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**EX SITU DISEASES AND THEIR CONTROL FOR REINTRODUCTION OF THE
ENDANGERED LION TAMARIN SPECIES (*LEONTOPITHECUS SPP.*)**

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Introduction

Lion tamarins represent a group of four species of *Leontopithecus spp.* (Fam: *Callitrichidae*) which are endemic to the Atlantic coastal rainforests of Brazil. As summarized recently in a species profile (WPTI 1994), these include the golden lion tamarin, (*L. rosalia*), from Rio De Janeiro, the golden-headed lion tamarin (*L. chrysomelas*) from southern Bahia, the black lion tamarin (*L. chrysopygus*) from Sao Paulo, and the rare black-headed (or black-faced) lion tamarin (*L. caissara*) most recently described as a new species near Sao Paulo and on the island of Superagui. The highest profile species of lion tamarin is the golden lion tamarin which has been propagated under captive conditions (*ex situ*), and reintroduced successfully into natural habitat over the last decade (KLEIMAN et al., 1990; BECK et al., 1991). This work was initiated over 20 years ago as a collaborative effort by the National Zoological Park and the Brazilian government and has expanded into an international program now involving the Rio De Janeiro Primate Center (CPRJ) and 140 American and European zoos, known as the International Golden Lion Tamarin Cooperative Research and Management Committee (GLTMC). The GLTMC is currently expanding to other continents and manages an *ex situ* breeding population of approximately 500 golden lion tamarins; this cooperative effort has produced a great deal of biological and biomedical data from both *ex situ* and *in situ* studies (BALLOU, 1994).

The purpose of this paper is to describe the biomedical aspects of this program which will include diseases identified and studied during *ex situ* propagation, and the control methods that were developed and used to prevent cointroduction of disease into the environment.

General Impact of Disease on Reintroduction

As reintroduction biology became established, the importance of disease problems affecting these programs surfaced in the 1980's and was highlighted by a number of losses attributed to disease outbreaks in several mammalian and avian species during reintroduction attempts (MONTALI and BUSH, 1992). Problems of this sort were formally addressed at a Workshop on Disease and Captive Conservation of Threatened Species spearheaded by the Captive Breeding Specialty Group of the IUCN held in Washington DC in 1991. This led to The International Conference on Implications of Infectious Disease for Captive Propagation and Reintroduction of Threatened Species which was held in Oakland, California in 1992 (WOLFF and SEAL, 1993). Among other tasks, risk assessment of theoretical models ranging from catastrophic to nondevastating infectious disease events as they related to reintroduction issues were developed (BALLOU, 1993); comprehensive protocols for monitoring, investigating and the surveillance of diseases under *ex situ* conditions and in wild habitats were established (MUNSON and COOK, 1993). A recent review, by VIGGERS et al. (1993), further emphasized other possible modes of disease transmission to non-target wild and domestic species, and the reverse transmission of natural population diseases to potentially susceptible *ex situ*-born animals; preventative techniques were vigorously stressed to negate disease pathogens from escaping into or back out of the environment.

In addition to infectious diseases, the introduction of genetically induced diseases could have a deleterious affect on natural populations. It was determined that *ex situ* animals also need be screened for congenital or

familial conditions, and any identified with or suspected of having such conditions should be excluded from reintroduction (BUSH et al., 1993).

Specific Infectious and Potential Genetic Diseases of Lion Tamarins

Lion tamarins are susceptible to a wide range of diseases communicable to *Callitrichidae* (POTKAY, 1992; MONTALI, 1994). Throughout the years of *ex situ* propagation, the following significant infectious (viral, bacterial and parasitic) and some potential genetically-based disease conditions were identified and studied in lion tamarins with the intention of eliminating or preventing any of them from being transmitted and perpetuated in wild populations:

Infectious Diseases

Significant viral diseases observed in lion tamarins include callitrichid hepatitis (CH) (MONTALI et al., 1993) and several other hepatitis viruses (RAMSAY and MONTALI, 1993). Herpes virus infections (CICMANEC, 1977), measles (anecdotal) and encephalomyocarditis virus (WELLS et al., 1986) have been reported in golden lion tamarins. Some viral agents, including coronaviruses of questionable pathogenicity also have been observed in lion tamarins with enteric diseases (RUSSELL et al., 1985).

Of numerous bacterial diseases that have been described in callitrichids, *Yersinia pseudotuberculosis* and *enterocolitica*, *Pasteurella multocida* tooth infections and sepsis (DUNCAN et al., 1993), *Salmonella enteritidis*, and *Aeromonas hydrophila* infections all occurred in golden lion tamarins at the National Zoological Park (NZIP) in Washington DC. Tuberculosis of undetermined type was reported to have occurred in golden lion tamarins (CRANDALL, 1964). *Campylobacter* also has been commonly isolated from golden lion tamarins at NZP without clinical disease but has been associated with inflammatory bowel disease in other callitrichid colonies (RUSSELL et al., 1985).

Parasitism

Acute pulmonary toxoplasmosis was reported in a group of golden lion tamarins at the San Diego Zoo (GRINER, 1983). More recently, disseminated toxoplasmosis developed in NZP golden lion tamarins on loan at three other zoos. Some of these animals were destined for reintroduction to natural habitats in Brazil but died or were removed while in quarantine before shipment.

Spirurid nematodes carried by cockroaches and coprophageous beetles represent an important group of helminths in lion tamarins. For example, *Pterygodermatites nycticeba* emerged as a significant parasite in *ex situ*-born golden lion tamarins at NZP and spread to other colonies (MONTALI, 1993A) but has been found also in other tamarin species. Another spirurid, *Trichospirura leptostoma*, has also been observed in the pancreatic duct but mainly as an incidental finding in the lion tamarins. The acanthocephalid *Prosthenorchis elegans* (thorny-headed worm), transmitted via the cockroach has also been the cause of golden lion tamarin mortalities (BENIRSCHKE, 1980). None of these parasites has been ever identified in any of the wild tamarins.

Potential Genetically-Based Conditions

A low percentage of congenital anomalies including facial and cephalic malformations, cleft palate and hydrocephalus were recognized in neonatal golden lion tamarins in SSP mortality records (MONTALI and RAMSAY, 1995). A relatively high frequency of retrosternal diaphragmatic defects leading to herniation occurred in the early *ex situ* propagation stages of golden lion tamarins (BUSH et al., 1980; MONTALI, 1993B), possibly associated with the use of over-represented founders occultly affected by the condition. Diaphragmatic defects were reported anecdotally in a golden-headed lion tamarin and also observed by DR. F. SIMON in a black lion tamarin originating from the wild, (Personal Communication, 1988). A true genetic mode could never be established for these diaphragmatic abnormalities. An inverted radiographic contrast peritoneogram was developed (BUSH and PHILLIPS, 1995) to screen out affected animals, and the incidence decreased markedly after breeding management changes. Hyperbilirubinemia resembling the Dubin-Johnson syndrome, a nonlethal disturbance in the metabolism of bilirubin in humans, was recently described in several cohorts of female golden lion tamarins (SCHULMAN et al., 1993). An unusually high prevalence of cystine choleliths and septate gallbladder occurs in lion tamarin species (PISSINATTI et al., 1992) with biliary obstruction an uncommon sequela; however, a familial trend was not established for this condition.

Disease Control Methods Employed in Lion Tamarins Destined for Reintroduction

The principle of disease control in the golden lion tamarin protocol has relied upon standard quarantine procedures during which animals destined for reintroduction undergo stringent health exams with appropriate clinical laboratory tests during set periods of observation. All of the anticipated infectious diseases and potential genetically-based conditions as discussed above are examined for. Since 1992, the reintroduction procedure for the golden lion tamarin has been a biphasic one in which *ex situ* propagated tamarin family groups are shipped from contributing zoos to a "pipeline" or staging zoo which provides free-ranging experience prior to shipment of the tamarins to Brazil. Therefore, all tamarins in the program now undergo two quarantine periods one of which includes appropriate vaccinations (e.g., in this case tetanus and rabies), tuberculin testing, routine hemogram, serum chemistry profiles and serological tests for callitrichid hepatitis, human and cebid herpesvirus infections and toxoplasmosis. Also, oral exams, body weight, a blood test for hyperbilirubinemia, and an inverted contrast peritoneogram must be performed to check for diaphragm defects as described above.

Tamarins destined for reintroduction must have no direct or indirect contact with any other nonhuman primates for two weeks prior to leaving the contributing zoo and for four weeks before entering Brazil from the staging zoo. During each of those quarantine periods, examination for ectoparasites, fecal screening for spirurid and other parasites and fecal cultures for pathogenic bacteria (salmonella, shigella, campylobacter, yersinia, etc.) must be performed according to the protocol. (Protocols with more detailed instructions for all of these procedures are available from golden lion tamarin studbook keeper, Dr. J.D. B a l l o u).

Results and Conclusions

During quarantine, tamarins with any signs of known disease problems or exposures were excluded from reintroduction. For example, several animals with positive CH, and toxoplasmosis titers were removed and a few hyperbilirubinemic animals and diaphragmatic defect suspects were eliminated. A group of NZP golden lion tamarins positive for campylobacter (but without clinical signs) were treated and held back until they no longer shed the organisms.

As of early 1994, approximately 125 golden lion tamarins live in the wild as the result of reintroduction efforts over a decade. About 100 of these animals were born in the wild to *ex situ* animals originating from Brazil (8%), Europe (38%), and North America (52%), that were reintroduced over the 10 year period. An attempt was made to keep the reintroduced and wild populations separate although some contact and interbreeding occurred between the two groups.

Early losses in the original reintroduced groups were rectified mainly by management changes with few animals lost to actual disease problems. Some sporadic losses occurred during transport to Brazil associated with undue weather, environmental factors or, rarely, care-taker error. Most of the reintroduction mortalities were lost to follow-up and attributed to tamarins' inexperience with foraging practices and predators; carcasses were usually not found or were in an advanced state of autolysis.

Survival rates increased dramatically in the latter years of reintroduction as more experience was gained with newer pre-release and release methodology. Some limited monitoring for CH and diaphragmatic defects were carried out on restored golden lion tamarin populations with no evidence of any positive CH titers (SCANGA et al., 1993) or effects from any deleterious genetic expression (Dr. M. BUSH, Unpublished Data, 1994).

Thus far, the golden lion tamarin has established a good record for successful reintroduction without contaminating wild populations or perpetuating any diseases acquired *ex situ*. It has been recommended as a useful model for other similar reintroduction programs (MONTALI and BUSH, 1995).

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Summary

Ex situ Diseases and their control for reintroduction of the endangered Lion Tamarin species (Leontopithecus spp.)

This paper provides an overview of important disease issues as they pertain to "reintroduction" biology and medicine, and describes the use of the endangered golden lion tamarin (*Leontopithecus rosalia*) as a model for disease-free reintroduction. Topics include general awareness and impact of disease on reintroduction programs with summaries of specific infectious (viral, bacterial, parasitic) and potential genetic diseases of lion tamarins. Disease control methods used for the successful reintroduction of golden lion tamarins into natural habitats in Brazil are also described.

Zusammenfassung

Bekämpfung von Erkrankungen bei gefährdeten Goldkopflöwenäffchen (Leontopithecus spp.) sowie deren Bedeutung für die Auswilderung

Die vorliegende Arbeit bietet einen Überblick über bedeutsame Erkrankungen mit besonderer Relevanz für die biologischen und veterinärmedizinischen Aspekte der Auswilderung. Die gefährdete Art des Goldkopflöwenäffchens (*Leontopithecus spp.*) dient hierbei als Modell für eine krankheitsfreie Auswilderung. Behandelt werden im einzelnen das vorhandene Bewußtsein zur Häufigkeit und zum Schweregrad von Erkrankungen sowie die Auswirkungen von Krankheiten auf Programme für die Auswilderung. Zusammenfassend werden einige für Goldkopflöwenäffchen spezifische (virusbedingte, bakterielle und parasitäre) Infektionskrankheiten sowie potentiell genetisch bedingte Erkrankungen behandelt. Abschließend wird über Methoden der Krankheitsbekämpfung im Zusammenhang mit der erfolgreichen Auswilderung von Goldkopflöwenäffchen in natürliche Lebensräume in Brasilien berichtet.

Résumé

Comment combattre les maladies dont souffrent les singes-lions (Leontopithecus spec.) et quelle est leur importance pour leur réintroduction

Le présent exposé offre un aperçu des maladies qui ont une signification toute particulière pour les aspects biologiques et vétérinaires de la "réintroduction" des animaux concernés. L'espèce à risque que représente le singe-lion (*Leontopithecus spec.*) sert, en l'occurrence, de modèle d'une réintroduction sans risque de maladie. Les objectifs comprennent en général la prise de conscience de la fréquence des maladies et de leur impact sur les programmes de réintroduction. En résumé, les auteurs décrivent plusieurs infections spécifiques (virales, bactériennes et parasitaires) ainsi que certaines maladies génétiques potentielles du singe-lion. A la fin de l'exposé sont présentées les méthodes de traitement des maladies en rapport avec le succès du programme de réintroduction des singes-lions dans leur environnement naturel au Brésil.

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