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Point to Ponder

Both Reintroduction and Recolonization Likely Contributed to the Re-establishment of a Fisher Population in East-central Alberta

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Abstract

Recently, Stewart *et al.* (2017) investigated the origins of contemporary fisher populations in the Cooking Lake Moraine (CLM) of east-central Alberta, Canada, where fishers (*Pekania pennanti*) from Ontario and Manitoba, Canada were reintroduced in the early 1990s. To address this objective, Stewart *et al.* (2017) compared microsatellite alleles from extant fisher populations in the CLM to those from Ontario, Manitoba, and other Alberta populations. They reported that the CLM population clustered with adjacent native Alberta populations, consistent with

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recolonization, but also that 2 of 109 microsatellite alleles in the CLM occurred only in the source populations from Ontario and Manitoba. Rather than allowing for the possibility that these alleles descended from reintroduced fishers, the authors speculated that they represented random mutations among fishers that recolonized the area naturally from nearby populations in Alberta, and concluded that the reintroduction had failed completely. We disagree with this conclusion for 2 reasons. We contend it is more likely that the 2 alleles represent a genetic signature from the individuals released during the reintroduction, rather than being the result of mutations. We further suggest that, irrespective of the genetic legacy of introduced fishers in the recovered population, the presence of reintroduced fishers in the CLM may have helped facilitate natural recolonization of the area by fishers from surrounding areas. In our view, Stewart *et al.*'s (2017) findings do not demonstrate conclusively that the reintroduction program failed; on the contrary, we argue that their findings indicate that reintroduced fishers likely contributed to the long-term persistence of fishers in the CLM. The uncertainty surrounding this case underscores the importance of genetic monitoring following reintroductions.

Key Words: Reintroduction Biology, Conservation Genetics, Wildlife Management, Species Recovery, Fisher, *Pekania pennanti*.

INTRODUCTION

During the early 1990s, Proulx *et al.* (1994) translocated 20 fishers (*Pekania pennanti*; 9 females and 5 males from near the towns of Aspley, Bancroft, and Boulter in Ontario, Canada, and 4 females and 2 males from near the town of Steinbach in Manitoba, Canada) to the Cooking Lake Moraine (CLM), a 400-km² area east of the city of Edmonton, Alberta, Canada (AB) where fishers were believed to have been extirpated for at least 50 years (Soper 1951; Proulx *et al.* 1994). Although fishers inhabited the CLM at the beginning of the 20th century (Hagmeier 1959; Hall and Kelson 1959), by the mid-1950s those populations had been decimated by strychnine poisoning, habitat loss, and unregulated trapping (Soper 1951, 1964; Badry *et al.* 1997).

Translocated fishers included 6 females and 3 males that were released in March 1990 (i.e., during the reproductive season), 5 females and 3 males in June 1990, and 1 female and 2 males in August 1991. Translocated animals were monitored from 1990 to 1992 during a study of their home range and habitat use (Badry *et al.* 1997). After the radio-telemetry study was completed, Badry (1994) reported that up to 3 females from the spring release, and 5 females and 3 males from the summer releases, were present in the reintroduction area. In addition, reintroduced mature male and female fishers had inhabited the same areas during the 1991 reproductive season. Badry (1994) also reported that 2 juvenile fishers were observed during the fall of 1993, including a male that was captured incidentally in a beaver (*Castor canadensis*) trap (Badry 1994). Thus, available evidence suggests that during the first few years after the translocations occurred, fishers interacted and potentially reproduced in the CLM.

STEWART *et al.*'s FINDINGS

Stewart *et al.* (2017) genotyped 147 individuals (40 from the CLM, 53 from other regions in Alberta, 29 from Ontario [only from the Bancroft area], and 25 from Manitoba) at 15 microsatellite loci to determine the genetic contribution of reintroduced individuals to the CLM population. They investigated the success of the reintroduction in terms of 3 possible non-mutually exclusive outcomes of the CLM reintroduction with regard to the genetic makeup of the contemporary CLM samples: I) genetic signature of reintroduction source populations (Ontario or Manitoba); II) alleles from adjacent Alberta populations; III) unique alleles not found in either reintroduction or neighboring populations. Importantly, success was equated with case I, which we argue below may be an overly narrow definition of success. We also dispute their interpretation of the data in terms of their refutation of outcome I.

Stewart *et al.* (2017) analyzed genotype frequencies using a standard assignment test and demonstrated that the extant CLM population clustered closely with adjacent Alberta populations, and not with Ontario or Manitoba populations, thereby supporting outcome II. However, 2 of the 109 microsatellite alleles detected among extant fishers in the CLM (*Ma-2* 173 and *Lut604* 136) only occurred in fishers from Ontario and Manitoba and, thus, appeared to be indicative of past interbreeding between reintroduced and recolonizing fishers (as in outcome I). However, Stewart *et al.* (2017) speculated that these alleles were the product of independent mutations and were not identical by descent to the Ontario alleles. They concluded on the basis of this speculation that the reintroduction was unsuccessful.

We accept the conclusion of the authors that the majority of the genomic background in the extant population was explained by recolonizing gene flow (outcome II) and not the

individuals translocated during the reintroduction. However, we disagree with their interpretation for the 2 alleles that matched the source populations for the reintroduction (which we believe supports outcome I) and, most importantly, their conclusion that the reintroduction was unsuccessful (based on a narrow definition of success).

In their argument against outcome I, Stewart *et al.* (2017) speculated that such mutations may have occurred simply because *Ma-2* and *Lut604* were composed of a large number of tandem repeats (Ellegren 2004). However, this explanation seems non-parsimonious. Many factors (e.g., repeat number, sequence of the repeat motif, length of the repeat unit, flanking sequence, interruption in the microsatellite, recombination rate, transcription rate [Schlötterer 2000]) affect the mutation rates of microsatellites, making it impossible to know the mutation rate without direct empirical evidence, and none was presented. More importantly, the absence of the 2 alleles in native Alberta populations, despite a large number of genotyped samples, indicates that such mutations were extremely unlikely. The improbability of their having arisen spontaneously belies the more parsimonious alternative that they originated from a population known to harbor those alleles and to have contributed alleles to that location in the past. Furthermore, although microsatellite mutation rates range from 10^{-6} to 10^{-2} per generation, which is considered much higher than base substitution rates (Schlötterer 2000), it seems quite unlikely that both alleles resulted from mutations in a naturally recolonized CLM fisher population that has existed for only 5 generations (ca. 5 years per generation for the fisher). Therefore, Stewart *et al.*'s (2017) conclusion that the 2 alleles that uniquely characterize the Ontario and Manitoba source populations resulted from random mutations appears, at best, arbitrary to us. On the contrary, we argue that the presence of diagnostic alleles in the CLM fisher population strongly suggests that some source individuals from Ontario and Manitoba persisted and contributed to the current gene pool, and to the re-establishment of a fisher population in the CLM.

As Stewart *et al.* (2017) concluded, fishers from other populations in Alberta clearly expanded naturally into the CLM area, which was not known to be occupied by fishers (Soper 1951, 1964; Banfield 1974) until the reintroductions that occurred in the early 1990s (Proulx *et al.* 1994). Fishers were neither reported by local naturalists (Proulx, 1989-1990, personal notes) nor captured by local trappers for decades in the CLM (F. Neumann, 1990, Alberta Fish & Wildlife Division, personal communication). During the 1990s, fishers recovered demographically throughout Alberta (Neumann 1993) and, not surprisingly, their distribution eventually included the reintroduction area. It is possible

that the presence of reintroduced fishers may have facilitated the natural recolonization of the CLM by fishers from surrounding areas. This would be expected if fishers tended to disperse preferentially to habitats occupied by conspecifics (Doty 1986; Stamps 1988; MacPherson *et al.* 2018). As a result, immigrants that bred with reintroduced fishers would produce fishers with a genetic admixture. Over time, however, with a greater genetic contribution from Alberta fishers and no genetic reinforcement from Ontario or Manitoba fishers, the genetic signature of descendants would likely be more akin to that of Alberta populations. Thus, the genetic characteristics of the original reintroduced fishers would have been diluted over time. Accordingly, we contend that the genetic admixture found among fishers in the CLM is more likely the result of hybridization between Ontario/Manitoba fishers that had been reintroduced in the early 1990s and Alberta fishers that immigrated into the CLM after the reintroduction. Perhaps 1 lesson that can be learned from this case study is the importance of regular genetic monitoring after reintroductions, which, had it been instituted, would have significantly advanced our understanding of the dynamics between reintroductions and recolonizations both in this case and in general.

Although genetic studies can be expensive and labor intensive, the inclusion of genetic data in management plans is necessary. Thus, genetic studies should be designed and implemented to test specific research hypotheses. For instance, 15 microsatellite loci are likely not enough to capture small genomic vestiges of the reintroduced population concealed in the genome ($2n = 38$ chromosomes), especially when the microsatellite regions are non-randomly distributed across the genome (Schlötterer 2000). Additional genome-wide studies with more microsatellite loci or next-generation sequencing technology (e.g., RADseq) may provide additional information about the origins of the CLM fisher population.

Additionally, if determining the genetic origins of CLM fishers is a priority for wildlife managers, then genetic studies involving maternally inherited genes (i.e., mitochondrial DNA) are needed to adequately address these questions because, in fishers, females are the philopatric sex (Aubry *et al.* 2004; Tucker 2013). Among genetic markers, only maternally inherited mitochondrial DNA is subject to the severe demographic constraint of direct descent from the original female. Consequently, the mitochondrial genome is subject to an extremely constrained evolutionary trajectory (Melnick and Hoelzer 1992; Prugnolle and de Meeus 2002; Ishida *et al.* 2011) and its addition to the analysis would provide for a more robust test of the genetic origins of extant fishers in the CLM than is possible using microsatellite data alone.

CONCLUSION

We conclude that Stewart *et al.*'s (2017) findings do not demonstrate conclusively that the reintroduction failed. On the contrary, we argue that their findings indicate that reintroduced fishers acted as founders that were augmented by fishers dispersing from other regions of Alberta, and that they contributed to the long-term persistence of fishers in the CLM. The uncertainty surrounding this case underscores the importance of genetic monitoring following reintroductions.

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