

Electronic Supplementary Material

Olah G*, Heinsohn RG, Brightsmith DJ, Espinoza JR, Peakall R (2016) Validation of non-invasive genetic tagging in two large macaw species (*Ara macao* and *A. chloropterus*) of the Peruvian Amazon. Conservation Genetics Resources

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Table S1. Microsatellite primer sequences and locus information for scarlet macaw (*Ara macao*) and red-and-green macaw (*Ara chloropterus*).

Locus name	Forward primer sequence 5'-3'	Reverse primer sequence 5'-3'	Repeat motif	*Fluorescent tag
SCMA02	*TCAACCTCCAGGTGCT TCC	TCCTTCAGTCACCAGC TTCA	(GT) ₂₁	NED
SCMA09	*CACTACCAGCAAGTAGC AGGC	TGAATTCTAACAAAGCA GCGG	(CA) ₂₀ CG (TA) ₃	VIC
SCMA14	*CGCATACTTTACACCCA CCA	TTGTGACAGGGCTAGG CAG	(AC) ₂₀	FAM
SCMA22	*AACTGTGATGAAGTTCG TGGC	CAACGGCTACACACAG TGCT	(TG) ₂₂	VIC
SCMA26	*AGCAAAGGTAAGGAGC AGCA	GGCACCTCTATCATCT ATTGCAG	(TG) ₂₁	VIC
SCMA27	*TTCTGCAGCAGTTCCA AA	TGGACTCTGTATTCCA GTCGC	(CA) ₂₂	FAM
SCMA30	*TTGCCAGGTCTTCTCT ACC	ACCACCTCTCTTGACT TGTAATTG	(CA) ₂₄	FAM
SCMA31	*TGTGCTCCCTACAGTTC CAA	AACGCTGAACTTGGTG TGGT	(AC) ₂₁	FAM
SCMA32	*GGCATGGCTCTTACTT GCT	TTGCCACTGAGGCTTC TACC	(TG) ₂₁	VIC
SCMA33	*GAGGCACTATTCTGGC AGC	GCTAAGCAGATTGTC TAAACATTCA	(AC) ₂₁	VIC
SCMA34	*TTTGGCAGTAGTCGGGA TTT	AACTTGGGAATACATC GCTGA	(AC) ₂₂	VIC

Table S2. Error reports for microsatellite markers used in this study for scarlet macaw (*Ara macao*) and red-and-green macaw (*Ara chloropterus*).

Presented are locus name, amplification failure rate (AF), number of repeated genotypes, allelic dropout at the smaller allele (A1), allelic dropout at the larger allele (A2), number of false alleles, and genotyping error rate (GE).

Locus	AF (%)	N repeats	Allelic dropout at A1	Allelic dropout at A2	False allele	GE (%)
AgGT17	0.4	55	1	0	0	1.8
SCMA 02	14.2	7	0	1	1	28.5
SCMA 09	1.7	49	0	0	0	0
SCMA 14	2.5	48	0	3	2	10.4
SCMA 22	1.9	43	0	0	0	0
SCMA 26	2.1	48	0	1	0	2
SCMA 27	6.3	15	2	0	2	26.6
SCMA 30	4.9	42	0	1	1	4.7
SCMA 31	18.6	50	0	0	0	0
SCMA 32	1.9	44	0	2	1	6.8
SCMA 33	1.5	53	0	0	0	0
SCMA 34	1.1	48	0	0	0	0

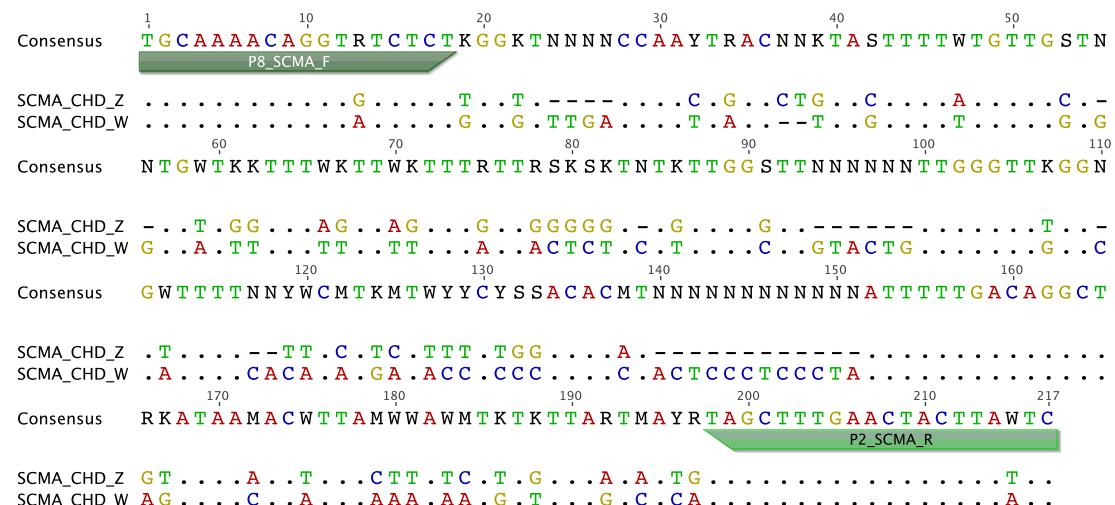
Table S3. Pairwise Shannon Partition Analysis and AMOVA between blood and feather samples of scarlet macaw (SCMA, *Ara macao*) and red-and-green macaw (RGMA, *Ara chloropterus*) in the Tambopata Research Center, Peru (999 permutations).

# loci	Species	N	Shannon		AMOVA	
			<i>sH(AP)</i>	P	<i>F_{ST}</i>	P
6 loci	SCMA	73	0.095	0.564	0.000	0.447
	RGMA	23	0.079	0.799	0.000	0.444
9 loci	SCMA	73	0.093	0.663	0.000	0.481
	RGMA	23	0.065	0.847	0.000	0.455
11 loci	SCMA	73	0.051	0.709	0.000	0.475
	RGMA	23	0.069	0.919	0.000	0.440

Figure S1. Binding sites of primers P8_SCMA_F and P2_SCMA_R on the consensus sequence of scarlet macaw (*Ara macao*) CHD Z and W genes.

P8_SCMA_F: TGCAAAACAGGTRTCTCT

P2_SCMA_R: GAWTAAGTAGTTCAAAGCTA



Geneious version 6.1 created by Biomatters. Available from
<http://www.geneious.com>

Figure S2. Species identification tests.

Red (circles) = red-and-green macaw (*Ara chloropterus*), RGMA (N = 313); blue (squares) = scarlet macaw (*Ara macao*), SCMA (N = 142); green (triangles) = unidentified bird species, UFO (N = 18).

(a) Probabilities of assignment to three genetic clusters identified by STRUCTURE.



(b) Bi-plot using pre-defined allele frequencies of the two taxa in assignment test of GenAlEx 6.5.

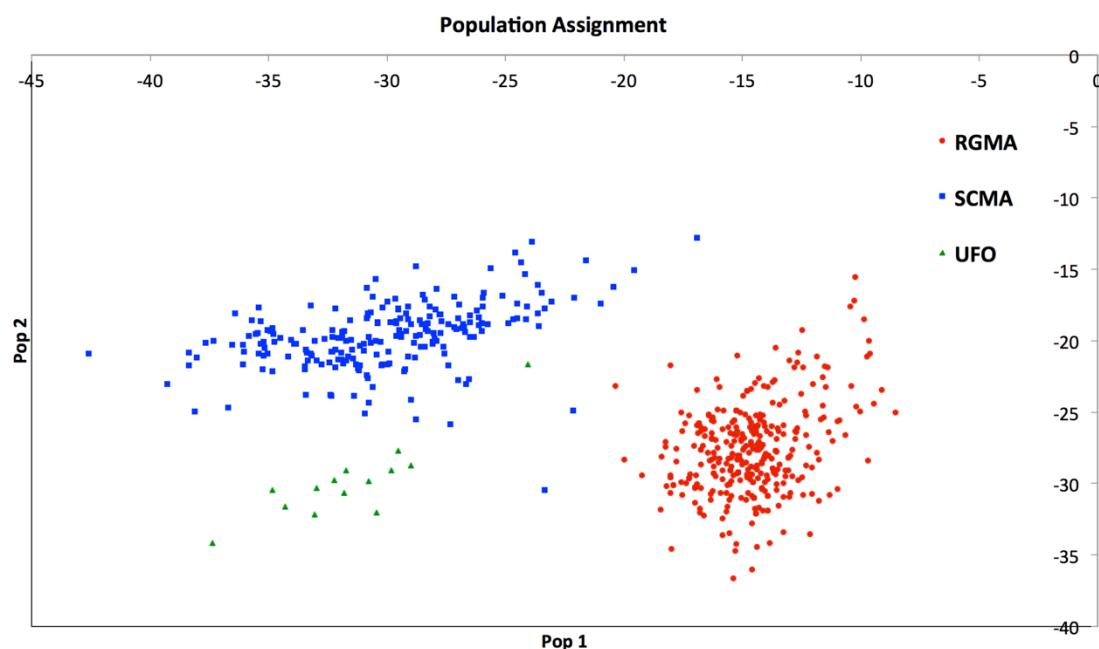


Figure S3. Complete genotype matches of scarlet macaw (*Ara macao*) blood and feather samples in Tambopata, Peru.

Each horizontal line represents a unique genotype with their ID on the right.

Each marker represents a sample with the name of the location collected (black circle = blood; triangle = feather), the distance (m) between locations, and the time (day) between collections on the right.

