

Floral reorientation: the restoration of pollination accuracy after accidents

W. Scott Armbruster^{1,2}  and Nathan Muchhala³ 

¹School of Biological Sciences, University of Portsmouth, Portsmouth, PO1 2DY, UK; ²Institute of Arctic Biology, University of Alaska Fairbanks, Fairbanks, AK 99775-7000, USA;

³Department of Biology, University of Missouri-St Louis, St Louis, MO 63121-4499, USA

Author for correspondence:

W. Scott Armbruster

Tel: +44 (0)1983 615361

Email: scott.armbruster@port.ac.uk

Received: 31 October 2019

Accepted: 11 January 2020

New Phytologist (2020) **227**: 232–243

doi: 10.1111/nph.16482

Key words: adaptive accuracy, floral orientation, flower–pollinator fit, late floral development, plant injuries, pollination, zygomorphic flowers.

Summary

- Plants sometimes suffer mechanical injury. The nonlethal collapse of a flowering stalk, for example, can greatly reduce plant fitness if it leads to ‘incorrect’ floral orientation and thus reduced visitation or poor pollination. When floral orientation is important for accurate pollination, as has been suggested for bilaterally symmetrical flowers, we predict that such flowers should have developmental and/or behavioural mechanisms for restoring ‘correct’ orientation after accidents.
- We made observations and conducted experiments on 23 native and cultivated flowering plant species in Australia, South America, North America and Europe.
- We found that flowers with bilateral symmetry usually have the capacity to reorient after accidents, and that this is manifested through rapid bending and/or rotation of pedicels or sexual organs or slower peduncle bending. Floral reorientation restores pollination accuracy and fit with pollinators. However, experimental floral misorientation in eight species with radially symmetrical flowers showed that, with one exception, they had little capacity to reorient their flowers, in line with expectations that the orientation of radially symmetrical flowers does not substantially affect pollination accuracy.
- Our results suggest that quick corrective reorientation of bilaterally symmetrical flowers is adaptive, highlighting a little-studied aspect of plant–pollinator interactions and plant evolution.

Introduction

That ‘accidents happen’ is an aphorism few would argue with. Zoology, as well as medicine, considers the capacity of animals to recover from mechanical injuries as a critical adaptation for mobility, survival and hence fitness (Frank *et al.*, 2017; Chang *et al.*, 2018). Less attention has been paid to recovery from accidental injury in plants, and virtually no research has addressed response to accidents involving flowers. Yet flowering stalks are often subject to accidental collapse, as when a scape blows down in the wind or coarse litter falls onto a stem, causing irreversible bending without severing the vascular system. Such events often push flowers into inappropriate orientations, even if the flowers are otherwise unharmed.

Embedded in the writings of Sprengel (1793), Darwin (1862), Robertson (1888) and, later, Berg (1960) and Stebbins (1974) is the concept that the orientation of flowers relative to the pull of gravity is critical to their proper function and is thus an adaptive trait. These characteristics are functionally important because the movement of flying pollinators and their ability to land on flowers is strongly influenced by their orientation relative to the direction of gravitational pull (Robertson, 1888; Fenster *et al.*, 2009). Orientation may also be important, particularly for laterally and

downwardly oriented flowers as a means to protect nectar from dilution and pollen from damage by rain, UV radiation or heat (Sprengel, 1793; Huang *et al.*, 2002; Aizen, 2003; Wang *et al.*, 2010; Haverkamp *et al.*, 2019; Lin & Forrest, 2019).

Floral orientation affects flower attractiveness and rates of visitation and pollination

Floral orientation can affect various aspects of pollination. Proper floral orientation can increase visitation rates by pollinators relative to that seen on misoriented flowers (Fulton & Hodges, 1999; Ushimaru & Hyodo, 2005; Ushimaru *et al.*, 2006; Wang *et al.*, 2014a). Floral misorientation may change the composition of the flower–visitor fauna, including increased visitation by poor pollinators and resource thieves (Wang *et al.*, 2014b).

In plants with bilaterally symmetrical (zygomorphic) flowers, misorientation of the flower relative to gravity may also affect flower–pollinator fit by causing pollinators to be misoriented, relative to the flower shape, when they land on the flower. This can reduce access to nectar (depressing attractiveness) and/or lower pollination efficiency because pollen is placed in the wrong location on pollinators relative to the expected site of stigma contact by other flowers in the population. Similarly, the stigmas of

misoriented flowers may contact pollinators in the wrong place relative to the expected position of pollen placed by other flowers in the population. The net result is lowering of both male and female components of reproductive fitness, as has been shown in a few empirical studies (Castellanos *et al.*, 2004; Ushimaru *et al.*, 2009; Wang *et al.*, 2014a, b).

Endress (1994) noted that the plane of symmetry in bilaterally symmetrical flowers is usually vertical, that is in line with the pull of gravity. This may reflect selection generated by interactions with pollinating animals, which also exhibit bilateral symmetry and behaviourally orient their planes of symmetry to the vertical, especially in flight. Indeed, floral orientation interacts functionally, in terms of pollination success, with floral symmetry (Neal *et al.*, 1998; Ushimaru & Hyodo, 2005; Ushimaru *et al.*, 2006, 2009; Fenster *et al.*, 2009; Nikkeshi *et al.*, 2015; Reyes *et al.*, 2016). In a review of floral-symmetry research, Neal *et al.* (1998) came to two relevant conclusions: bilaterally symmetrical flowers are usually laterally oriented (face sideways), and floral orientation usually exhibits less within-population variation in plants with bilaterally symmetrical flowers than in plants with radially symmetrical flowers (see also Nikkeshi *et al.*, 2015). Both observations are consistent with the operation of selection for accurate pollination in the evolution of bilaterally symmetrical flowers.

In this context, it is interesting to think about what kinds of flowers have experienced the strongest selection for particular orientations. Floral orientation may be a critical factor in the phenotypic and ecological specialisation (*sensu* Ollerton *et al.*, 2007) of flowers (e.g. Berg, 1960; Fenster *et al.*, 2009). Upwards-facing (vertical) flowers can be approached from many directions (Fig. 1a; Fenster *et al.*, 2009), and thus might not experience fitness decreases from small changes in orientation. By contrast, laterally oriented flowers with bilateral symmetry are approached from consistent directions by pollinators (Fig. 1b,c; Robertson, 1888; Fenster *et al.*, 2009); plants with such flowers may incur significantly reduced fecundity when their flowers vary from the average floral position ('floral misorientation') because they attract fewer pollinators or experience misplacement of anther and stigma contacts with the pollinator, relative to the rest of the population, as noted above.

Floral reorientation

If floral orientation is important to the proper functioning of flowers and, hence, plant reproductive fitness, we predict that plants have the capacity to reorient their flowers if they become misoriented by some accidental event, such as the bending of a peduncle or stem, at least in plant species with flowers lasting longer than it takes for reorientation to be manifested. Rapid, reversible, turgor-mediated reorientation of leaves relative to the sun (heliotropism) has been well documented and shown to increase photosynthesis (diaheliotropism), and/or reduce transpiration (paraheliotropism; Darwin, 1880; Ehleringer & Forseth, 1980). Similarly, stems and roots can, through differential growth, reorient relative to light (phototropism) and gravity (geotropism/gravitropism; Darwin, 1880; Hangarter, 1997; Raven & Johnson, 2002; Christie & Murphy, 2013). Flower and



Fig. 1 Examples of floral orientation and symmetry. (a) *Tricyrtis formosana* (Liliaceae), a species with upwards-facing flowers with radial symmetry; note that pollinators can approach and land on the flower from any direction (arrows). (b) *Dephinium glaucum* (Ranunculaceae), a species with laterally oriented flowers with bilaterally symmetrical calyces and corollas, but with essentially radially symmetrical androecia and gynoecia at the centre. Note that the bumble-bee pollinator can approach the flower and insert its proboscis from only one direction (arrow). (c) *Chamerion angustifolium* (Onagraceae), a species with laterally oriented flowers with radially symmetrical calyces and quasi-bisymmetric corollas; the pendent androecia and gynoecia are bilaterally symmetrical or asymmetric (i.e. one or no line of reflective symmetry).

flowering-shoot reorientation ought to represent natural extensions of such movement capabilities, yet this phenomenon has been remarkably little-studied. There has been some work on the movement of individual floral parts, such as petal closure in response to cold or nightfall (Darwin, 1862, 1880; Bynum & Smith, 2001; Armbruster *et al.*, 2006; Prokop & Fedor, 2016; Kemp & Ellis, 2019) or the movement of stamens to reduce intersexual interference (Sprengel, 1793; Martens, 1936; Ren, 2010; Ren & Tang, 2012; Armbruster *et al.*, 2014a). However, the only studies on whole-flower reorientation of which we are aware are descriptions of floral heliotropism in response to the sun's position (see review in van der Kooi *et al.*, 2019) and recent work by Yon *et al.* (2017) and Haverkamp *et al.* (2019) on the daily shift of floral orientation in *Nicotiana attenuata* from upwards by night (promoting pollination) to downwards by day (keeping the interior of flowers cooler). Despite the lack of

literature, however, we expect that, in the absence of constraints, many plants species have evolved the ability to adjust the orientation of flowers after mishaps knock them out of the alignment that best promotes pollination ('corrective floral reorientation').

The lack of work on reorientation of flowers in response to accidents is a motivation for empirical observations to assess the potential prevalence and significance of this phenomenon. Here we provide observations on 23 species of plants from multiple sites on four continents. We emphasise the taxonomic spread and diversity of mechanisms that plants employ to reorient flowers. Together, these observations suggest both multiple origins of the capacity to reorient flowers and its importance in successful plant reproduction in changing or unstable environments.

Adaptive accuracy of pollination

The effect of floral reorientation on plant reproductive success is best understood and quantified in terms of the adaptive accuracy of flowers with respect to pollination (e.g. Poblete Palacios *et al.*, 2019). Adaptive accuracy relates phenotype to fitness by including both trait precision and mean optimality (Armbruster *et al.*, 2004, 2009b; Hansen *et al.*, 2006). As applied to pollination, 'fundamental' floral accuracy (*sensu* Armbruster, 2014) links individual plant phenotypes and the population mean phenotype to inferred fitness. Previous studies of pollination accuracy have tacitly assumed that flowers have a characteristic orientation, and that accuracy is influenced by this orientation due to its effects on the positions of pollen placement on, and retrieval from, animal pollinators (see Armbruster, 2014; Armbruster *et al.*, 2014b). In a typical bilaterally symmetrical flower, the position of the anthers, relative to where the pollinator lands on, or enters, the flower or blossom (cluster of florets acting like a flower), determines where pollen is placed on the pollinator. Similarly, the position of the stigma determines where the stigma contacts the pollinator and picks up pollen. The optimal anther position for an individual flower is that position resulting in pollen being placed on the pollinator at the point where the stigmas of other flowers in the population are expected to contact the pollinator, as estimated by the mean stigma position for the population. Similarly, the optimal position of the stigma of an individual flower is the position that results in its stigmas contacting the pollinator in the site where pollen is expected to be deposited, as determined by the population-mean anther position.

Adaptive inaccuracy, the inverse of accuracy, captures the deviation of an individual or population from the theoretical or empirically estimated optimal phenotype. This is a measure of population-level or individual-level maladaptation, that is the 'phenotypic load' (by analogy with genetic load)

$$\text{Adaptive inaccuracy} = (\text{Trait mean} - \text{Optimum})^2 + \text{VAR}_{\text{trait}} + \text{VAR}_{\text{optimum}}, \quad \text{Eqn 1}$$

where the optimum for male floral function is the population-mean location of the stigmas, and the optimum for female function is the population-mean location of anthers (Armbruster *et al.*, 2004, 2009a,b; Hansen *et al.*, 2006).

Following this logic, the adaptive inaccuracy of the male component of pollination captures the fitness decrement associated with departure of the mean anther position in a flower, a plant or a population, from the optimum (difference squared) plus the imprecision (variance) in anther position. In turn, the adaptive inaccuracy of the female component of pollination fitness captures the mean deviation of the stigma of a flower, plant or population, from the optimal stigma position (difference squared) plus the imprecision (variance) in stigma positions. These measurements have usually been assessed in linear dimensions of the flowers, for example by relating pistil length to stamen length. However, the approach can be extended to two- (2-D) or three-dimensional (3-D) space using Euclidean distances (Armbruster *et al.*, 2014a).

Materials and Methods

We observed plant responses to both natural and manipulative experiments that shifted flowers from their normal orientation. We observed the effects of accidents changing floral orientation in *Tricyrtis formosana* (Liliaceae; UK), *Passiflora caerulea* (Passifloraceae; UK), *Delphinium glaucum* and *Aconitum delphinifolium* (Ranunculaceae; Alaska), and *Stylidium ciliatum* (Stylidiaceae; Western Australia). Manipulative experiments on *A. delphinifolium* and *S. ciliatum*, plus an opportunistic sample of 18 additional species (Tables 1, 2), involved misorienting flowers by tethering inflorescences or flowers so that flowers were either 45° (Ecuador) or 90° (elsewhere) off their normal orientation. In one case (*Pelargonium* sp.), we transplanted one plant to achieve the same effect. We then measured floral orientation in each species over the next few days after the manipulation. Experiments in Ecuador were conducted by NM and those in Australia, North America and Europe by WSA.

Where possible, 2-D adaptive inaccuracies, scaled to the squared means of flower diameter, were calculated from field measurements or via photographs, measured with IMAGEJ (Rasband, 1997–2018). We used Eqn 1, but omitted the optimum-variance term due to missing information (most species) or, in the case of *Chamerion angustifolium*, in order to make independent estimates of male and female inaccuracies (see Armbruster *et al.*, 2009a, 2014b). The positions of fertile parts were estimated in orthogonal 2-D space, and Euclidian distances from the optimal position were calculated. For most species, the original orientation was assumed to be optimal, and the starting positional imprecisions (variances) were not assessed or incorporated in the inaccuracy calculation (i.e. natural inaccuracy was assumed to be 0%, which corresponds to vertical orientation with no variation). Because adaptive inaccuracy is best represented as the population of Euclidian distances from the optimum, angular data were transformed into 2-D Euclidean distances based on unit circles (radius = 1; Ecuador data), where inaccuracies were scaled to the square of the mean diameter (= 4). For *C. angustifolium* (= *Epilobium angustifolium*, = *Chamaenerion angustifolium*; nomenclature following Elven, 2019), stigma positions and anther positions were separately assessed from photographs, and their natural (starting) imprecisions and

Table 1 Plant species used in manipulative experiments of floral misorientation.

Family	Species	Native (N) vs Cultivated (C)	Sample size (plants)	Sample size (flowers)	Location
Amaryllidaceae	<i>Agapanthus</i> sp.	C	2	24	Pittwater, New South Wales, Australia
Apocynaceae	<i>Mandevilla</i> sp.	C	2	9	Pittwater, New South Wales, Australia
Balsaminaceae	<i>Impatiens</i> sp.	C	4	4	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Campanulaceae	<i>Burmeistera sodiroana</i> Zahlbr.	N	1	1	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Campanulaceae	<i>Centropogon nigricans</i> Zahlbr.	N	1	1	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Caricaceae	<i>Carica papaya</i> L.	C	4	4	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Celastraceae	<i>Stackhousia huegelii</i> Endl.	N	1	6	Mundaring Shire, Western Australia
Ericaceae	<i>Cavendishia bracteata</i> (Ruiz & Pav. ex J.St.-Hil.) Hoerold	N	5	5	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Geraniaceae	<i>Pelargonium</i> sp.	C	1	20	San Luis Range, California, USA
Lamiaceae	<i>Salvia</i> sp.	N	5	5	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Liliaceae	<i>Tricyrtis formosana</i> Baker	C	3 (ramets)	7	Binstead, Isle of Wight, UK
Onagraceae	<i>Chamerion angustifolium</i> (L.) Holub.	N	4	19	Goldstream Valley, Alaska, USA
Orchidaceae	<i>Dactylorhiza fuchsii</i> (Druce) Soó	N	3	21	Binstead, Isle of Wight, UK
Passifloraceae	<i>Passiflora caerulea</i> L.	C	2	30	Binstead, Isle of Wight, UK
Ranunculaceae	<i>Aconitum delphinifolium</i> DC.	N	4	25	Goldstream Valley, Alaska, USA
Ranunculaceae	<i>Clematis pubescens</i> Huegel ex Endl.	N	1	12	Mundaring Shire, Western Australia
Ranunculaceae	<i>Delphinium glaucum</i> S. Wats.	N	3	60	Goldstream Valley, Alaska, USA
Rutaceae	<i>Citrus</i> sp.	C	5	5	Siempre Verde Cloud Forest Reserve, Pichincha, Ecuador
Scrophulariaceae	<i>Diascia</i> spp.	C	5	15	Binstead, Isle of Wight, UK
Scrophulariaceae	<i>Nemesia</i> sp.	C	4	13	Binstead, Isle of Wight, UK
Stylidiaceae	<i>Stylidium ciliatum</i> Lindl.	N	6	30	Mundaring Shire, Western Australia
Stylidiaceae	<i>Stylidium hispidum</i> Lindl.	N	1	5	Mundaring Shire, Western Australia
Stylidiaceae	<i>Stylidium amoenum</i> R.Br.	N	3	11	Mundaring Shire, Western Australia

inaccuracies were measured directly. The population mean stamen position (unmanipulated) was treated as the optimum for stigma position, and the population mean stigma position (unmanipulated) was treated as the optimum for stamen position. All distances were measured in proportions of sepal length (SL) and the resulting inaccuracies were scaled to $(2 \times SL)^2$, a joint measure of flower size and photographic magnification.

To assess the adaptive value of floral reorientation, the inaccuracies of the autonomously reoriented ('corrected') stigma and/or stamen positions were compared to inaccuracies of their initial positions immediately after the manipulation. Reorientation responses are thus also reported as the per cent improvement in accuracy afforded by reorientation relative to the total decrement caused by experimental tethering.

Under the hypothesis that floral reorientation is adaptive because it restores adaptive accuracy of misoriented flowers, we expected to see reorientation best developed in plants with bilaterally symmetrical flowers, which generally have more precise pollen placement and stigma contact with pollinators. To test this idea, we made two planned comparisons of reorientation after experimental misorientation. Each comparison was between species with bilaterally symmetrical flowers and species with more-or-less radially symmetrical flowers. The first comparison

involved assessing the proportion of flowers reorienting to within 30° of the initial orientation after 90° misorientation of flowers of four bilaterally symmetrical species and three radially symmetrical species (Australia). The species assessed were *Agapanthus* sp. (bilaterally symmetrical fertile whorls) and three species of *Stylidium* (all floral whorls bilaterally symmetrical) vs *Clematis* sp., *Stackhousia* sp. and *Mandevilla* sp. (all floral whorls essentially radially symmetrical). The second comparison involved measuring the angular recovery after 45° misorientation of flowers of four bilaterally symmetrical species and three radially symmetrical species (Ecuador; see Table 1). The species assessed were *Impatiens* sp., *Burmeistera sodiroana*, *Centropogon nigricans* and *Salvia* sp. (all floral whorls bilaterally symmetrical) vs *Citrus* sp., *Carica papaya* (all floral whorls radially symmetrical) and *Cavendishia bracteata* (radially symmetrical perianths but weakly bilaterally symmetrical fertile whorls).

Results

Mechanisms of floral reorientation

The survey species revealed phylogenetically widespread reorientation, at least in bilaterally symmetrical flowers. Several distinct

Table 2 Summary of floral responses to 'natural' and manipulative experiments misorienting flowers.

Family: species	Misorientation 'treatment'	Symmetry	Response	Change in mean ² -scaled pollination inaccuracy* (% restoration towards original accuracy)	Effect	Time taken for response or duration of observation
Amaryllidaceae: <i>Agapanthus</i> sp.	Manipulation	Radial perianth, bilateral androecium & gynoecium	Pedicle rotation	–	Accurate orientation usually restored	2–3 d
Apocynaceae: <i>Mandevilla</i> sp.	Manipulation	Radial	Pedicle rotation	0% change	No change in accuracy or pollinator foraging	10–30° rotation d ⁻¹
Balsaminaceae: <i>Impatiens</i> sp.	Manipulation	Bilateral	Pedicle rotation	14.6% → 2.87% (80.3%)	Accurate orientation mostly restored	1 d
Campanulaceae: <i>Burmeistera sodiroana</i>	Manipulation	Bilateral	Pedicle rotation	14.6% → 0.03% (99.8%)	Accurate orientation fully restored	2 d (was bud when misoriented)
Campanulaceae: <i>Centropogon nigricans</i>	Manipulation	Bilateral	Pedicle rotation	14.6% → 1.94% (86.7%)	Accurate orientation fully restored	2 d (was bud when misoriented)
Caricaceae: <i>Carica papaya</i>	Manipulation	Radial	No rotation	0% change	No change in accuracy or pollinator foraging	2 d
Celastraceae: <i>Stackhousia huegelii</i>	Manipulation	Radial	No rotation	0% change	No change in accuracy or pollinator foraging	3 d
Ericaceae: <i>Cavendishia bracteata</i>	Manipulation	Radial perianth, bilateral androecium	Pedicle rotation	14.6% → 10.0% (31.5%)	Small improvement in stamen accuracy	1–2 d
Geraniaceae: <i>Pelargonium</i> sp.	Manipulation	Bilateral	Peduncle bending	–	Accurate orientation fully restored	1–2 d
Lamiaceae: <i>Salvia</i> sp.	Manipulation	Bilateral	Pedicle rotation	14.6% → 0.24% (98.4%)	Accurate orientation fully restored	1 d
Liliaceae: <i>Tricyrtis formosana</i>	Natural 'experiment'	Radial	Pedicle bending	–	Upwards orientation restored	1–2 d
Onagraceae: <i>Chamerion angustifolium</i>	Manipulation	Calyx radial, corolla quasi-bisymmetric, androecium & gynoecium bilateral	Stamen and style bending (no pedicle rotation); (+ bending of peduncle tip)	Anthers: 31.7% → 3.40% (89.3%) Stigmas: 26.3% → 2.18% (91.7%)	Accurate anther and stigma positions mostly restored	Reorientation of fertile parts: 1–2 d Bending of peduncle tip: 2–4 d
Orchidaceae: <i>Dactylorhiza fuchsii</i>	Manipulation	Bilateral	Peduncle bending	73.6% → 7.83% (89.4%)	Accurate orientation restored in young flowers	1–2.5 d
Passifloraceae: <i>Passiflora caerulea</i>	Natural 'experiment'	Radial	No movement	–	No detectable upwards reorientation	No reorientation in 1–2 d lifetime of flower
Ranunculaceae: <i>Aconitum delphinifolium</i>	Natural 'experiment'	Bilateral	Pedicle bending	0% change	Optimal orientation for foraging bumble bees fully restored	?
<i>Aconitum delphinifolium</i>	Manipulation	Perianth bilateral, fertile whorls radial	Pedicle bending	0% change	Optimal orientation for foraging bumble bees fully restored	1 d
Ranunculaceae: <i>Delphinium glaucum</i>	Natural 'experiment'	Perianth bilateral, fertile whorls radial	Pedicle bending	0% change	Optimal orientation for foraging bumble bees fully restored	?
Ranunculaceae: <i>Clematis</i> aff. <i>pubescens</i>	Manipulation	Radial	No rotation	0% change	No change in accuracy or pollinator foraging	3 d of observation
Rutaceae: <i>Citrus</i> sp.	Manipulation	Radial	No rotation	0% change	No change in accuracy or pollinator foraging	2 d of observation

Table 2 (Continued)

Family: species	Misorientation 'treatment'	Symmetry	Response	Change in mean ² -scaled pollination inaccuracy* (% restoration towards original accuracy)	Effect	Time taken for response or duration of observation
Scrophulariaceae: <i>Diascia</i> sp.	Manipulation	Bilateral	Bending and twisting of pedicel	–	Accurate reorientation usually partial, often stopping at c. 30° from original position	2–3 d
Scrophulariaceae: <i>Nemesia</i> sp.	Manipulation	Bilateral	Bending and twisting of pedicel	180° treatment: 100% → 0.07% (99.9%) 90° treatment: 50% → 11.1% (77.8%)	Accurate reorientation in all but old flowers (one excluded in each experiment)	15–34 h 10 h
Stylidiaceae: <i>Stylidium ciliatum</i>	Manipulation and 'natural' experiment	Bilateral	Pedicel rotation, (+ bending peduncle tip)	50.0% → 2.33% (95.3%)	Accurate reorientation in all but old flowers	Pedicel rotation: 1–2 d Peduncle bending: 3–4 d
<i>Stylidium hispidum</i>	Manipulation	Bilateral	Pedicel rotation, (+ bending peduncle tip)	50.0% → 2.44% (95.1%)	Accurate reorientation in all but old flowers	Pedicel rotation: 1–2 d Peduncle bending: 3–4 d
<i>Stylidium amoenum</i>	Manipulation	Bilateral	Pedicel rotation, (+ bending peduncle tip)	50.0% → 7.12% (85.8%)	Accurate reorientation in all but old flowers	Pedicel rotation: 1–2 d Peduncle bending: 3–4 d

*First number is starting inaccuracy after experimental misorientation, and second number is finishing inaccuracy after corrective reorientation.

mechanisms were involved in reorientation of flowers and restoration of floral accuracy (Tables 2, 3). These mechanisms included movements of the peduncle (stalk of an inflorescence) or pedicel (stalk of an individual flower) and varied from species to species. Often two or more mechanisms operated in a single species. We recognised four distinct potential mechanisms of floral reorientation: peduncle bending, pedicel bending, pedicel rotation and bending/twisting of sexual organs.

Peduncle bending We observed bending of the inflorescence stalk leading to partial or full restoration of correct floral orientation (with minimal pedicel bending or rotation) in *Dactylorhiza fuchsii* (Orchidaceae; Fig. 2) and a bilaterally symmetrical *Pelargonium* species. We also observed restoration of floral orientation by bending of the peduncle tip in *Stylidium* spp., *A. delphinifolium*, *D. glaucum* and *C. angustifolium*. In these last four species, peduncle bending occurred as a slow response associated with new growth, in combination with other, faster reorientation processes.

Pedicel bending and/or rotation We observed corrective floral reorientation by pedicel bending in combination with some rotation in *A. delphinifolium* (Fig. 3), *D. glaucum*, *T. formosana*, *Salvia* sp., *B. sodiroana*, *C. nigricans* and *Impatiens* sp. We observed comparatively rapid reorientation of flowers primarily

through rotation of the pedicel (possibly in combination with some bending) in *Agapanthus* sp., *Stylidium* spp. (Fig. 4), *Diascia* spp. and *Nemesia* sp. (Table 2).

Bending and twisting of fertile parts only We observed reorientation of the fertile parts (but not the perianth) in *C. angustifolium*. This species has protandrous flowers with a radially symmetrical calyx and a quasi-bisymmetric corolla, but with fertile parts strongly deflected downwards and thus bilaterally symmetrical. Accurate repositioning (to the ventral location) of anthers was restored very quickly by filament bending or, in some cases, passive filament drooping, but with high imprecision (stamens with dehisced anthers did not reorient by drooping or bending). Accurate stigma positioning was restored over several days by style bending before spreading of the stigma lobes, but only in flowers that were not yet in the female stage at the time of manipulation. Female-stage flowers (with stigmatic lobes already spread) did not reorient styles to restore accurate stigma position.

Interactions between reorientation mechanisms

Although we pooled pedicel rotation and pedicel bending as a single mechanism of reorientation in the above description, these are actually two distinct mechanisms (Table 3). However, it is often hard to determine whether one, the other, or both are

Table 3 A classification of reorientation responses.

Type of response	Description	Example taxa
1. Peduncle bending (with minimal pedicel bending or rotation)	Base or tip of peduncle bends towards vertical leading to restoration of accuracy of all flowers or young flowers, respectively	Only peduncle bends: <i>Dactylorhiza fuchsii</i> (Orchidaceae) <i>Pelargonium</i> sp. (species with bilaterally symmetrical flowers) (Geraniaceae) Peduncle bends in combination with other mechanisms: <i>Chamerion angustifolium</i> (Onagraceae) <i>Aconitum delphinifolium</i> , <i>Delphinium glaucum</i> (Ranunculaceae) <i>Stylidium</i> spp. (Stylidiaceae)
2. Pedicel bending (with some rotation)	Bending of long pedicel restoring 'correct' floral orientation	<i>Tricyrtis formosana</i> (Liliaceae), <i>Aconitum delphinifolium</i> (Ranunculaceae) <i>Delphinium glaucum</i> (Ranunculaceae) <i>Diascia</i> spp. (Scrophulariaceae)
3. Pedicel rotation (possibly with some bending)	Twisting of short pedicel rotating flower to the 'correct' orientation	<i>Stylidium</i> spp. (Stylidiaceae) <i>Impatiens</i> sp. (Balsaminaceae) <i>Burmeistera sodiroana</i> , <i>Centropogon nigricans</i> (Campanulaceae) <i>Salvia</i> sp. (Lamiaceae)
4. Bending (and twisting?) of fertile parts only	Bending of filaments and styles restore 'correct' position of anthers and stigmas, respectively	<i>Chamerion angustifolium</i> (Onagraceae)
5. None	No significant change in orientation or change in orientation does not affect pollination accuracy	<i>Carica papaya</i> (Cariaceae) <i>Citrus</i> sp. (Rutaceae) <i>Clematis</i> sp. (Ranunculaceae) <i>Mandevilla</i> sp. (Apocynaceae) <i>Stackhousia</i> sp. (Stackhousiaceae)

operating. This is an area needing further research. When the pedicel is relatively short, it seems that primarily rotation is occurring. When the pedicel is longer, bending seems to predominate (e.g. Fig. 3; Table 3). However, ruling out the alternative mechanism is not possible without more detailed study.

We observed bending of the main stem in the transplant experiment with the *Pelargonium* with bilaterally symmetrical flowers

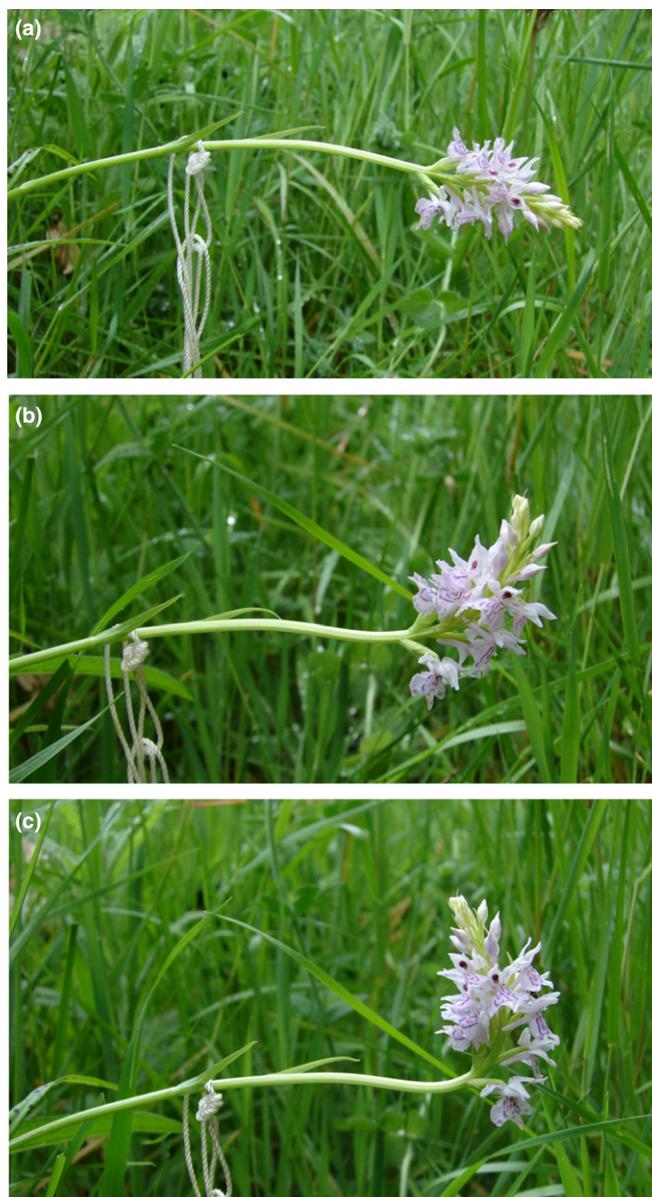


Fig. 2 *Dactylorhiza fuchsii* tethered on 13.06.2019, 13:00 GMT. (a) 13.06.2019, 13:15 GMT. (b) 14.06.2019, 10:15 GMT. (c) 15.06.2019, 09:28 GMT.

(Tables 1, 2). The direct effect of stem bending on floral orientation was negative, that is it moved the floral orientation away from the optimum. However, this effect was compensated for by further bending of the peduncle, ultimately resulting in the peduncle having undergone sigmoidal bending (Fig. 5). Bending of the stem led to restoring the original leaf orientation (presumably improving light capture), as well as restoring the growing axis to vertical. Although we have not observed any examples of bending of the stem as a mechanism for restoring floral orientation, it remains a possibility.

In *Stylidium* spp., *A. delphinifolium* and *D. glaucum*, we observed restoration of floral orientation by pedicel rotation or bending, in combination with a slower response of the peduncle bending upward. In *C. angustifolium*, slow peduncle bending occurred in combination with rapid reorientation of stamens and



Fig. 3 *Aconitum delphinifolium* tethered on 10.07.2019, 18:50 Alaska Summer Time. (a) 10.07.2019, 08:53 Alaska Summer Time. (b) 10.07.2019, 17:06 Alaska Summer Time. (c) 11.07.2019, 09:09 Alaska Summer Time.

styles. This combination of reorientation responses is probably common among plants with bilaterally symmetrical flowers borne in elongated inflorescences, such as spikes.

Effect of reorientation on floral adaptive accuracies of bilaterally symmetrical flowers

The corrective reorientation of flowers or fertile parts through rotation or bending led to dramatic reductions in adaptive inaccuracy (= improved accuracy) in most species (Table 2). In general, species with bilaterally symmetrical fertile whorls exhibited large improvements in pollination accuracy, with 77.8–99.9% recovery of the original accuracy through corrective reorientation within 10–48 h of experimental misorientation. In *C. angustifolia*, restoration of accuracy occurred through corrective reorientation of fertile parts rather than rotation of entire flowers. This led to recovery of accuracy by up to 89.3% for stamens and 91.7% for stigmas. By contrast, there was very little change in the pollination accuracy of flowers with bilaterally symmetrical perianths but with radial symmetry of fertile whorls (androecium and gynoecium), even though they showed whole-flower reorientation (*Aconitum*, *Delphinium*; Table 2). The advantage of rotation in such cases is more likely to be related to

pollinator attraction and flower handling (i.e. flower–pollinator fit with respect to reward access), which is not captured by the pollination-inaccuracy measurement.

Planned comparisons of reorientation in bilaterally vs radially symmetrical flowers

If floral reorientation is simply a consequence of normal plant development and shoot orientation rather than an adaptation related to improving pollination, the extent of floral reorientation in plants with radially symmetrical flowers should be similar to that seen in plants with bilaterally symmetrical flowers. The alternative hypothesis is that bilaterally symmetrical flowers show greater reorientation ability, as noted above.

In Australia, an average (\pm SE) of 3.70% (\pm 3.70%; $n=3$) of the flowers of species with radially symmetrical flowers recovered their orientation to within 30° of the original. By contrast, 95.5% (\pm 1.78%; $n=4$) of flowers on species with bilaterally symmetrical flowers showed recovery of orientation to within 30° of the original orientation (Fig. 6). Note that the difference was ‘significant’ with a parametric test (independent-samples t -test, not assuming equal variances, data subjected to angular transformation: $t=9.80$, $P=0.002$), and marginally ‘significant’ with a

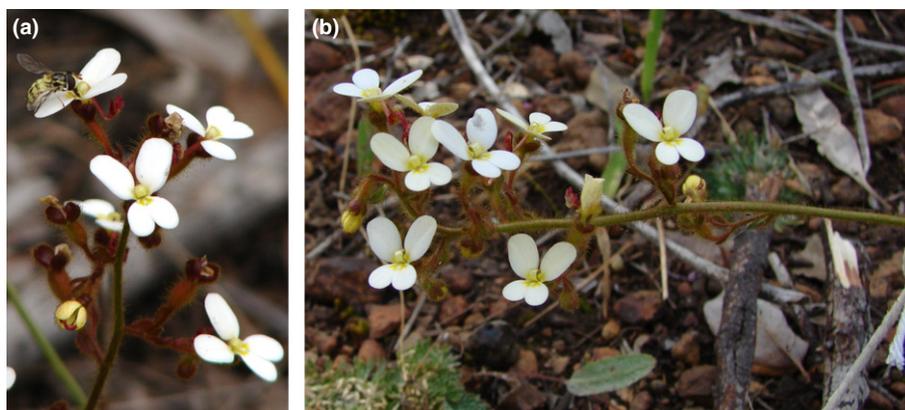


Fig. 4 Floral reorientation in *Styliidium ciliatum*. (a) Normal orientation. (b) Floral reorientation 2 d after floral scape was tethered to the horizontal.

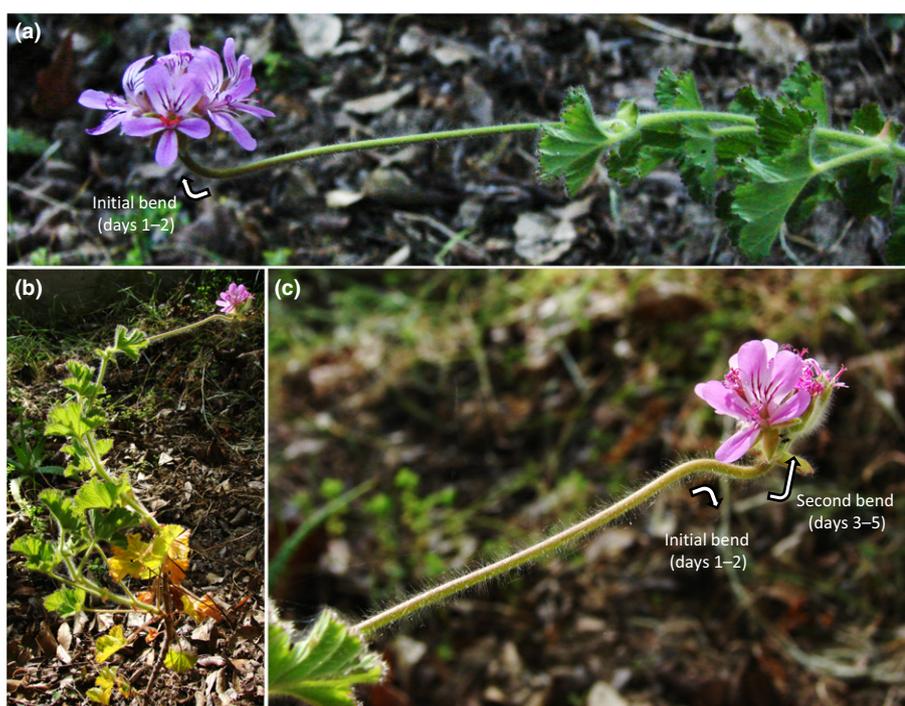


Fig. 5 *Pelargonium* sp. with bilaterally symmetrical flowers. Peduncle bending over 5 d leading to restoration and maintenance of correct floral orientation. Day 0 (not shown): transplanted so main shoot and inflorescence are horizontal (i.e. 90° off original orientation); both flowers and leaves are misoriented. (a) Days 1–2: peduncle bends 90°, restoring 'correct' orientation of weakly bilaterally symmetrical flowers; leaves still 'misoriented'. (b) Day 5: stem has reoriented 90°; leaves are now back to the original horizontal positions (presumably maximising photosynthesis); meanwhile the peduncle has bent another 180° in compensation, so the peduncle is now sigmoidal. (c) Close-up of peduncle showing sigmoidal bending and flowers in correct orientation.

nonparametric test (Mann–Whitney: $T = 10$, $P = 0.05$). However, 'significance' should not be interpreted strictly because the limited sample precluded phylogenetic correction (see Felsenstein, 1985; Armbruster, 1992).

In Ecuador, the reorientation responses were absent in the two radially symmetrical species (*C. papaya* and *Citrus* sp.). *Cavendishia bracteata* displayed an intermediate level of recovery of 9.4° in 24 h. This was markedly less than the four fully bilaterally symmetrical species (Table 2; Fig. 7). The mean corrective reorientation (\pm SE) was 5.18° (\pm 2.14°; $n = 3$) in species with radially symmetrical perianths, vs 35.2° (\pm 4.09°; $n = 4$) in species with bilaterally symmetrical perianths (Fig. 7, independent-samples t -test, not assuming equal variances: $t = 6.49$, $P = 0.002$). The same phylogenetic caution regarding 'significance' mentioned in the previous paragraph applies to these results as well.

Comparison of reorientation in two similar, radially symmetrical flowers

After conducting the above planned comparisons, we added two radially symmetrical, but phenotypically specialised flowers to this survey. *Passiflora* spp. generally bear 1-d flowers that have a complex arrangement of a corolla platform and corona fringe surrounding a nectar trough. The fertile parts are borne a consistent distance above the corolla platform so that nectar-seeking bees of only a certain size will contact the five stamens in the male phase or the three stigmas (at the same height) in the female phase. *Trycirtis formosana* is a distinctive lily with flowers almost perfectly convergent on *Passiflora* flowers, except that the nectar is held in a series of three pairs of saccate spurs at the base of the sepals, rather than in a trough (Table 1; Fig. 1a). In both cases, we could expect

Fig. 6 Planned comparison of reorientation of bilaterally and radially symmetrical flowers in Australia. Per cent of flowers returning to within 30° of original orientation 1–3 d after tethering to –90° (= vertical → horizontal).

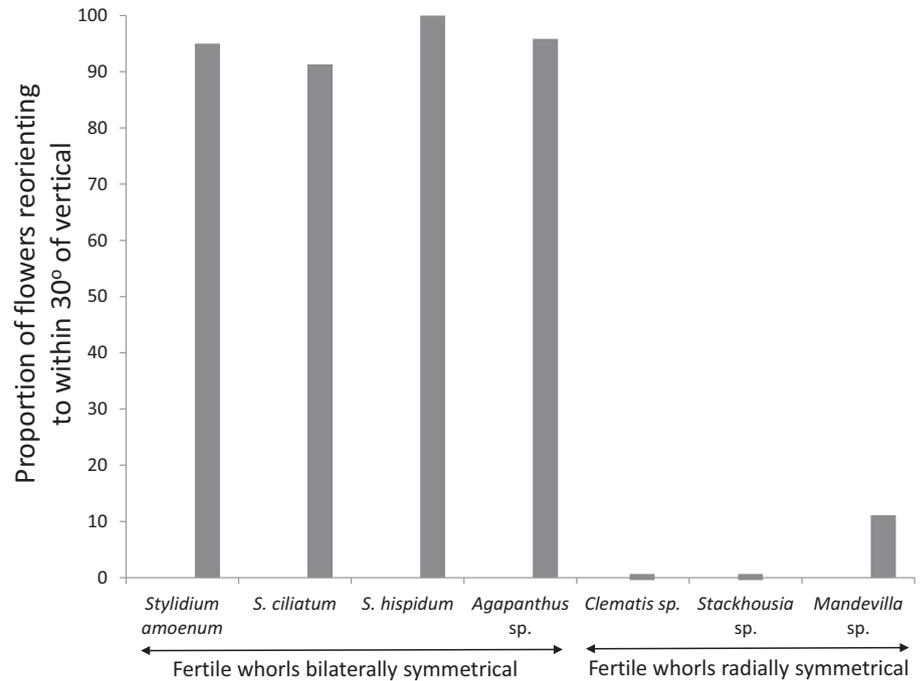
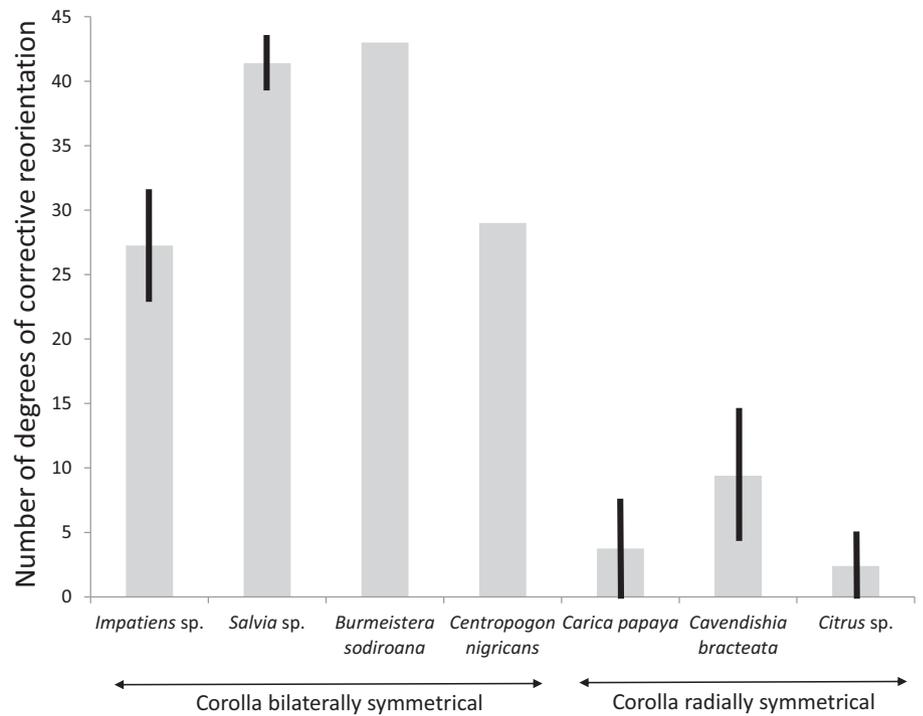


Fig. 7 Planned comparison of reorientation of bilaterally and radially symmetrical flowers in Ecuador. Mean number of degrees of return towards vertical 24 h (48 h for *Burmeistera* and *Centropogon*) after tethering to –45°. Bars are ± 1SD. *Burmeistera* and *Centropogon* are missing SD bars because only one flower was successfully tracked for each species.



floral function and bee pollination to work best when flowers face upwards. (Note that there is a radiation of floral orientations in *Passiflora*, with many hummingbird-pollinated and bat-pollinated species having laterally oriented flowers, often with bilaterally oriented fertile whorls, and some bee-pollinated and some bat-pollinated species having pendent flowers. Here we are referring only to classic bee-pollinated species such as *Passiflora caerulea* and *Passiflora foetida* with normally upward-facing flowers.)

Although the flowers and pollination of these two species are uncannily similar, the difference in the variation in their floral orientation could not have been greater. *Passiflora caerulea* had a large proportion of its flowers apparently misoriented and showed no tendency to reorient them correctively (Table 2). By contrast, *T. formosana* had almost all of its flowers facing perfectly upwards, and, when stems collapse from the vertical, flowers quickly reorient (usually within a day) by pedicel bending

(Table 2). This contrast may be related to one or both of two factors: *Passiflora* is a lax vine with short-lived (usually 1-d) flowers, and *Tricyrtis* is a rhizomatous herb with usually erect ramets and long-lived flowers (4+ d).

Discussion

Floral orientation is critical for accurate pollen placement and retrieval, especially in bilaterally symmetrical (= zygomorphic, = monosymmetric) flowers. However, mechanical accidents happen on occasion, such as misorientation of stems and scapes due to wind, herbivore damage or coarse litter falling from above. Even if a plant survives, fitness costs can be large if pollinators fail to visit misoriented flowers or if pollen is now placed on pollinators (or stigmas contact them) in the 'wrong' place relative to the location of stigma (and anther) contact by the rest of the population (i.e. if flowers experience reduced pollination accuracy). Because the fitness cost of floral misorientation is potentially so great, we expected to find plants to have evolved mechanisms to reorient flowers and thus restore pollination accuracy and pollinator fit after accidents. We found that most species with bilaterally symmetrical flowers exhibited one or a combination of distinct mechanisms of corrective floral reorientation (Table 3).

Experimental misorientation of flowers by 90° of rotation reduced the theoretical pollination accuracy of bilaterally symmetrical flowers by up to *c.* 50% of the maximal decrement (= 180° of rotation). Natural corrective reorientation of these flowers or flower parts returned pollination accuracy to within *c.* 5% of the starting maximum (Table 2; Fig. 6). Similar results were observed with experimental floral misorientation by 45° of rotation: the theoretical pollination accuracy of bilaterally symmetrical flowers was reduced by *c.* 15% of the maximum possible decrement. Natural corrective reorientation of these flowers returned pollination accuracy to within a few per cent of the starting maximum (Table 2; Fig. 7). Thus, our survey suggests that most plants with bilaterally symmetrical flowers have the capacity to reorient their flowers, returning pollination accuracy to within a few per cent of the original value within a day or two.

It is interesting that two species with bilaterally symmetrical perianths but radially symmetrical fertile parts, *Aconitum* and *Delphinium*, also showed dramatic corrective reorientation. For these species, we estimated that pollination accuracy would be unaffected by misorientation, because the stamens and stigmas are centrally located and their orientation would not affect sites of pollen placement or stigma contact. However, reorientation may be adaptive because attractiveness depends on pollinators handling the flowers and reaching the nectar efficiently. The nectar spur being in the 'wrong' place in misoriented flowers may reduce attractiveness (because of reduced nectar accessibility) and visitation rates. Hence attraction, not pollination efficiency, is likely to be a key factor in some species (i.e. those with radially symmetrical fertile whorls but bilaterally symmetrical perianths).

By contrast, species with bilaterally symmetrical fertile whorls but with radially or subradially symmetrical perianths (e.g. *Chamerion*, *Agapanthus* and, to a lesser extent, *Cavendishia*) do experience some improvement in pollination accuracy with

corrective reorientation of flowers or fertile floral parts. Visitation rates are presumably unaffected by rotation of the corolla, however. Indeed, *Chamerion* has not evolved the capacity to correctively reorient the perianth – only the fertile parts move correctively, whereas *Agapanthus* rotates the entire flower.

Not all radially symmetrical flowers are unable to reorient. Notably, *T. formosana* showed a strong and rapid response to misorientation, usually restoring upwards orientation of the misoriented flowers within a day. However, *P. caerulea*, with similarly complex flowers pollinated in a similar fashion, did not reorient its flowers. The difference may be explained by the *Tricyrtis* having multiday flowers and the *Passiflora* 1- or 2-d flowers. Reorientation in species with short-lived flowers may not occur fast enough to affect pollination fitness significantly. Indeed, all the species we examined in this study that had the ability to reorient flowers correctively had flowers lasting several days or longer.

We suggest that corrective reorientation of flowers after accidents is an underappreciated plant behaviour worthy of greater scrutiny. It would be valuable to next assess experimentally the extent to which reorientation increases pollinator visitation and/or pollen transfer relative to flowers that remain misoriented. Further research may reveal that other morphological and developmental features of flowers and inflorescences are unappreciated adaptations for floral reorientation after mechanical accidents. For example, the evolution of elongated pedicels could be favoured because they promote the capacity for rapid floral reorientation (see Figs 1b, 2). Future research on floral function and pollination should explore such possibilities, as well as extend the sampling of reorientation capabilities across a broader range of flowering plant species.

Acknowledgements

WSA acknowledges grant support from the Royal Society of London and from the British Council. WSA thanks Juliet Wege for help in identifying plants and Marjory Caw for hospitality during stays in Parkerville, WA. NM acknowledges the National Geographic Society for grant support and Daniela Proaño for assistance in the field.

Author contributions

WSA and NM conceived of the initial hypotheses and conducted fieldwork. WSA wrote the first draft, and both authors contributed revisions.

ORCID

W. Scott Armbruster  <https://orcid.org/0000-0001-8057-4116>

Nathan Muchhala  <https://orcid.org/0000-0002-4423-5130>

References

- Aizen MA. 2003. Down-facing flowers, hummingbirds and rain. *Taxon* 52: 675–680.
- Armbruster WS. 1992. Phylogeny and the evolution of plant–animal interactions. *BioScience* 42: 12–20.

- Armbruster WS. 2014. Floral specialization and angiosperm diversity: phenotypic divergence, fitness trade-offs and realized pollination accuracy. *AoB Plants* 6: plu003.
- Armbruster WS, Corbet SA, Vey AJM, Liu SJ, Huang SQ. 2014a. In the right place at the right time: *Parnassia* resolves the herkogamy dilemma by accurate repositioning of stamens and stigmas. *Annals of Botany* 113: 97–103.
- Armbruster WS, Hansen TF, Pélabon C, Bolstad GH. 2009a. Macroevolutionary patterns of pollination accuracy: a comparison of three genera. *New Phytologist* 183: 600–617.
- Armbruster WS, Hansen TF, Pélabon C, Pérez-Barrales R, Maad J. 2009b. The adaptive accuracy of flowers: measurement and microevolutionary patterns. *Annals of Botany* 103: 1529–1545.
- Armbruster WS, Pélabon C, Hansen T, Mulder CPH. 2004. Floral integration and modularity: distinguishing complex adaptations from genetic constraints. In: Pigliucci M, Preston KA, eds. *Phenotypic integration. Studying the ecology and evolution of complex phenotypes*. Oxford, UK: Oxford University Press, 23–49.
- Armbruster WS, Pérez-Barrales R, Arroyo J, Edwards ME, Vargas P. 2006. Three-dimensional reciprocity of floral morphs in wild flax (*Linum suffruticosum*): a new twist on heterostyly. *New Phytologist* 171: 581–590.
- Armbruster WS, Shi X-Q, Huang S-Q. 2014b. Do specialised flowers promote reproductive isolation? Realised pollination accuracy of three sympatric *Pedicularis* species. *Annals of Botany* 113: 331–340.
- Berg RL. 1960. The ecological significance of correlation pleiades. *Evolution* 14: 171–180.
- Bynum MR, Smith WK. 2001. Floral movements in response to thunderstorms improve reproductive effort in the alpine species *Gentiana algida* (Gentianaceae). *American Journal of Botany* 88: 1088–1095.
- Castellanos MC, Wilson P, Thomson JD. 2004. 'Anti-bee' and 'pro-bird' changes during the evolution of hummingbird pollination in *Penstemon* flowers. *Journal of Evolutionary Biology* 17: 876–885.
- Chang YH, Housley SN, Hart KS, Nardelli P, Nichols RT, Maas H, Cope TC. 2018. Progressive adaptation of whole-limb kinematics after peripheral nerve injury. *Biology Open* 7: bio028852.
- Christie JM, Murphy AS. 2013. Shoot phototropism in higher plants: new light through old concepts. *American Journal of Botany* 100: 35–46.
- Darwin CR. 1862. *On the various contrivances by which British and foreign orchids are fertilised by insects*. London, UK: Murray.
- Darwin CR. 1880. *The power of movement in plants*. London, UK: John Murray.
- Ehleringer J, Forseth I. 1980. Solar tracking by plants. *Science* 210: 1094–1098.
- Elven R (ed.). 2019. *Annotated checklist of the Panarctic Flora (PAF). Vascular plants*. [WWW document] URL <http://panarcticflora.org/> [accessed 1 January 2020].
- Endress PK. 1994. *Diversity and evolutionary biology of tropical flowers*. Cambridge, UK: Cambridge University Press.
- Felsenstein J. 1985. Phylogenies and the comparative method. *American Naturalist* 126: 1–25.
- Fenster CB, Armbruster WS, Dudash MR. 2009. Specialization of flowers: is floral orientation an overlooked first step? *New Phytologist* 183: 502–506.
- Frank ET, Schmitt T, Hovestadt T, Mitesser O, Stiegler J, Linsenmair KE. 2017. Saving the injured: rescue behavior in the termite-hunting ant *Megaponera analis*. *Science Advances* 3: e1602187.
- Fulton M, Hodges SA. 1999. Floral isolation between *Aquilegia formosa* and *Aquilegia pubescens*. *Proceedings of the Royal Society B: Biological Sciences* 266: 2247–2252.
- Hangerter RP. 1997. Gravity, light, and plant form. *Plant, Cell & Environment* 20: 796–800.
- Hansen TF, Carter AJR, Pelabon C. 2006. On adaptive accuracy and precision in natural populations. *American Naturalist* 168: 168–181.
- Haverkamp A, Li X, Hansson BS, Baldwin IT, Knaden M, Yon F. 2019. Flower movement balances pollinator needs and pollen protection. *Ecology* 100: UNSP e02553.
- Huang SQ, Takahashi Y, Dafni A. 2002. Why does the flower stalk of *Pulsatilla cernua* (Ranunculaceae) bend during anthesis? *American Journal of Botany* 89: 1599–1603.
- Kemp JE, Ellis AG. 2019. Cryptic petal coloration decreases floral apparency and herbivory in nocturnally closing daisies. *Functional Ecology* 33: 2130–2141.
- Lin SY, Forrest JRK. 2019. The function of floral orientation in bluebells: interactions with pollinators and rain in two species of *Mertensia* (Boraginaceae). *Journal of Plant Ecology* 12: 113–123.
- Martens P. 1936. Pollination et biologie florale chez *Parnassia palustris*. *Bulletin de la Société Royale de Botanique de Belgique* 68: 183–221.
- Neal PR, Dafni A, Giurfa M. 1998. Floral symmetry and its role in plant–pollinator systems: terminology, distribution, and hypotheses. *Annual Review of Ecology & Systematics* 29: 345–373.
- Nikkeshi A, Kurimoto D, Ushimaru A. 2015. Low flower-size variation in bilaterally symmetrical flowers: support for the pollination precision hypothesis. *American Journal of Botany* 102: 2032–2040.
- Ollerton J, Killick A, Lamborn E, Watts S, Whiston M. 2007. Multiple meanings and modes: on the many ways to be a generalist flower. *Taxon* 56: 717–728.
- Poblete Palacios JA, Soteras F, Cocucci AA. 2019. Mechanical fit between flower and pollinators in relation to realized precision and accuracy in the hummingbird-pollinated *Dolichandra cynanchoides*. *Biological Journal of the Linnean Society* 126: 655–665.
- Prokop P, Fedor P. 2016. Why do flowers close at night? Experiments with the lesser celandine *Ficaria verna* Huds. (Ranunculaceae). *Biological Journal of the Linnean Society* 118: 698–702.
- Rasband WS. 1997–2018. *ImageJ*. Bethesda, MD, USA: US National Institutes of Health. [WWW document] URL <https://imagej.nih.gov/ij/> [accessed 22 September 2019].
- Raven PH, Johnson GB. 2002. *Biology (6th edn)*. New York, NY, USA: McGraw-Hill.
- Ren M-X. 2010. Stamen movements in hermaphroditic flowers: diversity and adaptive significance. *Chinese Journal of Plant Ecology* 34: 867–875.
- Ren M-X, Tang J-Y. 2012. Up and down: stamen movements in *Ruta graveolens* (Rutaceae) enhance both outcrossing and delayed selfing. *Annals of Botany* 110: 1017–1025.
- Reyes E, Sauquet H, Nadot S. 2016. Perianth symmetry changed at least 199 times in angiosperm evolution. *Taxon* 65: 945–964.
- Robertson C. 1888. Zygomorphy and its causes. I–III. *Botanical Gazette* 13: 146–151, 203–208, 224–230.
- Sprengel CK. 1793. *Das Entdeckte Geheimnis der Natur im Bau und in der Befruchtung der Blumen*. Lehre, Germany: J. Cramer. Facsimile reprint 1972.
- Stebbins GL. 1974. *Flowering plants. Evolution above the species level*. Cambridge, MA, USA: Harvard University Press.
- Ushimaru A, Dohzono I, Takami Y, Hyodo F. 2009. Flower orientation enhances pollen transfer in bilaterally symmetrical flowers. *Oecologia* 160: 667–674.
- Ushimaru A, Hyodo F. 2005. Why do bilaterally symmetrical flowers orient vertically? Flower orientation influences pollinator landing behaviour. *Evolutionary Ecology Research* 7: 151–160.
- Ushimaru A, Kawase D, Imamura A. 2006. Flowers adaptively face down-slope in 10 forest-floor herbs. *Functional Ecology* 20: 585–591.
- van der Kooij CJ, Kevan PG, Koski MH. 2019. The thermal ecology of flowers. *Annals of Botany* 124: 343–353.
- Wang H, Tie S, Yu D, Guo Y-H, Yang C-F. 2014a. Change of floral orientation within an inflorescence affects pollinator behavior and pollination efficiency in a bee-pollinated plant, *Corydalis sheareri*. *PLoS ONE* 9: e95381.
- Wang H, Xiao C-L, Gituru RW, Xiong Z, Yu D, Guo Y-H, Yang C-F. 2014b. Change of floral orientation affects pollinator diversity and their relative importance in an alpine plant with generalized pollination system, *Geranium refractum* (Geraniaceae). *Plant Ecology* 215: 1211–1219.
- Wang Y, Meng LL, Yang YP, Duan YW. 2010. Change in floral orientation in *Anisodus luridus* (Solanaceae) protects pollen grains and facilitates development of fertilized ovules. *American Journal of Botany* 97: 1618–1624.
- Yon F, Kessler D, Joo Y, Llorca LC, Kim SG, Baldwin IT. 2017. Fitness consequences of altering floral circadian oscillations for *Nicotiana attenuata*. *Journal of Integrative Plant Biology* 59: 180–189.

See also the Commentary on this article by Goodwillie, 227: 8–9.