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Genetic Variation Within and Among Lion Tamarins

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ABSTRACT The golden lion tamarin *Leontopithecus rosalia rosalia,* one of the rarest and most endangered of New World primates, has been the focus of an intensive research and conservation effort for two decades. During that period, managed breeding from 44 founders has brought the captive population to over 400 individuals, a number that equals or exceeds the estimated number of free-ranging golden lion tamarins. The extent of genetic variation among golden lion tamarins was estimated with an electrophoretic survey of 47 allozyme loci from 67 captive and 73 free-ranging individuals. The amount of variation was low, compared to 15 other primate species, with 4% of the loci being polymorphic (P), and with an average heterozygosity $\overline{H}$ estimate of 0.01 in these callitrichids. Electrophoretic analyses of captive and free-ranging animals (N = 31) of two allopatric morphotypes, *Leontopithecus rosalia chrysopygus* and *L. r. chrysomelas,* were similar to the *L. r. rosalia* findings insofar as they also revealed limited genetic polymorphism. Computation of the Nei-genetic distance measurements showed that the three morphotypes were genetically very similar, although discernible differentiation had occurred at two loci. These data are consistent with the occurrence of recent reproductive isolations of these subspecies.

The golden lion tamarin, *Leontopithecus rosalia rosalia,* has been the focus of intensive conservation research efforts since 1965 (Coimbra-Filho and Mittermeier, 1977). There are today about 400 captive individuals derived from 44 founders in zoos throughout the world. Population size estimates for free-ranging animals suggest that fewer than 300 survive today (Kleiman et al., 1986). Research on the species has included detailed behavior studies, a highly successful captive breeding program (Kleiman, 1977a,b, 1978, 1981; Kleiman and Jones, 1977; Kleiman et al., 1982), socioecological field studies (Coimbra-Filho and Mittermeier, 1973; Coimbra-Filho, 1969; Kleiman et al., 1986), development of a protected reserve (Coimbra-Filho and Mittermeier, 1973, 1977, 1982), and a reintroduction project involving the release of captive-born animals into the wild (Kleiman et al., 1986). Although these investigations have succeeded in discovering important features of this species' natural history, many aspects of the biology and socioecology of lion tamarins have yet to be determined.

The golden lion tamarin is the best known of three distinct morphotypes of lion tamarins now restricted to isolated areas of southeastern Brazil (Fig. 1). Prior to incursion by man, each of these forms is thought to have been more widely distributed. Although the time of geographic isolation has not been established (Hershkovitz, 1977; Coimbra-Filho and Mittermeier, 1977, 1982; Rosenberger and Coimbra-Filho, 1984), *Leontopithecus'*

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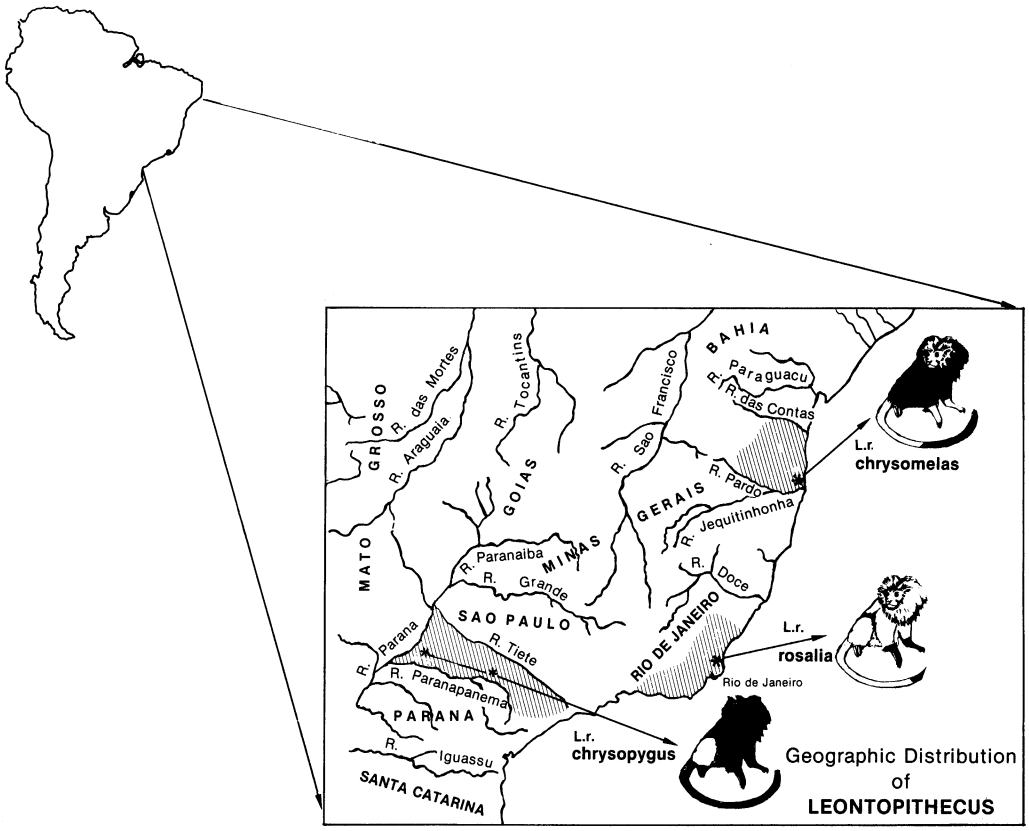

Fig. 1. Historically known geographic distributions are represented by the shaded area. Extant populations are represented by stars.

range might once have extended as far as Minas Gerais (Fig. 1) (Hershkovitz, 1977:825). Each of the three forms is easily distinguished by their distinctive coat colors. Primarily on the basis of this phenotypic trait, the group has been divided by a number of authors into the subspecies *Leontopithecus rosalia rosalia* (the golden lion tamarin), *L. r. chrysopygus* (the black lion tamarin), and *L. r. chrysomelas* (the golden headed lion tamarin) (e.g., Della Serra, 1951; Hershkovitz, 1977; Coimbra-Filho and Mittermeier, 1977; Kleiman, 1981). Recently, however, Rosenberger and Coimbra-Filho (1984) concluded that, based on discrete craniodental characteristics and other morphometric traits, enough ". . . genetically and adaptively important barriers have evolved . . ." to warrant separate species status for the three lion tamarins.

Whether these geographic isolates constitute biological species (cf. Mayr, 1963) has important consequences both for evolutionary biology and for the conservation and management of the endangered genus. From the conservationist's perspective, separate species status may dictate a different strategy regarding the genetic management of each group than might be developed for single geographic subspecies. In terms of our understanding of the evolutionary biology of this genus, the division of *Leontopithecus* into distinct species would suggest a long period of separation and concomitant ecological, genetic, and morphological divergence. Considering the refractory fossil record of this group (as, indeed, is generally the case for South American primates [Patterson and Pascual, 1972; Szalay and Delson, 1979; Martin et al., 1982]), such evolutionarily significant distinctions must rely on as many metrics as are available before taxonomically meaningful assignments can be made.

Decisions about the systematic assessment of the genus *Leontopithecus* are made more difficult by equivocal evidence from a num-

ber of areas. For example, although the three forms are currently allopatric, their phenotypic differences may reflect clinal variation of previously continuous populations (Coimbra-Filho and Mittermeier, 1972, 1973). There is also evidence that the forms produce hybrids (Coimbra-Filho and Mittermeier, 1976), although it is difficult to predict whether such hybrids would occur in natural settings.

The geographic range of all lion tamarins has become drastically reduced in historic times. The three forms of *Leontopithecus* are currently found in small oases of suitable habitat geographically isolated from one another by 600–1,000 km (Fig. 1). The northernmost form belongs to the *chrysomelas* group, known historically to have ranged south from the Rio das Contas and north from the Rio Jequitinhonha (Coimbra-Filho and Mittermeier, 1973, 1977; Hershkovitz, 1977; Kleiman, 1981). Today, *chrysomelas* populations persist in the state of Bahia, but deforestation is imminent. The *chrysopygus* form exists only in two restricted populations in the state of São Paulo, and the *rosalia* group is now found only in remnant forests Northeast of Rio de Janeiro City, although records indicate a more extensive range for this group as recently as 30 years ago (Coimbra-Filho and Mittermeier, 1977).

We present here an estimate of the amount and type of biochemical genetic variation as determined by isozyme electrophoresis of blood cells from the three morphotypes. Based on analysis of 47 loci in 171 individual samples, we determined that allozyme genetic variation is limited in both captive and wild populations of lion tamarins. Furthermore, the genetic distance between the three morphotypes is comparable with isolated populations of mouse, humans, or other mammalian species studied using similar methods.

MATERIALS AND METHODS

Subjects and data collection

A total of 171 lion tamarins was examined in this study. The majority of these animals (n = 67) are *L. r. rosalia* from captive colonies throughout the U.S. Within this subset of the sample, 23 of the original 27 founders of the U.S. captive population are represented by at least one descendant. The four unrepresented founders were animals whose contribution to the current gene pool was less than 0.71% (Ballou, 1985). Blood samples from 63 free-ranging golden lion tamarins were collected from the Poço das Antas Reserve in the state of Rio de Janeiro. Ten samples came from captive golden lion tamarins housed at the Centro de Primatologia do Rio de Janeiro (CPRJ-FEEMA) and consist of wild-born animals and first-generation offspring. Pedigrees for the U.S. and FEEMA-CPRJ population are well documented (Ballou, 1985). Samples from the eight *L. r. chrysomelas* and 16 *L. r. chrysopygus* are also from the captive colony at CPRJ-FEEMA. Samples from seven free-ranging black lion tamarins were collected by Claudio Padua in the Morro do Diabo and Caitetus reserves in the state of São Paulo.

Approximately 2 cc of whole blood was collected aseptically from the femoral vein of 154 animals using heparinized 3-cc syringes. Blood samples were prepared by standard techniques (O'Brien, 1980; O'Brien et al., 1980b) and stored at −70°C until electrophoresis. Blood and whole organs (liver, kidney, and spleen) were collected from 17 animals who had died from a variety of causes.

Electrophoresis

The electrophoretic conditions for the 47 enzymes and nonenzyme proteins examined in this survey are presented in Table 1. Histochemical stains used to resolve specific proteins have been described previously (O'Brien, 1980; O'Brien et al., 1980b; Harris and Hopkinson, 1976; Siciliano and Shaw, 1976). Albumin, transferrin, and adenosine phosphoribosyl transferase were resolved using 4.75% acrylamide gels on a Bio-Rad 220 gel system. The remaining enzymes were resolved using 12% starch gels (Electrostarch, Madison, WI) on a Buchler vertical electrophoresis system.

Genotypes were interpreted from electrophoretic phenotypes on the basis of mobility, number of bands, comparison with other mammalian electrophoretic studies, and subunit number of each enzyme (O'Brien et al., 1980a). For polymorphic loci, each animal's phenotype was compared to its pedigree (Ballou, 1985) to confirm Mendelian patterns of inheritance.

Data analysis

Several satistical parameters were used to determine the extent and character of genetic variation in lion tamarins. These are the proportion of polymorphic loci and average heterozygosity per locus per individual (Nei, 1975). Nei's (1972, 1978) genetic iden-

TABLE 1. Loci examined by electrophoresis in Leontopithecus

Enzyme	Gene symbol	IUB/IUPAC No.	Tissue[1]	Buffer system[2]
Acid phosphatase 1	ACP1	3.1.3.2	RBC, K	TC
Adenine phosphoribosyl transferase	APRT	2.4.2.7	RBC	TG
Adenosine deaminase	ADA	3.5.4.4	RBC, K	TC + TEB
Adenylate kinase	AK1	2.7.4.3	RBC, K	TC
Albumin	ALB		Plasma	TG
Aldolase A	ALDA	4.1.2.13	RBC, K	TEB, TC
Carbonic anhydrase 2	CA2	4.2.1.1	RBC, K	TEB
Catalase	CAT	1.11.1.6	RBC	TEB
Creatine phosphatase	CPKB	2.7.3.2	RBC, K	TEB
Diaphorase-1	DIA1	1.6.*.*	RBC, K	TEB
Diaphorase-4	DIA4	1.6.*.*	RBC, K	TEB
Esterase-1	ES 1	3.1.1.1	RBC, K	TC, TEB
Esterase-2	ES 2	3.1.1.1	RBC, K	TC, TEB
Esterase-3	ES 3	3.1.1.1	RBC, K	TC, TEB
α-1-Fucosidase	FUCA	3.2.1.51	K	TEB
α-Galactosidase	GALA	3.2.1.22	K	TEB
β-Galactosidase	GALB	3.2.1.22	K	TEB
Glucose-6-phosphate dehydrogenase	G6PD	1.1.1.49	RBC, K	TEB
Glucose phosphate isomerase	GPI	5.3.1.9	RBC	TEB, TC
Glutamate oxaloacetate transaminase	GOT1	2.6.1.1	RBC	TEB
Glutamate pyruvate transaminase	GPT	2.6.1.2	RBC	TEB, TC
β-Glucuronidase	Gus B	3.2.1.31	K	TEB
Glutathione reductase	GSR	1.6.4.2	K	TEB
Glyoxalase	GLO	4.4.1.5	RBC, K	TEB
Hemoglobin	Hb	—	RBC	TEB, TC
Hexokinase 1	HK1	2.7.1.1	RBC, K	TEB
Hexoaminidase A + B	HEX A, HEX B	3.2.1.30	K	TEB
Lactate dehydrogenase A	LDHA	1.1.1.27	RBC, K	TC
Lactate dehydrogenase B	LDHB	1.1.1.27	RBC, K	TC
Malate dehydrogenase 1	MDH1	1.1.1.37	RBC, K	TC
Malic enzyme I	MEI	1.1.1.40	K	TC
Mannose phosphate isomerase	MPI	5.3.1.8	K	TEB
Nucleoside phosphorylase	NP	2.4.2.1	RBC, K	TC
Peptidase A	PEPA	3.4.11	RBC, K	TEB
Peptidase B	PEPB	3.4.11	RBC, K	TEB
Peptidase C	PEPC	3.4.11	RBC, K	TEB
Peptidase D	PEPD	3.4.11	RBC, K	TEB
6-Phosphofructokinase	PFK	2.7.1.11	RBC	TEB
Phosphogluconate dehydrogenase	PGD	1.1.1.44	RBC, K	TC
Phosphoglyceromutase	PGAM	2.7.5.3	RBC, K	TC
Phosphoglucomutase 1	PGM1	2.7.5.1	RBC, K	TC
Phosphoglucomutase 2	PGM2	2.7.5.1	RBC, K	TC
Inorganic pyrophosphatase	PP	2.7.5.1	RBC, K	TC
Pyruvase kinase	PK	2.7.1.40	RBC, K	TEB
Superoxide dismutase I	SODI	1.15.11	RBC	TEB
Transferrin	TF	—	Plasma	TG
Triosephosphate isomerase	TPI	5.3.1.1	RBC, K	TEM

[1]RBC, red blood cells; K, kidney extract.
[2]Buffer systems are Tris citrate, pH 7.1 (TC); Tris borate EDTA, pH 8.6 (TEB); Tris-malaic, pH 7.4 (TEM); Tris glycine, pH 8.9 (TG).

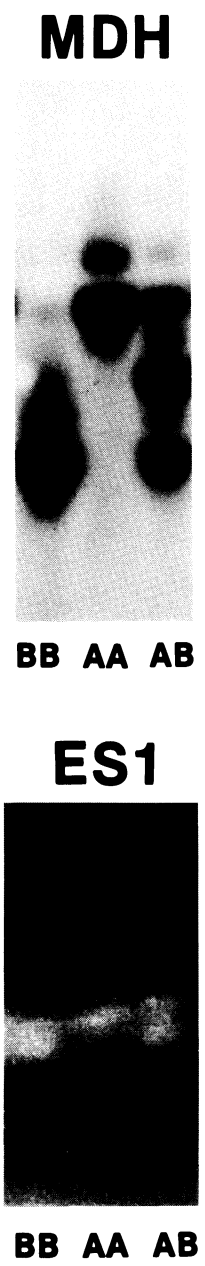

Fig. 2. Allelic isozyme phenotypes of polymorphic loci in *Leontopithecus*. A designates the most electropositive allele. MDH, malate dehydrogenase; ES1, esterase 1.

TABLE 2. Proportion of loci estimated to be polymorphic (P) and proportion of the genome estimated to be heterozygous ($\overline{H}$) in Leontopithecus

	No. Individuals	No. Loci	P (%)	$\overline{H}$
L. r. rosalia				
U.S. captive	67	47	4	0.01
Brazil	73	47	3	0.01
Total	140	47	3	0.01
L. r. chrysomelas	8	47	3	0.01
L. r. chrysopygus	23	47	3	0.003

tity and distance estimates were also calculated for each lion tamarin population.

RESULTS

An electrophoretic survey of 47 loci of *L. rosalia rosalia* revealed two polymorphic loci (*ES-1* and *MDH1*; Fig. 2) in a sample of 140 individuals (P = 0.04; average heterozygosity [$\overline{H}$] = 0.01, Table 2). These values are among the lowest reported to date for any primate population, captive or wild, examined by similar techniques (Fig. 3). Examining inbred and noninbred subpopulations indicated that no additional genetic variability was lost because of inbreeding within the sample. In fact, there was a slight excess of heterozygotes at the *MDH1* locus of the inbred group, although the trend was not significant and the allele frequencies did not deviate from Hardy-Weinberg expectations with 1 df (Table 3).

A second result of this survey was the striking similarity between the three geographically isolated groups. All animals were fixed for alleles of the same mobility at each of 45 monomorphic loci, regardless of their subspecific designation. Moreover, at least one allele was common to all three lion tamarins at the two polymorphic loci (Table 3). There were, however, discernible differences in frequencies of these alleles between *L. r. rosalia* and the other lion tamarins. Both of the polymorphic loci (*MDH* and *ES1*) varied in *L. r. rosalia*, whereas the *chrysomelas* and *chrysopygus* forms each showed variation at one locus only (*ES1*). The most common *MDH* allele in the *rosalia* group (the "a" allele) was absent in the other lion tamarins. *L. r. chrysomelas* was fixed for the *ES1* allele most rare in *L. r. rosalia*. The distinctions between *L. r. rosalia* and the other lion tamarins persisted in all subsets of the sample, i.e., in the

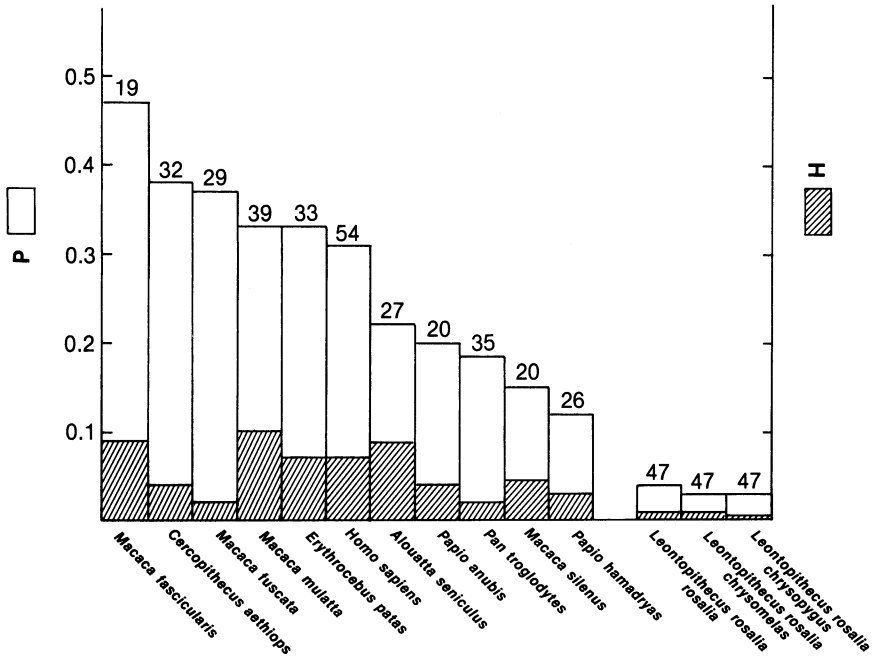

Fig. 3. Estimates of P (percent of tested loci that are polymorphic) and H (average heterozygosity) in primates. These estimtes are pooled from the electrophoretic literature on captive and free-ranging primates. *Macaca fascicularis*, *M. mulatta*, and *M. fuscata* are from Kawamoto et al. (1982), Nozawa et al. (1974), and Melnick et al. (1984). *Cercopithecus aethiops* is from Kawamoto et al. (1982), Lucotte et al. (1982), and Turner (1981). *Erythrocebus patas* is from Lucotte and Dandieu (1983) and Lewis (1984). *Homo sapiens* is from Harris and Hopkinson (1976), Harris (1966), and Nei and Roychoudhury (1982). *Alouatta seniculus* is from Pope (1983). *Papio anubis* and *P. hamadryas* are from Shotake et al. (1977) and Kawamoto et al. (1982). *Pan troglodytes* is from Bruce and Ayala (1978). *Macaca silenus* is from Jolly and King (1985). The numbers above each bar represent the number of loci included in this estimate.

U.S. and Brazilian colonies and in the wild-caught samples.

Both *MDH1* and *ES1* of *L. r. rosalia* were in conformance with the prediction of Mendelian transmission when subjected to pedigree analysis of captive animals. A χ^2 test for genetic equilibrium of allele frequencies for the three lion tamarins showed that *MDH1* was in equilibrium in each case (Table 3). *ES1* showed significant departure from the expectations of Hardy-Weinberg equilibrium in the captive U.S. populations and in the Brazilian population. This observation could be a consequence of the breeding structure of the populations resulting from captive management, although it might also be a sampling error because of the small number of individuals in certain phenotypic classes.

The genetic similarity between the three morphotypes of *Leontopithecus* was quantified using Nei's (1972, 1978) index of genetic distance, D, a statistic that measures the degree of allelic substitutions between populations based on electrophoretic mobility of several gene enzyme systems. Over the last decade, this metric has been applied to over 50 vertebrate genera (reviewed by Avise and Aquadro, 1981). The general, although not universal, observation is that the genetic distance between species or populations is roughly proportional to the time the taxa have been reproductively isolated. The distances between the three *Leontopithecus* groups (Table 4) is slight (0.007–0.03). This level of distance is comparable to that obtained from very recently isolated populations of mammal species and is actually less than distances derived between certain subspecies of mammals (Table 4; e.g., Bruce and Ayala, 1978; Nei, 1975; Rice and O'Brien, 1980).

TABLE 3. Genetic variation in Leontopithecus

	Allele frequency[1]					
	ES-1			MDH1		
	a	b	χ^2	a	b	χ^2
L. r. rosalia						
U.S. captive	0.05	0.95	6.28[2]	0.71	0.29	.04
Brazil (captive)	0.25	0.75	5.36[2]	0.90	0.10	.03
Brazil (wild)	0.0	1.0	—	0.98	0.02	.05
Total	0.04	0.96	51.61[2]	0.84	0.16	2.16
L. r. chrysomelas	0.50	0.50	2.0	0.0	1.0	—
L. r. chrysopygus						
Captive	0.95	0.05	0.003	0.0	1.0	—
Wild	1.0	0.0	—	0.0	1.0	—

[1]Allele frequencies: a is most electropositive allozyme.
[2]χ^2 revealed significant departure from expectations of Hardy-Weinberg equilibrium.

TABLE 4. Comparison of Nei-genetic distance within and between primates

	D	No. of Loci	Reference
Lion tamarins			
Leontopithecus rosalia rosalia—L. r. chrysomelas	.01	47	This study
L. r. rosalia—L. r. chrysopygus	.03	47	This study
L. r. chrysomelas—L. r. chrysopygus	.007	47	This study
Intraspecific comparisons			
Macaca mulatta (India)—*M. mulatta* (Thailand)	.007	29	Kawamoto et al. (1982)
M. fascicularis (Malaysia)—*M. fascicularis* (Indonesia)	.05	29	Kawamoto et al. (1982)
M. fuscata (troop YT)—*M. fuscata* (troop F)	.05	29	Nozawa et al. (1975)
Papio anubis (AK troop)—*P. anubis* (troop A)	.01	35	Shotake et al. (1977)
P. hamadryas (troop HBA)—*P. hamadryas* (Troop HBB)	.001	39	Shotake et al. (1977)
Pongo pygmaeus pygmaeus—P. p. abelii	.130	23	Bruce and Ayala (1978)
Homo sapiens (black)—*H. sapiens* (Asian)	.029	62	Nei and Roychoudhury (1982)
H. sapiens (black)—*H. sapiens* (white)	.027	62	Nei and Roychoudhury (1982)
H. sapiens (Asian)—*H. sapiens* (white)	.01	62	Nei and Roychoudhury (1982)
Interspecies comparisons			
Pan troglodytes—p. paniscus	.10	22	Bruce and Ayala (1978)
Hylobates lar—H. concolor	.13	21	Bruce and Ayala (1978)
Macaca mulatta—M. fascicularis	.10	29	Kawamoto et al. (1982)
M. mulatta—M. fuscata	.08	29	Kawamoto et al. (1982)
Papio anubis—P. hamadryas[1]	.02	29	Kawamoto et al. (1982)
Cercopithecus aethiops—C. sabaeus	.14	26	Lucotte et al. (1982)
C. sabaeus—C. pygerythrus	.13	26	Lucotte et al. (1982)
Intergeneric comparisons			
Homo sapiens—Pan troglodytes	.31	44	O'Brien et al. (1985)
Pan troglodytes—Gorilla gorilla	.25	44	O'Brien et al. (1985)
Gorilla gorilla—Pongo pygmaeus pygmaeus	.38	44	O'Brien et al. (1985)
Pongo pygmaeus pygmaeus—Hylobates lar	.54	21	Bruce and Ayala (1978)
Papio anubis—Cercopithecus aethiops	.88	29	Kawamoto et al (1982)
Cercopithecus aethiops—Macaca mulatta	.48	29	Kawamoto et al. (1982)
Other mammalian intraspecific comparisons: Socially subdivided populations			
Mus musculus	.02	46	Rice et al. (1980)
Swiss mice (three outbred laboratory colonies)	.02	46	Rice and O'Brien (1980)
Cynomys ludovidianus	.01	16	Chesser (1983)
Alces alces	.002	32	Ryman et al. (1980)

[1]Samples are from a hybrid zone in Ethiopia.

DISCUSSION

This survey detected a small amount of genetic variation (P = 0.04; $[\overline{H}]$ = 0.01) in the golden lion tamarin. Captive animals in both the U.S. and Brazilian colonies were more polymorphic and heterozygous than wild-caught animals (P = 0.03, $\overline{H}$] = 0.003). Variation was also reduced in samples from the golden headed and black lion tamarins ($[\overline{H}]$ = 0.01 and 0.003, respectively; P = 0.03 for both forms). Although the lion tamarins are not totally monomorphic, the level of genetic variability observed in this species is among the lowest reported for any primate, including those from other captive populations (Fig. 3). Even captive populations that are known to be severely inbred, such as the lion-tailed macaque (*Macaca silenus*; Jolly and King, 1985) have shown higher levels of P and $[\overline{H}]$ than the lion tamarins.

Pedigree analysis indicates that a moderate level of inbreeding has occurred among captive golden lion tamarins (mean inbreeding coefficient = 0.037; Ballou, 1985). The deleterious effects of inbreeding in zoo populations have been documented by Ralls et al. (1979), who demonstrated higher juvenile mortality in inbred offspring than noninbred offspring in several primates including golden lion tamarins (Ralls et al., 1979, 1980; Ballou and Ralls, 1982; Ralls and Ballou, 1982a,b). The incidence of infant mortality among captive inbred golden lion tamarins is 45% (Ballou, 1985), a high value and one that is consistent with the deleterious results of inbreeding. In contrast, the current infant mortality rates for noninbred animals is only 35% (Ballou, 1985). This figure is similar to infant mortality rates observed in other noninbred captive colonies of callitrichids, eg *Sanguinus oedipus* (Snowdon et al, 1985, S. Tardif, personal communication).

One goal of our research has been to develop sufficient markers for breeding management purposes. The evidence of inbreeding depression (Ralls and Ballou, 1982b) coupled with little isozyme polymorphism indicates that, although golden lion tamarins are not as genetically depauperate as, for example, the cheetah (O'Brien et al., 1983) or the northern elephant seal (Bonnell and Selander, 1974), the number of isozyme markers discovered here is limited. The low level of genetic variation observed in this sample of *Leontopithecus* underscores a critical problem in management of rare and endangered species: The establishment of a captive population is in itself a founder event, and, as such, the population is subject to potentially deleterious effects as a consequence of genetic drift. The situation is further confounded by the increased frequency of consanguineous matings that occur in captive situations.

This study represents the first electrophoretic survey of a callitrichid primate as well as the first survey of a primate whose mating system is monogamous and whose social organization is based on small family groups. Lifelong bonding of adult males and females reduces the number of allele combinations that would be available to animals whose mating system affords a larger mate selection throughout their reproductive years. It has been suggested that lower levels of genetic heterogeneity should be expected among pair-bonded animals, although individual demes should fix private alleles and become divergent from one another in this scheme (Bush, 1975; Wilson et al., 1975; Bush et al., 1977).

Levels of genetic variation are also complicated among the callitrichids by their propensity to produce twins. Although fraternal twins are usual in the few callitrichids examined thus far (in Hershkovitz, 1977), in the case of identical twins, only a single genotype is produced, increasing the level of genetic similarity among sibs and thus reducing the level of genetic variation in the demic pool. Further, the limited dispersal patterns of lion tamarins may increase the likelihood of consanguineous matings within an area, as has been observed in other free-ranging callitrichids (e.g., *Saguinus fuscicollis;* Terborgh and Goldizen, 1985).

Whether the low level of polymorphism and heterozygosity observed within lion tamarins is the result of founder effect or is a natural condition among wild lion tamarins or other callitrichids remains to be determined. Interestingly, the level of genetic distance between the three *Leontopithecus* mirrors genetic distance estimates of many avian congeners known to be good biological species (Avise and Aquadro, 1982). In addition to exhibiting similar degrees of genetic polymorphism, features of *Leontopithecus* mating strategy and patterns of ecological exploitation more closely parallel those of many birds than those of mammals.

The conventional taxonomic relationships within *Leontopithecus* have been based largely on morphologic considerations. Pelage coloration clearly demarcates the lion

tamarins into three forms (Coimbra-Filho and Mettermeier, 1972; Hershkovitz, 1977; Rosenberger and Coimbra-Filho, 1984). Additionally, the anatomically modified incisors and unique cranial shape of *L.r. chrysomelas*, the reduced premaxilla of *L. r. rosalia*, and the large body size of *L. r. chrysopygus* (Rosenberger and Coimbra-Filho, 1984) distinguish one form from another. Since these morphological characteristics may be important adaptations to microhabitat differences faced by each lion tamarin form, Rosenberger and Coimbra-Filho (1984) advocate the division of *Leontopithecus* into three separate species.

The derived genetic distances presented here suggest that the three forms of *Leontopithecus* are as similar biochemically as very briefly isolated populations of the same subspecies (Table 4). The genetic distance values between lion tamarins are less than those reported for many socially subdivided mammalian populations of the same species and for three human racial groups (Chakraborty et al., 1978; Nei and Roychoudhury, 1982; Table 4). Based on these distance estimates, Nei and Roychoudhury (1982) argue against even subspecific designation and emphasize that ". . . it is not appropriate to assign the rank of subspecies to the major races of man." Thus, when electrophoretic distance values for other polytypic groups of mammals are considered, distinct species designation for *Leontopithecus* is not supported, and even separate subspecies status appears unwarranted. We believe, however, that despite the low genetic distance values between the three lion tamarins, sound argument can be marshalled for the maintenance of the three subspecies.

When new species or subspecies arise in groups with low levels of genetic variability, each new group is expected to differ by fewer loci than more polymorphic species, such as man. Since none of the lion tamarins approach mammalian average levels of polymorphism and heterozygosity, comparing them to more polymorphic mammals with very different life histories obscures the importance of allelic differentiation between *Leontopithecus* morphotypes. The differentiation of alternate alleles at both polymorphic loci coupled with the morphological evidence suggests strong subspecific designation for *Leontopithecus*. We emphasize that this conclusion is consistent with a conservation strategy that monitors these three forms as distinct subspecific entities, since the biology of these callitrichids undoubtedly influences the observed levels of genetic variation.

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