of the forearm. The bone implant was taken from the hip and the patient had to remain hospitalized for five weeks. The Honduran Government paid approximately US\$ 40,000 for the medical treatment of this man.

Tapirs are normally shy animals that will avoid humans and will rapidly flee when encountered by people. Throughout Honduras tapirs are extremely difficult to find and see in their natural habitat, unless they are chased with hunting dogs, in which case the dogs will corner and surround the tapir until the hunter comes within shooting distance. Haddad et al. (2005) report on a fatal attack caused by a lowland tapir (Tapirus terrestris) in southeastern Brazil. In that case, the man who was killed by the tapir provoked the animal by grabbing and stabbing it. The tapir was later found nearby, dead from the wounds inflicted by the local farmers. To my knowledge, there is no other documented or published case of tapirs attacking humans in Honduras, although several hunters have related to me how provoked tapirs have attacked and killed their hunting dogs.

This event may motivate several reactions in the people from the village of Vallecito. Farmers armed only with machete (the great majority) will avoid trying to kill a tapir, while men with firearms will shoot a tapir on sight. A local hunter told me that a businessman from the city of Catacamas, Olancho, had requested a tapir's calf and was willing to pay him up to US\$ 500. Usually, in order to get the calf, hunters have to first kill the adult female. This hunter commented that after the attack, he was no longer interested in this kind of deals. On the other hand, there is a possibility that hunters and their dogs will more often organize tapir hunting expeditions that will now be positively seen by the local villagers.

In the SANP as in most of the rest of the country, deforestation and human population growth are severely reducing the tapir's habitat.

#### Nereyda Estrada M.Sc.

Proyecto Manejo Integrado de Recursos Naturales (USAID/MIRA) Country Coordinator, Honduras, IUCN/SSC Tapir Specialist Group (TSG) E-mail: nerestr@yahoo.com

#### Literature

Haddad, V., Assuncao, M. C., de Mello, R. C. and Duarte, M. R.. 2005: Wilderness & Environmental Medicine, 2(16): 97-100.

#### CONTRIBUTED PAPERS

# Population Dynamics and DNA Microsatellite Survey in the Lowland Tapir

Benoit de Thoisy<sup>1</sup>, Cécile Richard-Hansen<sup>2</sup>, François Catzeflis<sup>3</sup>, Anne Lavergne<sup>4</sup>

- TSG Member, Association Kwata, Cayenne, French Guiana. thoisy@nplus.gf
- Office National de la Chasse et de la Faune Sauvage, Kourou, French Guiana
- <sup>3</sup> Laboratory of Paleontology, University of Montpellier, France.
- <sup>4</sup> Institut Pasteur de la Guyane, Cayenne, French Guiana

#### Introduction

Located on the Eastern part of the Guianas shield, French Guiana is an administrative unit of France, covered by one of the largest remaining blocks of tropical rainforest, and with a very low rate of deforestation (Whitmore, 1997). Despite a still favorable context, the country suffers from lack of political interest in con-

servation of natural resources; for instance, only 3% of the territory is classified as nature reserves. The region also faces a dramatic increase of gold mining activities (Hammond  $et\ al.$ , 2006). Lastly, no hunting regulations are in effect. Several large species of birds and mammals are currently considered threatened, at least in the northern part of the country where most human populations are concentrated. In previous works, hunting pressure was quantified in 4 sites in the north of

the country, and results showed that the observed harvests were beyond maximal thresholds for several large primates species (de Thoisy *et al.*, 2005) and for the Lowland tapir (de Thoisy & Renoux, 2004).

We present the results of the first survey on microsatellite DNA polymorphism in the Lowland tapir, *Tapirus terrestris*, from French Guiana. Molecular approaches are gaining importance in species conservation, since molecular markers can provide reliable information on population dynamics, trends, and gene flow between areas (for studies in Neotropical mammals see Eizirik *et al.*, 2001; Norton & Ashley, 2004; Ruiz, 2005; for studies in Neotropical reptiles see de Thoisy *et al.*, 2006). Population genetics usefully complement field ecological and classical methods, such as surveys, assessment of habitats, and levels and impacts of threats.

#### **Material and Methods**

DNA was extracted with the phenol-chloroform procedure from tissue samples (n=37) collected in hunted animals all over the country (Figure 1). Five oligonucleotide primers developed for *T. bairdii* and *T. terrestris* (Ashley & Norton, 2004) were used: Tte15, Tte5, Tba21, Tba15, Tba23.

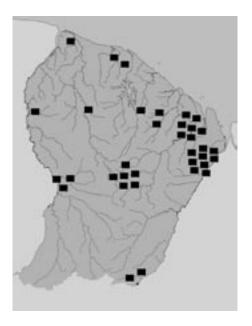


Figure I. Location of survey sites in French Guiana.

Population structuring was investigated with a Bayesian model-based clustering algorithm with STRUCTURE v.2 (Pritchard *et al.*, 2000). Genetic analyses were performed with GENEPOP v.3.4 (Raymond & Rousset, 1995). Genetic polymorphism was measured as observed number of alleles (A), observed

heterozygosity (Ho) and the heterozygosity expected under Hardy-Weinberg proportions (He). Deviation from Hardy-Weinberg equilibrium was tested using Fisher's exact test for fit of genotype proportion (Guo &Thompson, 1992) with the alternative hypothesis  $H_1 =$ heterozygote deficiency. The genetic structure of populations was examined by use of Fis and Fst (Weir & Cockerham, 1984). The significance of Fst was determined by a log-likelihood G-based test (Goudet et al., 1996). Presence of bottlenecks was investigated with the probability of heterozygote excess with a Wilcoxon test, and qualitatively by the shape of the distribution of pairwise differences in repeat numbers among all alleles at each locus, averaged across loci (BOTTLENECK 1.2.02, Cornuet & Luikart, 1996). The interlocus g-test and the Pk distribution method were used to evaluate the hypothesis of population expansion.

#### **Results**

The Bayesian model demonstrated with a high probability (p = 0.94) that the animals came from one single ancestral cluster. Further, the between-sample variation of alleles indicates no differentiation between northern and southern samples (Fst = -0.0014, p = 0.64 – Table 1), suggesting a single panmictic population. The allelic diversity in our sample ranged from 5 to 12, with high observed and expected heterozygosities (0.76 and 0.78 - respectively); the population is at Hardy-Weinberg equilibrium (Fis = 0.03, p = 0.95). This feature was confirmed by significant heterozygote excess (Wilcoxon test: p=0.03), which revealed a bottleneck estimated to have occurred 15-25 generations ago. In contrast, the second test, the loci pair-wise difference, showing a normal L-shape distribution, was not significant and suggests that this bottleneck was not severe. Lastly, both the interlocus-g test and the pK distribution revealed that the population is not expanding.

Table I. Average alleles/locus ( $\hat{A}$ ), expected (He) and observed (Ho) levels of heterozygosity and inbreeding coefficient (f).

Population (n)	(Â)	He	Но	f (Fis) / P-value
French Guiana (37)	8.00	0.78	0.76	0.03 / 0.95
Costa Rica* (15)	2.50	0.37	0.39	-0.06 / 0.69
Panama* (15)	3.30	0.43	0.41	0.03 / P>0.05

<sup>\*</sup> in Tapirus bairdii, Norton & Ashley 2004.

## Discussion and Conclusions

Gene diversity and heterozygocity values recorded in the French Guianan population of T. terrestris are comparable to those recorded in other healthy populations of large mammals, and higher than those recorded in fragmented populations of T. bairdii (Norton & Ashley, 2004) (Table 1). But the limited evidence of a rather recent bottleneck may nevertheless suggest a weakened population. These data represent our preliminary results on assessment of densities, showing reduced abundances in the northern part of the country, where threats to both the tapir and its forest habitats are higher (B. de Thoisy, unpub data). Also, the monitoring of game harvest showed, on the base of analysis of skulls, that almost 75 % of animals killed in this region were not adult (C. Richard-Hansen, unpub. data). Current pressures, including unmanaged hunting, the possibility to commercially market tapir meat, and the cryptic but nevertheless widespread hunting by gold miners, may have dramatic short-term consequences on tapirs in French Guiana.

The identification of a single population in French Guiana has nevertheless important conservation implications. Indeed, absence of structure in the genetic sample allows for a wide range of management options for the species, such as hunting periods and quotas. Large protected areas must also play a major role as hunting refugia for species management; i.e. the possibility that hunted areas may be adjacent to areas free of harvest. The latter could act as efficient sources for re-colonization, and may facilitate population recovery, as along with efficient conservation measures undertaken in hunted areas.

### **Acknowledgements**

The study was funded by the Association Kwata, and made possible with the logistic help of the Institut Pasteur de la Guyane.

#### Literature Cited

- Cornuet, J.M., Luikart, G., 1996. Description and power analysis of two tests for detecting recent population bottlenecks from allele frequency data. *Genetics* 144: 2001-2014.
- Eizirik E., Kim J.H., Menotti-Raymond M., Crawshaw P., O'Brien S.J., Johnson W. 2001. Phylogeography, population history and conservation genetics of jaguars (*Panthera onca*, Mammalia, Felidae). *Molecular Ecology* 10: 65-79.

- Goudet, J., 1999. FSTAT, a program to estimate and test gene diversities and fixation indices, version 2.8.
- Goudet, J., Raymond, M., De Meeüs T., Rousset, F., 1996. Testing differentiation in diploid populations. *Genetics* 144: 933-1940.
- Guo, S.W., Thompson, E.A., 1992. Performing the exact test of Hardy-Weinberg proportion for multiple alleles. *Biometrics* 48: 361-372.
- Hammond D.S., Gond V., de Thoisy B., Forget P.M., DeDijn B. Causes and consequences of a tropical forest gold rush in the Guiana Shield, South America. *Ambio*, in press.
- Norton J.E., Ashley M.V. 2004. Genetic variability and population structure among wild Baird's tapirs. *Animal Conservation* 7: 211–220.
- Pritchard, J.K., Stephens, M., Donnelly, P. 2000. Inferrence of population structure using multilocus gentype data. *Genetics* 155: 945-959.
- Raymond, M., Rousset, F., 1995. GENEPOP version 1.2: population genetics software for exact tests and ecumenicism. *Journal of Heredity* 86: 248-249.
- Reich, D.E., Goldstein, D.B., 1998. Genetic evidence for a Paleolithic human population expansion in Africa. Proceedings of the National Academy of Sciences 95: 8119-8123.
- Ruiz-Garcia M. 2005. The use of several microsatellite loci applied to 13 neotropical primates revealed a strong recent bottleneck in the Woolly Monkey (*Lagothrix lagotricha*) in Colombia. *Primate Report* 71: 27-55
- Shriver, M.D., Jin, L., Ferrel, R.E., Deka, R., 1997. Microsatellite data support an early population expansion in Africa. *Genome Research* 7: 586-591.
- de Thoisy B, Renoux F, Julliot C. 2005. Hunting in northern French Guiana and its impacts on primates communities. *Oryx* 39: 149-157.
- de Thoisy B, Renoux F. 2004. Status of the lowland tapir in French Guiana: hunting pressure and threats on habitats. Second International Tapir Symposium, TSG/SSC, Panama
- de Thoisy B, Hrbek T, Farias IP, Vasconcelos WR, Lavergne A. 2006. Genetic structure, population dynamics, and conservation of Black caiman (*Melanosuchus niger*). *Biological Conservation* 133: 474-482
- Weir, B.S., Cockerham, C.C., 1984. Estimating F-statistics for the analysis of population structure. *Evolution* 38: 1358-1370.
- Whitmore T.C. 1997. Tropical forest disturbance, disappearence, and species loss. Pages 3-12 in W.F. Laurance and R.O. Bierregaard Jr., editors. Tropical forest remnants: ecology, management and conservation of fragmented populations. The University of Chicago Press.