

TTPG PROJECTS

Genetic Testing of Galapagos Tortoises in the Private Sector.

In 2014, TTPG board member Jerry Fife was awarded a \$1,000 grant from the International Herpetological Symposium Grants Committee to help fund a study to identify the subspecies of Galapagos tortoises owned in the private sector. Most private owners of Galapagos tortoises do not know the origins of their tortoises. The project goals were to identify the subspecies and help encourage responsible management and breeding of Galapagos tortoises in the private sector.

Initially, the project costs were estimated at \$2,600 to test a panel of 10 tortoises. Project participants were expected to cover costs above the \$1,000 donated by IHS.

The Caccone Lab at Yale University, New Haven, CT is one of the only facilities with the capabilities of completing this testing. Dr. Adalgisa Caccone, Senior Research Scientist, Department of Ecology and Evolutionary Biology, Director YIBS-MSCG Center oversaw the testing. Unfortunately, due to the time required to complete this project, the minimum panel size increased from ten to fourteen tortoises and the costs rose from \$2,600 to \$6,300.

Thanks to the **Turtle and Tortoise Preservation Group (TTPG)** for their \$2,500 grant and the original \$1,000 IHS grant, funds were available to allow this project to continue and to become a reality.

Funding

- Jerry Fife
- James Badman
- Three Jay's Tortoise Sanctuary, LLC
- IHS Grant
- TTPG Grant
- Arizona State University



Blood samples were collected and submitted to Yale for testing, however due to issues with the buffer used to preserve the blood samples, new blood samples would need to be collected and tested. James Badman working with Colette Adams and the Gladys Porter Zoo also made contact with the Henry Doorly Zoo which believed they could test the original samples. In July, 2015 the following Galapagos tortoise blood samples were submitted for testing.

Number of Tortoises Tested

14 Yale University

30 Henry Doorly Zoo



Thanks to the efforts of James Badman, Drew Rheinhardt, Dr. Rich Funk, Wendy DeBevoise for their efforts to collect blood samples. Photo by Jerry Fife

Results

In December 2015, the results were received for the 14 samples submitted to Yale University. The Henry Doorly Zoo has agreed to test additional samples which are currently being collected. These results will be updated when available.

Summary of the 14 tortoises tested

- 2 Isabela/Santa Cruz hybrids (*C. porteri* mother)
- 6 Isabela/Santa Cruz hybrids (*Isabela* mother)
- 1 Isabela hybrid (*C. vicini* mother)

- 4 Purebred or hybrids (*C. guentheri*, *C. vandenburghi*, *C. microphyes*, & *C. vicina*)
- 1 Likely purebred *C. porteri*

Excerpts from the Yale Report

Mitochondrial (mtDNA) DNA Assignment

To enable assignment of the tortoises using *mtDNA* Yale sequenced a 697bp fragment of the mitochondrial control region locus. This locus has been demonstrated as useful for assigning captive individuals of unknown origin (Russello et al., 2007a; Russello et al., 2010, Benavides et al., 2012) and individuals of mixed ancestry from Volcan Wolf (Russello et al., 2007b; Poulakakis et al., 2008; Garrick et al., 2012; Edwards et al., 2013). To determine the presence of new haplotypes (a unique *mtDNA* sequence) and which sequences were identical to previously identified haplotypes, we combined the sequences from the 14 captive tortoises with our reference sequence database containing all known haplotypes recovered from Galápagos tortoises and used statistical parsimony as implemented in the program TCS v1.2.1 (Clement et al., 2000) to collapse identical sequences. No new haplotypes were revealed amongst the tortoises.

Conclusions Regarding Individual Assignment based on mtDNA and microsatellite loci

Based on mitochondrial and nuclear data, the tortoises tested are most likely from central or southern Isabela, where *C. guentheri*, *C. vandenburghi*, *C. microphyes*, and *C. vicina* roam, and the Santa Cruz species from western Santa Cruz, *C. porteri*. Two tortoises are Isabela/Santa Cruz hybrids, with a *C. porteri* mother, given the mtDNA assignment. Six are also Isabela/Santa Cruz hybrids but with an Isabela mother. One appears to be an Isabela hybrid with perhaps a *C. vicini* mother. Given that the Isabela species are hardest to distinguish as they are the most recently diverged in the Galápagos Archipelago, it cannot be determined whether four individuals are purebreds or hybrids between some combination of *C. guentheri*, *C. vandenburghi*, *C. microphyes*, and *C. vicina*, but both their mitochondrial and nuclear DNA originate from the island. One, on the other hand, is most likely a pure bred *C. porteri*.



Based on genetic results, current reproductive tortoises are in optimal breeding groups.
Photo by Jerry Fife