

Appendix 2: Primers and PCR protocols. Forward (F) and reverse (R) primers are given, all in 5'-3' order. Mitochondrial gene loci: 16S = 16S ribosomal RNA, CytB = cytochrome B oxidase, ND4 = NADH dehydrogenase subunit 4, COI = cytochrome C oxidase 1. Nuclear gene loci: c-mos = nuclear genomic protooncogene *c-mos*, Rag2 = recombination activating gene 2, PRLR = prolactin receptor.

Marker:	PCR protocol modifications:	Primer:	Primer Sequence:	Reference:
16S	annealing: 55°C	16sa-l (F)	CGCCTGTTTATCAAAAACAT	Palumbi et al. (1991)
		16sb-h (R)	CCGGTCTGAACTCAGATCACGT	
CytB	40 PCR cycles	L14910 (F)	GACCTGTGATMTGAAAAACCA YCGTTGT	Burbrink et al. (2000), de Queiroz et al. (2002)
		L14919 (F)	AACCACCGTTGTTATTCAACT	
		H16064 (R)	CTTTGGTTTACAAGAACAATGCTTTA	
		L14903 (R)	GACCTGTGATMTGAAAAACCA	
ND4	annealing: 55°C	ND4 (F)	CACCTATGACTACCAAAAGCTCATGTAGAAGC	Arévalo et al. (1994)
		Leu (R)	CATTACTTTTACTTGGATTTCACCA	
COI	denaturation: 40 sec annealing: 48.5°C	RepCOI-F	TNTTMTCAACNAACCACAAAGA	Nagy et al. (in press)
		RepCOI-R	ACTTCTGGRTGKCCAAARAATCA	
c-mos	40 PCR cycles	S77 (F)	CATGGACTGGGATCAGTTATG	Lawson et al. (2005)
		S78 (R)	CCTTGGGTGTGATTTTCTCACCT	
Rag2	annealing: 60°C, 45 sec	L562 (F)	CCTRADGCCAGATATGGYCATA C	Vidal & Hedges (2005)
		H1306 (R)	GHGAAYTCCTCTGARTCTTC	
PRLR	annealing: 50°C	PRLR_f1 (F)	GACARYGARGACCAGCAACTRATGCC	Townsend et al. (2008)
		PRLR_r3 (R)	GACYTTGTGRACCTCYACRTAATCCAT	

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