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Species boundaries, hybridization and gene flow

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The rise of genomics has spurred a renewed interest in hybridization and the permeability of species boundaries. However, these ideas are not new. Here I review early work by Patton and colleagues on hybridization, gene flow, and the nature of species boundaries in pocket gophers and argue that a focus on the underlying biology of the organism provides insights into hybridization and gene flow that are not obtainable from genomic data alone.

El auge de la genómica ha estimulado un renovado interés en la hibridación y la permeabilidad de los límites entre especies. Sin embargo, estas ideas no son nuevas. En este artículo, analizo los primeros trabajos de Patton y sus colegas sobre la hibridación, el flujo genético y la naturaleza de los límites entre especies en las tuzas y sostengo que un enfoque en la biología subyacente del organismo proporciona conocimientos sobre la hibridación y el flujo genético que no se pueden obtener a partir de datos genómicos únicamente.

Keywords: Population genetics; introgression; pocket gophers; *Thomomys bottae*.

The publication of a Neandertal genome sequence in 2010 included the startling discovery that most modern humans carry a small percentage of Neandertal DNA as a result of hybridization when the two species came into contact (Green *et al.* 2010). This conclusion was reached using a then-new statistical test (the ABBA-BABA test) that could be applied to whole-genome data to distinguish between shared variation due to unsorted ancestral polymorphism and shared variation due to gene flow. This discovery was followed by a torrent of papers showing that hybridization is common in many taxa and that species boundaries are permeable (Payseur and Rieseberg 2016). Hybridization became fashionable.

Of course, these topics are not new, and the recent studies were preceded by decades of empirical research on hybridization in a variety of taxa (*e. g.* Darwin 1859; Endler 1977; Arnold 1992). Jim Patton has been studying species boundaries, hybridization, and gene flow in gophers and other small mammals since the 1960's (Patton and Dingman 1968). Here I describe some of that work and highlight a few papers which I consider to be superb and which had a strong influence on my own research and thinking about hybridization and gene flow.

In a wonderful series of papers on pocket gophers over several decades, Patton documented morphological, cytogenetic, and genetic variation across Western North America in the genus *Thomomys* (*e. g.* Patton and Dingman 1968; Patton *et al.* 1972; Patton 1973; Patton and Yang 1977; Patton *et al.* 1979; Patton and Sherwood 1982; Hafner *et al.* 1983; Patton *et al.* 1984; Smith and Patton 1984; Patton and Smith 1990; Patton and Smith 1994). Most of this work focused on *T. bottae* but also included the closely related species *T. townsendii* and *T. umbrinus* with which *T. bottae* hybridizes. Patton was an early adopter of using starch gel electrophoresis to measure genetic variation as reflected in differences in allozyme frequencies (Patton *et al.* 1972), and he discovered that conspecific populations of *T. bottae* exhibited unusually high levels of genetic differentiation (Patton and Yang 1977). In fact, *T. bottae* is unusual among mammals in harboring an enormous amount of variation in karyotype, genes, and morphology. For example, within *T. bottae*, there are over 100 recognized morphological races (Patton and Smith 1990). This extreme variation stems from a variety of factors, including the large geographic range of *T. bottae* and the wide diversity of habitats and environments in which the species is found, from sea level to 13,000 feet. In addition, a fragmented distribution and comparatively low levels of dispersal provide opportunities for differentiation among populations. Patton was interested in describing the extent of hybridization among these different forms, as well as the amount of hybridization between *T. bottae* and closely related species where they come into contact.

Much of the initial interest in studying patterns of hybridization in gophers came from a desire to delineate species boundaries, rather than to study hybrid zone dynamics per se (Patton 1993). Nonetheless, these studies provided important insights into the frequency and consequences of hybridization among genetically differentiated taxa. Viewed through the lens of recent genomic studies of hybridization, Patton's work stands out for its deep focus on the biology of the organism. Pocket gophers are subterranean mammals, and much of their biology follows from this lifestyle. They are restricted to friable soils and consequently often have patchy distributions. They are solitary and live at low densities. Where taxa come into contact, they have mainly parapatric rather than overlapping distributions.

One of the clear patterns to emerge from Patton's work is that despite high levels of morphological and genetic differentiation between parapatric forms, hybridization is common where taxa meet. Most hybrid zones are narrow relative to the range of the hybridizing taxa, reflecting relatively low densities, limited movements, and restricted habitats available to gophers. Despite the prevalence of narrow hybrid zones, the consequences of hybridization differ substantially among different parapatric forms. For example, in central New Mexico *T. bottae ruidosae* and *T. b. actuosus* meet and hybridize along Nogal Canyon. These forms differ substantially in karyotype, morphology, and allozymes, with three fixed allozyme differences. Few F1 individuals are observed, but multiple backcross or later-generation intercross animals are seen (Patton *et al.* 1979). Moreover, evidence of mismatched allozyme alleles (in both directions) are seen in populations many miles from the narrow contact zone, consistent with significant introgression. In contrast, *T. bottae* and *T. umbrinus* hybridize in Sycamore Canyon in the Patagonia Mountains of southern Arizona. Again, the hybridizing forms differ substantially in karyotype, morphology, and allozymes, with three fixed allozyme differences. However, in this case, nearly all hybrids are F1's with little evidence of backcross progeny (Patton 1973). Moreover, histological studies of testes in F1 males reveal that these animals are either sterile or have greatly reduced fertility. Comparison of these two different hybrid zones (as well as others) illustrated the important and surprising result that the amount of genetic differentiation is not a good predictor of the amount of isolation in gophers. While the recent genomic data reveal a history of introgression between many taxa, such data tell us little about what actually happens when two taxa meet.

Patton recognized that species boundaries are difficult to define not only because of hybridization but also because the sorting of ancestral polymorphism and the genealogical relationships of small local populations can lead to biological species that are not monophyletic. In a series of papers, Patton and Smith (1981, 1989, 1994), along with work by Thaeler (1980) and Rogers (1991), provided an early empirical example that added to a growing recognition of the potential discordance between gene trees and species trees from both theory (*e. g.* Tajima 1983; Hudson 1992) and data (Avise *et al.* 1983; Avise 1989). This work anticipated the now-widespread understanding from genomic data that different genes may produce discordant trees (*e. g.* Degnan and Rosenberg 2009). *T. bottae* and *T. townsendii* have non-overlapping distributions in the northern Great Basin, and they hybridize in the Honey Lake Valley of northern California. Several studies showed that *T. bottae* is paraphyletic with respect to *T. townsendii*; in other words, there are some populations of *T. bottae* that are more closely related to *T. townsendii* than they are to other populations of *T. bottae* (Thaeler 1980; Patton and Smith 1989; Rogers 1991). Notably, Patton and Smith (1994) discovered not only that *T. bottae* is paraphyletic, but that different genes (mitochondrial and nuclear) revealed different phylogenies among the populations of these species, providing conflicting views of monophyly, paraphyly, and even polyphyly for species. These conclusions were possible only because of the detailed sampling that was performed as well as the use of different molecular markers, something that was still relatively uncommon in the early 1990's. Two quotes from this paper of 30 years ago seem particularly prescient.

"The complexities uncovered in this particular example are probably similar in virtually all other groups of pocket gophers and many other organisms. In other words, this case history is not likely to be an isolated incident, which can thus be ignored; rather, this pattern may be commonly observed for a wide range of organisms." And later: "… the more one knows about variation within and among populations, and thus the more detail that is available regarding intraspecific genealogy, the more likely it will be that the boundaries of species will be blurred…" (p. 23, Patton and Smith 1994). Indeed, blurry species boundaries now seem commonplace.

One of my favorite papers by Patton involves the study of gene flow among local populations of pocket gophers that are geographically close and not reproductively isolated. Conducted with his postdoc, Joanne Daly, this paper stands as one of the more thorough studies of gene flow in any mammal estimated using both direct and indirect methods (Daly and Patton 1990). In this study, they followed the movement of individual gophers between fields at the Hastings Natural History Reservation in Carmel Valley, California over several years using an impressive combination of approaches. All gophers were tagged, and blood was taken from all animals. Every animal in each of several populations was tracked. Gophers were trapped above ground using pitfall traps, and underground in burrows. Dispersal was recorded between established populations, as well as into fields in which all gophers had been removed. Observations were made almost continuously for eight months out of the year, for three consecutive years. The sex, age, and reproductive condition of all animals were recorded. Finally, all of the direct measures of dispersal were compared to indirect inferences of gene flow obtained from analysis of patterns of genetic differentiation from allozyme data generated in this study combined with data from a previous study of the same populations (Patton and Feder 1981). This combination of approaches revealed many insights into the nature of gene flow between local populations. For example, gophers often disperse above ground. Females disperse when young, but males disperse later. Notably, genetic data suggested that 8 to 18 migrants moved between populations each generation, a number substantially *above* the 1 to 6 individuals that were observed to be dispersing between established populations, but substantially *below* the 20 to 40 individuals that moved into unoccupied habitat. These observations are consistent with the idea that gene flow may occur from a combination of recolonization following local extinction as well as dispersal between established populations. Such a conclusion would have been impossible without the combination of direct and indirect approaches used by Daly and Patton. Another insight to emerge from this work is that even though females may disperse more than males, males may contribute more to gene flow through a heavily biased operational sex ratio. These and many other insights about dispersal and gene flow were possible because of an intense focus on individual animals in the field, insights that are not obtainable from genomic data alone.

Jim Patton is known to many as a preeminent mammalogist. From my brief comments on just a few of his many excellent papers, I hope to have conveyed that he also made lasting contributions to our understanding of basic issues in population genetics and evolutionary biology.

Jim Patton has had a profound influence on me both professionally and personally. I was fortunate to be an undergraduate at UC Berkeley in the early 1980s and to take many of the "ology" classes that were offered. Berkeley continues to offer these important classes, placing a premium on the experience that students get when exposed to field work and studies of organisms in their natural environment. The most memorable class of my college career was Jim Patton's mammalogy class. I was captivated by the science and by Jim's enthusiasm as a teacher. His class was rigorous and demanding, and as a professor, he was approachable. He always had high expectations, and students seemed to rise to the challenge. Like many people who teach mammalogy, I have modeled my own class after his. I was thrilled when he and Doug Kelt revised Lawlor's "Handbook to the Orders and Families of Living Mammals." We use the collections of the MVZ in my class, and my students are always amazed, as am I, by the sheer number of specimens with JLP tags on them, representing Orders and Families of mammals from every continent.

After taking his mammalogy class as a young student, I approached Jim to ask if I could get involved in research. He asked what I wanted to study. I said that I had no idea. He said something like "well, go figure it out and then come back!" Undeterred, I started talking with his graduate students and eventually ended up doing a little project on kangaroo rats. A few years later I had the unforgettable experience of joining him and Carol in the field in the altiplano of southeastern Perú. Jim has achieved a near-mythical status as a field biologist, and all I can say is that all the stories are true.

The aspect of Jim that I most admire is his support for others and his unfailing generosity. He is never too busy to stop and help others, and he treats everyone with the same kindness and respect, from young undergraduates to senior colleagues. He is, quite simply, a gem of a human. My experience is not unique – I know that Jim influenced generations of students and colleagues, and that our lives are richer because of him.

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Literature cited

Arnold, M .L. 1992. Natural hybridization as an evolutionary process. Annual Review of Ecology and Systematics 23:237-261.

Avise, J. C. 1989. Gene trees and organismal histories: a phylogenetic approach to population biology. Evolution 43:1191-1208.

Avise, J. C. *et al.* 1983. Mitochondrial DNA differentiation during the speciation process in *Peromyscus*. Molecular Biology and Evolution 1:38-56.

Daly, J. C., and J. L. Patton. 1990. Dispersal, gene flow, and allelic diversity between local populations of *Thomomys bottae* pocket gophers in the coastal ranges of California. Evolution 44:1283-1294.

Darwin, C. 1859. The origin of species by means of natural selection.

Degnan, J. H., and N. A. Rosenberg. 2009. Gene tree discordance, phylogenetic inference and the multispecies coalescent. Trends in Ecology and Evolution 24:332-340.

Endler, J. A. 1977. Geographic variation, speciation, and clines. Monographs in Population Biology 10, Princeton University Press. Princeton, U. S. A..

Green, R. E. *et al* . 2010. A draft sequence of the Neandertal genome. Science 328:710-722.

Hafner, J. C. *et al.* 1983. Contact zones and the genetics of differentiation in the pocket gopher *Thomomys bottae* (Rodentia: Geomyidae). Sytematic Zoology 32:1-20.

Hudson, R. R. 1992. Gene trees, species trees, and the segregation of ancestral alleles. Genetics 131:509-512.

Patton, J. L. 1973. An analysis of natural hybridization between the pocket gophers, *Thomomys bottae* and *Thomomys umbrinus*, in Arizona. Journal of Mammalogy 54:561-584.

Patton, J. L. 1993. Hybridization and hybrid zones in pocket gophers (Rodentia, Geomyidae). Pp. 290-308, *in* Hybrid Zones and the Evolutionary Process (Harrison, R. G., ed.). Oxford University Press. New York, U. S. A.

Patton, J. L., and R. E. Dingman. 1968. Chromosome studies of pocket gophers, genus *Thomomys*. I. The specific status of *Thomomys umbrinus* (Richardson) in Arizona. Journal of Mammalogy 49:1-13.

Patton, J. L., and S. Y. Yang. 1977. Genetic variation in *Thomomys bottae* pocket gophers: macrogeographic patterns. Evolution 31:697-720.

Patton, J. L., and J. H. Feder. 1981. Microspatial genetic heterogeneity in pocket gophers: Non-random breeding and drift. Evolution 35:912-920.

Patton, J. L., and M. F. Smith. 1981. Molecular evolution in *Thomomys*: phyletic systematics, paraphyly, and rates of evolution. Journal of Mammalogy 62:493-500.

Patton, J. L., and S. W. Sherwood. 1982. Genome evolution in pocket gophers (genus *Thomomys*). 1. Heterochromatin variation and speciation potential. Chromosoma 85:149-162.

Patton, J. L., and M. F. Smith. 1989. Population structure and the genetic and morphologic divergence among pocket gopher species (genus *Thomomys*). Pp. 284-304, *in* Speciation and its Consequences (Otte, D., and J. A. Endler, eds). Sinauer. Sunderland, Massachusetts.

Patton, J. L., and M. F. Smith. 1990. Evolutionary dynamics of *Thomomys bottae* pocket gophers, with emphasis on California populations. University of California Publications in Zoology 123:1-161.

Patton, J. L., and M. F. Smith. 1994. Paraphyly, polyphyly, and the nature of species boundaries in pocket gophers (Genus *Thomomys*). Systematic Biology 43:11-26.

Patton, J. L. *et al*. 1972. Genic Variation in Hybridizing Populations of Gophers (Genus Thomomys). Systematic Zoology 21:263-270.

Patton, J. L. *et al* . 1979. Hybrid zones in *Thomomys bottae* pocket gophers: genetic, phenetic, and ecologic concordance patterns. Evolution 33:860-876.

Patton, J. L. *et al*. 1984. Genetics of hybridization between the pocket gophers *Thomomys bottae* and *Thomomys townsendii* in northeastern California. The Great Basin Naturalist 44:431-440.

Payseur, B. A., and L. H. Rieseberg. 2016. A genomic perspective on hybridization and speciation. Molecular Ecology 25:2337-2360.

Rogers, M. A. 1991. Evolutionary differentiation within the northern Great Basin pocket gopher, *Thomomys townsendii*. II. Genetic variation and biogeographic considerations. Great Basin Naturalist 51:127-152.

Smith, M.F., and J. L. Patton. 1984. Dynamics of morphological differentiation: temporal impact of gene flow in pocket gopher populations. Evolution 38:1078-1087.

Tajima, F. 1983. Evolutionary relationship of DNA sequences in finite populations. Genetics 105:437-460.

Thaeler, C. S., Jr. 1980. Chromosome numbers and systematic relations in the genus *Thomomys* (Rodentia: Geomyidae). Journal of Mammalogy 61:414-422.

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