

Development of a forensic DNA profiling system and storage scheme to control illegal trade and traffic in CITES-listed parrots (Psittaciformes)

Luca Fumagalli, *Unité de Génétique Forensique, Centre Universitaire Romand de Médecine Légale, Ch. de la Vulliette 4, CH-1000 Lausanne 25 (Switzerland) and Département d'Ecologie et Evolution, Laboratoire de Biologie de la Conservation, Biophore, 1015 Lausanne (Switzerland)*

Key words

Wildlife forensics, DNA, Microsatellites, Individual genetic identification, Parentage analysis, Poaching

Aim of the study

The parrot family represents one of the bird group with the largest number of endangered species, as a result of habitat destruction and illegal trade (smuggling of eggs and wild animals, and laundering through captive breeding facilities). In this study, we isolated new species-specific DNA microsatellite markers in seven parrot taxa threatened with extinction (*Amazona brasiliensis*, *A. oratrix*, *A. pretrei*, *A. rhodocorytha*, *Anodorhynchus leari*, *Ara rubrogenys* and *Primolius couloni*), in order to develop a genetic identification and parentage DNA testing framework to determine illegal traffic.

Material and methods

Biological samples (blood, feathers and DNA extractions) were obtained from private aviaries in Switzerland, the University of Sao Paulo, Brazil (Prof. C. Y. Miyaki) and the Loro Parque Fundacion, Tenerife (Spain). From an enriched genomic library followed by 454 pyrosequencing, we isolated and tested 50 markers/species for suitability and polymorphism across an average number of 18 individuals/species.

Results and significance

We characterized a total of 95 new polymorphic microsatellite markers (mostly tetranucleotides) in the seven species. The mean number of alleles per species and across loci varied from 5.5 and 8.1, with the mean observed heterozygosities ranging from 0.63 to 0.85. Identity (P_{ID} and P_{IDsib}) and parentage (P_1 , P_2 and P_3) exclusion probabilities were highly discriminant.

Despite the potential of wildlife DNA forensics to determine with conclusive evidence Psittacidae illegal trade or harvest from the wild, the implementation of such analyses has been very limited to date for different reasons. First, the number of genetic markers (i.e. microsatellites) specifically developed for the species of interest is low, constraining the use of markers developed in other species (cross-amplification), with a frequent loss in marker specificity and resolution, and therefore loss of statistical power. Second, success rate in isolating microsatellites from parrots has always been reported as low. Third, because of the extreme rarity and therefore difficulty of sampling some species, compiling population DNA databases for the computation of the statistical certainty of a parental assignment is a very difficult task.

Without conclusive evidence it is difficult to prosecute illegal trade in wildlife. DNA analysis provides this evidence and can conclusively prove whether birds have or have not been captive bred. The high variability displayed by the microsatellite loci presented here demonstrates their potential utility to perform individual genotyping and parentage analyses. The data produced in this study will provide authorities with the ability to investigate suspected poachers and smugglers, investigating false parentage claims or establishing a link between trace evidence and an individual (e.g. in case of stolen birds, or when a bird is illegally transferred between different locations). In addition, the samples collected in this study will allow the setup for all these species of a population DNA database for probabilistic assignment, with the possibility of further genetic analyses when asked by law enforcement organizations.

Publications, posters and presentations

Cabrita, J.C.; Fumagalli, L. Characterization of polymorphic DNA microsatellite markers for forensic individual identification and parentage analyses in seven parrot species (family Psittacidae) threatened with extinction (*manuscript in progress*).

Project 3.13.01

Project duration November 2012 - April 2015