In summary, the follicular epithelium represents a beautiful example of how the combination of diverse signalling pathways creates cell diversity by using morphogen gradients. These gradients initiate spatially restricted gene expression domains that become stabilized by negative cross-regulation of their downstream targets. In the follicle cells, JAK/STAT signalling outcome differs depending on whether it interacts with EGFR signalling or not. Similarly, the result of EGFR pathway activation is conditioned by its interaction with either the Dpp/BMP or the JAK/STAT pathways. Finding out how the temporal- and spatial-specific information provided by these signalling pathways converge on their downstream targets and particularly how their activating and repressing inputs are integrated on the target gene enhancers is a fundamental area of research [13].

REFERENCES

1. Fregoso Lomas, M., De Vito, S., Boisclair Lachance, J.-F., Houde, J., and Nilson, L.A. (2016). Determination of EGFR signaling output by opposing gradients of BMP and JAK/STAT activity. Curr. Biol. *26*, 2572–2582.
2. Gonzalez-Reyes, A., Elliott, H., and St Johnston, D. (1995). Polarization of both major body axes in Drosophila by gurken-torpedo signalling. Nature *375*, 654–658.
3. Silver, D.L., and Montell, D.J. (2001). Paracrine signaling through the JAK/STAT pathway activates invasive behavior of ovarian epithelial cells in Drosophila. Cell *107*, 831–841.
4. Schupbach, T. (1987). Germ line and soma cooperate during oogenesis to establish the dorsoventral pattern of egg shell and embryo in Drosophila melanogaster. Cell *49*, 699–707.
5. McGregor, J.R., Xi, R., and Harrison, D.A. (2002). JAK signaling is somatically required for follicle cell differentiation in Drosophila. Development *129*, 705–717.
6. Manning, L.A., Weideman, A.M., Peercy, B.E., and Starz-Gaiano, M. (2015). Tissue landscape alters adjacent cell fates during Drosophila egg development. Nat. Comm. *6*, 7356.
7. Arbouzova, N.I., and Zeidler, M.P. (2006). JAK/STAT signalling in Drosophila: insights into conserved regulatory and cellular functions. Development *133*, 2605–2616.
8. Xi, R., McGregor, J.R., and Harrison, D.A. (2003). A gradient of JAK pathway activity patterns the anterior-posterior axis of the follicular epithelium. Dev. Cell *4*, 167–177.
9. Fregoso Lomas, M., Hails, F., Lachance, J.F., and Nilson, L.A. (2013). Response to the dorsal anterior gradient of EGFR signaling in Drosophila oogenesis is prepatterned by earlier posterior EGFR activation. Cell Rep. *4*, 791–802.
10. Zhao, T., Graham, O.S., Raposo, A., and St Johnston, D. (2012). Growing microtubules push the oocyte nucleus to polarize the Drosophila dorsal-ventral axis. Science *336*, 999–1003.
11. Starz-Gaiano, M., Melani, M., Meinhardt, H., and Montell, D. (2009). Interpretation of the UPD/JAK/STAT morphogen gradient in Drosophila follicle cells. Cell Cycle *8*, 2917–2925.
12. Starz-Gaiano, M., Melani, M., Wang, X., Meinhardt, H., and Montell, D.J. (2008). Feedback inhibition of Jak/STAT signaling by apontic is required to limit an invasive cell population. Dev. Cell *14*, 726–738.
13. Pinto, P.B., Espinosa-Vazquez, J.M., Rivas, M.L., and Hombria, J.C. (2015). JAK/STAT and Hox Dynamic Interactions in an Organogenetic Gene Cascade. PLoS Genet. *11*, e1005412.

Evolutionary Genetics: Small Genomic Regions Make a Big Impact

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http://dx.doi.org/10.1016/j.cub.2016.09.002

Biologists have long sought the genes that contribute to phenotypic and population divergence. Two new studies identify genomic regions involved in plumage coloration and migratory orientation.

Identifying the genes underlying ecologically and behaviorally important phenotypes in natural populations has long been a holy grail for evolutionary biologists. Despite a few early success stories, many involving genes in the melanocortin pathway that affect animal coloration [1], finding such genes has remained difficult [2]. However, the genomic revolution has led to the discovery of genes, or genomic regions, that underlie phenotypic variation.

A particularly promising approach for identifying these genomic regions is the study of hybrid zones between species where recombination facilitates genetic mapping of phenotypes [3]. Two papers published recently in *Current Biology* by Toews, Taylor *et al.* [4] and Delmore *et al.* [5] investigate hybrid zones between bird species and add to an emerging picture of the genomic architecture of hybridization and divergence.

Although the systems are different, both studies converge on similar results: a small number of genomic regions underlie the phenotypic traits of interest.

Toews *et al.* [4] studied a well-known but long- mysterious hybrid zone between phenotypically divergent blue- and golden-winged warblers in the Great Lakes region of North America (Figure 1A,B). Blue- and golden-winged warblers differ so extensively in plumage

Figure 1. Two pairs of hybridizing bird species.
Top left: Golden winged warbler (photo: Tom Johnson/Macaulay Library); top right: blue-winged warbler (photo: Robert Dorman/Macaulay Library); bottom left: Swainson's thrush; bottom right: thrush with light-level geolocator (thrush images: Kira Delmore).

traits that hybrids between the two species were once considered separate species referred to as Brewster's and Lawrence's warblers). The evolutionary history and causes of hybridization between these species has thus been of great interest for over a century [4], and this study brings modern genomic methods to bear on the issue. The surprising verdict is that the genomes of these taxa are largely homogeneous, with some of the lowest mean genome-wide divergence values reported in hybridizing birds. Demographic models show a continuous history of hybridization over several millennia, putting to rest the hypothesis that hybridization is a negative consequence of recent human intrusion. The persistence of unique phenotypic forms despite thousands of years of hybridization raises interesting new questions about conservation strategies for this threatened group.

Despite genome-wide homogeneity between warbler species, the authors were able to identify six small genomic clusters with fixed or nearly-fixed differences. They found a strong correlation between allele frequency differences in divergent genomic regions and divergence in plumage patterning, suggesting that the divergent regions contain genes involved in color differences between species. Many studies in wild populations have focused on coloration and have found genes of large effect underlying relatively discrete color patterns [6,7]. For example, a recent study of hybridizing hooded and carrion crows identified a single small genomic region responsible for differences in plumage color [8]. Toews *et al.* [4] also found relatively simple patterns of inheritance of multiple plumage patches. For example, the black throat patch present in golden-winged warblers appears to be a simple Mendelian recessive trait: only homozygous individuals express black patches. Intriguingly, some of the plumage patches are associated with separate genomic regions and vary independently of each other, whereas others appear to be linked. Complex plumage patterns are common across birds, yet little is known about their genetic basis. This study suggests that different patches can evolve and vary independently of each other, an exciting prospect for future work on phenotype evolution.

Delmore *et al.* [5] focus on another well-studied avian hybrid zone, that between two subspecies of Swainson's thrushes that hybridize along the Coast mountains of Western North America (Figure 1C,D). The hybrid zone corresponds to a migratory divide, where western birds migrate south to Mexico and Central America, and eastern birds migrate southeast to South America [9]. Unlike the warblers, this hybrid zone is comprised of two fairly well-differentiated subspecies that differ in plumage color as well as migratory route [5]. The authors addressed similar questions about the genomic architecture of hybridization as in the warblers but in a different evolutionary context and for traits that initially seem more complex and likely to involve more loci: migratory orientation and quantitative color differences.

Despite the complexity of the focal traits, Delmore *et al.* [5] use several lines of evidence to find both a genetic basis for, and divergent selection associated with, migratory route. First, the authors integrated genetic data with measurements of migratory behavior to show a correlation between ancestry and migratory direction. They then used genetic mapping to demonstrate strong associations between a cluster of SNPs on chromosome 4 and migratory route determined from geolocator data. In contrast to the genetic structure of plumage color in the warblers, migratory route in the thrushes has an additive genetic basis, meaning that heterozygous individuals exhibit intermediate migratory

routes. Moreover, the authors find selected regions of the genome that are not only associated with migratory orientation but also exhibit fixation of different alleles between subspecies. The combination of additive inheritance of migratory route and selection for alternative migratory alleles across a hybrid zone together provide important mechanistic support for a longstanding hypothesis: that hybrid individuals may inherit intermediate and maladaptive migratory routes. For example, additive genetic inheritance of migratory route in Swainson's thrushes means that first-generation hybrids would likely migrate directly over, rather than around, the Coast mountains, where the climate is much harsher and there are fewer resting points. Selection against such maladaptive migratory patterns should make migratory divides important to maintenance of species boundaries [10]. It is quite extraordinary that a behavioral trait as seemingly complex as migration has a strong genetic basis that can result in divergent selection between sympatric forms.

Both studies used an unbiased, anonymous sequencing approach to uncover intriguing candidate genes that are consistent with other studies of pigmentation [7] and migratory genetics [11]. Toews *et al.* [4] identified five candidate genes previously shown to be involved in feather development and coloration that are directly downstream of the only divergent regions found between the two warbler genomes. These genes include *ASIP*, which is involved in melanic coloration in mammals [12] and birds [13] and is perfectly associated with the black throat patch in warblers, and *BCO2*, which is strongly correlated with the extent of yellow plumage color and was found to be involved in yellow carotenoid metabolism. Delmore *et al.* [5] found a region containing *TYRP1*, a gene involved in eumelanin synthesis in other species [14], associated with quantitative color differences in thrushes. Together, these results further support a broadly conserved role of the melanocortin pathway in coloration and color patterning across diverse taxa [6,14]. Delmore *et al.* [5] also found several genes involved with migratory behavior in other animals co-localized in the cluster associated with migratory orientation in thrushes. These notably include *CLOCK*, a central component of the circadian clock in many species [11]. This result suggests that, similar to melanin-based coloration, there may be a suite of migratory genes that are common across taxa [4,11].

The relative roles of different evolutionary processes in contributing to hybridization and speciation remain debated. These include the importance of complete or near-complete allopatry in generating isolating barriers [15] and the roles of natural and sexual selection [16] as well as pre- and post-mating processes [17] in restricting gene flow in sympatry. Divergence localized at plumage loci in both hybrid zones suggests a potential role for mate choice and reproductive isolation based on color traits in both hybrid zones, and in the case of the thrushes, migratory behavior. There is indeed some evidence for assortative mating in thrushes [18], although the targets of mate choice in warblers remain an elusive and tantalizing question for future intrepid field biologists.

REFERENCES

1. Mundy, N.I. (2005). A window on the genetics of evolution: MC1R and plumage colouration in birds. Proc. R. Soc. B *272*, 1633–1640.
2. Rockman, M.V. (2012). The QTN program and the alleles that matter for evolution: all that's gold does not glitter. Evolution *66*, 1–17.
3. Buerkle, C.A., and Lexer, C. (2008). Admixture as the basis for genetic mapping. Trends Ecol. Evol. *23*, 686–694.
4. Toews, D.P.L., Taylor, S.A., Vallender, R., Brelsford, A., Butcher, B.G., Messer, P.W., and Lovette, I.J. (2016). Plumage genes and little else distinguish the genomes of hybridizing warblers. Curr. Biol. *26*, 2313–2318.
5. Delmore, K.E., Toews, D.P.L., Germain, R.R., Owens, G.L., and Irwin, D.E. (2016). The genetics of seasonal migration and plumage color. Curr. Biol. *26*, 2167–2173.
6. Hubbard, J.K., Uy, J.A.C., Hauber, M.E., Hoekstra, H.E., and Safran, R.J. (2010). Vertebrate pigmentation: from underlying genes to adaptive function. Trends Genet. *26*, 231–239.
7. Kronforst, M.R., Barsh, G.S., Kopp, A., Mallet, J., Monteiro, A., Mullen, S.P., Protas, M., Rosenblum, E.B., Schneider, C.J., and Hoekstra, H.E. (2012). Unraveling the thread of nature's tapestry: the genetics of diversity and convergence in animal pigmentation: Evolution and genetics of animal pigmentation. Pigment Cell Melanoma Res. *25*, 411–433.
8. Poelstra, J.W., Vijay, N., Bossu, C.M., Lantz, H., Ryll, B., Müller, I., Baglione, V., Unneberg, P., Wikelski, M., Grabherr, M.G., and Wolf, J.B. (2014). The genomic landscape underlying phenotypic integrity in the face of gene flow in crows. Science *344*, 1410–1414.
9. Delmore, K.E., Fox, J.W., and Irwin, D.E. (2012). Dramatic intraspecific differences in migratory routes, stopover sites and wintering areas, revealed using light-level geolocators. Proc. R. Soc. B. *279*, 4582–4589.
10. Irwin, D.E., and Irwin, J.H. (2005). Siberian migratory divides. In Birds of Two Worlds: the Ecology and Evolution of Migration, R. Greenberg, ed. (Johns Hopkins University Press), pp. 27–40.
11. Liedvogel, M., and Lundberg, M. (2014). In the genetics of animal movement and migration syndromes. In Animal Movement Across Scales, L.-A. Hansson, ed. (Oxford University Press), pp. 219–231.
12. Linnen, C.R., Poh, Y.P., Peterson, B.K., Barrett, R.D., Larson, J.G., Jensen, J.D., and Hoekstra, H.E. (2013). Adaptive evolution of multiple traits through multiple mutations at a single gene. Science *339*, 1312–1316.
13. Uy, J.A.C., Cooper, E.A., Cutie, S., Concannon, M.R., Poelstra, J.W., Moyle, R.G., and Filardi, C.E. (2016). Mutations in different pigmentation genes are associated with parallel melanism in island flycatchers. Proc. R. Soc. B *283*, 20160731.
14. Gratten, J., Beraldi, D., Lowder, B.V., McRae, A.F., Visscher, P.M., Pemberton, J.M., and Slate, J. (2007). Compelling evidence that a single nucleotide substitution in TYRP1 is responsible for coat-colour polymorphism in a free-living population of Soay sheep. Proc. R. Soc. B *274*, 619–626.
15. Feder, J.L., Egan, S.P., and Nosil, P. (2012). The genomics of speciation-with-gene-flow. Trends Genet. *28*, 342–350.
16. Safran, R.J., Scordato, E.S.C., Symes, L.B., Rodriguez, R.L., and Mendelson, T.C. (2013). Contributions of natural and sexual selection to the evolution of premating reproductive isolation: a research agenda. Trends Ecol. Evol. *28*, 643–650.
17. Butlin, R., Debelle, A., Kerth, C., Snook, R.R., Beukeboom, L.W., Castillo, C.R., Diao, W., Maan, M.E., Paolucci, S., Weissing, F.J., and van de Zande, L. (2012). What do we need to know about speciation? Trends Ecol. Evol. *27*, 27–39.
18. Ruegg, K., Anderson, E.C., and Slabbekoorn, H. (2012). Differences in timing of migration and response to sexual signalling drive asymmetric hybridization across a migratory divide. J. Evol. Biol. *25*, 1741–1750.