

Article

Could diffuse coevolution explain the generic eggshell color of the brown-headed cowbird?

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Abstract

The brown-headed cowbird (hereafter cowbird) is an avian brood parasite that produces an egg dissimilar to those produced by the majority of its diverse host community. The cowbird's generic egg may result from a Jack-of-all-trades strategy; however, the evolutionary mechanisms that select for their generic eggs are unclear. Here we propose that the cowbird's eggshell phenotypes have evolved via diffuse coevolution, which results from community-level selective pressures, rather than via pairwise coevolution that occurs between a particular host species and its brood parasite. Under diffuse coevolution the cowbird's host community, with varying eggshell phenotypes and recognition abilities, would select for a cowbird eggshell phenotype intermediate to those of its host community. This selection is exerted by hosts that reject cowbird eggs, rather than those that accept them; therefore, we expect cowbird eggshell colors can be approximated by both the phenotypes and rejection abilities of their host community. Here we use eggshell reflectance data from 43 host species to demonstrate that the cowbird eggshell phenotypes are reasonably predicted (within 2 just noticeable differences) by the eggshell phenotypes and rejection rates of their hosts. These findings suggest that cowbird eggshell phenotypes, and potentially those of other some generalist parasites, may evolve via diffuse coevolution. Importantly, this research provides insight into the underlying evolutionary processes that explain observed phenotypic variation and provides a framework for studying selection on both specialist and generalist parasites' traits.

Key words: cowbird, diffuse coevolution, egg rejection.

When a host of an avian brood parasite rejects its parasite's egg, it selects for counter-adaptations in the parasite, fulfilling a crucial role in its coevolution. Such coevolutionary interactions are characterized by the hosts' egg recognition abilities, which select for the parasite's reciprocal adaptations. These well-known evolutionary selection pressures give rise to the refined eggshell mimicry found in some avian brood parasites. For example, the common cuckoo *Cuculus canorus* is an obligate brood parasite that has diverged into a range of host-specific races. Females from each cuckoo host-race specialize in particular host species (Brooke and Davies 1988),

imposing the costs of parental care on their hosts (Hauber and Montenegro 2002; Kilner et al. 2004). In turn, these costs select for antiparasitic defenses in the host species that can occur at any stage of the nesting cycle (Grim 2007; De Mársico et al. 2019). When that host species adapts egg recognition abilities, they inadvertently select for improved eggshell mimicry of their cuckoo host-race (Dawkins and Krebs 1979; Brooke and Davies 1988). These interactions can lead to an escalating coevolutionary arms race with parasites evolving increasingly accurate mimicry and hosts evolving increasingly fine-tuned rejection behavior (Stoddard et al. 2011). Under this

form of “pairwise coevolution” the hosts’ role in selecting for the eggshell mimicry of various host-races is relatively clear.

Unlike cuckoo host-races that specialize on particular host species, many brood parasites have no apparent individual specialization and exploit a range of species (Friedmann et al. 1977; Winfree 1999; Stevens 2013). Thus, these brood parasites are generalists at both the species- and individual-level, whereas the common cuckoo is a generalist at the species-level and a specialist at the individual level (i.e., individuals comprise specific host-races, whereas multiple host-races comprise the species). As such, these “true generalist” brood parasites may produce a generic egg phenotype, rather than evolving eggshell mimicry of a particular host (Peer et al. 2000; Klippenstine and Sealy 2010; Rutledge et al. 2021). Contrary to examples of pairwise coevolution where the role of host egg rejection in shaping the eggshell phenotypes of specific cuckoo host-races has been documented (Stoddard and Stevens 2011), the role of host recognition (if any) in selecting for the generic eggs of true generalists is less clear (Winfree 1999; Peer et al. 2005).

The “Jack-of-all-trades” hypothesis was proposed to explain such generic parasite phenotypes (Feeney et al. 2014). This hypothesis states that laying a generic egg, with passable similarity to a range of hosts’ eggs, is a generalist strategy benefitting parasites by enabling them to exploit a range of hosts (Feeney et al. 2014). Such a strategy could explain why Horsfield’s bronze-cuckoo *Chalcites basalis*, a true generalist, has eggshell colors that are intermediate to at least 17 of their 27 hosts (Stoddard and Prum 2008; Feeney et al. 2014). Meanwhile, in North America, the brown-headed cowbird (*Molothrus ater*, hereafter cowbird) is a true generalist with over 200 host species (Friedmann and Kiff 1985; Gibbs et al. 1997; Lowther 2020). Its egg differs in eggshell ground coloration, spotting patterns, and size compared with the majority of its hosts (Banks and Martin 2001). Nonetheless, numerous studies have suggested that cowbirds possess a generic eggshell phenotype similar to the eggs of some of their hosts (Rothstein 1975; Peer et al. 2000; Peer and Sealy 2004; Feeney et al. 2014; Rutledge et al. 2021). However, whether the cowbird possesses a generic, intermediate, eggshell phenotype has yet to be definitively addressed.

Previous research suggests that intermediate eggshell phenotypes are expected to evolve when sympatric hosts share similar eggshell phenotypes, and when parasites encounter hosts in different areas or at different times (Sherratt 2002; Feeney et al. 2014). These conditions are likely for the cowbird, which breeds from mid-April to mid-July (Lowther 2020), and exploits such a wide range of hosts that its eggs could be similar to those of its various hosts by sheer coincidence (Grim 2005). In this system, each host that rejects a cowbird egg would select for cowbird eggs to converge on the eggshell phenotype of that host. The direction and strength of that selection depend on each host’s eggshell phenotype and recognition abilities (Moksnes and Honza 1993; Moskát and Honza 2002; Strausberger and Rothstein 2009). This process comprises selective pressures (e.g., egg rejection rates) that are exerted by a unique host community (rather than a single-host species), is known as “diffuse coevolution” (Janzen 1980). Under this model of coevolution, the decisions of an entire community of rejecting host species could select for the cowbird’s eggshell phenotype, resulting in eggshell characteristics that are intermediate to those of its host community (Figure 1).

Here, we provide the first test of whether diffuse coevolution selects for the cowbird’s eggshell coloration. Specifically, we analyzed data on avian eggshell ground coloration of the cowbird and its hosts and test whether the cowbird’s eggshell color can be

predicted by the distinct eggshell features and recognition abilities of their host community, in particular those hosts that reject cowbird eggs. That is, hosts that have adapted strong recognition abilities may select for very different cowbird eggshell phenotypes (Figure 1). Such divergent selection pressures on the cowbird’s eggshell coloration would limit the cowbird’s ability to counter-adapt its eggshell features (Figure 1E), contrary to the specialization of cuckoo host-races on particular host species.

Materials and Methods

Color and visual models

We compiled a dataset of 43 North American cowbird host species with experimentally demonstrated rejection rates, for which we then calculated the perceived differences in eggshell color (chromatic contrasts) and brightness (achromatic contrasts) between their eggshell colors and those of the cowbird’s egg (Supplementary Table S1). These estimates were based on the spectral reflectance measurements of museum eggshells (see Rutledge et al. 2021 for details) and represent the average eggshell ground coloration for each species, but in some cases, they may also incorporate the brown coloration of fine eggshell speckling (Hanley et al. 2015; Rutledge et al. 2021). Such an “averaging” of the fine brown speckling might be expected when viewed by the hosts, which typically have lower visual acuity than humans (Caves et al. 2018). In this study, we quantified avian-perceived quantum catches for the 4 photoreceptors of the average ultraviolet-sensitive viewer (Endler and Mielke 2005; Maia et al. 2019), under forest light (following, Rutledge et al. 2021; Supplementary Table S2). We then used receptor noise limited models to compare the perceived coloration of each host egg to that of the cowbirds’ egg (Vorobyev and Osorio 1998). In addition, we quantified achromatic contrast using double cone estimates, generated via the same neural noise limited avian visual models (Vorobyev and Osorio 1998; Siddiqi et al. 2004). For these estimates we used double cone sensitivity data (obtained from, Maia et al. 2019) for the blue tit *Cyanistes caeruleus*. We suspected that 3 of our hosts (Supplementary Table S1) may have violet-sensitive vision (Ödeen et al. 2011); therefore, to consider the possibility that our choice of visual system impacted our results, we repeated these analyses assuming the average violet-sensitive avian viewer (obtained from Maia et al. 2019) and double cone sensitivity for the chicken *Gallus gallus*, but these produced nearly identical findings. These model parameters were the best choices available, and importantly, minor errors in the choice of photoreceptor sensitivity make little difference on the model outcome (Lind and Kelber 2009; Olsson et al. 2018). The contrasts generated by these models were measured in just noticeable differences (hereafter JNDs), such that increasing values relate to increasing degrees of perceivable differences between 2 stimuli.

Although intermediate colors (e.g., those selected via diffuse coevolution) can be determined using coordinates within the tetrahedral color space (Feeney et al. 2014), distances within this space are not perceptually uniform and omit information on perceived luminance differences (Endler and Mielke 2005; Stoddard and Prum 2008). Yet, considering host responses to both chromatic and achromatic contrast is important because hosts are known to respond to variation in eggshell color and eggshell brightness (Lahti 2006; de la Colina et al. 2012), and their response to color variation can interact with eggshell brightness (Hanley et al. 2019a). Thus, in some cases, both chromatic and achromatic contrasts are likely important. Therefore, we opted to create a phenotypic space where perceived

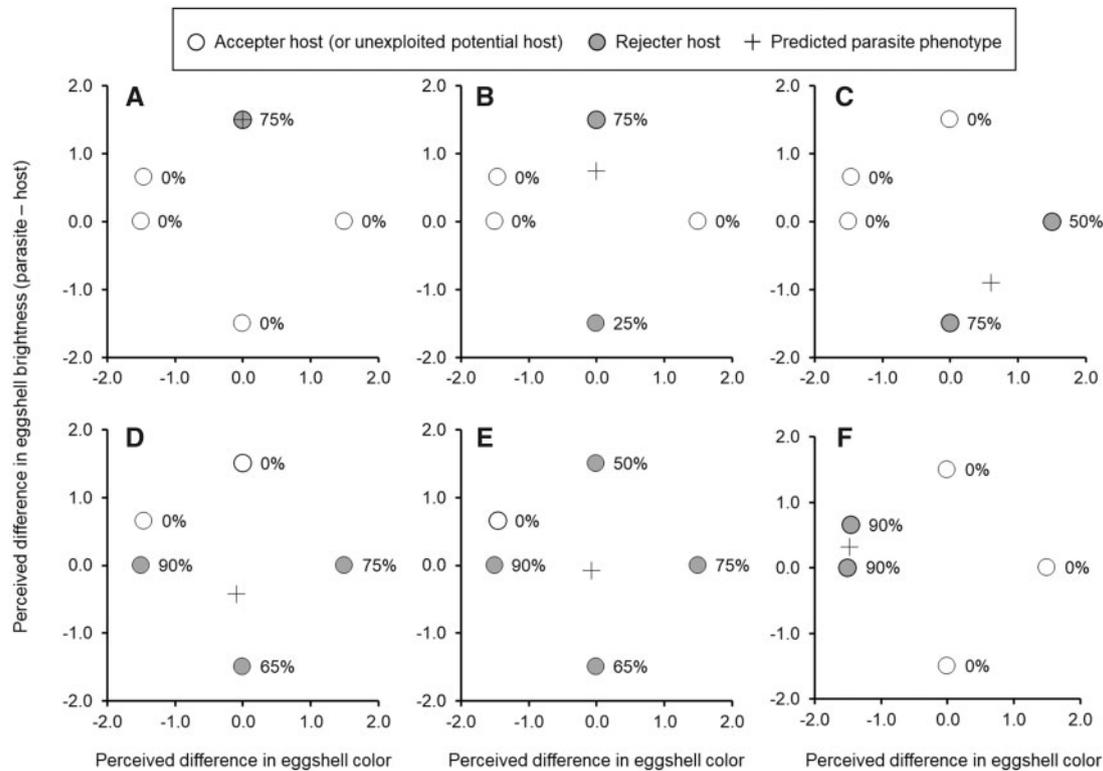


Figure 1. Hypothetical examples of phenotypic spaces illustrating the perceived differences between a parasite's eggshell and those of its hosts in terms of brightness (*y*-axis) and coloration (*x*-axis). Negative values on the *y*- and *x*-axes represent host eggs that are darker and browner than the parasite's, respectively (positive values on these axes represent brighter and bluer host eggs, respectively). The parasite's phenotype will evolve (+) in response to host selection for its appearance, which ultimately depends on the phenotypes of the rejecter hosts it exploits (dark circles) and the recognition abilities of those hosts (numbers to the right of each point refer to hypothetical rejection rates of each host). When the parasite exploits (A) a single-host species that rejects its egg (i.e., pairwise coevolution) at any rate (we illustrate 1 example, 75%), mimicry can evolve. In contrast, intermediate parasite phenotypes appearing generic with respect to those of their host community are expected under diffuse coevolution when the parasite exploits (B and C) 2, (D) 3, or (E) 4 host species that reject their eggs and have largely dissimilar eggshell phenotypes (i.e., spread out across the phenotypic space). Intermediate phenotypes are also expected in (F) parasites that typically specialize but are switching from one host to another, or periodically exploit a secondary host. Importantly, (A)–(F) illustrate distinct evolutionary scenarios that give rise to perfect (A) or imperfect (B–F) mimicry; they do not represent sequential steps of an arms race.

differences in color and luminance can be assessed (either jointly or separately). To do this, we compared the relative positions of each host's egg and cowbird's egg in the tetrahedral color space to determine whether the cowbird egg would be perceived as browner or bluer, before multiplying the chromatic contrast by -1 or 1 (Supplementary Figure S1), respectively (hereafter relative chromatic contrast). Similarly, we determined whether the perceived luminance of each host's egg was darker or lighter than the cowbird's egg and multiplied the achromatic contrast by -1 or 1 , respectively (hereafter relative achromatic contrast). This allowed us to generate a coordinate space, with the average cowbird eggshell phenotype set at the origin (Figure 2, hereafter phenotypic space), which summarizes virtually all chromatic and achromatic variation between eggshell phenotypes of hosts and parasites (Hanley et al. 2015) and that can be re-adjusted to center on other focal species (see below). This space allowed us to differentiate hosts that select for opposing cowbird eggshell phenotypes at identical rates (e.g., a host with an egg 3.5 JND browner than the cowbird and a host with an egg 3.5 JND bluer than the cowbird, both rejecting cowbird eggs at 50%, would select for an intermediate cowbird phenotype).

Egg rejection data

We obtained host-specific rejection rates for each host species based on experimentally determined rates of egg ejection and

abandonment/burying of cowbird eggs or models representing cowbird eggs (see sources in, Supplementary Table S1). When both forms of egg rejection rates were available, we added them to generate a total rejection rate for each species (hereafter rejection rate). We used these data to estimate each host's selection on the cowbird eggshell phenotype (Supplementary Table S1), rather than classifying hosts as either accepters or rejecters based on a threshold rejection rate, as is typical (Davies and Brooke 1989; Winfree 1999; Peer and Sealy 2004; Rutledge et al. 2021). Therefore, all hosts with rejection rates $>0\%$ rejected at a specific rate, while those with rejection rates equal to 0% accepted cowbird eggs. This resulted in 6 acceptor hosts and 37 rejecter hosts and allowed us to account for negligible to strong selection on cowbird phenotypes by the host community.

Calculation of intermediate phenotypes

If the cowbird's eggshell phenotype is being selected by diffuse coevolution, we expected that each rejecter host would select for cowbird eggs that are more similar to its own eggshell phenotype. Thus, if a cowbird population exploits 2 hosts that are equally skilled rejecters (i.e., reject at the same rate), the cowbird's eggshell phenotype should be roughly intermediate to the 2 hosts (e.g., Figure 1F). When only a single rejecter host is exploited, the parasite would be freed from these competing selection pressures, and host

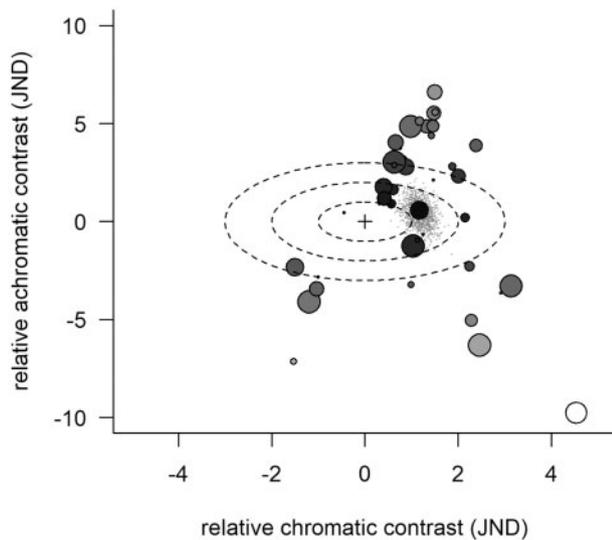


Figure 2. Host eggs (large spots) differ from cowbird eggs in both perceived chromatic variation (x -axis) and perceived achromatic variation (y -axis). Here we depict a phenotypic space, illustrating when host eggshell coloration would be perceived as bluer (positive) or browner (negative) than the coloration of the cowbird's egg on the x -axis and either brighter (positive) or darker (negative) than the cowbird's egg on the y -axis. This space is plotted in perceivable units (JNDs) relative to the cowbird's eggshell phenotype (centered at the origin, "+"). Spot size indicates the rejection rates of each host species, such that the largest spots represent the strongest responses. The concentric dotted lines represent 1, 2, and 3 JNDs, respectively. Each resampled host community predicted an intermediate cowbird eggshell phenotype (represented by a small grey dot, ".") within 2 JNDs of the cowbird's actual phenotype ("+"), forming a cloud of small gray dots to the right of the cowbird's own phenotype. Host communities were more likely to predict these intermediate phenotypes, and we illustrate the likelihood of these communities to predict any particular eggshell phenotype in our dataset (dark spots are more likely, whereas light spots are highly unlikely).

rejections could lead to eggshell mimicry (Figure 1A; Stoddard and Stevens 2011). Here we predicted the position of the cowbird's eggshell phenotype within the phenotypic space, considering the disparate eggshell phenotypes and selection pressures of its host community, by calculating the weighted centroid of a host community's phenotypic space:

$$\text{centroid } (x, y) = \begin{cases} x = \frac{\sum w_i x_i}{\sum w_i} \\ y = \frac{\sum w_i y_i}{\sum w_i} \end{cases}$$

where x_i and y_i are the relative chromatic contrast and achromatic contrast, the x and y coordinates of the phenotypic space, respectively, for the eggshell phenotypes of host i . The selection pressure (in this case rejection rate) of host i is represented by w_i .

We examined the differences within the phenotypic space using Euclidean distances:

$$d(x_n, y_n) = \sqrt{(x_0 - x_n)^2 + (y_0 - y_n)^2}$$

where the d is the measured Euclidean distance between the average cowbird phenotype (i.e., $x = 0$ and $y = 0$) and any point of interest n (e.g., an expected centroid) within the space. Such vector distances within this type of perceptual space have been used in the past in other contexts (Darst et al. 2006; Thorogood et al. 2017); however, in this study, we consider relative chromatic and achromatic

contrasts (e.g., bluer, browner, darker, or lighter than the parasite's egg) because this distinction is important for host egg recognition (Hanley et al. 2017, 2019b; Abolins-Abols et al. 2019; Manna et al. 2020). In this phenotypic space, a large Euclidean distance could correspond with a large chromatic or achromatic contrast (or both). While these distances are useful to quantify the differences between eggshell phenotypes and are technically in JNDs (the units on both axes), whether birds integrate ("mix") both chromatic and achromatic cues as "total-conspicuousness" (see Thorogood et al. 2017) and use that for decision-making is an area that requires further investigation (Kelber and Osorio 2010). Therefore, in this initial study, we tested both chromatic and achromatic contrasts alongside these Euclidean distances.

Statistical analyses

We used a resampling procedure to test whether the eggshell phenotypes of a cowbird's avian host community would provide a reasonable prediction of the cowbird's eggshell color (i.e., its position within the phenotypic space). This resampling procedure allowed us to simulate populations of varying host community composition (hereafter host communities), and therefore generate distributions of Euclidean distance and relative chromatic and achromatic contrasts, even with small sample sizes. To do this, we randomly sampled 80 species, with replacement, from our list of hosts. This sampling resulted in host communities with 36.53 ± 0.06 unique host species, which is slightly higher than comprehensive surveys of brown-headed cowbird host communities (Hahn and Hatfield 1995; Strausberger and Ashley 2005). This over-estimation was intended to account for the possibility that field studies have under-sampled the true host community, which is likely (Strausberger personal communication; Rothstein et al. 2002). These resampled host communities included both acceptor and rejecter hosts with a variety of egg phenotypes (i.e., eggs that varied in color, brightness, and spotting). For each host community, we calculated the expected coordinates of the cowbird's eggshell phenotype within the phenotypic space (i.e., the expected centroid), based on the eggshell phenotypes and rejection rates of each host in that community. We then calculated the Euclidean distance between the predicted centroid and the average observed cowbird eggshell phenotype (in this case, the origin), as well as the difference between the expected and observed eggshell phenotype along the x - and y -axes (the difference in relative chromatic and achromatic contrasts, respectively). These steps were repeated 1,000 times, each resample selecting a different host community.

To test the hypothesis that the cowbird's eggshell phenotype is an intermediate phenotype that can be estimated by its host communities, we used Wilcoxon signed-ranks test to determine whether cowbird eggshell phenotypes estimated by host communities were within 2 JNDs of the observed cowbird eggshell phenotype (the origin). Values on the order of 2 or 3 JNDs are often considered reasonable thresholds in natural settings (Spottiswoode and Stevens 2010, 2012; Stevens et al. 2013; Lind 2016), even though the theoretical discrimination threshold under ideal conditions is 1 JND. While the coordinates were estimated using relative chromatic and achromatic contrasts, we assessed how accurate this prediction was using absolute chromatic contrasts (i.e., estimates of -0.5 and 0.5 from the cowbirds' phenotype were both equally dissimilar). We report the percentage of host communities that predicted the cowbird eggshell phenotype within 2 JNDs, as well as the medians and interquartile ranges for the difference between the predicted and observed cowbird eggshell phenotype.

However, it is also possible that the host community could provide a reasonable estimate for any relatively intermediate eggshell phenotype. That is, the cowbird's phenotype matches this estimated intermediate phenotype due to pure coincidence. To address these possibilities, we tested whether the host community could predict *any* eggshell phenotype by repeating the steps above, each time centering the phenotypic space on a different host and then determining how well the other hosts' eggshell phenotypes and behaviors predicted that host's eggshell phenotype. The host community should, on average, more accurately predict the cowbird's eggshell phenotype than other eggshell phenotypes. This was done in the same manner as above, again selecting host communities 1,000 times and then measuring the distance between the origin (this time each host's eggshell phenotype) and the host community's prediction of each host's eggshell phenotype (Euclidean distance, and chromatic and achromatic contrast). We again used Wilcoxon signed rank tests to determine whether the host community more accurately predicted the cowbird's phenotype than other eggshell phenotypes in our dataset (a one-tailed test, i.e., the differences between other expected and observed phenotypes were greater than the difference between the expected and observed phenotype for the cowbird's eggshell). However, rather than comparing all 1,000 distance estimates for each species' phenotype, we compared each host's median estimated phenotype to the median estimated cowbird phenotype (i.e., $N = 43$).

Results

We found that the rejection rates and eggshell phenotypes of the cowbird's host community provided a reasonable approximation of the cowbird's eggshell phenotype. As predicted, the average cowbird phenotype was within 2 JNDs of the expected cowbird eggshell phenotypes (i.e., expected centroids), in terms of Euclidean distances (median = 1.35, interquartile range = 0.33, $P < 0.0001$), chromatic (median = 1.18, interquartile range = 0.28, $P < 0.0001$), and achromatic contrasts (median = 0.48, interquartile range = 0.61, $P < 0.0001$). The vast majority of expected phenotypes were within 2 JNDs of the average cowbird phenotype (98.5%, 100%, and 99.8% of cases for Euclidean distances, and relative chromatic and achromatic contrasts, respectively). These expected cowbird eggshell phenotypes were more accurate than the predictions for the majority of other eggshell phenotypes in the community in terms of total Euclidean distance (median = 3.62, interquartile range = 2.76, $P < 0.0001$) and relative achromatic contrast (median 3.1, interquartile range = 2.78, $P < 0.0001$); however, expected cowbird eggshell phenotypes were not more accurate in terms of chromatic contrast (median = 0.59, interquartile range = 0.95, $P = 0.99$), because the hosts in this dataset were generally bluer and, therefore, would select for bluer cowbird phenotypes (Figure 2).

Discussion

Here, we illustrate that the phenotypes and rejection behaviors of the brown-headed cowbird's host community provide a reasonable prediction of the cowbird's eggshell phenotype. Specifically, these findings show that the eggshell phenotypes and rejection behavior of rejecter hosts estimated cowbird eggshell phenotypes within 2 JNDs of the real cowbird phenotype. Thus, our findings suggest that diffuse coevolution could select for the eggshell phenotype of the generalist cowbird, and they highlight an evolutionary process by which a

"Jack-of-all-trades" strategy (Feeney et al. 2014) may arise as well as a predictive framework for studying this hypothesis.

Previous research found that the eggshell phenotype of the Horsfield's bronze-cuckoo *Chrysococcyx basalis* was intermediate to the eggs of its several host species (Feeney et al. 2014); however, few of these hosts exhibited strong rejection behavior (Langmore et al. 2005), and the role of host rejection response on the cuckoo's eggshell phenotype was not explored. Our study builds on this research by focusing on the underlying evolutionary process. By implicitly considering the eggshell phenotypes and rejection behaviors of the cowbird's host community, we demonstrate the role of host rejection behavior in shaping the cowbird's (a true generalist) eggshell phenotype. We found that by considering the eggshell phenotypes and rejection behaviors of host communities we could generally (>98% of the time) provide a good approximation of the cowbird's eggshell phenotype in terms of relative chromatic and achromatic contrasts, as well as Euclidean distances, within the phenotypic space.

Host eggshell phenotypes and rejection rates provided a better approximation of the cowbird's eggshell phenotype in terms of Euclidean distance and relative achromatic contrast than they would for other eggshell phenotypes in our dataset. However, hosts would also select for a bluer cowbird egg, undoubtedly because 86% of all hosts had bluer eggs than the cowbird's eggshell; however, we found similar results when we examined 84 host species with uncertain rejection rates (unpublished data). While the large proportion of hosts measured as bluer than the cowbird may be a by-product of sampling effort, it could suggest that there is selection pressure for hosts to lay eggs that are bluer than those of their parasite (Hanley et al. 2013), which could occur if hosts accept eggs bluer than their own while rejecting browner eggs, as has been shown in some hosts (Soler et al. 2012; Hanley et al. 2017, 2019b; Abolins-Abols et al. 2019; Manna et al. 2020). Additionally, there are possible selection pressures on cowbird eggshell phenotypes (i.e., preventing it from evolving bluer) that are independent of their hosts' selection that we have not yet taken into consideration. Nonetheless, although our relatively simple model of diffuse coevolution predicted a cowbird egg bluer than it really is, it still produced surprisingly accurate predictions of the cowbird's eggshell characteristics (within 2 JND; Figure 2).

Our findings show that diffuse coevolution could explain the imperfect mimicry of the cowbird eggshell, but other explanations cannot be ruled out. For example, cowbirds may selectively exploit accepter hosts with similar eggshell phenotypes and only parasitize rejecter hosts when preferred hosts are unavailable. It is reasonable to assume cowbirds will sometimes resort to sub-optimal hosts, as we know that host community composition may vary with geography and those female cowbirds tend to avoid laying eggs in nests that are already parasitized (White et al. 2007). Thus, when host choices are limited and/or competition for host nests is particularly intense, cowbird females may be limited to host nests that lack cowbird eggs, regardless of the host's suitability (Rivers et al. 2012). Furthermore, if cowbirds select their host communities, they may only parasitize that subset of rejecter hosts to which their generic egg has a passing resemblance (although we are unaware of any examples of this), or instead parasitize rejecter hosts with distinct eggshell features early in the laying sequence, causing misimprinting (Strausberger and Rothstein 2009). Although genetic evidence suggests that cowbirds do not generally specialize (Gibbs et al. 1997), there is evidence to suggest that there is at least some degree of host specificity (Woolfenden et al. 2003, 2004; Strausberger and

Ashley 2005) and geographic variation in host use (Hahn and Hatfield 1995; Woolfenden et al. 2004). Such host choice on the part of the cowbird could also result in a cowbird eggshell phenotype intermediate to its host community.

One important aspect of our proposed model is it can be empirically derived within cowbird populations (and their host communities) and is therefore readily adaptable to future investigations (e.g., incorporation of additional data on host rejection rates, host community composition, host intraspecific eggshell variation, etc.). Given our results, we suggest researchers explore the potential for diffuse coevolution to predict cowbird eggshell phenotypes in the field. Such future tests will be important as our models were built using species-specific information; however, host communities can vary considerably in their rejection rates (Moksnes and Honza 1993; Haas and Haas 1998; Moskát and Honza 2002; Strausberger and Rothstein 2009), and parasite populations can differ in their host preference between and within study sites (Hahn and Hatfield 1995; Woolfenden et al. 2004). These are likely important factors in determining the selective pressures acting on the cowbird's eggshell phenotype that we have not yet considered. Likewise, because host lists continue to grow and change as we learn more about brood parasites, it is probable that there are currently unknown hosts and hosts that the cowbird has yet to exploit (Rothstein et al. 2002) that will also affect the cowbird's eggshell phenotype. Therefore, future research can make continually more accurate predictions by updating the model's scaling factor (w_i , see Materials and Methods section) via field-collected data. For example, the model would be strengthened by incorporating the relative recognition abilities (Soler et al. 1999), suitability (Stokke et al. 2018), parasitism rates (Rivers et al. 2010), and abundances (Purcell and Verner 1999) of host species, once those data are available for the host community of a particular cowbird population. Incorporating these data may improve the accuracy of predicted cowbird egg color if hosts with relatively high rejection rates and comparatively dark blue-green eggs are infrequently used (see lower right corner of Figure 2). We encourage experiments where scaling factors are derived from fieldwork on distinct parasite populations and host communities. Such experiments could predict subtle differences in cowbird eggshell phenotypes at different localities, even if their host communities are the same (as their phenotypes, rejection rates, and parasitism rates may not be identical). Although collecting such data would be ambitious, we contend that the parasite's eggshell phenotype would simply be predicted by the product of the unique combination of eggshell characteristics and selection pressures of their host community (recognition abilities, suitability, parasitism rates, and abundances).

The processes resulting in the eggshell phenotypes of some true generalist parasites such as the cowbird and the refined mimicry of some specialists such as the cuckoo host-races are often considered entirely distinct (Rothstein 1990; Lanyon 1992; Winfree 1999; Stevens 2013). However, host selection of parasitic eggs is crucial for both diffuse and pairwise coevolution. Here we illustrate that a simple model can predict perfect mimicry when host communities consist of a single species or imperfect mimicry when host communities consist of multiple host species (Figure 1). For example, under this model, it is reasonable to expect an intermediate eggshell phenotype in a cuckoo host-race that is in the process of switching to a new host (Gibbs et al. 2000). Host-races typically parasitize a single-host species, but they occasionally exploit an additional sympatric local potential host species, which can result in 2 hosts influencing the eggshell phenotype of the parasite (Moksnes and Røskoft 1995;

Marchetti et al. 1998; Vogl et al. 2002; Koleček et al. 2021). Such a scenario was documented in a study investigating a population of great reed warbler *Acrocephalus arundinaceus* and reed warblers *Acrocephalus scirpaceus* parasitized by the common cuckoo (Drobnik et al. 2014). They found that although each hosts' eggshell coloration was distinct, the eggshell color of the cuckoo host-races were indistinguishable and intermediate to both hosts (see Figure 1F).

Unique host communities can select for the eggshell phenotypes of brood parasites through pairwise or diffuse coevolution (Janzen 1980; Brooke and Davies 1988; Ebert and Fields 2020). Our study suggests that diffuse coevolution may be the underlying mechanism generating the cowbird's intermediate eggshell appearance, and it provides a conceptual framework for studying the role of host selection on parasite eggshell phenotypes. More generally, this framework may help explain the evolution of imperfect mimicry via multiple models (Sherratt 2002; Kikuchi and Pfennig 2013). While promising, future research should define the selective pressures exerted by distinct host communities on the eggshell phenotypes of cowbirds parasitizing those communities. Overall, we call for others to consider diffuse coevolution as a promising mechanism that could provide further insight into the evolutionary processes that give rise to the eggshell phenotypes of brood parasites.

Supplementary Material

Supplementary material can be found at <https://academic.oup.com/cz>.

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Authors' Contributions

Initial concept (D.H.), initial draft (D.H. and B.M.J.), data collection (D.H. and S.L.R.), analyses (D.H.), all authors contributed to the manuscript prior to submission.

Conflict of interest

The authors declare no competing interests.

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