

# Induced twining in Convolvulaceae climbing plants in response to leaf damage

C. Atala and E. Gianoli

**Abstract:** Plant responses to herbivory include slow changes in growth patterns and biomass distribution. A recent study, however, showed that a convolvulaceous vine began twining sooner around a stake after 25% of the plant was defoliated (damaged). We evaluated whether this induced response is widespread within the Convolvulaceae, and made preliminary studies of its underlying mechanisms. Leaf damage was applied to seven twining vine species from the genera *Convolvulus*, *Calystegia*, and *Ipomoea*. We compared the twining rate (proportion of plants successfully climbing at a given time), growth rate, and twining geometry in the control and in damaged plants. We further evaluated the consequences of jasmonic acid application on the twining rate of *Ipomoea purpurea* (L.) Roth. Five out of the seven species tested showed an enhanced twining rate after leaf damage. Growth rate did not differ between damaged and undamaged plants in any species. The angle of ascent of the twining stem was lower in damaged plants during the first gyres. Jasmonic acid increased twining rate in *I. purpurea*, as did leaf damage. The induced twining was not due to increased growth, but to changes in the climbing process, and further mechanistic approaches should consider the jasmonate pathway. Induced twining may be common in the Convolvulaceae, and its occurrence in other families should be tested.

**Key words:** twining vines, herbivory, induced responses, jasmonic acid, leaf damage.

**Résumé :** Les réactions des plantes à l'herbivorie incluent des changements lents dans les patrons de croissance et de distribution de la biomasse. On a récemment constaté que la vigne de Convolvulaceae commence à s'enrouler plus tôt autour d'un tuteur après une défoliation à 25 %. Les auteurs ont déterminé l'étendue de cette réaction induite chez les Convolvulaceae et en ont examiné de façon préliminaire les mécanismes sous-jacents. Ils ont endommagé les feuilles de sept espèces de vignes volubiles des genres *Convolvulus*, *Calystegia* et *Ipomoea*. Ils ont ensuite comparé le taux d'enroulement (proportion des plantes grimpées à un moment donné), le taux de croissance et la géométrie de l'enroulement chez les plantes traitées et témoins. Ils ont également évalué les conséquences d'une application d'acide jasmonique sur les taux d'enroulement de l'*Ipomoea purpurea* (L.) Roth. Cinq des sept espèces examinées montrent un taux accru d'enroulement suite au dommage foliaire. Le taux de croissance ne diffère pas entre les plantes endommagées ou non au cours de la première spire. L'acide jasmonique augmente le taux d'enroulement chez l'*I. purpurea* tout comme le dommage foliaire. L'enroulement induit ne provient pas d'une augmentation de croissance, mais de modifications dans le processus de grimpe et l'on devrait considérer l'acide jasmonique pour en comprendre les mécanismes. L'induction de l'enroulement pourrait s'avérer commune chez les Convolvulaceae et l'on devrait vérifier sa présence chez d'autres familles.

**Mots-clés :** vigne volubile, herbivorie, réactions induites, acide jasmonique, dommage foliaire.

[Traduit par la Rédaction]

## Introduction

Plants attacked by herbivores can show changes in their growth patterns and the distribution of resources that minimize the detrimental effects of herbivore damage (Strauss and Agrawal 1999; Stowe et al. 2000; Tiffin 2000; Kessler

and Baldwin 2002). These responses are usually expressed in a timescale of weeks or months (McNaughton 1983; Juenger and Bergelson 1997; Lennartsson et al. 1998). Recently, however, Gianoli and Molina-Montenegro (2005) reported a rapid response in growth patterns in a climbing plant triggered by leaf damage that can reduce the risk of herbivory. They experimentally determined that leaf damage (25% of the plant defoliated) increases the climbing success of the twining vine *Convolvulus arvensis* L. (Convolvulaceae) within 24–48 h. Furthermore, field data showed that prostrate plants of *C. arvensis* suffer more herbivore damage than plants climbing onto an external support. It is unknown whether induced twining in response to leaf damage, as observed in *C. arvensis*, is a common feature of climbing plants, particularly within the Convolvulaceae.

The earlier twining of damaged vines could be the result of enhanced growth rate or changes in the climbing process. Increased growth after damage has been described as one of several compensatory responses to herbivory in plants (Trumble et al. 1993; Strauss and Agrawal 1999). The in-

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**Table 1.** Species included in the study, their life cycle, and growth rate of damaged and undamaged plants.

| Species                       | Life cycle | Growth rate for damaged (cm·d <sup>-1</sup> ) | Growth rate for undamaged (cm·d <sup>-1</sup> ) | <i>n</i> | <i>t</i> | <i>p</i> |
|-------------------------------|------------|---|---|----------|----------|----------|
| <i>Ipomoea purpurea</i>       | Annual     | 2.699±0.236                                   | 2.375±0.287                                     | 17       | 0.871    | 0.390    |
| <i>Ipomoea tricolor</i>       | Annual     | 5.068±0.423                                   | 5.608±0.428                                     | 20       | -0.898   | 0.375    |
| <i>Ipomoea nil</i>            | Annual     | 4.331±0.194                                   | 4.219±0.265                                     | 16       | 0.343    | 0.734    |
| <i>Ipomoea alba</i>           | Annual     | 6.486±0.546                                   | 7.278±0.560                                     | 11       | -0.670   | 0.492    |
| <i>Calystegia sepium</i>      | Perennial  | 1.658±0.202                                   | 1.761±0.225                                     | 10       | -0.339   | 0.739    |
| <i>Convolvulus arvensis</i>   | Perennial  | 0.433±0.084                                   | 0.420±0.104                                     | 15       | 0.099    | 0.922    |
| <i>Convolvulus hermanniae</i> | Perennial  | 1.743±0.291                                   | 1.770±0.220                                     | 7        | -0.956   | 0.440    |

**Note:** Growth rate (mean ± SE) of damaged and undamaged plants is compared (*t*-test for independent samples). Sample size (*n*) corresponds to individuals per treatment.

duced twining might also affect the climbing geometry of vines. The helical geometry of twining vines may be characterized by the radius of curvature and the angle of ascent (Putz and Holbrook 1991). While the radius of curvature is rather constant (Putz and Holbrook 1991; Silk and Holbrook 2005), the angle of ascent varies with the diameter of the supporting pole (Bell 1958; Putz and Holbrook 1991).

Mechanical wounding and herbivore damage often trigger jasmonate-dependant responses in plants (Karban and Baldwin 1997; McConn et al. 1997; Reymond et al. 2000; Bostock et al. 2001). These responses include the induction of chemical defenses (Baldwin 1996; Voelckel et al. 2001), trichomes (Traw and Bergelson 2003; Boughton et al. 2005), and even indirect defenses against herbivores (Heil et al. 2001; Thaler et al. 2002). Interestingly, it has been found that applying jasmonate to the surface of the plant induces tendrils coiling in the climbing plant *Bryonia dioica* (Falkenstein et al. 1991; Weiler et al. 1993; Bleichert et al. 1999). Consequently, jasmonate could be involved in the twining induction observed in *C. arvensis* (Gianoli and Molina-Montenegro 2005).

In this greenhouse study, we evaluated the effect of leaf damage on the twining rate (proportion of plants successfully climbing at a given time) of seven species belonging to three genera from the Convolvulaceae, a family where the climbing habit is a common feature (Cronquist 1981; Gianoli 2004). To further characterize this induced response, we evaluated whether damaged and undamaged plants differed in growth rate and in twining geometry. We also assessed the effect of foliar jasmonic acid (JA) application on the twining rate of one of the seven species studied.

## Materials and methods

### Cultivation of vines

Individuals of *Ipomoea purpurea* (L.) Roth., *Ipomoea tricolor* Car., *Ipomoea nil* (L.) Roth., *Ipomoea alba* L., *Convolvulus arvensis*, *Convolvulus hermanniae* L'Hér., and *Calystegia sepium* (L.) R. Br. were cultivated in a greenhouse for the experiment. Four of these species are annuals and three of them are perennials (Table 1). Whereas seeds from *I. purpurea*, *C. arvensis*, *C. hermanniae*, and *C. sepium* were collected in the field, in central Chile, seeds from the other species were obtained from commercial sources. To stimulate germination, seeds were put in jars with warm water for 24 h. Germinated seeds, i.e., those in which radicle emergence

was evident, were placed on damp paper in Petri dishes, and kept in a growth chamber at 25 °C. After the cotyledons were visible, seedlings were transplanted to plastic bags (1.5 L) full of commercial potting soil and were maintained in a greenhouse at Universidad de Concepción campus, Concepción, central Chile. Plants were watered to field capacity every other day. The experiment was conducted during the austral summer (November–January). The average temperature was 22.4 ± 9.0 °C during the day, and 13.7 ± 2.2 °C during night-time. PAR radiation usually exceeded 1000 μmol·m<sup>-2</sup>·seg<sup>-1</sup> between 1000 and 1700 h. Temperature was recorded using a data logger (HOBO data pendant, Onset Computer Corporation, Pocasset, Mass.) and light using a Li-250 light-meter (LI-COR Biosciences, Lincoln, Nebr.).

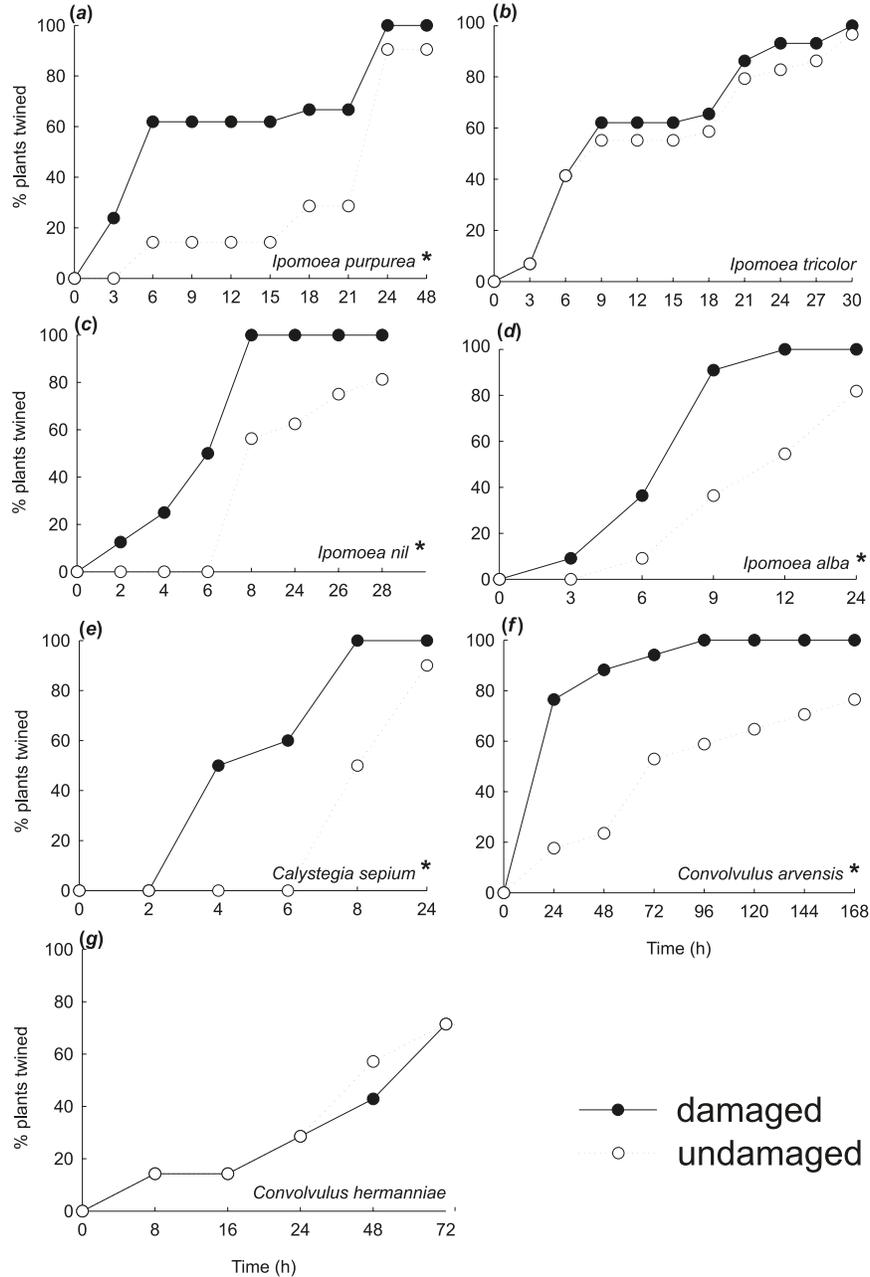
### Twining rate

When plants were 6–8 weeks old (five to six true leaves), they were allowed to twine around a stake. Single-stemmed, prostrate plants were provided with a vertical stake (1.1 cm in diameter) just in contact with the stem. Following an earlier study (Gianoli and Molina-Montenegro 2005), plants were defoliated by 25% (hereinafter, damaged) at the beginning of the experiment. This was achieved by using a pair of scissors to cut off 50% of the leaf area of half of the leaves. Leaves were clipped along the midvein. Control plants were not defoliated (hereinafter, undamaged). Undamaged and damaged plants were randomly placed on greenhouse benches. The number of replicates for each of the two treatments (damaged and undamaged) was as follows: *I. purpurea* (*n* = 21); *I. tricolor* (*n* = 29); *I. nil* (*n* = 16); *I. alba* (*n* = 11); *Calystegia sepium* (*n* = 10); *Convolvulus arvensis* (*n* = 17); and *C. hermanniae* (*n* = 7). After inflicting leaf damage, we recorded, at several times, the number of plants successfully twining (360° gyre) around the stake for both undamaged and damaged plants. The significance of differences in twining rate between treatments was determined using a Kolmogorov–Smirnov two-sample test. Although artificial damage and herbivore damage may induce different responses in plants (Lehtilä and Boalt 2004), some studies have found similar effects of both types of damage (Green and Ryan 1972; Enyedi et al. 1992). We chose artificial defoliation to control the magnitude and timing of damage.

### Growth rate

The same plants employed in the twining experiment

**Fig. 1.** Percentage of twined individuals in damaged (25% defoliated, filled circles) and undamaged (open circles) plants at different time intervals in seven Convolvulaceae species. (a) *Ipomoea purpurea* ( $n = 25$ ), (b) *I. tricolor* ( $n = 29$ ), (c) *I. nil* ( $n = 16$ ), (d) *I. alba* ( $n = 11$ ), (e) *Calystegia sepium* ( $n = 10$ ), (f) *Convolvulus arvensis* ( $n = 17$ ), (g) *C. hermanniae* ( $n = 7$ ). Significant differences between treatments in the distribution of the time of twining initiation are indicated (\*,  $p < 0.05$  Kolmogorov–Smirnov two-sample test).



were used to determine differences in growth rate. Growth rate was assessed by measuring the total length of the main stem from the base of the plant to the tip of the apex at the beginning of the experiment and 24 h later. This time interval spanned the time needed to observe induced twining (see Results). Measurements were conducted using a plastic thread. We carefully followed the path length of the main stem of the climbing plant with the thread, which was measured afterwards.

#### Angle of ascent

The angle of ascent is the angle between the stem of the

plant and the normal plane to the support (Putz and Holbrook 1991). From the seven species considered in this study, we chose three for measurements of the angle of ascent. We used *I. purpurea*, *I. tricolor*, and *I. nil* because of their larger sample size. We randomly selected plants from those used in the twining experiment. A few plants were excluded from the analysis because they were damaged after falling from the greenhouse benches owing to strong wind events. The angle of ascent was determined using image analysis software (Sigmascan 5, SPSS Inc., Chicago, Ill.) on pictures of the twining vines taken with a digital camera (Kodak Easyshare DX7630). Pictures were taken after the

**Table 2.** Repeated measures ANOVA for the effect of damage on the angle of ascent in *Ipomoea purpurea* ( $n = 16$ ), *I. tricolor* ( $n = 20$ ) and *I. nil* ( $n = 18$ ).

| Effect                    | SS     | df | MS     | F      | p      |
|---------------------------|--------|----|--------|--------|--------|
| <b><i>I. purpurea</i></b> |        |    |        |        |        |
| Damaged                   | 875.0  | 1  | 875.0  | 30.48  | <0.001 |
| Error                     | 602.9  | 21 | 28.7   | 0.63   |        |
| Gyre                      | 84.5   | 3  | 28.2   | 1.24   | 0.599  |
| Gyre×damaged              | 166.3  | 3  | 55.4   | —      | 0.303  |
| Error                     | 2818.0 | 63 | 44.7   |        |        |
| <b><i>I. tricolor</i></b> |        |    |        |        |        |
| Damaged                   | 1560.4 | 1  | 1560.4 | 16.873 | <0.001 |
| Error                     | 2312.0 | 25 | 92.5   | —      |        |
| Gyre                      | 5101.1 | 3  | 1700.4 | 31.632 | <0.001 |
| Gyre×damaged              | 3164.2 | 3  | 1054.7 | 19.621 | <0.001 |
| Error                     | 4031.6 | 75 | 53.8   |        |        |
| <b><i>I. nil</i></b>      |        |    |        |        |        |
| Damaged                   | 600.7  | 1  | 600.7  | 11.501 | 0.002  |
| Error                     | 1775.8 | 34 | 52.2   | —      |        |
| Gyre                      | 1060.9 | 2  | 530.4  | 23.495 | <0.001 |
| Gyre×damaged              | 336.4  | 2  | 168.2  | 7.449  | 0.001  |
| Error                     | 1535.2 | 68 | 22.6   |        |        |

**Note:** Gyre was the repeated factor.

fourth gyre was completed. Differences in angle of ascent between control and damaged plants for each species were determined using repeated measures ANOVA, considering “Treatment” as the main factor and “Gyre” as the repeated variable. We then conducted a Fisher LSD test to compare the angle of ascent between treatments in each gyre. Differences between gyres within a treatment were not evaluated because it was not the focus of the study. Differences between species were not addressed for the same reason. In *I. nil*, the data of the fourth gyre were almost completely lost due to plant damage (see above), so these data were omitted from the statistical analysis. Fourth gyre data for *I. nil* ( $n = 2$  per treatment) are shown for illustrative purposes only. The radius of curvature, which is a component of the twining geometry (Putz and Holbrook 1991), was not considered in this study because the species included show a constantly tight twining around the support and hence no radius variation was expected.

### Jasmonic acid

We chose *I. purpurea* for this experiment because of seed availability and the remarkable induced twining observed in the first set of experiments. Plants were set in the same way as in the twining experiment described above. Four treatments were considered ( $n = 14$  plants per treatment): JA, control, damaged, and undamaged. The JA treatment consisted in the foliar application of 1 mL of a racemic mixture of (+) and (−) JA  $1 \text{ mmol}\cdot\text{L}^{-1}$  (Sigma-Aldrich, St. Louis, Mo.) as reported earlier (Traw and Bergelson 2003). The aqueous solution was applied with a pipette on all fully expanded leaves before the start of the experiment, when plants had five to six true leaves. The leaves of control plants received 1 mL of water with a small amount of ethanol. Five drops of 100% ethanol were diluted in 500 mL water because the stock solution of JA contained the same amount of ethanol. Damaged and undamaged treatments

were applied as described for the twining experiment. Likewise, we recorded the time at which the stem completed a  $360^\circ$  turn around its support, and measured stem growth rate. The significance of differences in twining rate between treatments was determined using a Kolmogorov–Smirnov two-sample test as in the first experiment, and differences in growth rate were compared using a one-way ANOVA.

## Results

### Induced twining

Five of the seven species showed induced twining after leaf damage (Fig. 1; Kolmogorov–Smirnov test). Thus, all species but *C. hermannieae* and *I. tricolor* showed significant differences in the distribution of the times to successful twining between damaged and undamaged plants (Kolmogorov–Smirnov test). These statistical results, along with the information shown in Fig. 1, allow us to conclude that damaged plants had a greater twining rate than undamaged plants. Some species showed very rapid responses that were evident just 4–6 h after the start of the experiment (Fig. 1). All responsive species showed induced twining within 24 h.

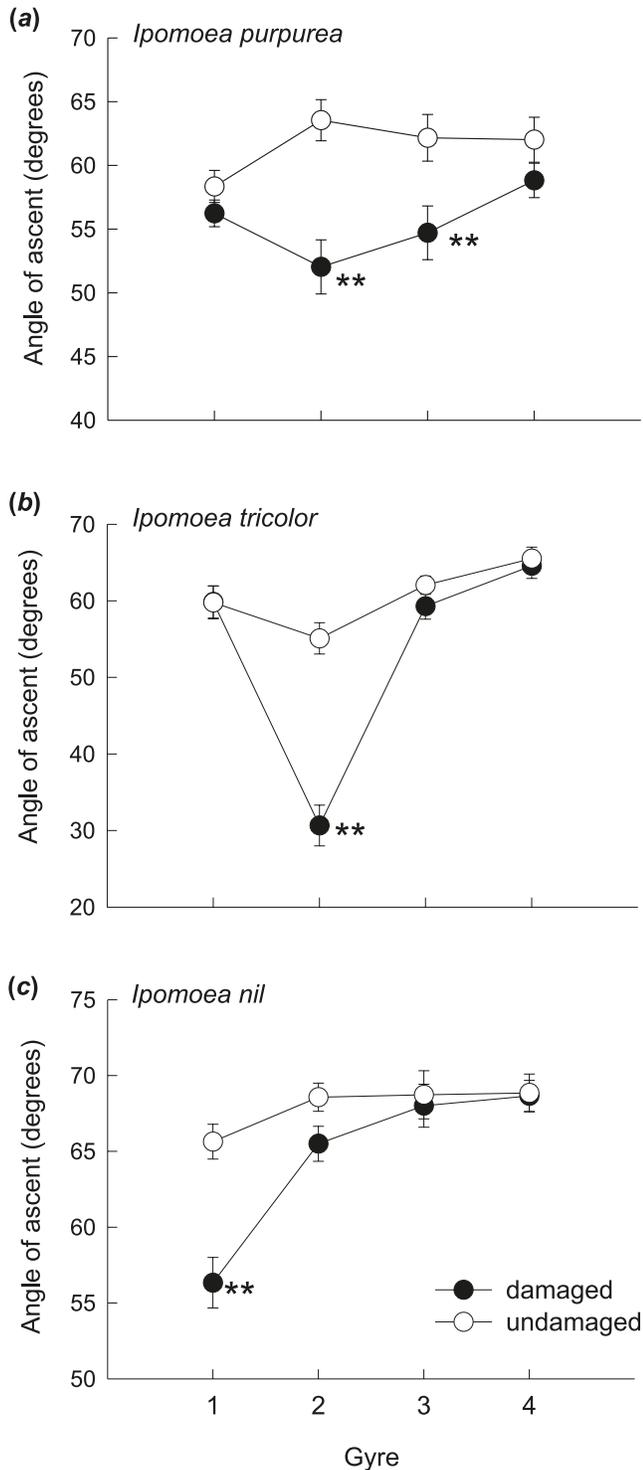
### Growth rate

We found no difference in growth rate between undamaged and damaged plants in any species ( $t$ -test,  $p > 0.05$ ; Table 1). This implies that any difference in twining rate between treatments is not a consequence of differences in growth. As expected, annual species showed higher growth rates than perennial species (Table 1): whereas perennials had growth rates ranging from  $0.4 \text{ cm}\cdot\text{d}^{-1}$  in *C. arvensis* to  $1.7 \text{ cm}\cdot\text{d}^{-1}$  in *C. hermannieae*, annuals grew up to  $6 \text{ cm}\cdot\text{d}^{-1}$  (data not shown).

### Angle of ascent

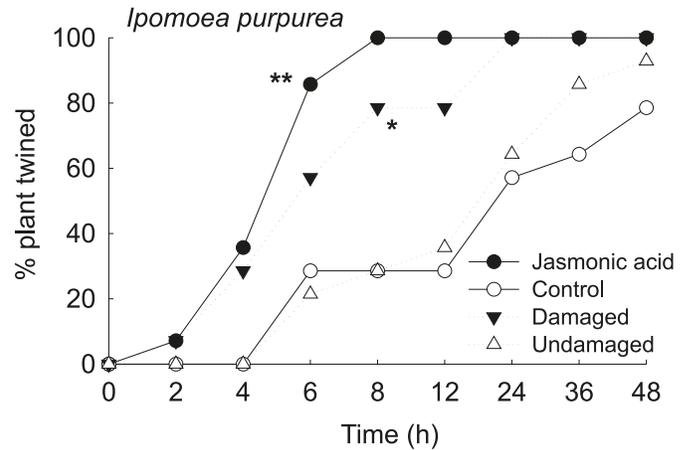
The angle of ascent was affected by leaf damage in all

**Fig. 2.** Angle of ascent of the main stem during the first four gyres in damaged (25% defoliation, filled circles) and undamaged (open circles) plants of the twining vines. (a) *Ipomoea purpurea* ( $n = 16$  per treatment), (b) *I. tricolor* ( $n = 20$  per treatment), and (c) *I. nil* ( $n = 18$  per treatment). Significant differences between treatments within the same gyre are indicated (\*\*,  $p < 0.01$ ; Fisher LSD test).



three species evaluated (Table 2). Damaged plants showed a reduced angle of ascent within the first three gyres, but species differed in the particular gyres affected (Fig. 2). In

**Fig. 3.** Percentage of twined individuals of *Ipomoea purpurea* at different time intervals in four treatments: (filled circles) jasmonic acid, aqueous solution of JA with traces of ethanol; (open circles) control, water with traces of ethanol and undamaged; (filled triangles) damaged, 25% defoliation; and (open triangles) undamaged, no defoliation, no water. Significant differences in twining rate between JA and control treatments, and between damaged and undamaged treatments are indicated ( $n = 14$  plants per treatment; \*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ; Kolmogorov–Smirnov two sample test). No differences were found between the jasmonic acid and damaged treatments (Kolmogorov–Smirnov two sample test,  $p > 0.10$ ), or between the control and undamaged treatment (Kolmogorov–Smirnov two sample test,  $p > 0.10$ ).



*I. purpurea*, these were the second and third gyres. In the second gyre the angle of ascent of damaged plants was roughly 10° lower than that of undamaged plants. In the fourth gyre differences between treatments tended to disappear. In *I. tricolor*, the angle of ascent of damaged plants was almost half of that of undamaged plants in the second gyre. In the third gyre the angle of ascent in damaged plants returned to undamaged-like values, and this trend continued in the fourth gyre. In *I. nil*, the angle of the first gyre in damaged plants was significantly lower compared with undamaged plants, but in the second, third and fourth gyres the angles did not differ between treatments.

**Jasmonic acid**

JA application induced twining in *I. purpurea* as did leaf damage. Both treatments (JA and damaged) showed a greater twining rate than their respective controls (control and undamaged, respectively, Kolmogorov–Smirnov test,  $p < 0.05$ ; Fig. 3). There were no significant differences in twining rate between JA and damaged (Kolmogorov–Smirnov two-sample test,  $p > 0.10$ ) and between the control (water + traces of ethanol) and undamaged plants (Kolmogorov–Smirnov two-sample test,  $p > 0.10$ ). Stem growth rate did not differ across treatments (one-way ANOVA;  $F_{[3,52]} = 0.463$ ,  $p > 0.70$ ).

**Discussion**

Our results suggest that induced twining after leaf damage may be a common feature of the Convolvulaceae. Gianoli and Molina-Montenegro (2005) first detected such induced response in *C. arvensis* and we have found it in *Calystegia*

and *Ipomoea* species. We did not find the response in *C. hermannaiae* and *I. tricolor*. One possible explanation for this lack of induced twining could be that the damage inflicted (25% leaf-area loss) was not enough to elicit the response. It has been shown in other systems that a damage threshold must be surpassed to elicit induced responses (Gianoli and Niemeyer 1997; Underwood 2000). We observed very rapid twining induction in some species, even within 4–6 h, which is faster than that reported for *C. arvensis* (Gianoli and Molina-Montenegro 2005). This may be partially explained by the fact that those were annual species, which showed a growth rate 4–6 times greater than that of *C. arvensis*.

Successfully climbing onto a support is essential for vine fitness in natural populations (Putz 1984; Puntieri and Pysek 1993). In this study we found that leaf damage can influence twining dynamics. The ecological significance of the induced twining reported herein might be questioned, because in most species the differences between damaged and undamaged plants disappeared in less than 24 h (but see *C. arvensis*, where such differences held after 7 d). However, even such a short advantage may be important during early ontogenetic stages of plants, when they can be highly susceptible to the attack of ground herbivores (Hanley et al. 1995; Green et al. 1997; Watkinson 1997).

The enhanced climbing of damaged plants was due to changes in the twining process itself, and not to differences in growth rate. Twining vines have the innate tendency to form a helix of smaller radius and larger torsion than is possible for a given support (Scher et al. 2001). This is based on observations of twining vines, which once removed from their supporting pole tend to coil tighter (Silk and Hubbard 1991). The angle of ascent of twining vines decreases with increasing support diameter (Putz and Holbrook 1991) and mechanical stimuli can slow vine circumnutation (Brown 1993). We found that leaf damage increased twining rate and altered the helical geometry. A reduction in the angle of ascent occurred during the first gyres after damage, and vanished as the plant continued to twine around its support. The biological significance or ecological advantage for wounded vines of producing gyres with a lower angle of ascent remains to be elucidated. Combining the results of studies conducted with vines climbing onto supports of different diameter (Putz and Holbrook 1991; Scher et al. 2001), it could be argued that lower angles of ascent may be associated with marginally greater twining forces — those responsible for torsion — and significantly smaller twining loads — those allowing the stem to hug the pole. However, experimental evidence for this hypothetical relationship is lacking.

The mechanism underlying the induced responses reported herein is yet to be discovered. We found that JA application enhanced twining rate in *I. purpurea*, and preliminary results indicate that this is also true for *C. arvensis* (C. Atala and E. Gianoli, unpublished data, 2007). These results suggest that the induced twining in the Convolvulaceae could be mediated by the JA pathway and agree with earlier findings that JA-related compounds are inducers of tendrils coiling in the climber *Bryonia dioica* (Falkenstein et al. 1991; Weiler et al. 1993; Blechert et al. 1999). In particular, the biologically active molecule involved in tendrils

coiling, and presumably in enhanced stem twining, is the JA precursor 12-oxophytodienoic acid (OPDA) (Stelmach et al. 1999). It remains to be elucidated how OPDA elicits the differential growth across the cross-section of the stem, with greater cell turgor and elongation in the stem side away from the support, which is supposed to underlie vine circumnutation (Silk 1989). JA and related octadecanoids are ubiquitously involved in plant defensive responses to wounding (Karban and Baldwin 1997). Leaf damage by herbivores and artificial defoliation rapidly increase JA levels (Bostock 1999; Reymond et al. 2000) and the exogenous application of jasmonates emulates several wound- or herbivory-induced chemical and physical defenses (Thaler 1999; Traw and Bergelson 2003; Boughton et al. 2005).

The climbing habit seems to be a key innovation within flowering plants because of the greater diversity of climbing vs. nonclimbing sister taxa (Gianoli 2004). Although the climbing habit has been widely studied (Darwin 1875; Peñalosa 1982; Putz and Holbrook 1991; Gianoli 2001; Schnitzer and Bongers 2002; Gianoli 2004; Schnitzer 2005), we are still far from a full understanding of the mechanisms underlying it. If the induced twining after leaf damage reported herein for the Convolvulaceae occurs in climbers from other plant families, it might be conceivable that herbivory avoidance could be yet another ecological feature associated with the climbing habit, apart from the widely accepted benefits in terms of light capture and structural cost-saving (Darwin 1875; Putz 1984; Putz and Chai 1987; Hegarty and Caballé 1991; Gianoli 2002; Niklas 1992).

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## References

- Baldwin, I.T. 1996. Methyl jasmonate-induced nicotine production in *Nicotiana attenuata*: inducing defenses in the field without wounding. *Entomol. Exp. Appl.* **80**: 213–220. doi:10.1007/BF00194760.
- Bell, P.R. 1958. Twining of the hop (*Humulus lupulus* L.). *Nature* (London), **181**: 1009–1010. doi:10.1038/1811009a0.
- Blechert, S., Bockelmann, M., Fülllein, M., von Schrader, T., Stelmach, B., Niesel, U., and Weiler, E.W. 1999. Structure-activity analyses reveal the existence of two separate groups of active octadecanoids in elicitation of the tendril-coiling response of *Bryonia dioica* Jacq. *Planta*, **207**: 470–479. doi:10.1007/s004250050506.
- Bostock, R.M. 1999. Signal conflicts and synergies in induced resistance to multiple attackers. *Physiol. Mol. Plant Pathol.* **55**: 99–109. doi:10.1006/pmpp.1999.0218.
- Bostock, R.M., Karban, R., Thaler, J.S., Weyman, P.D., and Gilchrist, D. 2001. Signal interactions in induced resistance to pathogens and insect herbivores. *Eur. J. Plant Pathol.* **107**: 103–111. doi:10.1023/A:1008703904253.
- Boughton, A.J., Hoover, K., and Felton, G.W. 2005. Methyl jasmonate application induces increased densities of glandular tri-

- chomes on tomato, *Lycopersicon esculentum*. J. Chem. Ecol. **31**: 2211–2216. doi:10.1007/s10886-005-6228-7. PMID: 16132222.
- Brown, A.H. 1993. Circumnutation: from Darwin to space flights. Plant Physiol. **101**: 345–348. PMID:11537497.
- Cronquist, A. 1981. An integrated system of classification of flowering plants. Columbia University Press, New York, N.Y.
- Darwin, C. 1875. On the movements and habits of climbing plants. John Murray, London, UK.
- Enyedi, A.J., Yapalni, N., Silverman, P., and Raskin, I. 1992. Signal molecules in systemic plants resistance to pathogens and pest. Cell. **70**: 879–886. doi:10.1016/0092-8674(92)90239-9. PMID:1525827.
- Falkenstein, E., Groth, B., Mithöfer, A., and Weiler, E.W. 1991. Methyljasmonate and  $\alpha$ -linolenic acid are potent inducers of tendril coiling. Planta, **185**: 316–322.
- Gianoli, E. 2001. Lack of differential plasticity to shading of internodes and petioles with growth habit in *Convolvulus arvensis* (Convolvulaceae). Int. J. Plant Sci. **162**: 1247–1252. doi:10.1086/322950.
- Gianoli, E. 2002. Maternal environmental effects on the phenotypic responses of the twining vine *Ipomoea purpurea* to support availability. Oikos, **99**: 324–330. doi:10.1034/j.1600-0706.2002.990213.x.
- Gianoli, E. 2004. Evolution of a climbing habit promotes diversification in flowering plants. Proc. R. Soc. Lond. Ser. B. Bio. Sci. **271**: 2011–2015.
- Gianoli, E., and Molina-Montenegro, M. 2005. Leaf damage induces twining in a climbing plant. New Phytol. **167**: 385–390. doi:10.1111/j.1469-8137.2005.01484.x. PMID:15998392.
- Gianoli, E., and Niemeyer, H.M. 1997. Characteristics of hydroxamic acid induction in wheat triggered by aphid infestation. J. Chem. Ecol. **23**: 2695–2705. doi:10.1023/A:1022554708782.
- Green, T.R., and Ryan, C.A. 1972. Wound-induced proteinase inhibitor in plant leaves: a possible defense mechanism against insects. Science (Wash.), **175**: 776–777. doi:10.1126/science.175.4023.776. PMID:17836138.
- Green, P.T., O'Dowd, D.J., and Lake, P.S. 1997. Control of seedling recruitment by land crabs in rainforest on a remote oceanic island. Ecology, **78**: 2474–2486.
- Hanley, M.E., Fenner, M., and Edwards, P.J. 1995. An experimental field study of the effects of mollusc grazing on seedling recruitment and survival in grassland. J. Ecol. **83**: 621–627.
- Hegarty, E.E., and Caballé, G. 1991. Distribution and abundance of vines in forest communities. In The biology of vines. Edited by F.E. Putz and H.A. Mooney. Cambridge University Press, Cambridge, UK. pp. 313–337.
- Heil, M., Koch, T., Hilpert, A., Fiala, B., Boland, W., and Linsenmair, E. 2001. Extrafloral nectar production of the ant-associated plant, *Macaranga tanarius*, is an induced indirect, defensive response elicited by jasmonic acid. Proc. Natl. Acad. Sci. U.S.A. **98**: 1083–1088. doi:10.1073/pnas.031563398. PMID:11158598.
- Juenger, T., and Bergelson, J. 1997. Resource and pollen limitation of compensation to herbivory in scarlet gilia, *Ipomopsis aggregata*. Ecology, **78**: 1684–1695.
- Karban, R., and Baldwin, I.T. 1997. Induced responses to herbivory. University of Chicago Press, Chicago, Ill.
- Kessler, A., and Baldwin, I.T. 2002. Plant responses to insect herbivory: The emerging molecular analysis. Annu. Rev. Plant Biol. **53**: 299–328. doi:10.1146/annurev.arplant.53.100301.135207. PMID:12221978.
- Lehtilä, K., and Boalt, E. 2004. The use and usefulness of artificial herbivory in plant–herbivore studies. In Insects and ecosystem function. Ecological studies 173. Edited by W.W. Weisser and E. Siemann. Springer Verlag, Heidelberg, Germany. pp. 257–265.
- Lennartsson, T., Nilsson, P., and Tuomi, J. 1998. Induction of over-compensation in the field gentian, *Gentiana campestris*. Ecology, **79**: 1061–1072.
- McConn, M., Creelman, R.A., Bell, E., Mullet, J.E., and Browse, J. 1997. Jasmonate is essential for insect defense in *Arabidopsis*. Proc. Natl. Acad. Sci. U.S.A. **94**: 5473–5477. doi:10.1073/pnas.94.10.5473. PMID:11038546.
- McNaughton, S.J. 1983. Compensatory plant growth as a response to herbivory. Oikos, **40**: 329–336. doi:10.2307/3544305.
- Niklas, K.J. 1992. Plant biomechanics: an engineering approach to plant form and function. University of Chicago Press, Chicago, Ill.
- Peñalosa, J. 1982. Morphological specialization and attachment success in two twining lianas. Am. J. Bot. **69**: 1043–1045. doi:10.2307/2442901.
- Puntieri, J.G., and Pysek, P. 1993. The effects of physical support and density on biomass production and size hierarchies of *Galium aparine* populations. Oikos, **66**: 279–284.
- Putz, F.E. 1984. The natural-history of lianas on Barro Colorado Island, Panama. Ecology, **65**: 1713–1724. doi:10.2307/1937767.
- Putz, F.E., and Chai, P. 1987. Ecological studies of lianas in Lambir National Park, Sarawak, Malaysia. J. Ecol. **75**: 523–531.
- Putz, F.E., and Holbrook, N.M. 1991. Biomechanical studies of vines. In The biology of vines. Edited by F.E. Putz and H.A. Mooney. Cambridge University Press, Cambridge, UK. pp. 73–98.
- Reymond, P., Weber, H., Damond, M., and Farmer, E.E. 2000. Differential gene expression in response to mechanical wounding and insect feeding in *Arabidopsis*. Plant Cell, **12**: 707–719. doi:10.1105/tpc.12.5.707. PMID:10810145.
- Scher, J.L., Holbrook, N.M., and Silk, W.K. 2001. Temporal and spatial patterns of twining force and lignification in stems of *Ipomoea purpurea*. Planta, **213**: 192–198. doi:10.1007/s004250000503. PMID:11469583.
- Schnitzer, S.A. 2005. A mechanistic explanation for global patterns of liana abundance and distribution. Am. Nat. **166**: 262–276. doi:10.1086/431250. PMID:16032578.
- Schnitzer, S.A., and Bongers, F. 2002. The ecology of lianas and their role in forests. Trends Ecol. Evol. **17**: 223–230. doi:10.1016/S0169-5347(02)02491-6.
- Silk, W.K. 1989. On the curving and twining of stems. Environ. Exp. Bot. **29**: 95–99. doi:10.1016/0098-8472(89)90042-7.
- Silk, W.K., and Holbrook, N.M. 2005. The importance of frictional interactions in maintaining the stability of the twining habit. Am. J. Bot. **92**: 1820–1826. doi:10.3732/ajb.92.11.1820.
- Silk, W.K., and Hubbard, M. 1991. Axial forces and normal distributed loads in twining stems of morning glory. J. Biomech. **24**: 599–606. doi:10.1016/0021-9290(91)90292-U. PMID:1880143.
- Stelmach, B.A., Müller, A., and Weiler, E.W. 1999. 12-Oxophytodienoic acid and indole-3-acetic acid in jasmonic acid-treated tendrils of *Bryonia dioica*. Phytochemistry, **51**: 187–192. doi:10.1016/S0031-9422(99)00017-5.
- Stowe, K.A., Robert, J.M., Hochwender, C.G., and Simms, E. 2000. The evolutionary ecology of tolerance to consumer damage. Annu. Rev. Ecol. Syst. **31**: 565–595. doi:10.1146/annurev.ecolsys.31.1.565.
- Strauss, S.Y., and Agrawal, A.A. 1999. The ecology and evolution of plant tolerance to herbivory. Trends Ecol. Evol. **14**: 179–185. doi:10.1016/S0169-5347(98)01576-6. PMID:10322530.
- Thaler, J.S. 1999. Induced resistance in agricultural crops: Effects of jasmonic acid on herbivory and yield in tomato plants. Environ. Entomol. **28**: 30–37.

- Thaler, J.S., Farag, M., Pare, P., and Dicke, M. 2002. Jasmonate deficient tomato mutant has reduced direct and indirect defense. *Ecol. Lett.* **5**: 764–774. doi:10.1046/j.1461-0248.2002.00388.x.
- Tiffin, P. 2000. Mechanism of tolerance to herbivore damage: what do we know? *Evol. Ecol.* **14**: 523–536. doi:10.1023/A:1010881317261.
- Traw, M.B., and Bergelson, J. 2003. Interactive effects of jasmonic acid, salicylic acid, and gibberellin on induction of trichomes in *Arabidopsis*. *Plant Physiol.* **133**: 1367–1375. doi:10.1104/pp.103.027086. PMID:14551332.
- Trumble, J.T., Kolodny-Hirsh, D.M., and Ting, I.P. 1993. Plant compensation for arthropod herbivory. *Annu. Rev. Entomol.* **38**: 93–119. doi:10.1146/annurev.en.38.010193.000521.
- Underwood, N. 2000. Density dependence in induced plant resistance to herbivore damage: threshold, strength and genetic variation. *Oikos*, **89**: 295–300. doi:10.1034/j.1600-0706.2000.890210.x.
- Voelckel, C., Schittko, U., and Baldwin, I.T. 2001. Herbivore-induced ethylene burst reduces fitness cost of jasmonate- and oral secretion-induced defenses in *Nicotiana attenuata*. *Oecologia*, **127**: 274–280. doi:10.1007/s004420000581.
- Watkinson, A.R. 1997. Plant population dynamics. *In* *Plant Ecology*, 2nd edn. Edited by M.J. Crawley. Blackwell Scientific Publications, Oxford, UK. pp. 359–400.
- Weiler, E.W., Albrecht, T., Groth, B., Xia, Z.Q., Luxem, M., Liss, H., Andert, L., and Spengler, P. 1993. Evidence for the involvement of jasmonates and their octadecanoid precursors in the tendril coiling response of *Bryonia dioica*. *Phytochemistry*, **32**: 591–600. doi:10.1016/S0031-9422(00)95142-2.