leaf loss was caused by generalist grasshoppers and mammals; a specialist chrysomelid beetle was unaffected by clipping neighboring sagebrush. We chose pairs of sagebrush plants growing within 15 cm of each other in that preliminary experiment and did not determine the range of distances over which communication between sagebrush individuals occurs.

Previous work had not established whether sagebrush exhibits systemic induced resistance among branches within an individual when one branch has been experimentally damaged. Many plants are capable of responding systemically to herbivores using chemical signals that move internally through vascular connections (Karban and Baldwin 1997, De Bruxelles and Roberts 2001), although a growing number of species are being found to be composed of branches that are not well integrated (Watson and Casper 1984, Watson 1986, Vuorisalo and Hutchings 1996, Orians and Jones 2001). Such plants exhibit only localized, sectorial exchange of nutrients, hormones, and secondary chemicals thought to mediate interactions between plants and their attackers (Davis et al. 1991, Shulaev 1995, Orians et al. 2000, 2002). Limited vascular connections also have been shown to constrain induced responses to herbivores (Orians 2005). Only those leaves or branches with direct vascular connections to damaged tissue became more resistant to herbivores (Jones et al. 1993, Viswanathan and Thaler 2004). Sagebrush is known to be highly sectorial, so that localized pruning of branches results in root stunting only immediately beneath the clipped branches; other branches are not affected (Cook and Stoddart 1960). This pattern led Cook and Stoddart to conclude "that sagebrush reacts in the same manner as a straight-grained tree" and that "the sagebrush plant separates into a number of apparently self-supporting units." Airborne cues emitted by damaged sagebrush

may be more effective at generating systemic resistance than cues transmitted internally.

Here we asked the following questions: (1) Does sagebrush exhibit systemic induced resistance throughout a plant when one branch is experimentally clipped? (2) How far are nearest conspecific neighbors of sagebrush individuals? (3) Over what distances can we detect communication between sagebrush neighbors that affects resistance to herbivory? In other words, combining questions 2 and 3, do most sagebrush individuals experience cues from neighbors that could produce meaningful effects of communication? (4) Is airflow necessary for communication to affect resistance between branches within an individual? (5) Is airflow necessary for communication to affect resistance between different individuals?

#### METHODS

Sagebrush (*Artemisia tridentata*) is the dominant plant of the Great Basin region of western North America (see Plate 1). It comprises over 90% of the plant biomass at some sites, particularly those grazed by livestock (Pickford 1932, Young et al. 1988). Our experiments were conducted at two field sites dominated by sagebrush along the interface of the Sierra Nevada range and the Great Basin. Our first site was on the south flood plain of Convict Creek at the Sierra Nevada Aquatic Research Laboratory (SNARL) near Mammoth Lakes, California (37°36'57" N, 118°49'47" W), at an elevation of 2160 m. In our first induction experiment at SNARL (question 1), we did not take wind direction or plant age into account. This site is walled in by steep glacial moraines so that wind flow is consistently downslope, from the west. This topography allowed us to control which plants were upwind or downwind of others in subsequent experiments of communication (questions 3–5). Sagebrush plants used in our communication experiments at SNARL were relatively similar in age, having germinated as the unintended consequence of watering during the summers of 1997–1999 and at the edge of a prescribed burn conducted by the U.S. Forest Service on 7 January 1999 (R. Karban, *personal observation*). Our second site was on the floodplain of Sagehen Creek (Sagehen) near Truckee, California (39°26.67' N, 120°11.30' W) at an elevation of 1830 m. Wind direction is not consistent at Sagehen.

Sagebrush has an extensive list of herbivores (Wiens et al. 1991) including vertebrates and insects of almost every feeding guild. Sagebrush at both study sites experiences relatively increased folivory during the spring (at the time of snowmelt) and again during the autumn by deer, *Odocoileus hemionus*. Folivory is relatively less intense during the summer and is primarily by grasshoppers. The most common summer folivore at both sites is the grasshopper *Cratypedes neglectus*, although many other grasshopper species feed on sagebrush. At SNARL, other common grasshopper species include *Trimerotropis fontana*, *Conozoa sulci-*



PLATE 1. Sagebrush is the most abundant and the defining plant of the Great Basin biome of North America. It grows in nearly monospecific stands of densely packed individuals with canopies that extend close to those of neighbors (see Fig 3). Photo credit: K. Shiojiri.

*frons*, *Cordillacris occipitalis*, *Cratypedes lateritius*, and *Melanoplus sanguinipes*, while *Trimerotropis fontana* and *Camnula pellucida* are abundant at Sagehen.

*Does sagebrush exhibit systemic induced resistance?*

We tested the hypothesis of systemic induced resistance by selecting 52 large sagebrush plants at SNARL. All plants had at least two major branches that were connected aboveground. One branch was haphazardly designated as the treatment branch and the other as the assay branch. We did not control or record the orientation of the branches with respect to wind direction or distance between their canopies; distances ranged from  $\sim 1$  to 150 cm. On 17 July 2005, we clipped the distal edge of all leaves of the treatment branch from 26 plants with scissors and left the treatment branch unclipped for the other 26 control plants. On 2 August we recorded the proportion of leaves on the assay branch of each plant that had been damaged by naturally occurring herbivory. In order to meet the assumptions of ANOVA, we transformed these proportions using the arcsine transformation, although figures report the actual proportions. We compared the mean proportion of leaves with herbivore damage on assay

branches of plants with experimentally clipped or unclipped treatment branches.

*What proportion of individuals communicate?*

In order to begin to assess whether communication between sagebrush individuals is important in nature, we surveyed a population of plants and determined how far apart their nearest conspecific neighbor grew. We then compared this distribution to an assessment of the distance over which communication could be detected in the field. One hundred focal plants were selected at 20-m intervals along a 2-km transect that ran parallel to Sagehen Creek in a dry meadow through habitat dominated by young sagebrush (5–15 years old). Every 20 m, the closest sagebrush plant was located, and the distance from that focal plant to its nearest conspecific neighbor was recorded.

To determine how far communication was effective, we selected and marked 250 pairs of young sagebrush plants with canopies that were close to one another (from touching to 1 m apart) at SNARL. Pairs were selected so that open space separated their foliage along a line oriented east to west, although they were not necessarily nearest neighbors in other directions. We

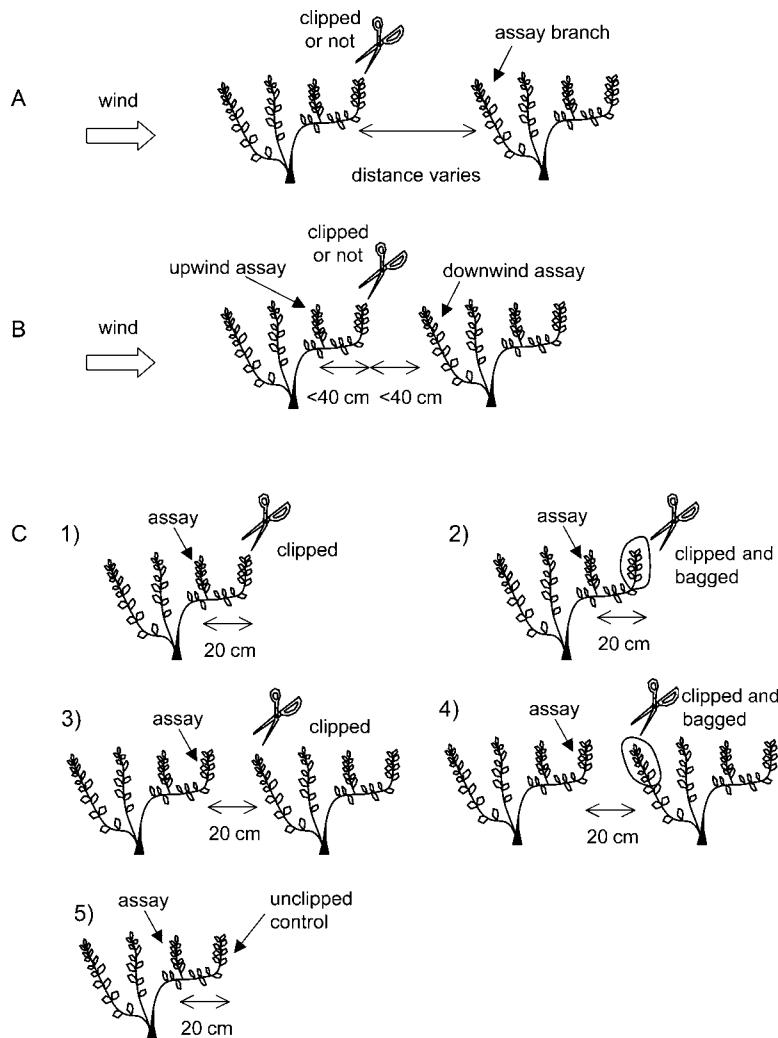


FIG. 1. Schematic diagram of experimental designs. (A) Experiment at Sierra Nevada Aquatic Research Laboratory (SNARL), where wind consistently blew from the west and the distance between plants was varied. (B) Experiment at SNARL in which herbivore damage was assayed on both an upwind branch on the focal plant and a downwind branch on a neighboring plant. (C) Experiment at Sagehen involving five treatments: (1) clipped branch and assay branch on the same plant, (2) clipped branch enclosed in a plastic bag and assay branch on the same plant, (3) clipped branch and assay branch on different plants, (4) clipped branch enclosed in a plastic bag and assay branch on different plants, (5) unclipped control branch and assay branch. Wind direction was not consistent during this experiment.

recorded the distance between each pair of plants. Pairs of plants were separated by at least 2 m from other pairs to minimize the possibility of contamination of treatments. We clipped foliage in early May, corresponding with snowmelt, from one branch of the upwind bush most adjacent to the downwind bush for half of the pairs (Fig. 1A). To make branches with clipped foliage, we removed most of the distal half of all leaves with scissors and allowed cut material to fall to the ground. The upwind bush of the other pairs was not clipped and these pairs served as controls. We recorded the number of leaves with herbivore damage on the adjacent branch of each downwind bush during June and July. Since branch size was an important factor affecting the number of damaged leaves, the total number of leaves on the

downwind branch was recorded and damage was standardized for a branch with 100 leaves. This damage assay was used in all of the subsequent experiments.

We compared the mean standardized number of leaves that were damaged by herbivores on branches with clipped or unclipped upwind neighbors at various distances. We suspected that neighbors within 20 cm might behave differently than those farther away. We divided the population of pairs of plants into three distance categories, with plants 0–20 cm, 21–60 cm, and >60 cm apart. We conducted a two-way ANOVA of the standardized number of damaged leaves with treatment (clipped or unclipped neighbor) and distances between plants (three categories) as main effects. Treatment means were compared a posteriori using Tukey's hsd.

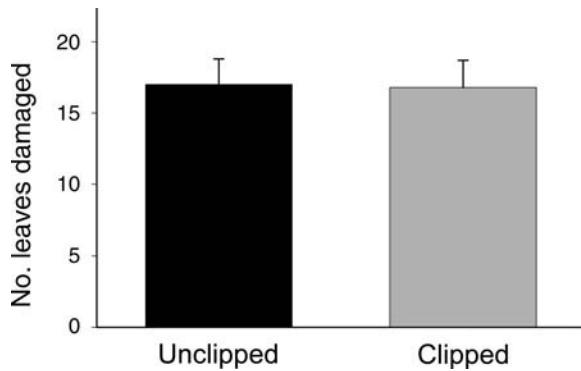


FIG. 2. Systemic induced resistance. Standardized number of leaves damaged (per 100 leaves) by herbivores on branches connected to focal branches that were either clipped or unclipped. Histogram bars show means and SE.

We did not conduct an ANCOVA with distance as a covariate because we could not describe the relationship between distance and number of damaged leaves with a linear model (Neter et al. 1996, Cottingham et al. 2005).

#### *Is air contact required?*

In order to test the hypothesis that volatile cues are required for communication, we took advantage of the fact that the prevailing wind at SNARL blows from one direction to compare communication between upwind and downwind pairs. Our a priori prediction was that we would detect communication in the downwind plant (branch) when the upwind plant (branch) was experimentally damaged, but not the reverse. We selected 240 pairs of branches for this experiment. Each pair had a focal branch that was randomly assigned to be clipped for half of the cases, or to be an unclipped control for the other half. As in the previous experiment, we clipped the foliage of one branch of those plants assigned to be clipped in May, coinciding with snowmelt. We assayed communication by comparing naturally occurring leaf damage during summer to assay branches that were either on *another* plant within 40 cm *downwind* of the focal plant or were within 40 cm *upwind* of the *same* focal plant (Fig. 1B). We performed a two-way ANOVA with clipping treatment and direction as crossed main effects. We tested our a priori prediction that only the branch downwind of clipped focal neighbors would experience reduced natural levels of herbivory with planned contrasts.

We conducted a second set of experiments that tested the hypothesis that airborne cues emitted by clipped sagebrush are more effective at generating systemic resistance than cues transmitted internally. We manipulated air contact between branches of the same and different plants using plastic bags to restrict airflow instead of relying on prevailing winds. These experiments were conducted at Sagehen, where wind direction is inconsistent. We selected 150 pairs of branches that

were within 20 cm of each other and randomly assigned 30 pairs to be in each of the following treatments (Fig. 1C). (1) We clipped foliage from one branch and assayed naturally occurring damage on another branch of that same plant. (2) We performed a treatment similar to the first treatment except that we blocked airflow by clipping foliage inside of a plastic bag and then sealed the bag around the clipped branch with a wire twist-tie. (3) We clipped foliage from one branch and assayed naturally occurring damage on a branch of a different plant. (4) We performed a treatment similar to the third treatment except that we blocked airflow from the clipped branch with a plastic bag. (5) We identified unclipped controls. We clipped the distal edge of approximately one-third of the leaves of each branch assigned to the clipping treatment on 23 June, 19 July, and 11 August. This clipping regime removed less leaf area each time compared to earlier experiments, but we clipped three times instead of once. We assayed herbivory on 28 September, later in the season than in previous experiments. If volatile communication were necessary for systemic induced resistance, we would have three a priori predictions: (1) blocking air contact will result in no systemic induced resistance (treatments 2 and 5 will not differ), (2) when air contact is allowed, systemic induced resistance will be detectable (treatment 1 will have less damage than treatments 2 and 5), and (3) when air contact is allowed, communication will cause induced resistance in a branch of a neighboring plant (treatment 3 will have less damage than treatments 4 and 5). We tested these predictions with planned contrasts following a one-way ANOVA.

Enclosing branches in plastic bags can cause unexpected consequences that are unrelated to blocking volatile cues. We evaluated whether such experimental artifacts could influence our response variable, herbivore damage to adjacent branches. We tested this possibility by enclosing one branch of 30 bushes in plastic bags on 8 June and recording damage on adjacent assay branches on 5 August. No branches were experimentally clipped in this experiment. We found no detectable artifacts of our bags on damage by herbivores to adjacent branches (standardized number of leaves with damage for branches adjacent to bagged branches =  $4.96 \pm 0.62$  leaves; for branches adjacent to unbagged control branches =  $5.43 \pm 0.63$  leaves;  $F_{1,58} = 0.30$ ,  $P = 0.59$ ).

## RESULTS

#### *Does sagebrush exhibit systemic induced resistance?*

We found no evidence of systemic induced resistance in sagebrush (Fig. 2). Levels of natural herbivore damage measured on assay branches were not detectably different for plants with experimentally clipped branches or unclipped branches ( $F_{1,51} = 0.02$ ,  $P = 0.90$ ). In this experiment, the assay branch and the treatment branch (clipped or unclipped) shared vascular connections aboveground, although the distance and orientation between branches was not controlled or recorded.

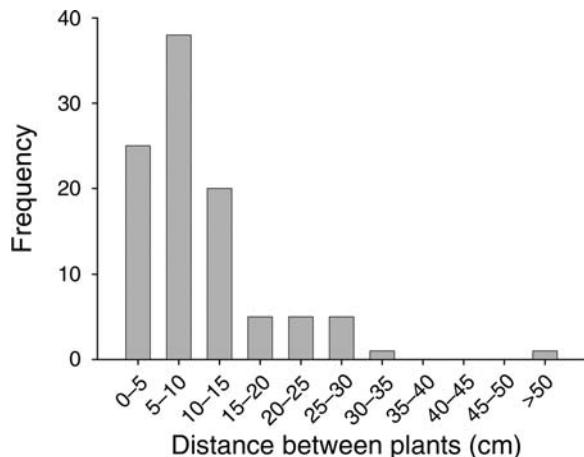


FIG. 3. The frequency distribution of distances between focal plants and their nearest conspecific neighbors at Sagehen.

*What proportion of individuals communicate?*

Most sagebrush plants at Sagehen had nearest neighbors that were relatively close together (Fig. 3). Most individuals (88/100) had a nearest neighbor within 20 cm and all individuals (100/100) had a nearest neighbor within 60 cm.

Since tobacco responded to cues released by sagebrush over distances of up to 10–20 cm (Karban et al. 2003), we suspected that communication between sagebrush might occur over similar distances. A post hoc visual inspection of leaf damage at SNARL suggested that damage to the downwind (assay) plant of the pairs broke into three distance categories: 0–20 cm, 21–60 cm, and >60 cm apart (Fig. 4). An analysis of the data categorized in this way revealed a significant effect of clipping the upwind plant on damage experienced by the downwind plant (clipping treatment  $F_{1,244} = 9.97$ ,  $P = 0.002$ ), although neither distance nor the interaction between distance and clipping were significant (distance  $F_{2,244} = 0.90$ ,  $P = 0.41$ , clipping  $\times$  distance  $F_{2,244} = 2.31$ ,  $P = 0.10$ ). The response to distance and clipping resembled a step function. When pairs were closer than 60 cm, the downwind neighbor experienced reduced damage relative to plants with unclipped neighbors (Fig. 4; Tukey’s hsd). However, when pairs were farther than 60 cm apart, there was no detectable effect of clipping the upwind neighbor on levels of damage experienced by the downwind plant.

*Is air contact required?*

As in the previous experiment, all plants experienced relatively little leaf herbivory when we compared branches upwind and downwind from clipped neighbors at SNARL. Of the 240 pairs that we started with, 222 were still alive at the end of summer. We compared the herbivore damage experienced by a shoot upwind but on the same plant as a clipped or unclipped focal shoot (upwind assay in Fig. 1B). In the same experiment we

examined damage on a shoot of a different plant that was downwind of a clipped or unclipped focal shoot (downwind assay in Fig. 1B). We predicted that damage would be reduced only for the downwind shoot with a clipped neighbor. This prediction was upheld (Fig. 5). The downwind shoot experienced ~50% less damage near a clipped neighbor than near an unclipped neighbor (a priori contrast:  $F_{1,218} = 4.92$ ,  $P = 0.03$ ). As predicted, we could detect no difference in damage experienced by the upwind shoot with clipped and unclipped neighbors (a priori contrast:  $F_{1,218} = 0.009$ ,  $P = 0.92$ ).

Our experiment at Sagehen tested the hypothesis that air contact was necessary for communication to induce resistance to herbivory among branches within an individual bush as well as between bushes. We hypothesized that only those branches that received volatile cues from neighboring clipped branches would experience less herbivory than controls. This hypothesis was supported by the data (Fig. 6). Overall, our treatments affected levels of leaf damage during the summer and autumn (whole model  $F_{4,138} = 5.56$ ,  $P < 0.001$ ). When air contact was blocked by plastic bags, we detected no reduction in herbivory on unclipped branches of clipped plants, relative to controls (planned comparison of treatments 2 vs. 5,  $F_{1,138} = 0.40$ ,  $P = 0.53$ ). When air contact was possible, herbivore damage was reduced by 27–33% on unclipped branches of clipped plants relative to controls (planned comparison of treatments 1 vs. 2, 5,  $F_{1,138} = 5.50$ ,  $P = 0.02$ ). This indicates that air contact was required for systemic induced resistance within single bushes. The same result was found for communication between bushes. When air contact was possible between bushes, herbivore damage was reduced by 54% on unclipped branches close to clipped ones on neighboring plants relative to controls (planned com-

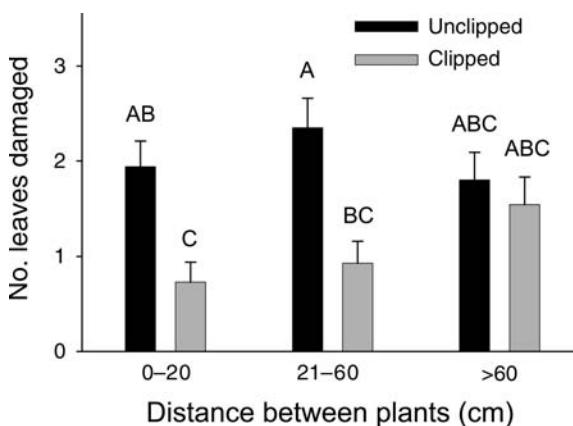


FIG. 4. Over what distances does communication occur? Standardized number of leaves damaged (per 100 leaves) by herbivores for downwind assay branches separated from clipped or unclipped neighbors by three distance categories as shown in Fig. 1A. Histogram bars show means and SE; letters above the bars indicate statistical differences among means using Tukey’s hsd.

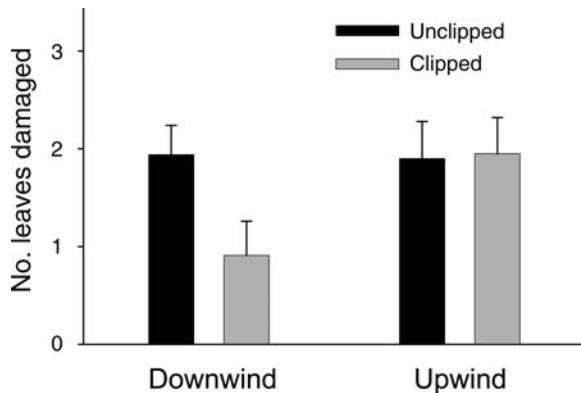


FIG. 5. Standardized leaf damage for either upwind branch of the same plant or downwind branch of a neighbor plant. The focal plant was either clipped or unclipped, as shown in Fig. 1B. Histogram bars show mean and SE.

parison of treatments 3 vs. 4, 5,  $F_{1,138} = 15.75$ ,  $P < 0.001$ ).

#### DISCUSSION

Cues from experimentally clipped sagebrush caused neighboring tobacco to increase resistance to herbivores and to produce more flowers and fruits in most years than tobacco with unclipped neighbors (Karban and Maron 2002). However, this induced resistance is probably not important ecologically since it occurred over limited distances and most wild tobacco individuals do not grow in such close proximity to sagebrush neighbors. In contrast, communication between sagebrush plants was detectable up to 60 cm (Fig. 4), well within the range over which many sagebrush individuals have nearest neighbors (Fig. 3). Volatile cues are released for several hours after clipping (Preston et al. 2004), although communication between sagebrush plants consistently caused decreased levels of herbivory over the summer season. In all cases, levels of leaf loss to herbivory were low during the summer. Resistance caused by communication will only be ecologically important to the plant if the difference in herbivory translates into differences in plant fitness, abundance, or distribution. These parameters must be measured over many years before the ecological impact of this effect can be evaluated meaningfully for sagebrush, a slow-growing, long-lived desert shrub. We have recently initiated a long-term experiment at SNARL to measure effects of induced resistance on plant survival and reproduction.

Experiments at SNARL, where airflow is largely unidirectional, suggested that air contact was essential for communication between sagebrush individuals. Only downwind branches near clipped neighbors experienced reduced leaf damage (Fig. 5). This result was consistent with experiments that physically prevented airflow between sagebrush and tobacco (Karban et al. 2000). In the sagebrush experiments at SNARL, the downwind

assays were on different plants than those that were clipped, although upwind assays were on the same plants (Fig. 1B). This first design confounded wind direction with plant identity. The experiment at Sagehen separated these two effects (Fig. 1C). Branches near clipped neighbors experienced less herbivory whether they were on the clipped individual or on a different plant in that experiment (Fig. 6). This reduction in herbivory occurred only when airflow was permitted, confirming that airflow is required for communication to induce resistance.

We failed to find evidence of systemic induced resistance among sagebrush branches when leaves of one branch were experimentally clipped (Fig. 2). Although we cannot conclude definitively that systemic induced resistance does not occur, these negative results appear difficult to reconcile with our finding of induced resistance when neighboring individuals were clipped. However, these seemingly contradictory results can be explained by the hypothesis that sagebrush does not communicate effectively using internal cues to induce systemic resistance, but uses an external volatile cue instead. Airflow was required for increased resistance between neighboring individuals (eavesdropping), and airflow was required for signaling between neighboring branches on the same plant (Figs. 5 and 6). In the experiments examining systemic resistance in sagebrush (Fig. 2), there was no control over the distance or direction separating branches. As such, air contact may or may not have occurred between branches of individual plants. This result, that air contact is necessary for systemic induced resistance among branches of sagebrush, raises other puzzling questions.

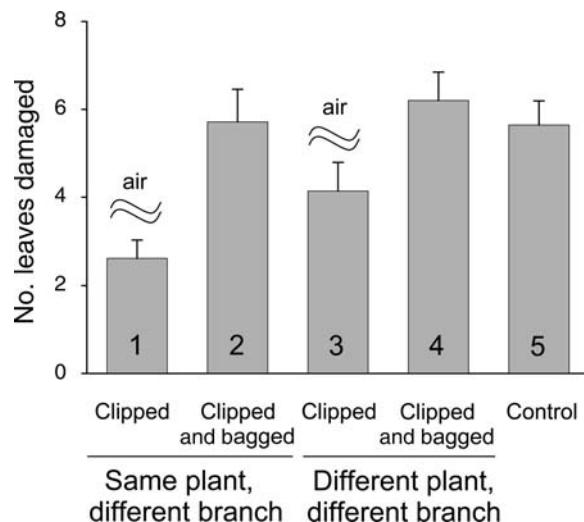


FIG. 6. Average standardized number of leaves damaged for assay branches near clipped, clipped and bagged, or unclipped neighbors and SE. Numbers at the base of histogram bars correspond to treatments in Fig. 1C. Air symbols above histogram bars indicate treatments with air contact between clipped and assay branches.

Why should communication between branches of a single individual require airflow rather than occur via vascular connections, as it has been found to do in some other plants (e.g., Baldwin 1989, Pearce et al. 1991)? One possible explanation is that sagebrush is composed of relatively independent branches that are largely autonomous with regard to hormonal signals and defensive compounds. Previous observations of the consequences of clipping sagebrush suggested that it was not well integrated by vascular connections (Cook and Stoddart 1960). It is likely that plant hormones involved in plant defense and secondary chemicals that help protect leaves are not very mobile through the vasculature of *A. tridentata*. Since sagebrush probably cannot rely on an internal transduction system, it may accomplish this signaling by using external, volatile cues. *A. tridentata* was unusual, though not unique, among species of *Artemisia* in producing and emitting large quantities of volatile methyl jasmonate (Hildebrand et al. 1998). Highly sectorial movement of dye was found generally for shrubs growing in Mediterranean and desert habitats (Waisel et al. 1972). If internal signals do not move readily among branches of sagebrush, perhaps an external volatile signal allows communication among otherwise isolated units within an individual.

The first plants that were observed to respond to cues released by sagebrush were individuals of tomato (Farmer and Ryan 1990) and wild tobacco (Karban et al. 2000). Tobacco plants appeared to suffer less herbivory and experienced increased reproductive success near clipped sagebrush; this observation begged the question: why would sagebrush release cues that helped unrelated plants? The results reported here provide one plausible explanation for emissions of cues by sagebrush. The cues may be released to enable an individual sagebrush plant to coordinate and regulate its own defenses against herbivores and perhaps to control other functions as well. In principle, selection at the level of the individual can favor releases of volatile cues if such releases benefit the emitter. Effects on other individuals of the same or different species are not necessarily the result of adaptation. Of course, release of volatile cues may play other possible roles in signaling to kin, allelopathy, attracting predators of herbivores, etc., and these will be considered in the future.

#### CONCLUSION

These experiments indicate that systemic induced resistance did not occur among branches of a sagebrush individual without air contact. With air contact, branches became more resistant to herbivory when neighboring branches of the same or a different individual were clipped. Sagebrush individuals grew sufficiently close to neighbors so that such communication may be ecologically important, although experiments conducted over longer time frames are required to evaluate the impact of this induced resistance on plant fitness, abundance, and distribution. Since volatile

communication between individuals has been documented convincingly only for sagebrush and alder, it is much too early to know how common this phenomenon is in nature. There is also field evidence for communication between individuals of *Salix sitchensis* (Rhoades 1983) and *Betula pubescens* (Haukioja et al. 1985), as well as a variety of laboratory experiments (Dicke and Bruin 2001). Results from these other systems may be explained by mechanisms other than communication (Karban and Baldwin 1997, Dicke and Bruin 2001). However, more detailed experiments involving these species would be informative and would provide a sense for how general volatile communication that affects herbivory might be. Since branches of sagebrush exhibited limited communication by vascular traces, sagebrush may rely on airborne volatile cues to coordinate responses among branches. Sagebrush is certainly not unusual in having limited integration of branches; indeed all woody plants are sectorial to a greater or lesser extent (Harper 1977, Watson and Casper 1984, Sprugel et al. 1991). Sectoriality appears to be particularly common among shrubs that grow in dry environments (Waisel et al. 1972). We predict that other plants that are restricted in their vascular connections may also employ volatile signaling systems.

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