What Maintains Flower Colour Variation within Populations?

Yuval Sapir, M. Kate Gallagher and Esther Senden

Natural selection acts on phenotypic trait variation. Understanding the mechanisms that create and maintain trait variation is fundamental to understanding the breadth of diversity seen on Earth. Flower colour is among the most conspicuous and highly diverse traits in nature. Most flowering plant populations have uniform floral colours, but a minority exhibit within-population colour variation, either discrete (polymorphic) or continuous. Colour variation is commonly maintained by balancing selection through multiple pollinators, opposing selection regimes, or fluctuating selection. Variation can also be maintained by heterozygote advantage or frequency-dependent selection. Neutral processes, or a lack of selection, may maintain variation, although this remains largely untested. We suggest several prospective research directions that may provide insight into the evolutionary drivers of trait variation.

Within-Population Flower Colour Variation

Phenotypic trait variation is the raw material upon which natural selection acts. Variation is increased by genetic recombination or mutation and is eroded as selection eliminates all but the most favourable genetic combinations [1]. While the mechanisms that create and maintain trait variation among populations or species have been well studied [2,3], less attention has been paid to cases where phenotypic variation is maintained within populations. Individuals within the same population should experience similar selection regimes leading to trait homogeneity. Investigating the mechanisms that counteract this homogenising process can provide a deeper understanding of how diversity is created and why within-population variation in potentially adaptive traits is so rarely maintained. Here we explore these questions in relation to flower colour. Flower colour sits at the intersection of ecological interactions, genetics, and evolution and thus provides an excellent system to understand the causes of variation and, more importantly, what maintains or erodes it.

In most species, flowers (or floral structures) within populations are monomorphic, all having the same colour. But in some species flower colour can also be polymorphic, exhibiting two or more discrete colours, as in the case of Linanthus paryae [4], or continuous, displaying colours on a continuous colour scale as is the case in several Iris species [5] (Figure 1 and Table 1 for examples of different types of colour variation). Discrete colour polymorphism may result from genetic variation at a single locus (or several loci) [6,7], whereas continuous colour variation can result from either multilocus genetic variation with additive effects or differential expression of pigment biosynthesis genes [8,9] (Box 1).

In this review, the term ‘colour variation’ includes both polymorphic (discrete) and continuous colour categories. Moreover, while both types of colour variation may stem from environmentally dependent phenotypic plasticity, here we focus on genetically based, heritable colour variation associated with floral visual signalling. Other types of colour variation, including variation in colour patterns [10,11], rare colour mutants resulting from a lack of pigment synthesis [6,12], or colour change at the individual flower level [13,14], while interesting, fall outside the scope of this review.

Highlights

Phenotypic trait variation is the raw material of natural selection. Within-population flower colour variation can be a useful characteristic to understand fundamental evolutionary processes that create and maintain trait variation.

Flower colour variation has largely been studied at the species level. Understanding the mechanisms that create and maintain within-population flower colour variation is fundamental to understanding the breadth of diversity seen on Earth.

Most research on within-population flower colour variation explores how balancing selection maintains variation through multiple selection regimes. Yet, theory suggests many other possible mechanisms.

Flower colour is traditionally considered an adaptation to pollinator visual perception. In populations with either discrete or continuous colour variation, however, nonpollinator agents of selection may play an important role.

The role of random or neutral processes in maintaining variation remains largely untested but should not be ruled out.

Exploring a broader array of mechanisms in the study of trait variation is needed to disentangle random processes from selection-driven processes in creating biodiversity.

*Correspondence: sapir@tauex.tau.ac.il (Y. Sapir).
Twitter: @yuval_sapir (Y. Sapir) and @kate_mkg (M.K. Gallagher).
Flower colour is the product of biochemical processes that are influenced by a combination of genetic, physiological, and environmental factors. While flower colour is largely determined by the identity and composition of pigments in floral tissues (Box 1), the colour’s chromatic traits, including dominant wavelength (hue), spectral purity (saturation), and intensity (brightness) [15], can also be influenced by other factors, including soil or cell pH [16,17], water availability [18], or petal (surface) structure [19,20]. How flower colour is perceived, however, can be quite different from what the colour is, depending on the visual system of the perceiver.

Flower colour is a key visual signal used to attract pollinators, but visual systems vary widely among pollinator guilds [21]. The resulting differences in spectral sensitivity influence which colours pollinators can perceive, as well as their ability to detect differences among colours or even to detect specific colours against the background colours in an entire habitat [22]. Thus, flower colour diversity in angiosperms is thought to be an adaptation to the vast diversity of animal pollinators.

Flowers use colour to signal the presence of a potential reward to animal pollinators, typically nectar and pollen [15,23]. The association of flower colour with floral rewards affects pollinator behaviour, which, in turn, directly affects plant fitness. Monomorphic flower colour usually arises from directional selection (see Glossary), typically driven by pollinators, which leads to the loss of genetic and phenotypic variation from a population [24–26]. Monomorphic flower colour can also arise through positive frequency-dependent selection. In this case, frequency-dependent foraging by pollinators leads to the loss of low-frequency morphs by conferring a fitness advantage to the common morph, giving rise to monomorphism [24,27].

These two mechanisms, directional selection and positive frequency-dependent selection, are the primary ways that monomorphic flower colour is maintained in populations. Yet, transitions between colours are common across ecosystems and phylogenetic clades [28,29]. Given that colour shifts are common, why do relatively few species maintain stable colour variation within populations [30,31]? Numerous mechanisms that drive and maintain variation have been proposed [32,33], but only some have been explored in the context of flower colour. Recently, several studies on flower colour variation have investigated the formation of such variation and explored the possible mechanisms that maintain flower colour variation within populations. Here we focus on the ecological and genetic processes that drive and maintain within-population flower colour variation.

Mechanisms That Maintain Flower Colour Variation

**Heterozygote Advantage**

A simple genetic mechanism that can maintain flower variation is the Mendelian heritability system of one locus with two alleles, where heterozygotes are as fit as or more fit than both the homozygotes (overdominance [34–37]). For example, heterozygote plants of the alpine orchid, *Gymnadenia rhellicani*, form an intermediate colour phenotype. Bee and fly pollinators each visit one of the homozygotes but will also visit the heterozygote; thus, the overall visitation rate to the heterozygote gives it higher fitness than either homozygote [37]. In another case of the Japanese *Sisyrinchium* sp., flower colour dimorphism is maintained due to higher fitness of the white heterozygote than either dominant or recessive homozygous colour morphs, although the dominant homozygote is also white [34]. Nonetheless, the cause of this heterozygote advantage is not known. Maintaining variation through heterozygote advantage requires understanding the ecological factors and possible pleiotropic effects that drive higher heterozygote fitness, which may be why it has only rarely been documented [35,36,38].

Glossary

**Assortative mating:** also referred to as "positive assortative mating or homogamy," a mating pattern in which individuals with similar phenotypes mate with one another more frequently than would be expected under a random mating pattern. Assortative mating leads to the loss of low-frequency morphs by conferring a fitness advantage to the common morph, giving rise to monomorphism [24,27].

**Disassortative mating:** when individuals with dissimilar genotypes or phenotypes mate with one another more frequently than would be expected under random mating. Disassortative mating alters allele frequencies and tends to stabilise them at intermediate levels.

**Frequency-dependent selection:** when the fitness of an allele (or trait) in the population depends upon its frequency. In positive frequency-dependent selection, the fitness of a phenotype or genotype increases as it becomes more common. In negative frequency-dependent selection, the fitness of a phenotype or genotype decreases as it becomes more common.

**Overdominance:** also referred to as "heterosis," a condition in genetics where the phenotype of the heterozygote lies outside the phenotypical range of both homozygous parents. Overdominance can also be described as heterozygote advantage, wherein heterozygous individuals have a higher fitness than homozygous individuals.

**Pleiotropy:** where one gene influences two or more phenotypic traits.
Balancing Selection

**Balancing selection** occurs when phenotypic traits are the target of different selection regimes [33]. While each selection regime alone may cause homogenization of a particular phenotype through directional selection, their combination leads to variation. Balancing selection on allele frequencies requires that an allele is beneficial or deleterious as a function of the selection regime and that alterations in these selection regimes will change the relative fitness of this allele [33]. We explore the evidence for balancing selection leading to stable within-population flower colour variation as caused by multiple selection regimes or fluctuating selection.

Figure 1. Within-Population Flower Colour Variation: Examples for Different Levels of Variation – Monomorphism (A, B), Dimorphism (C–E), Polymorphism (F), and Continuous Variation (G, H). (A) Monomorphic yellow flowers of *Senecio glaucus*, Bessor, Israel. (B) Monomorphic population of *Anemone coronaria*, Shokeda, Israel. (C) Monomorphic population of *Erucaria rostrata*, Mamshit, Israel. (D) Blue and orange flowers of *Lysimachia arvensis*, Hermon, Israel. (E) Blue and yellow flowers of *Iris lutescens*, Navas, France. (F) Blue and white flowers of *Linanthus parryae*, California, USA. (G) Dark-purple tube (left) and white tube (right) flowers of *Linum pubescense*, Carmel, Israel. (H) Polymorphic population of *Anemone coronaria*, Megido, Israel. (I) Continuous colour variation of *Castilleja miniata*, California, USA. (J) Flower colour variation in *Iris petrana*, Yeruham, Israel. Photo credits: Yuval Sapir (A, C, E, I), Yael Orgad (B, D, H), Naomi Fraga (F), Merav Lebel (G), Sarah Jacobs (I), and Hilla Ziv (J).
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Floral colour variation may represent an adaptive compromise to selection by multiple agents. When multiple selection agents act in the same direction, the outcome may be accelerated and stronger directional selection, leading to monomorphism [39]. But two or more selection agents acting in different directions may lead to flower colour variation, either chromatic or achromatic.

### Biotic Agents

**Multiple Pollinators.** When preferences for flower colour vary among individual pollinators or pollinator guilds, colour constancy can maintain colour variation [40,41]. Within populations of the same species, colour constancy leads to **assortative mating** that maintains colour polymorphism, as is the case with *Anemone pavonina* subsp. *candida* (hawkmoth) and subsp. *collina* (hummingbird) [42]. Moreover, divergent preferences among pollinators can create both discrete colour polymorphism and continuous colour variation [42,43], depending on the extent of the selection regimes and the detection thresholds of each pollinator. It seems likely that pollinator-mediated selection on colour will lead to continuous colour variation in cases where colour differences are smaller than the detection threshold of the main pollinators [44].

**Multiple Behaviours.** Some pollinators exhibit different behaviours for different colour morphs of the same flower species, thus maintaining the polymorphism by visiting the different colour morphs.

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*Variation types categorised as dimorphic (two colour morphs), polymorphic (more than two discrete colour morphs), and continuous (gradual colour shades).

Species names are listed as they were in the original papers. Family names are listed according to http://www.plantsoftheworldonline.org/.

Categorised as desert, Mediterranean, temperate, arctic, montane, or tropic.
Box 1. The Molecular Basis of Flower Colour

**Pigment Biosynthesis**

Flower colour is the result of pigment accumulation in petal tissue. Three floral pigment types exist: carotenoids, flavonoids, and betalains.

Carotenoids predominate the evolution of flowering plants. They are involved in photosynthesis and are precursors for two plant hormones [99], suggesting that their role in producing yellow to orange flower colours is secondary. Carotenoids are synthesised in plastids and are lipid soluble. The pathway consists of core enzymes and is strongly regulated through post-transcriptional mechanisms as well as external factors, including stress, diurnal oscillations, and mycorrhizal interactions [100].

Flavonoids are polyphenolic secondary metabolites, among which anthocyanins are the most common. Anthocyanins range from red, to pink and purple, to blue [101]. Anthocyanins are synthesised in the cytosol and transported and accumulated in the cell vacuole and cell wall [102]. The anthocyanin biosynthesis pathway (ABP) and its subpathways each produce different pigments that are regulated by three transcription factors, among which the R2R3-MYB family is considered most important [103]. Alterations in the activity of a single R2R3-MYB can lead to regulatory-induced changes of the up- and downstream genes along the pathway, as well as changes in anthocyanin pigment composition [104]. Betalains are red-violet or yellow-orange, water-soluble, nitrogen-containing compounds derived from the amino acid tyrosine [105]. They occur exclusively in the Caryophyllales and do not co-occur with anthocyanins [106]. The betalain pathway is regulated by R2R3-MYB transcription factors, similar to the flavonoid pathway [107].

**Mechanisms Underlying Colour Variation and Polymorphism**

Among all pigments, anthocyanins are responsible for the widest range of flower colour variation and are most often associated with colour-polymorphic species [30]. Shifts in flower colour are hypothesised to result from either mutation in structural ABP genes or transcriptional regulation of those genes [108], with the latter thought to be more common [98]. Anthocyanins occur in both floral and vegetative tissues, and in addition to altering colour, they also function as stress-induced secondary metabolites that are important for the mitigation of stresses, including oxidative stress, herbivory, drought, and frost [109]. Due to their role in mitigating stress, shutdown of the entire pathway can have severe negative fitness effects [110]. Within-population colour variation may therefore be the outcome of a balance between genetic mutations, direct selection on colour, and selection through the pleiotropic effects of colour genes associated with environmental heterogeneity [28,54,111].

The absorption of light by pigments, and thus the colour observed, is also influenced by the diversity of pigments and their relative concentration in the cell [112,113]. Copigmentation of anthocyanins with flavonoids, flavones, or carotenoids can be important in modifying pigment colour [114,115]. Besides pigmentation, vacuolar pH, as well as cellular structures, can modify the observed colour signal [16,112].

Flower colour is thus the result of biochemical processes that are influenced by a combination of molecular, physiological, and ecological factors. Understanding how pigment synthesis is regulated, by studying the molecular basis of flower colour shifts, may shed light on the origin and maintenance of the dazzling palette of colour variation observed in nature.

[45]. For example, *Linum pubescens* flowers are dimorphic with either dark purple or white tubes. Its bee-fly pollinators, *Usia bicolor*, visit purple-tubed flowers in the morning for nectar feeding and visit white-tubed flowers in the afternoon for mating rendezvous [46]. Consequently, both colour morphs have similar fitness, which explains the persistence of the colour dimorphism over years and across populations [47]. Divergent selection by different behaviours of the same pollinator has also been observed in a few studies in South African daisies [48]. In general, however, this is not a commonly reported mechanism for maintaining within-population colour polymorphism, and it has never been associated with continuous colour variation.

**Mutualists and Antagonists.** Colour polymorphism may result from conflicting selection simultaneously exerted by mutualists and antagonists [49–51]. In *Raphanus sativus*, both pollinators and herbivores prefer light-coloured morphs; thus, dark morphs are maintained by avoiding predation despite lower pollination rates [51]. Similarly, floral colour polymorphism in *Claytonia virginica* is maintained through positive pollinator-mediated selection for redder flowers, whereas white flowers receive less damage from herbivores and pathogens [49]. Hence, differential selection for one colour by mutualists and for another colour by antagonists may maintain colour polymorphism.
Different Selection through Male and Female Function. Variance in selection regimes through male and female function can drive colour dimorphism. This is a relatively common phenomenon in animals [52] but rarely reported for flower colour. We have found only one such example, and only for pollen colour in the hermaphrodite species *Campanula americana*. In this species, female fitness is higher in light pollen plants due to higher fruit set, whereas dark pollen plants receive more pollinator visits and exhibit elevated pollen performance under heat stress, resulting in a male fitness advantage [53]. It remains unclear whether flower colour dimorphism can be attributed to differential selection through male and female function in plants.

Abiotic Agents
Abiotic factors exert selection on flower colour, either through the direct effect of floral pigment defence against environmental stresses [54] or indirectly due to a genetic correlation between flower colour and another trait (pleiotropy [55]). Within-population colour variation may arise when abiotic factors exert selection in a contrasting direction compared with biotic selection agents. For example, contrasting selection by herbivores and drought indirectly maintains flower colour dimorphism in the selfing species *Boechera stricta* through pleiotropic traits in the vegetative tissues [18]. Similarly, in several bird-pollinated *Protea* species in South Africa, white morphs cope better with environmental stressors but have lower herbivory defences than co-occurring pink morphs [56]. In these cases, pollinators do not exhibit a preference for either colour morph and thus play no part as selection agents on colour.

In some species, the frequency of within-population colour variation differs geographically, presumably driven by associated shifts in selection agents. This is likely the case for two Eastern Mediterranean *Anemone* species, *Anemone pavonina* and *Anemone coronaria*, where populations in drier regions have only scarlet flowers, whereas those in mesic or colder regions are polymorphic [57,58]. The scarlet flowers are pollinated exclusively by glaphyrid beetles, which, due to their colour perception, prefer scarlet-coloured flowers [59]. Thus, in drier regions, the combination of beetle pollination and drought tolerance linked to the scarlet allele results in monomorphic scarlet populations. In mesic regions, however, *Anemone* are likely wind pollinated, and therefore populations are likely colour polymorphic due to random mating and a lack of beetle-mediated selection. Other abiotic factors that can drive colour polymorphism include water availability (blue and scarlet *Lysimachia arvensis* [60,61]) and frost (white and blue *Parrya nudicaulis* [62]). UV radiation explains one of the few examples of continuous flower colour variation, ranging from white to dark pink, in *Silene vulgaris* [63]. Compared with other mechanisms, examples of abiotic selection agents driving flower colour variation are relatively abundant. It remains unclear, however, whether this prevalence reflects a reality in nature or confirmation bias.

Fluctuating Selection
Spatial or temporal variation in selection may lead to maintenance of phenotypic variation, especially if there is generation overlap [64–66]. That said, fluctuating selection creating flower colour polymorphism has only ever been detected in the white-blue desert annual *Linanthus panyae*. Interestingly, this colour dimorphism was initially thought to result from neutral evolutionary processes [67], but later it was determined that the polymorphism resulted from fluctuating selection exerted by temporal and spatial variation in spring precipitation [4,68]. Fluctuating selection due to interannual variation in water availability may also drive gynoecium colour dimorphism in *Butomus umbellatus* [69]. Possibly, fluctuating selection has only rarely been detected because it requires long-term datasets alongside understanding the abiotic adaptation in relate to colour (pleiotropy). We suggest that determining the relative importance of fluctuations in either abiotic or biotic selection is important for understanding the evolution of adaptive colour variation.
Frequency-Dependent Drivers

In most cases described earlier, the processes that maintain flower colour variation involve selection acting on the individual, independent of population composition. Some mechanisms, however, depend on the relative frequency of colour morphs, which can lead to frequency-dependent selection due to disassortative mating and adaptive foraging.

Disassortative Mating

Whether pollinator-mediated disassortative mating can drive colour polymorphism depends on the learning ability of the pollinators and whether that learning subsequently changes their behaviour [70–72]. Pollinator shifts among colour morphs is more likely if reward quality or quantity also varies between morphs and if the cost of shifting among morphs is small [72]. Moreover, if flower colour variation is associated with another advertisement trait (e.g., scent), pollinators will tend to be more consistent [73], which may promote positive frequency-dependent selection and reduce trait variation.

Negative frequency-dependent selection (NFDS) occurs when the fitness of one colour morph depends on the abundance of another morph, with the result that the less abundant morph has a per capita reproductive advantage leading to the maintenance of both morphs [74]. In theory, NFDS could arise in early-flowering deceptive plants that use naive pollinators, but as yet there is no evidence to support this hypothesis [75,76]. In fact, the only example of NFDS maintaining colour polymorphism that we know of is the rewardless orchid, Dactylorhiza sambucina [77], and that result could not be replicated, even in the same species [78,79].

Adaptive Foraging of Pollinators

The adaptive foraging behaviour among individual pollinators may also maintain colour polymorphism. Niche partitioning among individual pollinators is predicted to occur in communities with both high floral diversity and high resource competition [72,80]. Under such conditions, conspecific pollinators may adopt individual foraging strategies to specialise on different, equally rewarding colour morphs [81,82]. When spatiotemporal fluctuations in rewards hampers pollinators’ abilities to learn which flowers are the most beneficial, they are predicted to adopt a ‘majoring–minoring’ behaviour – that is, to devote a small fraction of each foraging bout to sampling of novel floral morphs [83]. Thus, in populations with high floral colour diversity, a polymorphism may be maintained if individual pollinators constantly visit different morphs (i.e., niche partitioning) while also ‘minoring’ frequently to track resource changes through time. While adaptive foraging behaviour could theoretically explain existing colour polymorphisms, this hypothesis remains untested.

Neutral or Random Processes

So far, all of the drivers and processes we have discussed assume that flower colour is adaptive or that polymorphism is the outcome of an adaptive process. Neutral or random processes can also lead to within-population flower colour variation. For example, genetic analyses of colour dimorphic Iris lutescens populations revealed that the relative frequency of yellow versus purple flowers within populations is affected either by drift or by gene flow, but not by ecological factors [84]. Such neutral processes may result from increases in genetic variation accompanied by a lack of selection. The lack of selection by pollinators in Sphaeralcea polychrome, for example, explains the maintenance of continuous colour variation because intrapopulation variation in flower colour is lower than the discrimination threshold of the pollinators [44].

Alternatively, molecular evolution of genes related to colour formation may involve neutral increases in trait variation driven by recurring mutations that produce new alleles at a faster rate than selection removes them, thus shifting the mutation–selection balance. The rate of shifts in
flower colour due to mutations has largely been documented between species [85,86], but some evidence suggests that mutation rates may also vary among populations [29]. While species-level colour modifications through mutations of either core pigment biosynthesis genes or transcription factors are relatively common and predictable [6,87–89] (Box 1), the extent of mutation-driven colour variation within populations is largely unknown but is expected to be different from that among populations [29,90]. Future studies should explore the molecular evolution of colour-related genes among colour morphs within populations to address this gap.

Neutral flower colour variation not only is hard to detect but also may be masked by other processes, some of which are not neutral. Migration or gene flow among populations in a colour-polymorphic species can counteract the removal of polymorphism by selection. Given that levels of colour polymorphism vary among populations [4,84], it is plausible that migration can balance directional selection and maintain within-population flower colour variation, but this hypothesis requires explicit testing. Notably, neutral accumulation of colour maladaptive morphs may be maintained despite strong directional selection. This is expected in long-lived perennial plants with overlapping generations, where polymorphic alleles are sustained in the population even if selection is strong [91]. However, as far as we know, this has never been documented at the population level.

Flower Colour Polymorphism – Open Questions

Is Flower Colour Polymorphism Adaptive or Neutral?

Flower colour variation is traditionally considered to be an adaptation to pollinator visual perception. As such, most studies exploring the evolutionary drivers of flower colour variation have focused on pollinator-mediated selection, sometimes with an additional selection agent (e.g., [18,42,49,50]). The role of random or neutral processes in maintaining colour variation has rarely been tested [92]. Within-population flower colour variation is relatively uncommon. If neutral processes are responsible for flower colour variation, the relatively small fraction of within-population variation may reflect correspondingly low rates of heritable changes in flower colour in most species. Moreover, even if such changes appear in the population, it may create a temporary polymorphism that will eventually be lost through genetic drift [93]. Alternatively, if selection governs colour polymorphism, its rarity may stem from the rarity of conditions that enable adaptive polymorphism. The wide range of selection-related mechanisms that could maintain flower colour variation, from multiple selection agents to pleiotropy, suggest that adaptive colour variation should be more abundant. However, neutrality or randomness is hard to test. It is usually considered the null hypothesis, whereas the adaptive explanation is the dominant hypothesis, which may have created a publication bias. The molecular drivers of flower colour shifts have been well studied at the species level and above (e.g., [6,89,94]), but little is known about the population-level dynamics. Advances in population genomics may allow us to test the extent to which mutation rates affect within-population phenotypic variation and thus may facilitate testing the neutrality hypothesis.

Are All Kinds of Colour Variation Similar?

Most of the studies we found in our review of the literature were of discrete colour variation, whereas only a few studied continuous colour variation (Table 1). Two of those were associated with selection exerted by abiotic factors [63,95], and the other two attributed continuous colour variation to a lack of pollinator-mediated selection [44,66]. Drivers of continuous flower colour variation may either correspond with variation in the expression of pigment-related genes or variation in the composition of pigments and copigmentation. Although our understanding of these mechanisms as drivers of continuous colour variation is limited, we can hypothesise that such variation requires either continuous selection regimes (e.g., pleiotropic adaptation along environmental gradients [63,85]) or a lack of selection [44,66]. As it stands, however, this hypothesis remains untested.
Why Is Colour Polymorphism So Rare? Or Is It?

Some of the mechanisms that drive flower colour variation seem more common or well documented than others. Flower colour is considered a pollination-relevant trait. However, a recent meta-analysis found that the strength of both pollinator- and nonpollinator-mediated selection on floral traits tends to be similar [36]. We found many examples where the abiotic environment plays an important role in maintaining colour variation (Table 1) [97]. Indeed, many examples came from environments with extreme or heterogeneous conditions (e.g., high elevation or Mediterranean climate), where it may be beneficial to maintain genetic variation, potentially resulting in comparatively higher incidences of polymorphic traits. If so, colour polymorphisms may be distributed unequally among ecosystems: rare in one and more common in the other. If this hypothesis is true, however, we would expect to find a positive correlation between trait-polymorphic populations and environmental heterogeneity, as well as corresponding adaptive processes maintaining such polymorphisms. Conversely, if there is no such correlation and thus no difference in rates of polymorphic versus monomorphic populations across environments, it may indicate that polymorphism arises by chance or at least that it arises as frequently through nonadaptive mechanisms as through adaptive mechanisms. In this case, we might also expect that polymorphisms should arise relatively frequently but not persist very long on an evolutionary time scale.

In order to test these hypotheses, we first need to determine the fraction of species that have within-population flower colour variation. To date, such data only exist for the British flora [31], the Mediterranean basin flora [30], for the orchid family [98], and for the genus *Iris* [5]. In the British flora, 17.4% of species are defined as polymorphic [31], but these numbers are probably inflated as the descriptions do not discriminate between within-population polymorphism and the occurrence of the occasional rare colour forms or variation between populations [31]. In the genus *Iris*, 26% of species are colour variable; however, colour variation is a derived trait in only a few clades of species [5] and so likely does not represent the rate of polymorphism across angiosperms. In the absence of a comprehensive global database of colour variation, the extent of within-population flower colour variation remains an open question.

**Concluding Remarks**

Within-population flower colour polymorphism is useful to understand fundamental evolutionary processes that create and maintain trait variation, and in particular it presents an opportunity to disentangle random processes from selection-driven processes leading to adaptive character combinations. The vast majority of research in this area has focused on balancing selection. Yet, theory suggests that many possible mechanisms could drive and maintain genetic polymorphism [32] (see Outstanding Questions). Studies testing the extent to which random or neutral processes influence trait variation are particularly scarce, and few attempts have been made to connect population genetic theory with these mechanisms [52]. We believe that introducing more theory to the study of flower colour polymorphism and exploring processes beyond balancing selection are necessary in order to further understand the evolutionary ecology of flower colour. Moreover, despite the attraction of colourful flowers to humans and pollinators, the importance of within-population flower colour variation as a model to understand the micro-evolutionary process has largely been overlooked. We suggest that understanding the global extent of the phenomenon, quantifying the selection regimes, and determining the underlying genetic basis of flower colour and its changes will provide insight necessary to develop the evolutionary theory regarding drivers of variation and its maintenance in nature.

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Declaration of Interests
No interests are declared.

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