

Adaptive divergence in seed color camouflage in contrasting soil environments

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Summary

- Although adaptive plant population divergence across contrasting soil conditions is often driven by abiotic soil factors, natural enemies may also contribute. Cryptic matching to the native soil color is a form of defensive camouflage that seeds can use to avoid detection by seed predators.
- The legume *Acmispon wrangelianus* occurs across a variety of gray–green serpentine soils and brown nonserpentine soils. Quantitative digital image analysis of seed and soil colors was used to test whether genetically based seed color is a closer match to the color of the native soil than to the color of other nearby soils.
- Lineages bear seeds that more closely match the color of their native serpentine or non-serpentine soil type than the opposing soil type. Further, even within a soil type, lineages bear seeds with a closer color match to the soil at their native site than to other sites.
- The striking concordance between seed and native soil color suggests that natural selection for locally camouflaged seed color morphs, probably driven by seed predators, may maintain adaptive divergence in pigmentation, despite the opportunity for migration between soil environments.

Introduction

‘When we see leaf-eating insects green, and bark-feeders mottled-grey; the alpine ptarmigan white in winter, the red-grouse the colour of heather, and the black-grouse that of peaty earth, we must believe that these tints are of service to these birds and insects in preserving them from danger.’

Darwin (1859) *The Origin of Species*

Heterogeneous selection as a result of variation in soil conditions can drive adaptive plant population divergence and result in ecotypes locally adapted to contrasting soil conditions despite the absence of physical barriers to gene flow (Linhart & Grant, 1996). Although abiotic soil factors are an important source of divergent selection (Kruckeberg, 1954; McNeilly, 1968; Brady *et al.*, 2005), little is understood about the contributions of natural enemies to patterns of divergent selection across soil conditions (Cremieux *et al.*, 2008). The effects of natural enemies can be spatially variable if the efficacy of prey defense is context dependent. Camouflage, the use of protective coloration to reduce the risk of detection by enemies (Stevens, 2007), is a highly context-dependent defense that depends upon the match of a prey’s appearance to the coloration of a given environment (Endler, 1978).

A locally cryptic distribution of heritable color camouflage is strongly suggestive of heterogeneous natural selection via visually cued predation. There is both empirical (Hargeby *et al.*, 2004; Sandoval & Nosil, 2005; Nosil & Crespi, 2006; Vignieri *et al.*, 2010) and theoretical (Nilsson & Ripa, 2010) support for predation as a selective agent driving intraspecific polymorphic color

camouflage. The degree of phenotypic divergence in color camouflage is related directly to the level of gene flow between habitats, with higher levels of gene flow requiring stronger selection to maintain color divergence (Mullen & Hoekstra, 2008; Nosil, 2009; Rosenblum & Harmon, 2011). Habitat-specific selection on color crypsis can be very strong, especially where driven by visually cued avian predators (Kettlewell, 1958; Clarke & Murray, 1962; Muggleton, 1978; Vignieri *et al.*, 2010). In plants, camouflage of conspicuous structures, such as bracts (Klooster *et al.*, 2009), trichomes (Weins, 1978; Lev-Yadun, 2006) and vegetative surfaces (Weins, 1978; Ellis *et al.*, 2006), may allow plants to match the color of background substrates to reduce detection by herbivorous natural enemies. However, few studies have examined the natural microevolutionary origins of population divergence in seed coloration as a form of camouflage, despite the prominent role played by variation in seed color in evolutionary and agricultural genetics (Mendel, 1866; Winkel-Shirley, 2001; Armstead *et al.*, 2007).

Seed predators are an important class of natural enemies of plants that can consume a large proportion of a plant’s

reproductive output (Janzen, 1971b). Post-dispersal seed predation can drastically reduce seed survival (Maron & Simms, 1997, 2001; Kauffman & Maron, 2006; Bricker *et al.*, 2010). Many plant communities are seed limited and so this predation can exert a strong influence on population demography (Turnbull *et al.*, 2000; Bricker *et al.*, 2010). For example, seed predators can have a stronger effect than catastrophic fire on post-dispersal seed survival and regeneration in forests (Zwolak *et al.*, 2010). A variety of seed defenses have evolved in response to selection as a result of seed predation, including toxins for chemical defenses (Janzen, 1969), protective tissues for mechanical defenses (Elliot, 1974), masting to satiate predators (Janzen, 1971a) and cryptic coloration for visual concealment (Cook *et al.*, 1971). Despite the ecological impacts of seed predators and evolutionary impacts on seed defense traits, we know surprisingly little about how variation in the selection imposed by seed predators may affect the local adaptation of seed defenses.

Visually cued seed predators, such as birds (Marone *et al.*, 2008), are important agents of selection on seed camouflage as they feed more rapidly and persistently upon less cryptic seeds (Jones *et al.*, 2006). For example, coastal populations of dove weed experience high seed predation by doves and are variable in coloration and well camouflaged, whereas desert populations, where doves do not occur, have monomorphic, less well-camouflaged seeds (Cook *et al.*, 1971; Cook, 1972). The seeds of some pines are polymorphic in color and seed colors may offer differential fitness in the presence of birds post-fire against a mosaic background of ash and exposed soil (Nystrand & Granstrom, 1997; Saracino *et al.*, 2004). Background color matching has therefore been demonstrated to affect seed fitness in nature, supporting the selective advantage of seed color crypsis in the presence of avian seed predators. However, locally adaptive population divergence for cryptic seed color camouflage has not been demonstrated in plants.

Large-seeded plants, such as legumes, are particularly vulnerable to post-dispersal seed predators (Maron & Crone, 2006), and may therefore exhibit strong evolutionary responses to seed predators. *Acmispon wrangelianus* is a native annual legume inhabiting open grassland environments throughout California. Seeds develop in leguminous pods, which dehisce explosively. Seeds disperse onto the soil surface within meters of the maternal plant (Lau *et al.*, 2008), which leaves them spatially clumped and exposed on the soil surface, putting them at high risk of predation (Brown, 1975; Mittelbach, 1984). *Acmispon wrangelianus* displays polymorphic seed color, with lineages bearing seeds of different colors, ranging from shades of gray to brown. *Acmispon wrangelianus* occurs on a variety of moderately disturbed or open soil environments, from gray-green, physiologically harsh serpentine soils to brown, more fertile nonserpentine soils. Outcrops of serpentine soils are commonly embedded in a matrix of various nonserpentine soils; however, neither soil condition is homogeneous in chemistry or color. The selective environment of this mosaic of soils is highly complex (Baythavong *et al.*, 2009). We know little about the capacity of selection to generate fine-scale genetic differentiation within soil types in these open patchy environments, where gene flow may counter the effects of selection, or how the strength of adaptive differentiation could differ

across conditions. Vegetation on the physiologically stressful serpentine soil is sparser than on nonserpentine soils at the McLaughlin Reserve (Harrison, 1999; Harrison *et al.*, 2003), and open habitats are often seed limited and subject to higher effects from seed predators (Maron & Simms, 1997; Maron & Crone, 2006; Maron & Kauffman, 2006; Denham, 2008), which could drive closer color camouflage on serpentine soils. Alternatively, if harsh serpentine soils support smaller populations, low serpentine effective population sizes and swamping gene flow from nonserpentine populations could result in weaker color camouflage on serpentine soils (Kawecki, 1995; Leimu & Fischer, 2008).

As a prerequisite to the investigation of locally adaptive *A. wrangelianus* seed color camouflage, I first verified that there was significant color variation among the seeds and soils from different collection sites. To determine whether the spatial distribution of color morphs across soil environments coincides with the pattern of adaptive divergence expected to result from selection by visually cued seed predators, I examined the following questions. (1) *Ecotypic color camouflage*. Do lineages bear seeds with a closer color match to their native serpentine or nonserpentine soil type (excluding the native site) than to the opposing soil type? Do serpentine and nonserpentine soil populations differ in the accuracy of this color match? (2) *Localized color camouflage*. Do lineages bear seeds with a closer color match to the soil at their native site than at other sites of the same serpentine or nonserpentine soil type? Do serpentine and nonserpentine soil populations differ in the accuracy of this color match?

Materials and Methods

Collections

Acmispon wrangelianus (Fisch. & C.A. Mey) D.D. Sokoloff (formerly *Lotus wrangelianus*) is a highly self-fertile small forb that grows in dense and distinct patches (Lau, 2006). It produces seeds of *c.* 2–3.5 mm in the largest diameter at maturity, weighing *c.* 2–6 mg each (Fig. 1). Most *Acmispon* lineages have hard seed coats and, as with most legumes, the testa prevents the seeds from imbibing water, which enforces dormancy (Rolston, 1978; Baskin *et al.*, 2000). After dispersal, *A. wrangelianus* seeds form a persistent seed bank within the soil (Levine & HilleRisLambers, 2009). Soil disturbance, oscillations in soil temperature or other factors that may scarify or crack the seed coat can cause the seed to break dormancy and initiate germination (Quinlivan, 1961; Jayasuriya *et al.*, 2009; Dalling *et al.*, 2011).

Maternal lineages and soil samples were collected at persistent *A. wrangelianus* populations from three serpentine soil and three nonserpentine soil sites at the University of California's Donald and Sylvia McLaughlin Natural Reserve, California, USA (Fig. 2; Supporting Information Table S1). *Acmispon wrangelianus* populations and soils at these sites have been monitored in a long-term ecological study for over a decade (Harrison, 1999; Harrison *et al.*, 2003; Porter & Rice, 2012). Mature seed pods were collected from plants selected haphazardly within a patch at a site, with an average of 18 maternal families collected per site. At three haphazardly selected locations within each site, six 200-g samples

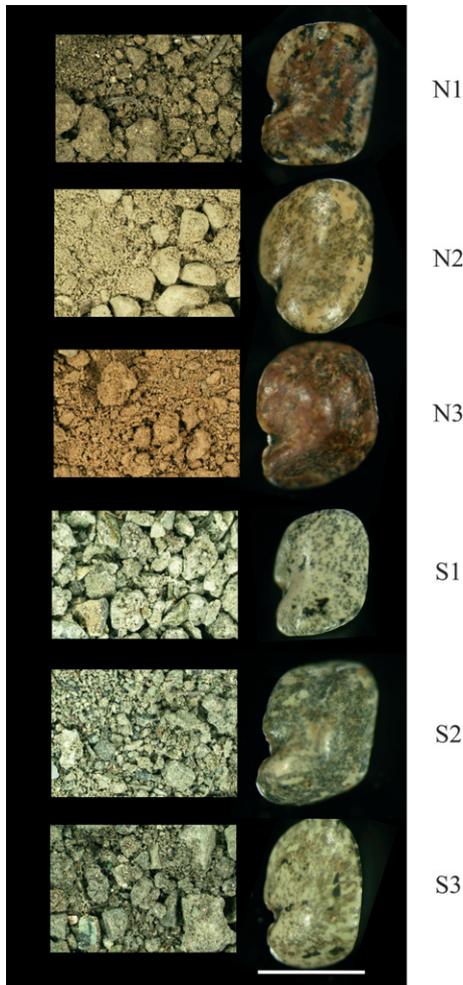


Fig. 1 Photographs showing soil and *Acmispon wrangelianus* seed color matches. Soil is a mix of six samples from a site; seed from each site was grown for a generation in potting soil in the glasshouse. Top to bottom: three nonserpentine soil sites (N1–N3) and three serpentine soil sites (S1–S3). Bar (for seeds), 2 mm.

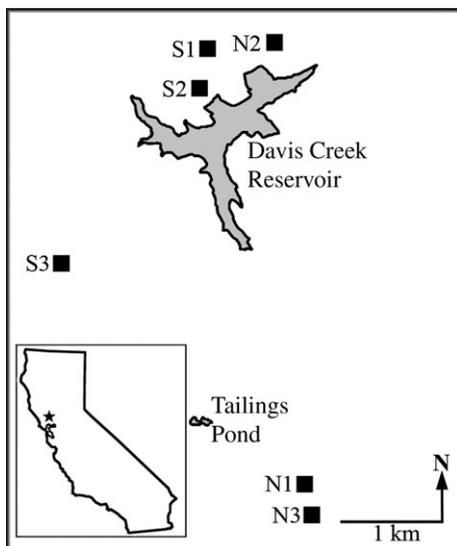


Fig. 2 Map of three nonserpentine soil (N1–N3) and three serpentine soil (S1–S3) collection sites for *Acmispon wrangelianus* at the McLaughlin Reserve (star on inset map of California, USA).

of soil sifted to 1 cm were collected. These samples were air dried and sifted to 2 mm for image analysis (Fig. 1).

To ensure that the observed variation in the color of seeds was caused by genetic, rather than environmental, effects, field-collected seeds were grown for one generation in potting soil in a glasshouse, and the seeds from these glasshouse plants were used to generate color measurements. One seed from each of the 109 lineages in total was grown in a randomized position in a glasshouse and allowed to self-fertilize. Seed color and pattern appeared to be highly consistent within a lineage. One mature seed from each common garden maternal plant was selected at random for image analysis.

Color measurements

The colors of seeds and soils were measured as reflectance values quantified from calibrated digital images captured via a dissecting microscope (Olympus, Model# SZX16; Olympus, Center Valley, PA, USA). Reflectance information from digital images using spectral standards is strongly correlated with values generated by spectrometry (Stevens, 2007). Measurements from digital images were utilized because of the ability to enlarge seed images through a microscope and to average colors within a seed image that often contained variable and complex patterns of color distribution caused by speckles and blotches (Fig. 1). To ensure a consistent light environment, images were captured in an otherwise dark room. Two fiber optic light boxes were set to illuminate each sample from four angles to reduce the effects of shadow on the images, and were run at the maximum setting for at least 1 h before photography. A consistent image exposure time and focal distance were set using color standards (X-Rite Mini ColorChecker; Grand Rapids, MI, USA), Munsell Color) for calibration, so that no pixel values in the image exceeded 245 out of a possible 255 in the red, green or blue (RGB) color channels. White point balancing was disabled on the imaging system. Six gray reflectance standards (X-Rite Mini ColorChecker, colors 19–24), which reflect a known amount of light equally at all wavelengths, were photographed during each imaging session to allow downstream adjustments for digital bias in color measurement. Seeds and soil samples were individually photographed with a size standard. All images were saved in an uncompressed Tagged Image File Format (TIFF).

Digital quantification of color was performed using ImageJ analysis software (Abramoff *et al.*, 2004). Seed images were separated from the neutral background and the average pixel values for the RGB color channels were recorded. A similar protocol was followed for soils and images of the gray color standards, except that, for these images, there was no background coloration to remove. The response of the camera's RGB channels to increasing radiance was measured as nonlinear and so a linearization correction was applied to raw RGB values calibrated to true values for the six gray color standards (*sensu* Stevens *et al.*, 2007). Linearized RGB values were then equalized with respect to the gray reflectance standards to produce broad-band values of reflectance in the long-wave (LW), medium-wave (MW) and short-wave (SW) parts of the spectrum (*sensu* Stevens *et al.*,

2007). Linearization and equalization were accomplished using standard equations (Westland & Ripamonti, 2004; Stevens *et al.*, 2007) to yield reflectance values in the three broad-band parts of the spectrum (LW, MW and SW) corresponding to the camera's sensors. This calibration of the seed and soil color data compensated for artifacts introduced by the microscope's imaging system and effects of the illuminants, to yield accurate reflectance values. The average discrepancies between calibrated reflectance values and true standardized reflectance values for a sample of 17 X-Rite Mini ColorChecker color standards ranged from <1% on the green channel to 4.3% on the blue channel.

To visualize the digital quantification of color, a canonical discriminant analysis (CDA) (PROC CANDISC, SAS Institute, 2008) was used to reduce the dimensionality of the reflectance values to allow a two-dimensional projection. CDA derives canonical variates (linear combinations of measurement variables) with the highest possible multiple correlation with defined groups to maximally separate the groups. Seed and soil samples were classified into 12 predefined groups constituting seed or soil samples from each of the six sites of origin, with the LW, MW and SW reflectance values from each sample as dependent variables. The first two canonical discriminants explained 97.6% of the color variance among samples, and these were plotted for all samples (109 seed samples and 36 soil samples). Canonical discriminant 1 loaded heavily on LW values and largely separated seeds and soils, whereas canonical discriminant 2 loaded heavily on SW values and largely separated seeds and soils on serpentine sites from those on nonserpentine sites (Fig. 3). CDA was used solely to visualize seed and soil reflectance values; the multivariate analyses of variance described below were used to test the

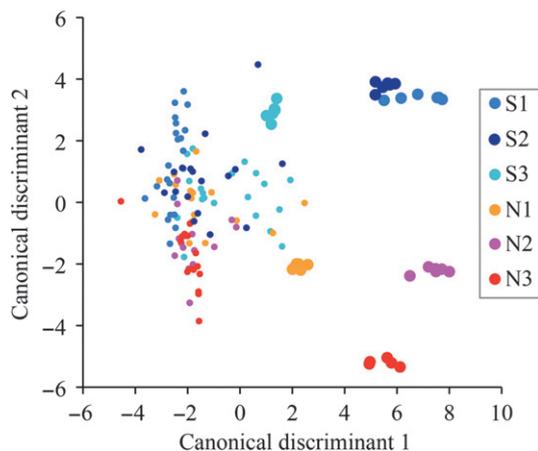


Fig. 3 Visualization of quantitative color data. A two-dimensional projection of long-wave (LW), medium-wave (MW) and short-wave (SW) reflectance values derived from digital images, plotted as canonical discriminants from a canonical discriminant analysis of *Acmispon wrangelianus* seeds (small circles) and soils (large circles) originating from six sites (color legend on right; serpentine soil sites S1–S3 and nonserpentine soil sites N1–N3). Canonical discriminant 1 loads heavily on LW values and canonical discriminant 2 loads heavily on SW values. Canonical discriminant 1 largely separates seeds and soils, whereas canonical discriminant 2 largely separates seeds and soils of serpentine sites from those of nonserpentine sites.

experimental hypotheses described in the Introduction because of the power of contrasts for addressing a priori hypotheses.

The predominant visual predators of *A. wrangelianus* seeds are currently unknown, although species occurrence records at the McLaughlin Reserve suggest a diversity of potential predators (Table S2). Potentially relevant seed-eating animals include numerous species of granivorous birds, as many birds use tetrachromatic vision in sophisticated color vision systems (reviewed in Bennett & Thery, 2007), granivorous insects, such as harvester ants, as ant species can exhibit di- or, possibly, trichromatic color vision and some are capable of color discrimination (Kretz, 1979; Cammaerts & Cammaerts, 2009; Camlitepe & Aksoy, 2010; Aksoy & Camlitepe, 2012), and granivorous rodents, as species of both rats and mice can be dichromats capable of color discrimination (Jacobs *et al.*, 2001, 2004). Because specific species of seed predators in this system have not yet been identified, and because of the diversity of visual systems in potential seed predators, color was quantified as reflectance spectra and not transformed to correspond to any particular receiver's specific visual system. Therefore, these reflectance spectra do not measure perceptual differences for particular clades of predators, which could differ in sensitivity to particular wavelengths (Endler & Mielke, 2005; Stevens, 2011). Despite this constraint, this study provides a crucial first step by using digital methods to quantify the match of seed reflectance spectra to those of the native soil substrate via methods that have become standard in investigations of coloration (Rudh *et al.*, 2007; Lindstedt *et al.*, 2010) and color matching of the substrate (Whiteley *et al.*, 2009; Clarke & Schluter, 2011).

Color is processed independently of brightness in many vertebrate and invertebrate visual systems (reviewed in Endler & Mielke, 2005). To account for variation in overall brightness among samples, each LW, MW and SW reflectance value was standardized into a proportion of the total reflectance for a sample (e.g. proportional value for LW = LW/(LW + MW + SW)). Reflectance values were then transformed into chromaticity coordinates: X_1 and X_2 values in a trichromatic (triangular) color space (Kelber *et al.*, 2003; Appendix A3: A3–A4). As the primary goal of the analysis was to test for color matching, these chromaticity coordinates (a measure of color only) were examined in downstream analyses. The brightness of each sample was examined as the mean reflectance over the measured spectrum $((LW + MW + SW)/3)$ (Lindstedt *et al.*, 2010). However, unlike color, trends in brightness did not show a signature of localized color matching of seeds and soils, and were not considered further.

Analysis

As a prerequisite to the investigation of locally adaptive color camouflage, I first verified that there was color variation among the seeds and soils from different sites and contrasting soil types. Multivariate analysis of variance (MANOVA in the General Linear Models (GLM) procedure; SAS Institute, 2008) was used to test whether seeds from different sites (nested in soil types) and serpentine or nonserpentine soil types differed in color, as measured by chromaticity coordinates. Within the MANOVA framework,

Pillai's Trace was used to generate an *F*-test. Multivariate normality of the X_1 and X_2 chromaticity coordinates was evaluated graphically by comparing the squared Mahalanobis distance between observations and the centroid to a chi-squared distribution in a quantile–quantile plot (Gnanadesikan, 1977). The distribution of seed, but not soil, X_1 and X_2 values is consistent with assumptions of multivariate normality (Fig. S1). This suggests that the *P* values for soil color may not be robust. However, examination of the CDA indicates that the deviation from multivariate normality is caused by a lack of overlap among measurements between sites, indicating strong soil color differences between sites.

A second MANOVA was used to test whether seeds more closely matched the color of their native soil type and native site than they did other soils, and whether serpentine and nonserpentine seeds displayed different degrees of matching. Here and below, the chromaticity coordinates from the six images of soil per site were averaged as a measure of soil color at a site. The pair-wise Euclidean distance (*D*) in color space between the chromaticity coordinates of each seed and that of each soil was calculated. Smaller seed–soil color distances in multivariate trichromatic color space therefore indicate a closer color match. One seed from each of 109 *A. wrangelianus* lineages was compared with six soil means, yielding six distance measurements for each seed, for a total of 654 color distance values. For each seed, these distances followed a convention, whereby D_1 denotes the comparison with soil from the native site, D_2 and D_3 denote comparisons with soil from non-native sites within the seed's native soil type (serpentine or nonserpentine), and D_4 , D_5 and D_6 denote comparisons with soil from sites of the opposite soil type. Each seed was considered an experimental unit (i.e. subject) and compared with six soils, and so these color distances were treated as six within-subject response variables in the MANOVA to account for the nonindependence of the six comparisons for each seed. This MANOVA model included two fixed, between-subject predictors: soil type, indicating whether a lineage was native to serpentine or nonserpentine soil, and site nested in soil type, indicating a seed's native site. As above, multivariate normality of the X_1 and X_2 values was confirmed graphically by comparing the squared Mahalanobis distance between observations and the centroid to a chi-squared distribution in a quantile–quantile plot (Gnanadesikan, 1977) (Fig. S1).

In this MANOVA, two a priori orthogonal contrasts were used to examine whether seeds were closer in color to their native soil

types and sites than to foreign ones. The first contrast was used to test for ecotypic color camouflage: that is, whether seeds were a closer color match to their native serpentine or nonserpentine soil type than to the opposing soil type. It determined whether the distance in color space between a seed and soil was smaller for the native soil type, excluding the native site, than for the foreign soil type (the mean of D_2 and D_3 vs the mean of D_4 , D_5 and D_6). The second contrast was used to test for localized color camouflage: that is, whether seeds were a closer color match to soil at their native site than at other sites of the same serpentine or nonserpentine soil type. It determined whether the distance in color space between a seed and soil was smaller for the native site than for other sites within the native soil type (D_1 vs the mean of D_2 and D_3). To determine whether serpentine or nonserpentine seeds or seeds from different sites differed in the strength of color matching to the native soil type or native site, the between-subject predictors (soil type and site) were examined for both contrasts. All MANOVA analyses were run on the data in both units of reflectance and as chromaticity coordinates, and the results of the statistical models were nearly indistinguishable.

Results

Acmispon wrangelianus lineages exhibited significant genetic variation in seed color. Lineages collected from different sites differed in seed color ($F_{8,206} = 9.03$, $P < 0.0001$) (Table 1), and lineages collected from serpentine soil and nonserpentine soil differed in seed color ($F_{2,102} = 39.21$, $P < 0.0001$) (Table 1). Similarly, soils collected from different sites appeared to differ in color ($F_{8,60} = 262.84$, $P < 0.0001$) (Table 1), and soils collected from serpentine and nonserpentine sites differed in color ($F_{2,29} = 9004.25$, $P < 0.0001$) (Table 1), although the *P* values associated with the tests for soils may not be robust as discussed above.

Ecotypic color camouflage

Acmispon wrangelianus displayed adaptive divergence in seed color camouflage between serpentine and nonserpentine soil populations. The distance in color space between a seed and a soil was smaller for sites from a seed's native soil type, excluding the native site, than for sites of the foreign soil type, as determined by an a priori contrast in a MANOVA ('Intercept'; Table 2).

Table 1 Multivariate analysis of variance (MANOVA) testing the effects of sources of variation on chromaticity coordinates for *Acmispon wrangelianus* seed and soil color

Factor	Fixed effects	Numerator df	Denominator df	Pillai's Trace	<i>F</i>	<i>P</i>
Seeds	Soil	2	102	0.4346	39.21	< 0.0001
	Site	8	206	0.5192	9.03	< 0.0001
Soils	Soil	2	29	0.9984	9004.25*	< 0.0001*
	Site	8	60	1.9445	262.84*	< 0.0001*

Sources of variation include whether lineages or soil samples were collected from serpentine or nonserpentine soils ('Soil') and the particular site of collection ('Site').

**F* and *P* values are approximate as a result of soil color deviation from multivariate normality.

Table 2 Multivariate analysis of variance (MANOVA) testing the effects of sources of variation on the Euclidean distance in color space between *Acmispon wrangelianus* seeds and the soils of the home soil type (excluding the home site) vs the away soil type

Fixed effects	Numerator df	Denominator df	Pillai's Trace	F	P
Intercept	1	103	0.3595	57.83	< 0.0001
Soil	1	103	0.0232	2.46	0.1201
Site	4	103	0.2372	8.01	< 0.0001

Sources of variation include the orthogonal contrast denoting the distance in color space between seeds and soils of either native or opposing soil type, excluding the home site (Intercept), and the interaction of this transformed variable with soil type (Soil) and site (Site).

Therefore, seeds exhibited ecotypic differentiation between soil types in color camouflage whereby seeds more accurately matched soils from sites of the same soil type than soils from sites of the opposing soil type (Fig. 4).

Seeds from different sites, but not different soil types, differed in the accuracy of this ecotypic seed color camouflage. The magnitude of the difference in color distance from a seed to soils from sites of the native vs opposing soil type varied among sites ('Site'; Table 2), indicating variation among populations for the degree of seed color camouflage to the native soil type (Fig. 5a).

However, the degree of ecotypic seed–soil color matching did not differ between populations native to serpentine or nonserpentine soils ('Soil'; Table 2).

Localized color camouflage

The distribution of *A. wrangelianus* seed color is consistent with an evolutionary response to very local selection on seed color camouflage. Seeds more closely matched the soil color of their native site than other sites from their native soil type. The distance in color space between a seed and a soil was smaller for soil from a seed's native site than for foreign sites within the native soil type ('Intercept'; Table 3). Therefore, seeds displayed locally cryptic color camouflage within a soil type ('Site Within Soil Type'; Fig. 4).

Seeds from different sites and different soil types differed in the accuracy of this local seed color camouflage to the native site. The magnitude of the difference in color distance from a seed to soils from sites of the native vs opposing soil type varied among sites ('Site'; Table 3), indicating variation among populations for the degree of seed color camouflage to the native site. In addition, the degree of matching to the soil at the native site differed between populations native to serpentine or nonserpentine soils. Serpentine populations exhibited a closer match to the soil at the native site than did those from nonserpentine soil ('Soil'; Table 3).

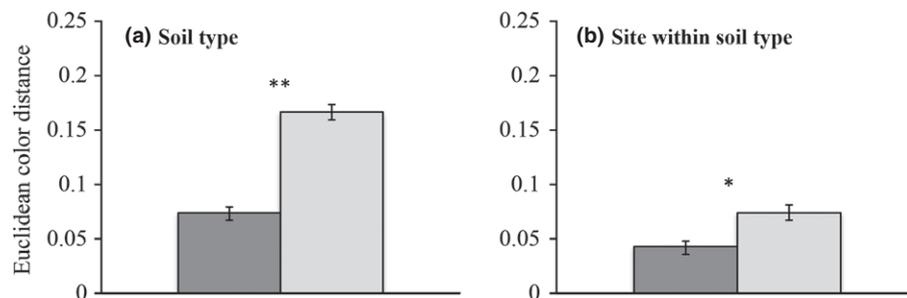


Fig. 4 Match of *Acmispon wrangelianus* seed color to native soil color. Euclidean distances between seed and soil chromaticity coordinates in trichromatic color space (\pm SE) in a multivariate analysis of variance to test whether seeds more closely match: (a) the native soil type, excluding the native site (dark gray), rather than the foreign soil type (light gray) ('Soil Type') and (b) the native site (dark gray) rather than other sites of the same soil type (light gray) ('Site Within Soil Type'). Significant differences are indicated as follows: *, $P < 0.05$; **, $P < 0.0001$.

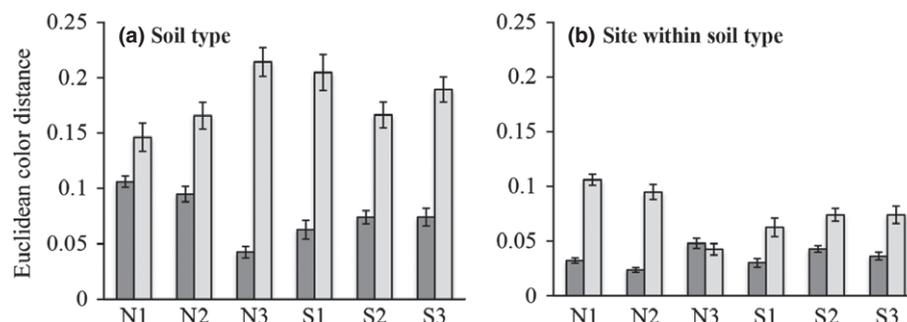


Fig. 5 Match of *Acmispon wrangelianus* seed color to native soil color by site. Euclidean distances between seed and soil chromaticity coordinates in trichromatic color space (\pm SE) in a multivariate analysis of variance to test whether seeds more closely match: (a) the native soil type, excluding the native site (dark gray), rather than the foreign soil type (light gray) ('Soil Type') and (b) the native site (dark gray) rather than other sites of the same soil type (light gray) ('Site Within Soil Type'), for lineages originating from the three non-serpentine soil sites (N1–N3) and three serpentine soil sites (S1–S3). The six sites differ significantly in the magnitude of the color difference in both (a) and (b) ($P < 0.001$).

Table 3 Multivariate analysis of variance (MANOVA) testing the effects of sources of variation on the Euclidean distance in color space between *Acemispom wrangelianus* seeds and soils of the home site vs other sites of the same soil type

Fixed effects	Numerator df	Denominator df	Pillai's Trace	F	P
Intercept	1	103	0.0525	5.71	0.0187
Soil	1	103	0.1231	14.46	0.0002
Site	4	103	0.7231	67.25	<0.0001

Sources of variation include the orthogonal contrast denoting the distance in color space between seeds and soils of either the native site or foreign sites within the native soil type, and the interaction of this transformed variable with soil type, and site nested in soil type.

Discussion

Acemispom wrangelianus exhibits intraspecific genetic divergence, whereby lineages bear seeds with a striking concordance to the native soil color. This suggests that natural selection for locally camouflaged color morphs maintains adaptive divergence in pigmentation despite the relatively short distances (*c.* 0.5–5 km) separating the sites in this study. The divergent distribution of color morphs across soil environments coincides with the pattern expected to result from selection caused by visually cued seed predators. Selection against mismatched color morphs may counter gene flow and hence promote further adaptive divergence, despite the opportunity for migration between soils (Nosil *et al.*, 2008).

Populations native to serpentine and nonserpentine soils exhibit soil type-specific differentiation in seed coloration. Overall, serpentine seeds more closely match soils from sites on serpentine than nonserpentine soil, and nonserpentine seeds more closely match soils from sites on nonserpentine than serpentine soil. The evolution of polymorphic crypsis can favor reproductive isolation and evolutionary divergence between populations originating in different habitat types. Assuming that seed predation would be stronger against seeds with a greater color mismatch, predation could impose stronger selection against migrants between populations of different soil types than migrants between populations of the same soil type. Selection against less cryptic immigrants can reduce gene flow between populations of contrasting habitat type, maintaining genetic isolation by adaptation (Bolnick & Nosil, 2007; Nosil *et al.*, 2008).

Although color matching occurs at the level of serpentine and nonserpentine soil types, it also occurs at the level of local, subtler differences in soil color within a soil type. Seeds more closely match soil color from the native site than the soil color of other sites of the same soil type. Seed–soil color matching thus appears to have evolved in response to localized shifts in soil color beyond the general distinctions between the gray serpentine or brown nonserpentine soil. Seed–soil color matching at this fine scale suggests that locally cryptic seed color camouflage may be a widespread phenomenon driven by mosaics of selection on subtle aspects of seed pigmentation. This site-specific matching suggests that locally cryptic seed coloration may not be restricted to the unusually strong contrast in soil colors across the boundaries of

serpentine soil outcroppings, and may be widespread in plants on a variety of soil chemistry environments.

Selection against mismatched color morphs inhabiting contrasting environments has often been found to be asymmetric (Hoekstra, 2004, Kettlewell, 1958), which could drive closer color matching in one environment than another. However, seeds from serpentine and nonserpentine soils do not appear to differ in the strength of ecotypic color camouflage. Habitat-associated differences in rates of predation may not result in differences in the strength or efficacy of selection on color camouflage if the relative fitness effects of predation are similar across habitats. For example, avian predators of rodents impose different rates of predation between habitats; however, in terms of relative fitness, it is equally disadvantageous for prey to be mismatched in either habitat (Vignieri *et al.*, 2010). In addition, the impact of potentially greater predation rates in the sparsely vegetated serpentine habitat could be counterbalanced by gene flow from larger populations on nonserpentine soil into smaller serpentine populations.

Overall, seeds tend to more closely match the soil of their native site as opposed to other sites of the same soil type. However, populations of seeds from some sites are more closely matched to the color of their native soil type and site than others and, overall, populations from serpentine sites are more closely matched to the native site within this soil type than are nonserpentine populations. Site-to-site variation in the strength of color camouflage could reflect mosaics in the efficacy of selection exerted by seed predators. Future work could disentangle how ecological factors, such as patch size, gene flow, distance to contrasting soil color and predation rates, correlate with the accuracy of a population's color match, to suggest which factors have strong effects on the efficacy of selection for localized color camouflage.

Dormancy in seed banks can dampen the ecological effects of seed predation by buffering plant populations from predator-driven fluctuations in recruitment (Maron & Crone, 2006). However, seed banks may preserve the strong localized evolutionary effects of seed predation by providing a cumulative memory of its effects. The finding of locally cryptic seed color camouflage provides evidence suggesting that post-dispersal seed predators are an important agent of selection affecting the lifetime fitness of plants (Turnbull *et al.*, 2000). Seed predation may be an especially effective agent of selection in annual seed-banked species, such as *A. wrangelianus*, because individuals often spend a much greater proportion of their lives as seeds than as vegetative plants. The cumulative risk of predation during the life history phase in the seed bank may be high, even if bouts of predation are infrequent. In addition, seed predation is a form of herbivory with maximal fitness costs to the dormant plant: if a seed is consumed, an individual is very likely to die outright, rather than experience some degree of reduction in fitness.

Seed coloration may be a relatively inexpensive form of defense that can respond quickly to selection. Seed color in legumes can be conferred by a handful of well-defined genes that regulate flavonoid and anthocyanin biosynthetic pathways in the maternal tissues during seed coat development (Vandenberg & Slinkard,

1990; McClean *et al.*, 2002). Relatively few genes of major effect underlie patterns of locally adaptive color camouflage polymorphisms in other locally cryptic organisms, such as some vertebrates (Hoekstra, 2006; Rosenblum & Harmon, 2011). The potential for both strong selection and a rapid response to selection may explain how this genetic differentiation can occur on such a fine scale, despite the potential for swamping caused by gene flow. Future work should investigate whether color crypsis has evolved in the presence of strong or weak levels of gene flow between patches, which could allow an estimation of the strength and target of selection that could establish the observed patterns (Gray & McKinnon, 2007).

Avian seed predation probably contributes to selection for local seed color camouflage, as birds have been shown to preferentially consume seeds most divergent from the color of the substrate under controlled and field conditions (Nystrand & Granstrom, 1997; Saracino *et al.*, 2004; Jones *et al.*, 2006) and usually have well-developed color vision (Endler & Mielke, 2005). However, other seed-eating animals present at the McLaughlin Reserve with the potential to perceive color, such as harvester ants or even rodents (Kretz, 1979; Jacobs *et al.*, 2001, 2004; Cammaerts & Cammaerts, 2009; Aksoy & Camlitepe, 2012), cannot be ruled out as potential sources of selection, especially given the high densities of some species at the study sites. The determination of the particular visually cued predators driving selection for color camouflage, the spatial and temporal patterns of seed consumption, and the selective advantage for locally matched seed color will be important steps towards the documentation of natural selection on seed color camouflage and the development of robust perceptual models of color discrimination (e.g. Spottiswoode & Stevens, 2012) for appropriate predators under ecologically relevant illumination (e.g. Chiao *et al.*, 2000). In the developing field of seed defense theory, a correlate of plant defense theory specific to seeds and their antagonists (Dalling *et al.*, 2011), seed camouflage may represent an important axis of seed defense.

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References

- Abramoff MD, Magelhaes PJ, Ram SJ. 2004. Image processing with ImageJ. *Biophotonics International* 11: 36–42.
- Aksoy V, Camlitepe Y. 2012. Behavioural analysis of chromatic and achromatic vision in the ant *Formica cunicularia* (Hymenoptera: Formicidae). *Vision Research* 67: 28–36.
- Armstead I, Donnison I, Aubry S, Harper J, Hörttensteiner S, James C, Mani J, Moffet M, Ougham H, Roberts L *et al.* 2007. Cross-species identification of Mendel's locus. *Science* 315: 73.
- Baskin JM, Baskin CC, Xiaojie L. 2000. Taxonomy, anatomy and evolution of physical dormancy in seeds. *Plant Species Biology* 15: 139–152.
- Baythavong BS, Stanton ML, Rice KJ. 2009. Understanding the consequences of seed dispersal in a heterogeneous environment. *Ecology* 90: 2118–2128.
- Bennett ATD, Thery M. 2007. Avian color vision and coloration: multidisciplinary evolutionary biology. *American Naturalist* 169: S1–S6.
- Bolnick DL, Nosil P. 2007. Natural selection in populations subject to a migration load. *Evolution* 61: 2229–2243.
- Brady KU, Kruckeberg AR, Bradshaw HD. 2005. Evolutionary ecology of plant adaptation to serpentine soils. *Annual Review of Ecology Evolution and Systematics* 36: 243–266.
- Bricker M, Pearson D, Maron J. 2010. Small-mammal seed predation limits the recruitment and abundance of two perennial grassland forbs. *Ecology* 91: 85–92.
- Brown JS. 1975. A preliminary study of seed predation in desert and montane habitats. *Ecology* 56: 987–992.
- Camlitepe Y, Aksoy V. 2010. First evidence of fine colour discrimination ability in ants (Hymenoptera, Formicidae). *Journal of Experimental Biology* 213: 72–77.
- Cammaerts MC, Cammaerts D. 2009. Light thresholds for colour vision in workers of the ant *Myrmica sabuleti* (Hymenoptera, Formicidae). *Belgian Journal of Zoology* 139: 40–49.
- Chiao CC, Osorio D, Vorobyev M, Cronin TW. 2000. Characterization of natural illuminants in forests and the use of digital video data to reconstruct illuminant spectra. *Journal of the Optical Society of America, A, Optics, Image Science, and Vision* 17: 1713–1721.
- Clarke B, Murray J. 1962. Changes of gene-frequency in *Cepaea nemoralis*: the estimation of selective values. *Heredity* 17: 467–476.
- Clarke JM, Schluter D. 2011. Colour plasticity and background matching in a threespine stickleback species pair. *Biological Journal of the Linnean Society* 102: 902–914.
- Cook AD. 1972. Polymorphic and continuous variation in seeds of dove weed, *Eremocarpus setigerus* (Hook) Benth. *American Midland Naturalist* 87: 366–376.
- Cook AD, Atsatt PR, Simon CA. 1971. Doves and dove weed—multiple defenses against avian predation. *BioScience* 21: 277–281.
- Cremieux L, Bischoff A, Smilauerova M, Lawson CS, Mortimer SR, Dolezal J, Lanta V, Edwards AR, Brook AJ, Tscheulin T, *et al.* 2008. Potential contribution of natural enemies to patterns of local adaptation in plants. *New Phytologist* 180: 524–533.
- Dalling JW, Davis AS, Schutte BJ, Arnold AE. 2011. Seed survival in soil: interacting effects of predation, dormancy and the soil microbial community. *Journal of Ecology* 99: 89–95.
- Darwin C. 1859. *On the origin of species by means of natural selection or the preservation of favoured races in the struggle for life*. London, UK: John Murray.
- Denham AJ. 2008. Seed predation limits post-fire recruitment in the waratah (*Telopea speciosissima*). *Plant Ecology* 199: 9–19.
- Elliot PF. 1974. Evolutionary responses of plants to seed eaters: pine squirrel predation on Lodgepole pine. *Evolution* 28: 221–231.
- Ellis AG, Weis AE, Gaut BS. 2006. Evolutionary radiation of “stone plants” in the genus *Argyrodema* (Aizoaceae): unraveling the effects of landscape, habitat, and flowering. *Evolution* 60: 39–55.
- Endler JA. 1978. A predator's view of animal color patterns. *Evolutionary Biology* 11: 319–364.
- Endler JA, Mielke PW. 2005. Comparing entire colour patterns as birds see them. *Biological Journal of the Linnean Society* 86: 405–431.
- Gnanadesikan R. 1977 *Methods for statistical data analysis of multivariate observations*. New York, NY, USA: John Wiley.
- Gray SM, McKinnon JS. 2007. Linking color polymorphism maintenance and speciation. *Trends in Ecology & Evolution* 22: 71–79.
- Hargeby A, Johansson J, Ahnesjo J. 2004. Habitat-specific pigmentation in a freshwater isopod: adaptive evolution over a small spatiotemporal scale. *Evolution* 58: 81–94.
- Harrison S. 1999. Native and alien species diversity at the local and regional scales in a grazed California grassland. *Oecologia* 121: 99–106.

- Harrison S, Inouye BD, Safford HD. 2003. Ecological heterogeneity in the effects of grazing and fire on grassland diversity. *Conservation Biology* 17: 837–845.
- Hoekstra HE. 2006. Genetics, development and evolution of adaptive pigmentation in vertebrates. *Heredity* 97: 222–234.
- Hoekstra HE, Drumm KE, Nachman MW. 2004. Ecological genetics of adaptive color polymorphism in pocket mice: geographic variation in selected and neutral genes. *Evolution* 58: 1329–1341.
- Jacobs GH, Fenwick JA, Williams GA. 2001. Cone-based vision of rats for ultraviolet and visible lights. *Journal of Experimental Biology* 204: 2439–2446.
- Jacobs GH, Williams GA, Fenwick JA. 2004. Influence of cone pigment coexpression on spectral sensitivity and color vision in the mouse. *Vision Research* 44: 1615–1622.
- Janzen DH. 1969. Seed-eaters versus seed size, number, toxicity and dispersal. *Evolution* 23: 1–27.
- Janzen DH. 1971a. Escape of *Cassia grandis* L. beans from predators in time and space. *Ecology* 52: 964–979.
- Janzen DH. 1971b. Seed predation by animals. *Annual Review of Ecology and Systematics* 2: 465–492.
- Jayasuriya K, Baskin JM, Baskin CC. 2009. Sensitivity cycling and its ecological role in seeds with physical dormancy. *Seed Science Research* 19: 3–13.
- Jones KA, Krebs JR, Whittingham MJ. 2006. Interaction between seed crypsis and habitat structure influences patch choice in a granivorous bird, the chaffinch *Fringilla coelebs*. *Journal of Avian Biology* 37: 413–418.
- Kauffman MJ, Maron JL. 2006. Consumers limit the abundance and dynamics of a perennial shrub with a seed bank. *American Naturalist* 168: 454–470.
- Kawecki TJ. 1995. Demography of source–sink populations and the evolution of ecological niches. *Evolutionary Ecology* 9: 38–44.
- Kelber A, Vorobyev M, Osorio D. 2003. Animal colour vision – behavioural tests and physiological concepts. *Biological Reviews* 78: 81–118.
- Kettlewell HBD. 1958. A survey of the frequencies of *Biston bechularia* (L.) (Lep.) and its melanic forms in Great Britain. *Heredity* 12: 51–72.
- Klooster MR, Clark DL, Culley TM. 2009. Cryptic bracts facilitate herbivore avoidance in the mycoheterotrophic plant *Monotropis odorata* (Ericaceae). *American Journal of Botany* 96: 2197–2205.
- Kretz R. 1979. Behavioral analysis of color vision in the ant *Cataglyphis bicolor* (Formicidae, Hymenoptera). *Journal of Comparative Physiology* 131: 217–233.
- Krueckeburg AR. 1954. The ecology of serpentine soils 3: plant species in relation to serpentine soils. *Ecology* 35: 267–274.
- Lau JA. 2006. Evolutionary responses of native plants to novel community members. *Evolution* 60: 56–63.
- Lau JA, McCall AC, Davies KF, McKay JK, Wright JW. 2008. Herbivores and edaphic factors constrain the realized niche of a native plant. *Ecology* 89: 754–762.
- Leimu R, Fischer M. 2008. A meta-analysis of local adaptation in plants. *PLoS ONE* 3: 1–8.
- Levine JM, HilleRisLambers J. 2009. The importance of niches for the maintenance of species diversity. *Nature* 461: 254–257.
- Lev-Yadun S. 2006. Defensive functions of white coloration in coastal and dune plants. *Israel Journal of Plant Sciences* 54: 317–325.
- Lindstedt C, Talsma JHR, Ihalainen E, Lindstrom L, Mappes J. 2010. Diet quality affects warning coloration indirectly: excretion costs in a generalist herbivore. *Evolution* 64: 68–78.
- Linhart YB, Grant MC. 1996. Evolutionary significance of local genetic differentiation in plants. *Annual Review of Ecology and Systematics* 27: 237–277.
- Maron JL, Crone E. 2006. Herbivory: effects on plant abundance, distribution and population growth. *Proceedings of the Royal Society B: Biological Sciences* 273: 2575–2584.
- Maron JL, Kauffman MJ. 2006. Habitat-specific impacts of multiple consumers on plant population dynamics. *Ecology* 87: 113–124.
- Maron JL, Simms EL. 1997. Effect of seed predation on seed bank size and seedling recruitment of bush lupine (*Lupinus arboreus*). *Oecologia* 111: 76–83.
- Maron JL, Simms EL. 2001. Rodent-limited establishment of bush lupine: field experiments on the cumulative effect of granivory. *Journal of Ecology* 89: 578–588.
- Marone L, de Casenave JL, Milesi FA, Cueto VR. 2008. Can seed-eating birds exert top-down effects on grasses of the Monte desert? *Oikos* 117: 611–619.
- McClellan PE, Lee RK, Otto C, Gepts P, Bassett MJ. 2002. Molecular and phenotypic mapping of genes controlling seed coat pattern and color in common bean (*Phaseolus vulgaris* L.). *Journal of Heredity* 93: 148–152.
- McNeilly T. 1968. Evolution in closely adjacent plant populations III: *Agrostis tenuis* on a small copper mine. *Heredity* 23: 99–108.
- Mendel G 1866. Versuche über Pflanzen-Hybriden. *Verhandlungen des Naturforschenden Vereines Brünn* 4: 3–47 (first English translation in 1901, J. R. Hortic. Soc. 26: 1–32; reprinted in *Experiments in Plant Hybridization*. Harvard University Press, Cambridge, MA, USA, 1967).
- Mittelbach GG. 1984. Experimental studies of seed predation in old-fields. *Oecologia* 65: 7–13.
- Muggleton J. 1978. Selection against melanic morphs of *Adalia bipunctata* (two-spot ladybird): a review and some new data. *Heredity* 40: 269–280.
- Mullen LM, Hoekstra HE. 2008. Natural selection along an environmental gradient: a classic cline in mouse pigmentation. *Evolution* 62: 1555–1569.
- Nilsson J, Ripa J. 2010. The origin of polymorphic crypsis in a heterogeneous environment. *Evolution* 64: 1386–1394.
- Nosil P. 2009. Adaptive population divergence in cryptic color pattern following a reduction in gene flow. *Evolution* 63: 1902–1912.
- Nosil P, Crespi BJ. 2006. Experimental evidence that predation promotes divergence in adaptive radiation. *Proceedings of the National Academy of Sciences, USA* 103: 9090–9095.
- Nosil P, Egan SP, Funk DJ. 2008. Heterogeneous genomic differentiation between walking-stick ecotypes: “Isolation by adaptation” and multiple roles for divergent selection. *Evolution* 62: 316–336.
- Nystrand O, Granstrom A. 1997. Post-dispersal predation on *Pinus sylvestris* seeds by *Fringilla* spp: ground substrate affects selection for seed color. *Oecologia* 110: 353–359.
- Porter SP, Rice KJ. 2012. Tradeoffs, spatial heterogeneity, and the maintenance of microbial diversity. *Evolution*. doi:10.1111/j.1558-5646.2012.01788.x.
- Quinlivan B. 1961. Effect of constant and fluctuating temperatures on permeability of hard seeds of some legume species. *Australian Journal of Agricultural Research* 12: 1009–1015.
- Rolston MP. 1978. Water impermeable seed dormancy. *Botanical Review* 44: 365–396.
- Rosenblum EB, Harmon LJ. 2011. “Same same but different”: replicated ecological speciation at white sands. *Evolution* 65: 946–960.
- Rudh A, Rogell B, Hoglund J. 2007. Non-gradual variation in colour morphs of the strawberry poison frog *Dendrobates pumilio*: genetic and geographical isolation suggest a role for selection in maintaining polymorphism. *Molecular Ecology* 16: 4284–4294.
- Sandoval CP, Nosil P. 2005. Counteracting selective regimes and host preference evolution in ecotypes of two species of walking-sticks. *Evolution* 59: 2405–2413.
- Saracino A, D’Alessandro CM, Borghetti M. 2004. Seed colour and post-fire bird predation in a Mediterranean pine forest. *Acta Oecologica-International Journal of Ecology* 26: 191–196.
- SAS Institute. 2008. *SAS software version 9.3*. Cary, NC, USA: SAS Institute.
- Spottiswoode CN, Stevens M. 2012. Host–parasite arms races and rapid changes in bird egg appearance. *American Naturalist* 179: 633–648.
- Stevens M. 2007. Predator perception and the interrelation between different forms of protective coloration. *Proceedings of the Royal Society B: Biological Sciences* 274: 1457–1464.
- Stevens M. 2011. Avian vision and egg colouration: concepts and measurements. *Avian Biology Research* 4: 168–184.
- Stevens M, Parraga CA, Cuthill IC, Partridge JC, Troscianko TS. 2007. Using digital photography to study animal coloration. *Biological Journal of the Linnean Society* 90: 211–237.
- Turnbull LA, Crawley MJ, Rees M. 2000. Are plant populations seed-limited? A review of seed sowing experiments. *Oikos* 88: 225–238.
- Vandenberg A, Slinkard AE. 1990. Genetics of seed coat color and pattern in lentil. *Journal of Heredity* 81: 484–488.
- Vignieri SN, Larson JG, Hoekstra HE. 2010. The selective advantage of crypsis in mice. *Evolution* 64: 2153–2158.
- Weins D. 1978. Mimicry in plants. *Evolutionary Biology* 11: 365–403.
- Westland S, Ripamonti C. 2004. *Computational colour science using MATLAB*. Chichester, UK: John Wiley & Sons Ltd.

- Whiteley AR, Gende SM, Gharrett AJ, Tallmon DA. 2009. Background matching and color-change plasticity in colonizing freshwater sculpin populations following rapid deglaciation. *Evolution* **63**: 1519–1529.
- Winkel-Shirley B. 2001. Flavonoid biosynthesis. A colorful model for genetics, biochemistry, cell biology, and biotechnology. *Plant Physiology* **126**: 485–493.
- Zwolak R, Pearson DE, Ortega YK, Crone EE. 2010. Fire and mice: seed predation moderates fire's influence on conifer recruitment. *Ecology* **91**: 1124–1131.

Supporting Information

Additional supporting information may be found in the online version of this article.

Fig. S1. Quantile–quantile comparison plots for seed colors, soil colors and Euclidean color distances.

Table S1 Coordinates and soil calcium : magnesium ratios for sites at the McLaughlin Reserve, California, USA

Table S2 Potential seed predator species at the McLaughlin Reserve, California, USA

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