

ASIATIC LION

Panthera leo persica



Population & Habitat Viability Assessment
P.H.V.A. and Global Animal Survival Plan Workshops
18-21 October 1993, Baroda, India

Report

May 1995

N.V.K. Ashraf, R. Chellam,
S. Molur, D. Sharma and S. Walker, Editors

A Collaborative Workshop

Municiple Corporation of Baroda/Sayyaji Baug Zoo
Forest Department of Gujarat
Zoo Outreach Organisation / CBSG, India
Wildlife Institute of India
North American Asiatic Lion Species Survival Plan
American Zoo Association Conservation Endowment Fund
Purina Big Cat Survival Fund



भारतीय वन्यजीव संस्थान
Wildlife Institute of India

CONTENTS

I. Overview

Executive Summary
Extract of Important Recommendations

II. Working Group Reports

Wild Population

- Habitat (Gujarat, Madhya Pradesh and Rajasthan sub-groups)
- Prey-Base Requirements
- Population Modelling
- Translocation
- Monitoring and Research
- Lion-Human Interactions

Captive Population

- Global Animal Survival Plan (GASP)

Management Science

- Diseases and Veterinary Research
- Reproductive and Genetics Research

Management of Human factor

- Ecodevelopment
- Public Education

III. Reference materials and appendices

List of participants and thanks

Appendices

- Report of Veterinary Workshops
- Relationship of Genetics and Reproduction Relative to the Management and Conservation of the Asiatic Lion, Dr. D.E. Wildt
- Central Zoo Authority Project with CCMB
- Meeting on Controlled Breeding of Lion, Central Zoo Authority
- Wildlife Institute of India note on translocation (from WII News)
- Proposed IUCN Resolution Statement on Animal Genome Resource Banking for Species Conservation
- GRB Guidelines and Protocol

P.H.V.A. Workshop for Asiatic lion

Panthera leo persica

18-21 October 1993, Baroda

Report

May 1995

Section I

Introductory material

P.H.V.A. Workshop for Asiatic lion

Panthera leo persica

18-21 October 1993, Baroda

Report

May 1995

Section II

Working Group Reports

Executive Summary

The Asiatic lion is a large, predatory carnivore which used to range over much of the Indian subcontinent and surrounding area. It is an animal whose size, strength and nobility have earned it identification with emperors and kings. It is an important cultural and historical symbol for India, having been selected as the emblem of the Government of India. The present status of the species is that it survives as a solitary, relatively small population of around 300 animals in a single relatively small area of 1400 square km. which is intensively managed. The behavioral and biological characteristics of the animal are such that it requires a large area to permit normal social interaction with its conspecifics as well as containment in a protected area away from human habitation.

The Asiatic lion has been of concern for many years as the population is said to have diminished to a scant twenty to one hundred or so animals. Previously, reintroduction and translocation efforts had been undertaken to try and establish another population but these efforts were not successful, due to lack of proper planning and methodology. Recent research has underscored the speculation that even the wild population may be suffering from inbreeding depression.

The Wildlife Institute of India and the Government of India have supported research to study the ecology of the Gir population and take up the matter of finding an alternative habitat for Asiatic Lion. One of the major tasks of this Workshop being to pursue this initiative, the suitability of several sites for lion translocation were assessed on the basis of prey population and other habitat factors. They were ranked as follows in order of suitability: Kuno, Sitamata, Darrah-Jawahar Sagar, Kumbalgarh, Barda.

The Asiatic Lion Population and Habitat Viability Assessment workshop was conducted with different working groups in parallel sessions with plenary sessions for presentation, review, and integration of the individual reports. Written draft reports prepared by all the working groups form the body of this report.

Individual working groups were:

1. Habitat (Gujarat, Madhya Pradesh and Rajasthan sub-groups)
2. Prey-Base Requirements
3. Population Modelling
4. Translocation
5. Monitoring and Research
6. Lion-Human Interactions
7. Captive Population
8. Diseases and Veterinary Research
9. Reproductive and Genetics Research
10. Ecodevelopment
11. Public Education

The three Habitat Sub-groups assessed the suitability of the following sites: Barda (Gujarat), Sitamata, Darrah-Jawahar Sagar, Kumbalgarh (Rajasthan), and Kuno

(Madhya Pradesh); for lion translocation using 11 parameters. The sites were assessed on the basis of prey populations (both including and excluding livestock) by the **Prey Base Requirements Group**.

The proposed translocation sites were ranked as follows for suitability as a habitat for lions, based upon a synthesis of the results of the above working groups:

1. Kuno
2. Sitamata
3. Darrah-Jawahar Sagar
4. Kumbalgarh
5. Barda

The **Populations Modelling Group** confirmed through computer simulations that growth rates and probability of extinction of the Gir lion population are critically linked to age of female first reproduction and first year mortality rates which are strongly influenced by habitat and prey availability. In addition the viability of the population depends on the carrying capacity of the Gir Forest stabilising at a range between 200-250 animals. It was further established that significant reductions and changes in the size and structure of the population due to catastrophic disease-event would be devastating. The modelling exercise provided statistical indicators that establishment of a second population will reduce the risk of extinction of the Asiatic lion significantly. It could be demonstrated also by the modelling exercise that the existing population will not be harmed by the removal of sufficient animals to translocate to the alternative site.

The **Translocation Group** has delineated a methodology to be followed for release of the lions, according to Guidelines of the IUCN/SSC Reintroduction Specialist Group. This includes genetic and demographic selection of stock, veterinary screening, research, monitoring, training and education-ecodevelopmental activity for Pre-translocation phase, Planning, preparation and release phase, and Post-release phase.

The **Monitoring and Research Group** has stressed the need for constant monitoring of the lion population in Gir and also at the site(s) where lions are to be translocated. Maintaining long-term records and intensively studying the numerous lion groups using radio telemetry are recommended. The Workshop participants as a group felt the need for a continuous research program with a permanent research base in Gir. Research on prey species, other carnivores like leopard and striped hyena, and animal-habitat relationships have also been recommended.

The **Lion-Human Interactions Group** considered all the possible types of interaction between lions and human beings, (including the *Maldharis* and villagers who live inside the protected area as well as villagers in the border area outside the p.a.) and analyzed the circumstances and consequences of such interactions. The need for population management of lions outside the Gir forest has been stressed, as well as relocation of the *Maldharis* from the Gir protected area. Recent studies of case-by-case physical interaction underscore the need for an innovative education and awareness program for the people living adjacent to the forest boundary.

The **Captive Population Group** has very clearly outlined the objectives of the captive breeding program and also has fixed a regional limit on the number of lions that can be held in captivity. Maintaining the purity of the stock of Asiatic lions and retaining the maximum amount of genetic diversity are amongst the major goals identified. This would involve facilitating the integration of wild caught problem lions outside the Gir Forest in captive breeding programs. All lions would have transponders implanted in them to enable definite identification. A detailed protocol for breeding and husbandry of the lions has also been provided.

The **Diseases and Veterinary Research Group** has compiled a comprehensive list of all diseases reported from captive and free ranging lions and identified areas which need research attention. Some of the surveys for diseases and parasites assume great importance and immediate relevance as these are currently the major threats facing the lions in Gir forest.

The **Reproductive and Genetics Research Group** has summarized the information available on the reproductive biology of Asiatic lions and identified subjects that need research attention. Detailed outlines and justification have been provided for the proposed research. Emphasis has been placed on the need for setting up a Genome Resource Bank (GRB) within India for insuring the preservation of genetic diversity. Developing artificial insemination techniques as part of the suggested research programs was also suggested. The group highlighted the urgent need to systematically sample the free-ranging lion population to assess the genetic diversity in the population. A few genetic management strategies are suggested, especially for the captive population.

The **Ecodevelopment Group** looked at a wide variety of possible initiatives which could give a better life for the people living in and around the Gir forest and at the same time reduce their dependence on the natural resources of this tract. These included grass fodder development, soil-moisture conservation measures, energy-related activities, employment generation activities, regular program of immunization of livestock, provision of separate water troughs for livestock, relocation of *Maldharis*, and eco-tourism.

The **Public Education Group** stressed the need to educate the population at large on general conservation values and of endangered species in particular. Various strategies have been outlined to achieve this, such as educating village leaders, recognition, motivation and training of individuals and organisations presently doing effective awareness work, and identification of most effective media for imparting nature education to different target groups in order to provide them with attractive and accurate baseline information.

The overwhelming consensus of the Workshop was that an alternative habitat for the Asiatic lion must be established with all possible speed, but without compromise of the accepted strategies and principles governing systematic and scientific reintroduction. This should be done simultaneously with strengthening effective protection and management of the Gir Forest and assuring the viability of the captive population and alternative genetic resources.

SUMMARY OF RECOMMENDATIONS

HABITAT MANAGEMENT

Gir habitat: Gir habitat management includes following forestry measures. It was recommended that afforestation programmes in grasslands and savanna areas in Gir should be avoided in future. It was also recommended that the sub-climax stage of vegetation (highly suitable for lion habitat) should be retained by opening the canopy and thinning of teak stands. Lopping and pollarding of coppicing species may be done so as to lower the browse level for ungulates (after the resettlement of remaining *Maldharis*).

Gir habitat may be increased where there is scope for doing so. Corridors to Babra Vidi, Mitiyala, Ambardi Reserve Forest, Malanka Vidi, Kannada hill, Ghatvad, and Girnar may be developed and these areas should be assigned special status. The adjoining Chachai-Pania sanctuary (38.9 sq.km) may be considered as part of Gir and more attention given to it for prey base and water resources development.

Gir habitat may be further protected from disturbance by measure to control traffic in the sanctuary. It is recommended that roads in Gir should never be tarred to contain heavy flow of traffic. Meter gauge tracks for railways should be maintained instead of going for broad gauge and trains should not be allowed to pass through the protected area after sunset.

Water management. Intensive water hole management should be done for even distribution of water (one water hole per 5 sq. km) and for increasing preybase. Whatever precipitation that falls in the area should be harnessed against days of drought and ground water resource should be more extensively developed. For example, when repairing roads, soil should be dug out by making rectangular pits in level lands along the road which will retain washed out soil and allows part of rain water to percolate into the ground. Water quality should be monitored during drought conditions.

Alternative habitat: Habitat Synthesis: An assessment of proposed translocation sites for the Asiatic lion indicated the suitability of the proposed sites for lion translocation was ranked in the following order 1. Kuno, 2. Sitamata, 3. Darrah, 4. Kumbalgarh, 5. Barda

PREY – BASE REQUIREMENT

The Prey base Working group suggested different estimates according to biomass versus prey numbers, and including or excluding lion preferences for each species of wild ungulate.

Scenario I: 70% wild prey and 30% livestock

Scenario II: 100% wild prey

Scenario III: Prey consumption solely as a function of prey availability.

The lions are assumed to show no preference for wild prey over livestock, and to have equal access to both categories of prey. This analysis suggests that several sites might support viable lion population and it might be desirable to consider more than one site

for translocation. Translocated populations would not necessarily be allowed to grow open ended but regulated by active population management.

POPULATION MODELLING

The growth rates and probability of extinction of the Gir Forest lion population are most sensitive to the age of female first reproduction and first year mortality rates. Both of these parameters are strongly influenced by nutrition and population density as it affects conflicts which result in the deaths of cubs.

If the carrying capacity of the Gir Forest is 200 or less the probability of extinction of the population will increase significantly. The range of 200-250 appears to be important for the viability of this population. Thus habitat availability and nutrition are close to critical values for this population. This interpretation is reinforced by the observation that prides of lions are moving outside of the Park.

The population is vulnerable to a catastrophic disease event such as has recently occurred in Africa. This would put the population to an increased risk of extinction as a result of normal environmental variation. Also another reduction in population size would lead to more rapid inbreeding. Lions appear vulnerable to inbreeding depression as reflected in measures of sperm characteristic and perhaps cub mortality. It will be valuable to collect blood samples from all animals removed from the Gir Forest for serological studies as a basis to monitor possible introduced diseases. The risk of extinction of the Asiatic lion will be significantly reduced with the establishment of a second viable wild population. The presence of a captive population provides additional insurance until the second wild population is established.

This population can easily sustain removal of sufficient adult animals for translocation to another site to start another population. Removal of younger animals or cubs would have even less effect on the viability of the population. Translocation projects can be designed in terms of the behavioral and genetic requirements. It is likely that removed animals would be rapidly replaced in the population from natural recruitment. Given the baseline conditions that have been explored in these simulation models, further modelling can be done to test ideas about the best strategy for a successful translocation with minimum or no effects on the Gir Forest population.

TRANSLOCATION

The Translocation Working Group followed guidelines of IUCN/SSC Reintroduction Specialist Group and suggested 1) a pre-translocation phase with feasibility studies and background research, identification of suitable release stock including appropriate genetic assessment and due consideration to the human element; 2) a planning, preparation and release phase should first insure governmental and funding approval and establishment of well-defined institutional support after which an actual release strategy with strategic measures regarding transport quarantine, etc may be made; 3) a Post release phase for monitoring, demographic, ecological, genetic and behavioral study of the released stock, and periodical review of the project.

MONITORING AND RESEARCH

Monitoring the size and structure of the free-ranging lion population(s) is essential to understand their population dynamics. Monitoring should include the techniques of Individual recognition, Radio telemetry; Cross-sectional censuses. Research should consist of basic demographics parameters, Social organization and dispersal patterns, monitoring of lion diet through collection of scat, of prey populations, of leopards and striped hyenas, and of vegetation.

LION-HUMAN INTERACTIONS

The Lion-human interactions working group recommended lion population management outside the park by appointing village wildlife watchers, relocation of *Maldharis* from Gir p.a.; implementation of the recommendation from studies carried out over the years (e.g. Central Committee on Tourism in Protected Areas, 1984 and Experts Committee, appointed by Government of Gujarat, 1990-1993)

CAPTIVE MANAGEMENT

A genetically pure, healthy captive population of between 400 to 600 animals should be developed taking care to provide for the genetic diversity and demographic stability for the long term.

REPRODUCTIVE AND GENETICS RESEARCH

For *ex situ* (zoo) breeding programs, assisted reproduction (techniques like artificial insemination) should be developed for overcoming problems associated with sexual incompatibility, cases of organic infertility and aged or under-represented founders unable to contribute to species preservation. This will also be useful for implementing the controlled breeding strategy with fewer complications such as transport of large animals.

Genetic Resource Banks (GRBs), repositories containing germplasm, blood product, tissues and DNA from selected, free living individuals to provide insurance against future human-induced or natural catastrophes, allowing the interactive movement of biological materials between living populations should be started in a systematic fashion in the range country, that is, India.

VETERINARY RESEARCH

It is recommended to initiate veterinary research on captive as well as wild populations of the endangered Asiatic lion. A full time researcher with veterinary background should be employed on this long term project. A complete disease diagnostic laboratory at Sakkarbaug Zoo which is located about 60 km away from Gir should be established as well as small laboratory facility at Sasan itself, primarily for preserving collected specimen. Research topics for wild and captive animals were identified by the workshop group.

Free ranging population at Gir: 1. Investigation on the prevalence of macroparasites; 2. Investigation on the prevalence of antibodies against specific microbial infections; 3. Research on posterior weakness.

Captive population: 1. Establishing the normal physiological values of the Asiatic lion; 2. Research on the probable causes for juvenile mortality; 3. Investigation of epidemiology and therapy of myiasis (maggot infestation)

ECODEVELOPMENT

The following items for providing for the local communities have been proposed; Grass fodder development, soil-moisture conservation measures, energy-related activities, employment generation activities, regular program of immunization of livestock in and around Gir, provision of separate water troughs for domestic livestock, relocation of *Maldharis*, community development facilities, eco-tourism, local NGO's, research monitoring and evaluation.

PUBLIC EDUCATION

Recommendations for nature education program included creating awareness in the local community of the value of the biodiversity in Gir forests, its benefits and the dangers of losing it. It was also recommended to educate the village leaders to organize a "Forest Protection Team" which can be utilised for solving some of the conservation problems of Gir. Recognition of the persons and groups currently working effectively will motivate them further and among them some identified persons may be provided training opportunities in education and interpretation. The importance of using appropriate methodologies according to the season in conducting nature activities was stressed. Identification of the most appropriate press media for educating different target groups and providing them with well-thought out material should be done. Creating ecological awareness and improving attitudes towards environment and conservation can be done with active participation from the community, including community leaders, school students, local industrial houses, religious and political leaders and other association and staff desirable for achieving educational goals.

HABITAT WORKING GROUP

Working Group members: Pushp Kumar (Facilitator), S. Chavan, N.S. Bundela, M.K. Misra, V.K. Sinha, C.D. Patel, J.P. Vasava, R.S. Shekhawat, V.M. Parasharya, V. Bhuva. N. Shah, Diwakar Sharma

The habitat group was divided into three subgroups – Gujarat, Rajasthan and Madhya Pradesh.

It was decided to tabulate the important habitat parameters and compare these for the five proposed sites in the three states. Gir Wildlife Sanctuary and National Park (the current home) was taken as a representative habitat and the highest rank was given to it. This is with the exception of the disturbance factor which was quite high here and – as a result – received only a low score of four.

The ranking given to the proposed areas represents the consensus reached by all members of the habitat group. The intention is to help the persons surveying these sites.

Table 1: Comparison of habitat parameters among the proposed sites for lion reintroduction.						
Habitat Parameters	Gir	Barda	Kuno	Sitamata	Kumbhal	Darraharh
Area & Shape	10	3	7.5	8.5	2	5
Climate	10	2	6	7	6	6
Temperature	10	9	6	6.5	6	6
Terrain	10	1	8	9	2	9
Water	10	5	7	6	2	7
Vegetation	10	2	7	8	7	7
Canopy Cover	10	3	8	6	7	7
Shrub Forest	10	2	7	4	7	7
Riverine Forest	10	1	6	4	3	3
Disturbance	04	1	6	4	3	3
Prey base	10	1	9	3	4	3
Total	104	30	77.5	66	49	63

HABITAT SUB-GROUP FOR GUJARAT

Members : S.A. Chavan, Dr. C.D. Patel, J.P. Vasava, R.V. Asari, Shivbhadrasinhji, B.M. Parasharya, R. Tiruvengadam, Pushp Kumar, B.J. Pathak, Nita Shah, Diwakar Sharma

Gir Sanctuary

Total Area: 1412.13 sq.km

Geology: Deccan Trap basalt; limestone, alluvium soil

Soils: Red, brown and black cotton; loam, silty-loam, sandy-loam

Climate: Semi-arid

Temperature: Min 5⁰ C and Max 47⁰ C

Rainfall: Average annual rainfall is 900 mm. It is about 1000 mm at Sasan in west and 650 mm at Jasadhar in east Gir. Rainfall is very variable with droughts occurring at least once in 4 – 5 years. Cyclones rarely affect this tract.

Topography: Gently undulating to hilly with a few extensive plains.

Vegetation: Western Gir broadly has dry deciduous teak, mixed deciduous teak, thorn scrub forests. Eastern Gir has savannah, thorn scrub, mixed deciduous forests. Canopy is more closed in western Gir compared to eastern Gir with teak and mixed deciduous forest.

Water status: Six principal rivers that flow are Hiren, Shingawada, Machundri, Jatardi, Adak, Raval. Besides these, several other streams drain the area. General drainage is towards west, southwest, and south.

Reservoirs: Four reservoirs within the sanctuary, Hiren, Shingawada, Machundri, and Raval.

Forage: Leaves of trees like *Acacia* spp, *Terminalia crenulata*, *Diospyros melanoxylon*, *Zizyphus mauritiana*, *Bauhinia racemosa*, *Ficus* spp., *Mitragyna parviflora*, etc. and herbs, shrubs like *Helicteres isora*, *Desmodium*, *Tephrosia*, *Achyranthes aspera*, etc. Main palatable grasses include *Apluda mutica*, *Themeda quadrivalvis*, *Sahema nervosum* and *Dicanthium annulatum*.

Cover: Most important plant species providing cover to lions and wild ungulates are *Carissa congesta*. Besides this *Ficus bengalensis* gives shelter to most of the carnivores, herbivores and birds.

Wildlife Population (1990):

Lion	284
Chital	27600
Sambar	1764
Nilgai	1524
Wildpig	2200

Threats

Grazing: At present there are 14,000 livestock distributed in about 60 Nesses in which about 360 *Maldhari* families reside. In addition to this, livestock from villages adjacent to the forest come in for grazing. At present, people are allowed to collect grass from the peripheral areas.

Fire (Mainly ground fires): Incidence more severe in central Gir. Main causes of fire are negligence by fire line workers, M.F.P. and firewood collectors and intentional kindling of fire.

Minor Forest Products: People collect M.F.P., firewood and grass. Somnath and Satadhar temple authorities collect billi (*Aegle marmelos*) leaves.

Poaching: This is insignificant because of extensive wireless network and protection.

Encroachment: In the peripheral areas of Gir, proper boundary demarcation is poor which encourage intentional encroachment.

Corridors: Corridors which were previously existing have been lost due to cultivation and human habitation

Roads: Five major roads pass through Gir including two state highways through central Gir.

Railways: Meter-gauge railway passes through western Gir. Since the train is pulled by steam engine the lighted coal causes occasional fires. In addition, every year two to three instances of lion or other wildlife getting run over by trains are recorded.

Temples: Temples located in Gir are near important water sources. These temples attract hordes of devotees and tourists, causing severe disturbances to the wildlife. The tourists dispose their garbage into the streams and carry out rituals which spoil the aesthetic value of the area, while depriving the wildlife from using the water source.

Barda Sanctuary

Total area:	192 sq.km.
Geology:	Deccan Trap basalt and associates
Soil:	Black cotton soil and in patches rocky areas devoid of topsoil
Topography:	Hilly
Climate:	Semi-arid
Temperatures:	10°C min. 40°C max.
Rainfall:	125 mm – 250 mm

Vegetation: Acacia, bamboo in patches, *Manilkara hexandra*, *Ixora arborea*, *Diospyros melanoxylon*, *Zizyphus*, *Lantana*, etc.

Water sources: No major river but both natural and artificial old tanks present; three reservoirs on the periphery outside the sanctuary meant to provide drinking water to Porbandar, Bhanvad and Ranavar. Drought and scarcity of water is a major problem.

Forage: Availability is very poor as *Lantana* occupies most of the areas.

Threats:

Grazing: There are 60 nesses with 600 *Maldhari* families and 4000 to 5000 cattle. As there is very poor grass growth, *Maldharis* lop trees to feed their cattle. At the beginning of the century 110 tree species were reported by Mr. Jay Krishna Indrajai but now, because of overgrazing, drought and other biotic pressures, almost 50% of tree species have become locally extinct.

Fire: With the poor grass growth fire incidence is negligible. M.F.P. collection is negligible but collect *rayan* (*Manilkara hexandra*)

Firewood: Many illegal distilleries producing country liquor collect firewood with the help of the *Maldharis*.

Poaching: Very depleted prey base and hence poaching is negligible.

Encroachment: Cement industries on the periphery encourage encroachment and increased biotic pressure by mining, and human and vehicular movement.

Corridors: Previously, Barda was well connected with Alech Hills allowing free movement of lions into Girnar and Gir. Now the corridor is highly fragmented due to roads, agricultural fields, and habitations. This situation rules out scope for corridor development.

Temple: The Kileshwar Temple though within the Sanctuary does not attract a large number of devotees.

Recommendations for Gir Habitat Management: (Gujarat sub-group)

1. Avoid afforestation programmes in grasslands and savanna areas in Gir in future.
2. Attempt should be made to acquire additional areas of Tulsishyam trust within the Protected Area.
3. Roads in Gir should never be tarred to contain heavy flow of traffic. Road side slopes should be stabilized by a cover of fast growing floral species. For road repairing, soil should be dug out by making rectangular pits in level lands along the road. Such pits will retain washed out soil and allow part of rain water to percolate into the ground.
4. The present status of railway as a meter gauge track should be maintained. No train should be allowed to pass through Gir after sun set.
5. Dry rubble wall around Gir should be maintained.
6. A more vigilant and expanded fire-watch squad with extensive communication network is essential.
7. There is scope to increase lion habitat outside Gir sanctuary by developing corridors to Babra Vidi, Mitiyala, Ambardi Reserved Forest, Malanka Vidi, Kannada hill, Ghatvad, and Girnar. These areas should be assigned special status.
8. Resettlement of remaining *Maldhari* families on the fringe areas outside Gir in a phased manner.
9. Opening of canopy and thinning of teak stands to keep the vegetation in sub-climax stage which is suitable as lion habitat.
10. Lopping and pollarding of coppicing species after the removal of remaining *Maldharis*. This will lower the browse level for the ungulates.
11. Intensive water hole management for even distribution of water and for increasing prey base. At least one water hole should be provided per 5 sq.km.
12. The problem of erratic rainfall can be tackled by harnessing whatever precipitation that falls in the area.
13. Barring a few dug wells and hand pumps, the ground water resources are largely under developed. A conjunctive use of surface and ground water will provide an

important means of water resource management.

14. Water quality should be monitored during drought conditions

15. The adjoining Chachai-Pania sanctuary (38.9 sq.km.) should be considered as part of Gir and more attention should be paid to it for prey base and water resources development.

HABITAT SUB-GROUP FOR MADHYA PRADESH

Members – N.S. Bundela, M.K. Mishra, V.K. Sinha and D. Sharma

Kuno Wildlife Sanctuary:

Area: 345 sq.kms

Proposed to add 265 sq.kms. Scope to extend to 2000 sq.kms. Corridor/further expansion has good possibilities because area is surrounded by similar type of forest in three directions (except east)

Geology: Vindhya hills – primarily sandstone tract which can support aquifers.

Soil: Alluvial along Kuno river, deep soil; sandy loam on slopes gully/ravine formation moderate to heavy approximately 10% of the area.

Topography: Plain area about 40% moderate slope about 30% steep slopes about 30%. Altitude range: 275-550 m

Climate: Semi-arid

Rainfall: Average 602.75 mm. Moderate drought once in every six years

Temperature: Max. 48°C; Min. 4°C

Water sources: Perennial water pools in river beds along 120-125 km. length. One perennial spring, 8 water pools outside river. 10 Artificial water holes. Water points are well distributed.

Vegetation: Mixed dry deciduous forest is composed of species like *Anogeissus pendula*, *Acacia catechu*, *Boswellia serrata*, *Sterculia urens*, *Lannea grandis*, *Zizyphus spp.* Teak is reported from 5-9 sq.km.

Canopy: *Dalbergia latifolia*, *Zizyphus mauritiana*, *Acacia leucophloea*, *Anogeissus latifolia*, *Bauhinia racemosa*, *Ficus spp.*, *Dendrocalamus spp.*, *Aegle marmelos*, *Emblica*, *Flacourtia*, *Bridelia*, *Diospyros*, *Lannea*.

Shrub: *Capparis*, *Zizyphus nummularia*, *Helicteres isora*.

Ground: Palatable grasses (1.5 – 2m tall), weed like *Cassia tora* and *Solanum spp.* are also found.

Riverine tract: *Terminalia arjuna*, *Syzigium cumini*, *Mitragyna*, *Ficus glomerata*, *Vitex negundo*. Thickets of *Vitex* and *Syzigium* are present. Patches of bamboo thickets are also found.

Threats: No road or railways in present sanctuary. One cave temple visited by about 1000 people once in a year for 15 days. No major or minor projects. One district road, one narrow gauge railway, and a canal (all 15 kms long) just outside the proposed area. No major or minor project has been proposed.

Biotic factors: 21 villages having 5000 people and 6500 livestock in present sanctuary. 30 villages with 8000 people and 10000 livestock in the proposed area. 90% of the populations are tribal. 25%-30% of the area is affected by fire annually. About 150 nomadic graziers are in the proposed area from August to November. No minor forest products are collected in the present sanctuary. Diospyros leaves, Emblica and Zizyphus fruits and some medicinal plants are collected for a limited period of 15 days in the present and proposed areas.

Human Population: Sparse density. Six villages are needed to be relocated outside immediately.

Wildlife population (1992):

Tiger	9
Leopard	10
Black buck	1400
Chinkara	1800
Chital	3500
Sambar	700
Nilgai	2000
Wild boar	2500
Langur	2000

HABITAT GROUP FOR MADHYA PRADESH – Recommendations:

1. Although the extent of the sanctuary at present is 345 sq.km. it can be extended to ~2000 sq.km. in case of lion introduction. An addition of 65sq.km. has already been proposed.
2. Run off rainwater is to be checked by ways of adopting soil and water conservation measures to maintain and improve eco-systems and control of ravine formation.
3. Water holes should be increased in number
4. Active patrolling to be introduced in present and proposed area.
5. Competition with tiger has to be studied along with the monitoring of introduced lions. In case of adverse interactions, the tiger could be removed.
6. People in the adjoining villages should be asked to surrender their fire arms and regulations regarding the fire arm license have to be strictly enforced.

Darrah-Jawahar Sagar Wildlife Sanctuary

Area: 500 sq.km. Buffer: 600 sq.kms in form of Bharsrodgarh WLS, and other areas.

Remarks:

1. Rehabilitation of Girdharpura, Lakshmipura, Darrah, Mashalpur and Chandbaori villages outside the proposed area.
2. Perennial source of water in form of Chambal River.
3. Corridor between Darrah WLS and Jawahar Sagar WLS will have to be developed and maintained
4. Likely resistance from the local Gujjars.
5. Low prey base; allow the population of sambar and chital to build up over a period of time and also by translocation some animals from other areas.
6. Tigers are not reported from the area.

Table 2. Comparison of habitat parameters among the proposed sites for lion reintroduction.

Habitat Parameters	Gir	Barda	Kuno	Sitamata	Kumbalgarh	Darrah
Area & Shape	10	3	7.5	8.5	2	5
Climate	10	2	6	7	6	6
Temperature	10	9	6	6.5	6	6
Terrain	10	1	8	9	2	9
Water	10	5	7	6	2	7
Vegetation	10	2	7	8	7	7
Canopy Cover	10	3	8	6	7	7
Shrub Forest	10	2	7	4	7	7
Riverine Forest	10	1	6	4	3	3
Disturbance	04	1	6	4	3	3
Prey base	10	1	9	3	4	3
Total	104	30	77.5	66	49	63

HABITAT SUBGROUP FOR RAJASTHAN

Members: Nita Shah, R. S. Shekhawat

[Please refer to the comparative table for habitat parameters]

Sitamata Wildlife Sanctuary

Area: 500 sq.km.; Buffer: 800 sq.km. of Pratapgadh forest division.

Remarks:

1. Extension of the sanctuary to the adjoining forest areas of Chitrimata, Rampura, Panchaguda and Umarkota forest blocks and identifying a viable area within the WLS and buffer zones for reintroduction of the Asiatic lion.
2. Rehabilitation of Arampura, Maeda, Dhar, Pal and Ambaretti villages outside the proposed zone.
3. Ensuring effective protection.
4. Need to build up the ungulate population of four-horned antelope, chinkara, sambar, chital and nilgai to improve the prey base.
5. Foresee no resistance from the local people.
6. No tiger reported from the area, no probability of competition with existing carnivores i.e., leopards, hyenas and jackals.

Kumbalgarh Wildlife Sanctuary

Area: 586 sq.kms. Buffer 475 sq.kms of Raoli Todgarh Sanctuary.

Remarks:

1. No perennial source of water in form of rivers.
2. Shape of the WLS is linear with only 5-15 kms. width.
3. Low prey base; need to build up the ungulate population; (sambar and chital) or translocate them from other areas.
4. The WLS is not a suitable alternate home for the lions for the reasons stated above.

Table 1. Comparison of Habitat Characteristics between Gir and the Proposed Sites for Lion Reintroduction

Habitat Parameters	Gir	Barda	Kuno	Sitamata	Kumbalgarh	Darrah-Jawahar Sagar
Locality Factor						
Area	1412 sq.km	190 sq.km	2000 sq.km. (345 sq.km now)	1300 sq.km (500 sq.km)	1060 sq.km (586 sq.km)	1100 sq.km (500 sq.km)
Geology	Deccan trap basalt	Deccan trap basalt	Vindhyan sandstone tract	Aravalli, granites	Aravalli sandstone	Vindhyan
Climate	900mm; 5 ⁰ -47 ⁰ C	200mm; 10 ⁰ -40 ⁰ C	602mm; 4 ⁰ -48 ⁰ C	650mm; 5 ⁰ -43 ⁰ C	600mm; 2 ⁰ -48 ⁰ C	600mm; 5 ⁰ -45 ⁰ C
Vegetation	Dry deciduous teak and <i>Anogeissus teak</i> , <i>Acacia catechu</i>	<i>Acacia</i> associations <i>Acacia nilotica</i> <i>Manilkara</i>	Dry deciduous <i>Anogeissus pendula</i> and <i>Acacia catechu</i> <i>Boswellia serrata</i> , <i>Zizyphus spp.</i> <i>Diospyros</i> , <i>Bauhinia</i>	Dry deciduous teak forest, teak – bamboo, <i>Lannea</i> <i>Diospyros</i> , <i>Anogeissus latifolia</i> , <i>Boswellia serrata</i>	Dry deciduous <i>Anogeissus pendula</i> <i>Acacia catechu</i> , <i>A. latifolia</i> , <i>Boswellia</i> <i>Lannea</i>	Dry deciduous <i>Anogeissus pendula</i> , <i>A. latifolia</i> , <i>A. catechu</i>
Shrub cover	Medium	Dense	Sparse	Dense	Sparse	Sparse
Ground cover	Dense, extensive	Poor growth	Dense good growth Extensive	Dense in 200 sq.km. area, restricted	Good growth Extensive	Good growth Extensive
Riverine Tract	Good riverine vegetation with 40% canopy cover. Long impenetrable patches of <i>Carissa</i> in shrub layer. <i>Syzygium</i> , <i>Pongamia</i> , <i>Holoptelia</i> is dominant species.	Small extent of thickets in patches	<i>Terminalia arjuna</i> , <i>Syzygium cumini</i> , <i>Mitragyna</i> , <i>Ficus</i> , <i>Carissa</i> , <i>Vitex negundo</i> Patches of bamboo	50km length <i>Syzygium</i> , <i>Terminalia</i> , <i>Mitragyna</i> Continuous thickets of cover	Fragmented patches of <i>Syzygium</i> , <i>Terminalia</i>	Similar to Kumbalgarh

Table 2: Comparison of habitat parameters among the proposed sites for lion reintroduction						
Habitat Parameters	Gir	Barda	Kuno	Sitama a	Kumbal garh	Darraharh
Area & Shape	10	3	7.5	8.5	2	5
Climate	10	2	6	7	6	6
Temperature	10	9	6	6.5	6	6
Terrain	10	1	8	9	2	9
Water	10	5	7	6	2	7
Vegetation	10	2	7	8	7	7
Canopy Cover	10	3	8	6	7	7
Shrub Forest	10	2	7	4	7	7
Riverine Forest	10	1	6	4	3	3
Disturbance	04	1	6	4	3	3
Prey base	10	1	9	3	4	3
Total	104	30	77.5	66	49	63

Comparison of habitat parameters among the proposed sites for lion reintroduction.

**Table 1. Prey Base – availability of prey under different scenarios
Scenario 1:70/30; Scenario 2: all wild; Scenario 3 prey availability**

Site	Biomass		Nos.		Biomass		Nos		Biomass		Nos.	
	PrPf	None	PrPf	None	PrPf	None	PrPf	None	PrPf	None	PrPf	None
Kuno	122	87	127	110	99	55	125	82	120	88	123	104
Darrah	201	160	273	164	15	6	15	9	223	182	291	300
Sitamata	203	162	272	162	19	7	15	7	224	183	290	299
Kumbalgarh	70	52	86	57	22	10	24	15	74	57	90	89

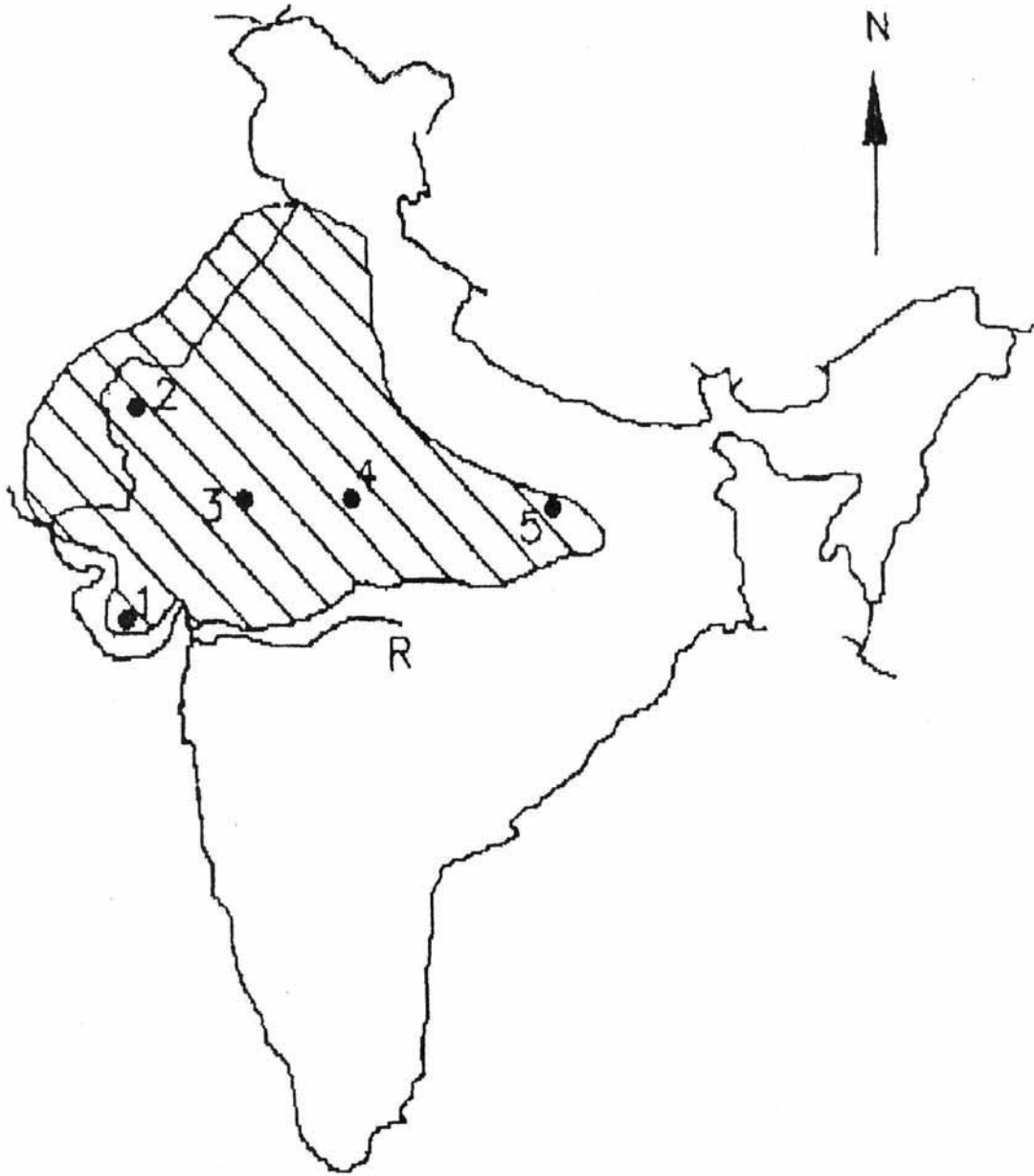
PrPf : lions show same preferences across wild prey species as Gir.

None: lions take wild prey in proportion to their abundance

Numbers: lion numbers depend on the number of prey animals in each site.

Biomass: lion numbers depend on the biomass of prey animals in each site.

Prey base report: Prey availability – three scenarios and their parameters



The Historic range of Asiatic lions in India.*

* This map was obtained from Dr. Ravi Chellam, Scientist, Wildlife Institute of India.

PREY – BASE REQUIREMENT

Members: V. Bhuva, S. Tikedar, C. Packer, Ravi Chellam and S. Chakraborty

An evaluation of the proposed translocation sites:

The Prey Base Working Group considered the data on ungulate populations in the Gir forest and compared it with the existing data for the 4 other sites. Available census figures were used for wild ungulates and livestock in each area and estimated the approximate carrying capacity for lions in each site by extrapolating from the Gir data. For each site 12 different estimates were calculated. These were based on three alternative scenarios according to different preferences of lions for livestock compared to wild ungulates. Within the scenario the group calculated four different estimates according to biomass versus prey numbers, and including or excluding lion preferences for each species of wild ungulate.

Scenario I: 70% wild prey and 30% livestock

The most recent data from Gir show that the lion diet is composed of 70% wild prey and 30% livestock. Since wild ungulates comprise only about 47% of the total prey animals, the Gir lions show a 1.25 times preference for wild ungulates over domestic stock. This scenario assumes that the lions in the new sites would have similar access to livestock and the same preference for wild ungulates as at Gir.

Scenario II: 100% wild prey

Here it has been assumed that lions gain no access to livestock. Thus their population numbers would depend entirely on the wild species. We assume here that with a similar restriction at Gir, the lion population would be reduced to 70% of its current total.

Scenario III: Prey consumption solely as a function of prey availability.

In this case, the lions are assumed to show no preference for wild prey over livestock, and to have equal access to both categories of prey.

Table I summarizes the results of these alternatives. It can be seen that for most sites, the estimated lion population sizes are very sensitive to assumptions concerning the utilization of livestock. At two sites, Darrah and Sitamata, the estimated population size varies from less than 10 to over 300 lions, depending on the access of lions to livestock.

Across scenarios, Kuno shows the lowest variation in lion population size (55 to 127) and the highest carrying capacity in a diet derived purely from wild ungulates (55 to 125). Across scenarios, the other 3 sites show considerable variation in their projected carrying capacity for lions [Darrah (6 to 300) Sitamata (7 to 299), Kumbalgarh (10 to 90)] and their carrying capacity is extremely low when projected for a diet derived purely from wild ungulates [Darrah (6 to 15), Sitamata (7 to 19), Kumbalgarh (10 to 24)].

Not much significance should be attached to the projected lion numbers because of the large number of assumptions and the rough nature of the census data. These numbers merely provide guidance to evaluate the relative suitability of each site. As best as we can guess at this stage, Kuno appears to be the most suitable site. This is largely due to the higher level of availability of wild ungulates.

Presently at Darrah and Sitamata, the wild ungulate density is very low, probably due to the high density of livestock. Hence there is a possibility of the wild ungulate population recovering if the livestock is excluded from these habitats. However, this recovery could take up to 20 years.

This analysis suggests that several sites might support viable lion population and it might be desirable to consider more than one site for translocation. Translocated populations would not necessarily be allowed to grow open ended but regulated by active population management. These populations would act as an insurance against the extinction of free ranging lions. To maximize genetic variation, the translocated populations should be part of a metapopulation and the gene flow should be maintained by periodic switching of males between the subpopulations.

See Table 1 (end of Report)

Habitat synthesis: An assessment of proposed translocation sites for the Asiatic lion:

As part of the Asiatic lion PHVA, working groups were constituted to assess the five proposed translocation sites on prey availability and various habitat parameters. This report attempts to summarize the findings of all these groups and to subjectively rate the suitability of the proposed sites for lion translocation. The sites considered were – 1. *Sitamata*; 2. *Kumbalgarh*; 3. *Darrah* (all in Rajasthan); 4. *Kuno* (in Madhya Pradesh); 5. *Barda* (Gujarat).

The findings of both the prey and habitat assessment groups are presented in detail elsewhere in this workshop report. It emerges that given the present knowledge and status of conservation of these sites we would rate the sites, in the following order of suitability for lion translocation.

1. Kuno
2. Sitamata
3. Darrah
4. Kumbalgarh
5. Barda

This group recognizes that an intensive field survey should be undertaken to validate these results.

Table 1. Prey Base – availability of prey under different scenarios Scenario 1:70/30; Scenario 2: all wild; Scenario 3 prey availability												
Site	Biomass		Nos.		Biomass		Nos		Biomass		Nos.	
	PrPf	None	PrPf	None	PrPf	None	PrPf	None	PrPf	None	PrPf	None
Kuno	122	87	127	110	99	55	125	82	120	88	123	104
Darrah	201	160	273	164	15	6	15	9	223	182	291	300
Sitamata	203	162	272	162	19	7	15	7	224	183	290	299
Kumbalgarh	70	52	86	57	22	10	24	15	74	57	90	89

PrPf : lions show same preferences across wild prey species as Gir.

None: lions take wild prey in proportion to their abundance

Numbers: lion numbers depend on the number of prey animals in each site.

Biomass: lion numbers depend on the biomass of prey animals in each site.

Prey base report: Prey availability – three scenarios and their parameters

Population modelling Report of the Working Group

Members: *U.S. Seal, S. Brewer, S. Molur, Ajith Kumar Bhowmik, S. Tikedar*

The simulation models were constructed with the DOS versions VORTEX 5.1 and 7.0 and run on IBM compatible machines with either a 486 or pentium processor and a math coprocessor. The software and a manual are available on request.

Age of first reproduction

The average age of adult lions at the birth of the first litter is dependent upon the population studied and whether the animals are captive or wild. Data from wild African lion populations and the captive Asiatic lions at Sakkarbaug Zoo supports 3 years as the average age for females and males for both subspecies. Reproduction may delay one year in the Gir forest lion population. The choice of age of first reproduction for females will affect generation time for females and the estimated population growth rate (r). Population scenarios were modeled for female ages of first reproduction set at either 3 or 4 years to evaluate the sensitivity of the results to this parameter.

The age of first successful breeding for males in the wild is usually later than the age of physiological capability which is about 3 years in captive lions. The age for wild male African lions is 5 years. The average age for Gir forest lions is uncertain but may be between 5 and 8 years. This choice will not affect population growth rate since lions are a polygynous species. This age will affect the generation time for males, the rate of loss of heterozygosity in the population, and the effective population size.

Sex ratio at birth

The only data available for the Asian lion are from captive births. The studbook records births of which were males and females. This yield a ratio of 0.38 (biased towards females) rather than the expected ratio of about 0.50 (equal numbers of males and females). We used the studbook data to set the ratio at 0.38 in the scenarios modeled.

Life expectancy

VORTEX models all living adult animals as potentially part of the breeding pool. The maximum life expectancy is about 15 years for wild lions. Animals may live longer in captivity and in the wild but this probably is maximum age of reproduction for animals in the wild population.

Litter sizes and proportion of females producing a litter

Asiatic lion litters have not been observed at birth in the wild. Data from captive lion births indicate a mean litter size of about 2.5 cubs at birth. Estimates of mean litter size at the time of observation in the wild, usually at 3-6 months of age, have varied from 1.5 to 3.5 cubs. Small prides with 1-2 adults' females usually have only one cub per litter surviving to 1 year of age. Every adult female Asiatic lion is thought to have a problem with survival of their litters because the

young cubs are vulnerable to being killed in social strife. About 60% of adult females do not produce a litter in a given year. Based upon captive data, litter sizes of living cubs are distributed from 1 to 5 in the following proportions: 0 = .60, 1 = .08, 2 = .15, 3 = .10, 4 = .60, and 5 = .01.

Mortality

Cub mortality was estimated in three ways. First, four litters from 4 wild females had 9 observed cubs (3-6 months of age) of which 3 died (33%) by one year. It is likely that additional mortality had occurred from birth to the age at which these cubs were observed. At a mean litter size of 2.5, four litters would have produced 10 cubs and the mortality from birth to one would then be 40%, second, at the Sakkarbaug Zoo there have been 74 deaths in 205 births (36%) by the age of one year. Third, it is estimated that there is differential survival of cubs in small (1-2 females) and large (3-5 adult females) prides. One cub on average will survive in the small pride and 2 will survive in the large pride. It is estimated that 3 of 4 prides in the Gir Forest are in the small category.

Adult mortality

Adult mortality was extrapolated from data on animals taken to the zoo for health reasons (putative wild deaths) and skins deposited at the zoo. From 1988 through September 1993 there were 60 animals removed representing an average of 10 per year. Given the census estimate of 284 lions in the Gir forest and a stable age distribution, 122 animals would be in the adult age classes. 10 deaths per year is 8% mortality. This was modelled at 10% to account for deaths in the wild that may not have been detected through these collections.

There was a general impression that sub-adult mortality was low but data are not available. Also, there were no data to separate mortality by sex. The following estimates were used in the models. Sensitivity analyses examined the effects of using the ages of 3 and 4 for the age of female first reproduction and 10 and 15% mortality for sub-adult mortality.

females		
	0-1	35%
	1-2	10%
	2-3	10%
	3-4	10%
adult		10%

males	0-1	35%
	1-2	10%
	2-3	10%
	3-4	10%
	4-5	10%
	5-6	10%
adult		10%

Catastrophes

There is at present no consensus regarding the potential occurrence or severity of Catastrophes. There does seem to be agreement that the most recent drought did not negatively affect the lions. We examined the effects of an event occurring with a 10% probability, a modest 10 increase in mortality, and either a 10% or 50% reduction in reproduction in that year. Recent disease epidemics in the Serengeti lion population has killed an estimated 1/3 to 1/2 of that population.

Initial population size

The officially conducted census estimates 284 animals. Models were also run at a lower population size of 200.

Capacity

Carrying capacity was modeled at 300 and at 250 animals.

Inbreeding

The inclusion of an inbreeding depression factor (heterosis, 3.14 lethal equivalents) has minimal effect on the population at this size. If conditions are such that it fluctuates in size or goes through a bottleneck, as happened early in the century then this factor would increase in importance. Molecular studies indicate that there is very little genetic heterozygosity in the Gir Forest population when compared to wild populations of the African lion.

Results of Modelling

Introduction

The scenarios developed for simulation modelling were intended to provide an overview of the population dynamics of the Gir Forest lion population under current management conditions and current estimates of population parameters, to provide estimates of the risk of extinction of the population over the next 100 years allowing for environmental variation and a possible rare catastrophe, to test the estimates of the population parameters for internal consistency, to test the effects of changes in management, to test possible consequences of removing animals from the population to establish a second population, and to assist in devising plans for the removal of animals for a second population while posing a minimum risk to the Gir population.

The interactions of short duration (6 animals every 3 years for 10 years) and long duration (every 3 years for 100 years) adult animal removal from the population (simulating the effects of removals to start a new population), a catastrophe, carrying capacity ($K=300, 200, \text{ or } 250$), first year mortality (year 0-1=35% or 50%), mortality from age 1-2 years (1-2 = 10% or 15%), and age of first reproduction (female AFR = 3 or 4 years; male AFR = 5 or 6 years) on population growth rate (r), probability of extinction in 100 years (P_e) and final mean population size (N) at 100 years were examined over the range of suggested possible values with the results presented in Tables 1-3 and Figures 1-21. The results of these scenarios provide insight into which population parameters may be most significant for the survival of the lion population.

The figures are arranged with 4 groups of 4 scenario results (a total of 16 scenarios summarized) in each figure. Each set of 4 results is for a combination of female and male Age of First Reproduction as indicated on the x-axis. Within each set of results the first pair compares the outcomes at 50 or 35% neonatal mortality for $K = 300$. The second pair makes the same comparison for $K = 200$. The tables are in sets of 4 (2 per page) comparing the interactions of effects of high (long duration) or low (short duration) harvest rates and of a high or low catastrophe in terms of effect on reproduction. The first four Tables (1-4) present the results of these interactions on population growth rate (r). The second set (Table 4-8) present the effects of the same sets of interactions on the probability of extinction, and the third set (Tables 9-12) the effects on mean population size of the surviving population at 100 years. All of the scenarios in the first set of 12 tables were with mortality at age 1-2 = 10%. The set of 6 Tables (Tables 13-18) were with 1-2 year mortality = 15% and comparing the effects of low and high catastrophe effects at a low harvest rate. The high harvest rate had already been shown to increase the P_e in many of the scenarios at 1-2 year mortality = 10% so was not examined further since the increase in 1-2 year mortality would only further increase the probability of extinction.

Age of first reproduction

The minimum condition for survival of a population is a positive growth rate of value of ' r '. However, populations with a positive ' r ' are still vulnerable to extinction under fluctuating environmental conditions. The stochastic population growth rate (r) provides an indication of whether the population may increase if positive or may decline and eventually become extinct. The age of first reproduction for male lions, whether 5 or 7 years, made no difference in the population growth rate under any of the conditions simulated (Figures 1-4). In contrast, increasing the age of first reproduction from 3 to 4 years in females resulted in about a 30% reduction in the population growth rate (Figures 1-4). Determination of this age, and its variation, will be more important for females in the Gir Forest population than for males. The probability of extinction was higher with the higher first age of reproduction for females under all scenarios (Figures 5-8). Extinctions did occur even though the population growth rates were all positive. The projected mean population sizes were lower with the higher age of first reproduction (Figures 9-12). Mean population sizes were closest to carrying capacity of $AFR = 3$ but were affected more strongly by the 0-1 year mortality than the female AFR.

Carrying capacity

A decrease in the carrying capacity from 300 to 200 animals had no effect on the stochastic population growth rate under these conditions (Figures 1-4). The probability of extinction (P_e) was higher at the lower K , under all conditions, when the cub mortality was 50% (Figures 5-8). The mean population size of the surviving populations at 100 years were limited by K but were less than K . Under some conditions the mean population size was only 50% of K reflecting population size limitation by other parameters than K . Values for a carrying capacity of 250 (Figures 19-21) were intermediate between 300 and 200.

First year mortality

An increase in the annual mortality rate from 35 to 50% for the 0-1 year age class resulted in more than 50% decline in the growth rate ('r'), however the 'r' values remained positive (Figures 1-4). The 'r' values were near zero when the females AFR was increased to 4 years. The probability of extinction increased with the increase of cub mortality to 50% and was further increased at the lower carrying capacity. Mean population sizes at 100 years were also significantly lower with the 50% cub mortality rate.

Second year mortality

Increase in 1-2 year old mortality rates to 15% (Figures 13-18) had the expected effects of reducing population growth rates by 10-20% and increasing the risk of extinction particularly at the high cub mortalities. There was little effect on the 100 year mean population sizes.

Catastrophe

Catastrophes, of different intensity of effects on reproduction, were simulated to occur about once every 10 years (probability of occurrence = 10%). The low catastrophe scenario include only a 10% reduction in reproduction in the year of occurrence (Figures 1,3,5,7,9 and 11), while the high scenario experienced a 50% reduction in reproduction (Figures 2,4,6,8,10 and 12). There was a small reduction in growth rate, and an increase in probability of extinction, and a small reduction in final mean population size in the high catastrophe effect scenarios.

Harvest effects

There was little effect on the lion population growth rate with high harvest compared to the low harvest under these conditions (Figures 1 & 2 compared to Figures 3 & 4). Addition of the high catastrophe scenario produced a further small reduction in population growth rate (Figures 2 & 4). An increase in the level of harvest had a significant effect on the P_e in all of the scenarios with 50% cub mortality. The combination of high harvest and high catastrophe produced high extinction rates, ranging from 5 to 60% with 50% cub mortality. Reduction of cub mortality to 35% virtually removed the risk of extinction over the 100 years of the simulations.

Summary and recommendations

1. The growth rates and probability of extinction of the Gir Forest lion population are most sensitive to the age of female first reproduction and first year mortality rates. Both of these parameters are strongly influenced by nutrition and population density as it affects conflicts which result in the deaths of cubs.
2. If the carrying capacity of the Gir Forest is 200 or less the probability of extinction of the population will increase significantly, the range of 200-250 appears to be important for the viability of this population. Thus habitat availability and nutrition are close to critical values for this population. This interpretation is reinforced by the observation that prides of lions are moving outside of the Park.

3. The population is vulnerable to a catastrophic disease event such as has recently occurred in Africa. This would put the population increased risk of extinction as a result of normal environmental variation. Also another reduction in population size would lead to more rapid unbreeding. Lions appear vulnerable to inbreeding depression as reflected in measures of sperm characteristic and perhaps cub mortality. It will be valuable to collect blood samples from all animals removed from the Gir Forest for serological studies as a basis to monitor for possible introduced diseases.
4. The risk of extinction of the Asiatic lion will be significantly reduced with the establishment of a second viable wild population. The presence of a captive population provides additional insurance until the second wild population is established.
5. This population can easily sustain removal of sufficient adult animals for translocation to another site to start another population. Removal of younger animals or cubs would have even less effect on the viability of the population. Translocation projects can be designed in terms of the behavioral and genetic requirements. It is likely that removed animals would be rapidly replaced in the population from natural recruitment. Given the baseline conditions that have been explored in these simulation models, further modelling can be done to test ideas about the best strategy for a successful translocation with minimum or no effects on the Gir Forest population.

Table 1. Simulation Modeling of the Gir Forest Asiatic Lion Population. Interaction of age of first reproduction in females (3 or 4 years) and males (5 or 6 years), cub mortality (35 or 50%), catastrophes (frequency = 10%, impact on survival 0.9, and impact of reproduction 0.9 or 0.5) and carrying capacity (300 or 200) on stochastic population dynamics. Three adult females and 3 males were removed from the population every 3 years over the 100 years (**high harvest scenarios**). Each scenario was repeated 200 times and run for 100 years.

File	AFR	Catas	r	SD(r)	PE	N	SD	H	TE
K = 300, 0-1 Mort = 35%									
001	3, 5	.9	0.0748	0.116	0.00	288	17.9	94.4	0
002	3, 6	.9	0.0746	0.116	0.00	285	23.8	94.5	0
003	4, 5	.9	0.0468	0.108	0.00	277	32.9	94.8	0
004	4, 6	.9	0.0462	0.109	0.00	279	29.9	94.8	0
005	3, 5	.5	0.0680	0.125	0.00	281	27.5	94.3	0
006	3, 6	.5	0.0678	0.126	0.00	284	24.2	94.4	0
007	4, 5	.5	0.0428	0.116	0.00	269	37.1	94.5	0
008	4, 6	.5	0.0408	0.118	0.00	260	52.0	94.4	0
K = 300, 0-1 Mort = 50%									
009	3, 5	.9	0.0332	0.122	0.03	251	54.0	93.8	0
010	3, 6	.9	0.0341	0.121	0.02	246	61.3	94.1	0
011	4, 5	.9	0.0110	0.120	0.20	181	87.1	92.5	0
012	4, 6	.9	0.0092	0.119	0.25	184	88.5	92.3	0
013	3, 5	.5	0.0286	0.129	0.06	222	73.2	93.2	0
014	3, 6	.5	0.0298	0.129	0.04	214	78.6	93.2	0
015	4, 5	.5	0.0055	0.130	0.37	155	93.6	91.3	0
016	4, 6	.5	0.0027	0.129	0.42	150	81.9	91.4	0
K = 200, 0-1 Mort = 35%									
017	3, 5	.9	0.0717	0.118	0.00	187	17.2	91.8	0
018	3, 6	.9	0.0716	0.120	0.00	189	17.3	92.0	0
019	4, 5	.9	0.0452	0.111	0.02	179	28.0	91.8	0
020	4, 6	.9	0.0445	0.111	0.00	179	27.6	92.1	0
021	3, 5	.5	0.0680	0.127	0.00	186	21.9	92.0	0
022	3, 6	.5	0.0664	0.127	0.00	185	21.1	91.8	0

023	4, 5	.5	0.0387	0.122	0.04	167	39.4	91.7	0
024	4, 6	.5	0.0394	0.121	0.02	166	38.4	91.5	0
K = 200, 0-1 Mort = 50%									
025	3, 5	.9	0.0335	0.125	0.08	157	47.2	90.8	0
026	3, 6	.9	0.0332	0.123	0.04	154	47.1	90.7	0
027	4, 5	.9	0.0075	0.127	0.45	107	56.4	88.5	0
028	4, 6	.9	0.0073	0.123	0.40	104	58.9	89.6	0
029	3, 5	.5	0.0230	0.133	0.24	134	60.0	90.3	0
030	3, 6	.5	0.0257	0.132	0.17	136	53.1	89.7	0
031	4, 5	.5	0.0016	0.132	0.58	91	57.2	87.4	93
032	4, 6	.5							

Table 2. Simulation Modeling of the Gir Forest Asiatic Lion Population. Interaction of age of first reproduction in females (3 or 4 years) and males (5 or 6 years), cub mortality (35 or 50%), catastrophes (frequency = 10%, impact on survival 0.9, and impact of reproduction 0.9 or 0.5) and carrying capacity (300 or 200) on stochastic population dynamics. Three adult females and 3 males were removed from the population every 3 years over only 10 years (low harvest scenarios). Each scenario was repeated 200 times and run for 100 years.

File	AFR	Catas	r	SD(r)	PE	N	SD(N)	H	TE
K = 300, 0-1 Mort = 35%									
101	3, 5	.9	0.074	0.1177	0	292.41	17.13	94.51	0
102	3, 6	.9	0.0754	0.1175	0	292.66	16.42	94.57	0
103	4, 5	.9	0.0464	0.1102	0	288.18	20.1	94.88	0
104	4, 6	.9	0.0457	0.1083	0	281.7	24.92	94.93	0
105	3, 5	.5	0.0707	0.1248	0	289.16	23.36	94.31	0
106	3, 6	.5	0.0684	0.1256	0	288	21.7	94.5	0
107	4, 5	.5	0.0405	0.1175	0	274.91	33.62	94.74	0
108	4, 6	.5	0.0411	0.118	0	278.03	32.53	94.64	0
K = 300, 0-1 Mort = 50%									
109	3, 5	.9	0.035	0.1209	0	263.52	42.49	94.48	0
110	3, 6	.9	0.0345	0.1218	0	268.56	41.41	94.34	0
111	4, 5	.9	0.0125	0.1159	0	216.26	76.92	93.29	0
112	4, 6	.9	0.0127	0.1168	0	219.86	74.29	93.15	0
113	3, 5	.5	0.0285	0.1269	0	254.48	50.85	93.91	0
114	3, 6	.5	0.0288	0.1282	0	253.72	51.84	94.03	0
115	4, 5	.5	0.0077	0.1243	0	192.84	81.61	91.98	0
116	4, 6	.5	0.0048	0.1262	0.02	175.05	87.87	91.21	0
K = 200, 0-1 Mort = 35%									
117	3, 5	.9	0.0741	0.1202	0	193.86	12.3	92.15	0
118	3, 6	.9	0.0736	0.1196	0	192.52	14.23	92.07	0
119	4, 5	.9	0.0466	0.1123	0	187.42	19.44	92.48	0
120	4, 6	.9	0.0468	0.1116	0	187.1	19.52	92.65	0
121	3, 5	.5	0.0674	0.1287	0	189.88	15.96	91.75	0
122	3, 6	.5	0.0658	0.1296	0	191.39	16.32	92.02	0
123	4, 5	.5	0.0409	0.121	0	181.93	23.57	92.29	0
124	4, 6	.5	0.0422	0.1206	0	181.58	24.44	92.45	0
K = 200, 0-1 Mort = 50%									
125	3, 5	.9	0.0332	0.1239	0	174.8	30.83	- 91.72	0
126	3, 6	.9	0.034	0.1237	0	176.43	30.28	92.01	0
127	4, 5	.9	0.011	0.1209	0	144.79	47.72	90.62	0
128	4, 6	.9	0.0102	0.1207	0.01	136.07	53.55	90.41	0
129	3, 5	.5	0.029	0.1294	0	165.27	37.66	91.52	0
130	3, 6	.5	0.0276	0.1298	0	167.56	41.14	91.54	0
131	4, 5	.5	0.0057	0.1277	0.01	122.99	53.7	89.03	0
132	4, 6	.5	0.0059	0.1288	0.02	125.61	55.62	89.15	0

Table 3. Simulation Modeling of the Gir Forest Asiatic Lion Population. Interaction of age of first reproduction in females (3 or 4 years) and males (5 or 6 years), cub mortality (35 or 50%), catastrophes (frequency=10%, impact on survival 0.9, and impact of reproduction 0.9 or 0.5) and carrying capacity (300 or 200) on stochastic population dynamics. Three adult females and 3 males were removed from the population every 3 years over only 10 years (low harvest scenarios). The mortality of the 1-2 year age class was increased to 15% as compared to 10% in Tables 1 and 2. Each scenario was repeated 200 times and run form 100 years.

File	AFR	Catas	r	SD(r)	PE	N	SD(N)	H
K = 300, Mort: 0-1 = 35%, 1-2 = 15%								
201	3, 5	.9	0.0653	0.1191	0	287.55	20.17	94.52
202	3, 6	.9	0.0662	0.1187	0	288.96	19.12	94.6
203	4, 5	.9	0.0387	0.1106	0	277.4	29.9	94.69
204	4, 6	.9	0.0405	0.1118	0	275.5	32.96	94.8
205	3, 5	.5	0.0599	0.1272	0	284.48	24.23	94.52
206	3, 6	.5	0.0602	0.127	0	283.52	26.22	94.57
207	4, 5	.5	0.0352	0.1197	0	273.26	32.98	94.54
208	4, 6	.5	0.0349	0.1208	0	269.46	40.41	94.56
K = 300, Mort: 0-1 = 35%, 1-2 = 15%								
209			0.0274	0.1232	0	259.35	49.22	94.01
210			0.0275	0.1235	0	254.2	51.64	94.35
211			0.003	0.1224	0.03	177.29	86.84	91.06
212			0.004	0.1201	0.02	178.71	85.4	91.58
213			0.0235	0.1302	0	242.24	59.17	93.57
214			0.0187	0.1293	0	229.82	72.46	93.38
215			-0.0014	0.1313	0.03	149.13	86.9	89.07
216			-0.0014	0.1319	0.04	160.5	88.5	89.83
217			0.0636	0.1225	0	192.74	13.61	92.2
218			0.0657	0.1219	0	192.42	14.59	92.27
219			0.0392	0.1145	0	183.56	23.57	92.38
220			0.0386	0.1148	0	184.63	21.45	92.51
221			0.0592	0.1308	0	188.5	20.18	91.95
222			0.0598	0.1314	0	189.89	16.29	91.93
223			0.0354	0.1225	0	184.12	24.28	92.01
224			0.0333	0.1224	0	178.79	24.75	92.32
225			0.0261	0.1271	0	167.04	39.87	91.66
226			0.0264	0.126	0	171.33	34.38	91.6
227			0.0046	0.1229	0	124.04	55.53	89.45
228			0.0031	0.1249	0.04	126.36	53.92	89.82
229			0.0185	0.1337	0.01	150.3	46.4	90.61

230			0.0181	0.1343	0.02	148.23	48.27	90.68
231			-0.0004	0.1316	0.02	105.15	58.11	87.23
232			-0.0032	0.135	0.07	101.5	54.73	87.04
233			0.0658	0.1192	0	239.7	19.37	93.5
234			0.0652	0.1197	0	240.59	17.89	93.56
235			0.0402	0.1125	0	232.74	26.62	93.9
236			0.0398	0.1127	0	230.42	27.35	93.96
237			0.0584	0.1281	0	234.3	23.91	93.47
238			0.0596	0.1293	0	232.1	25.41	93.58
239			0.0348	0.1212	0	225.39	30.18	93.55
240			0.0353	0.1208	0	224.5	32.09	93.55
241			0.0247	0.1267	0	207.89	46.6	92.84
242			0.0257	0.124	0	214	42.79	93.23
243			0.0035	0.1218	0.02	151.71	70.95	90.38
244			0.0034	0.1237	0	144.97	70.55	89.91
245			0.0197	0.132	0	191.76	56.02	92.09
246			0.0177	0.1318	0	191.84	60.35	92.23
247			-0.0023	0.1326	0.05	124.13	69.53	88.99
248			-0.001	0.1307	0.02	125.65	72.11	89.28

Figure 1

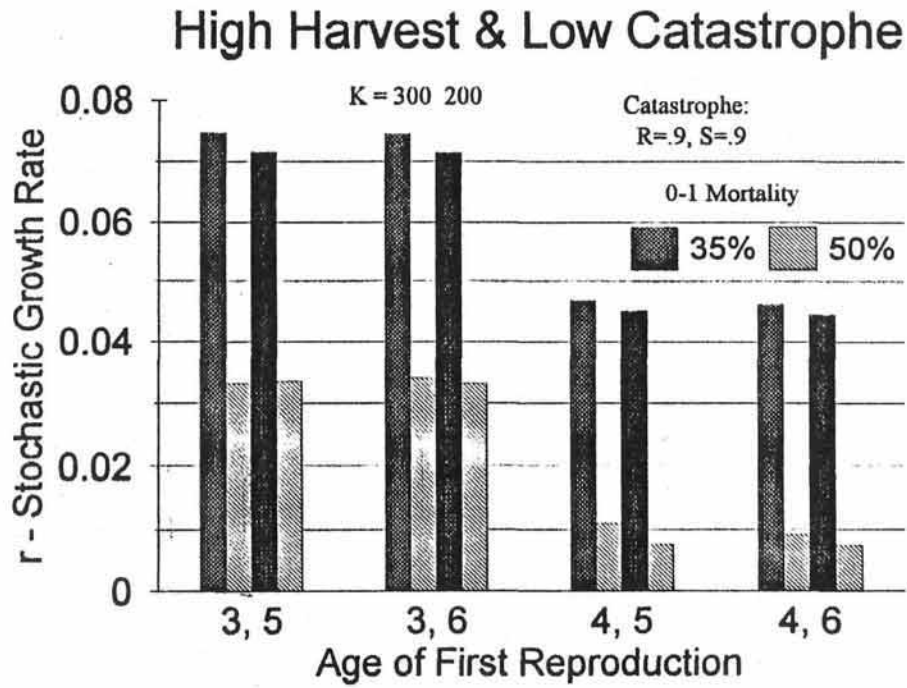


Figure 2

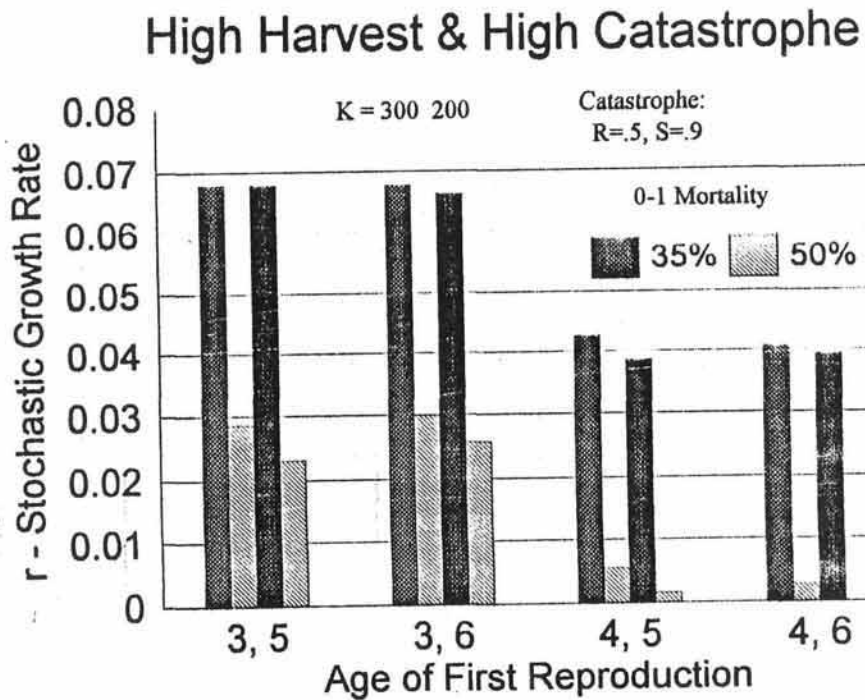


Figure 3

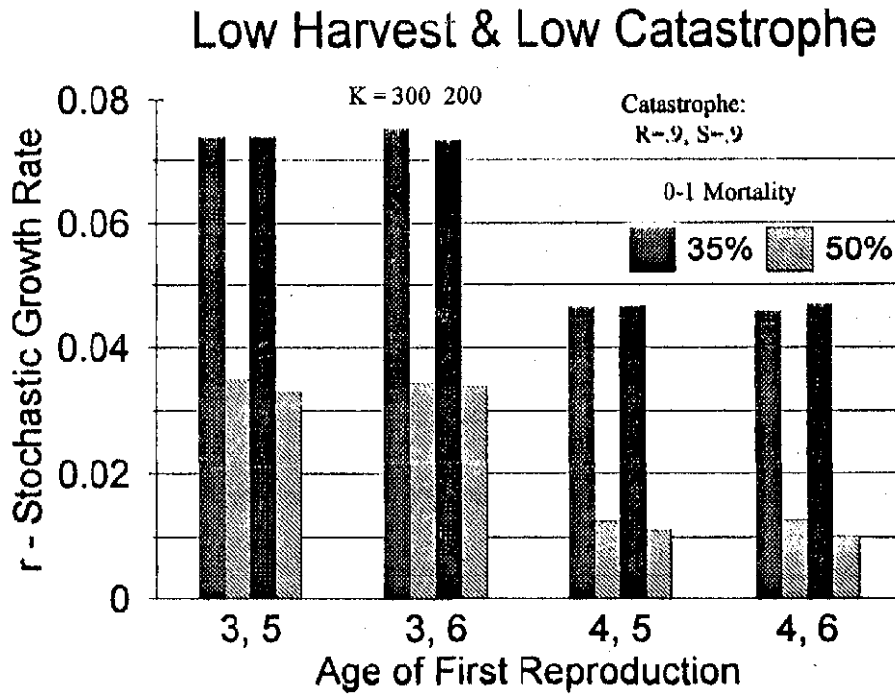


Figure 4

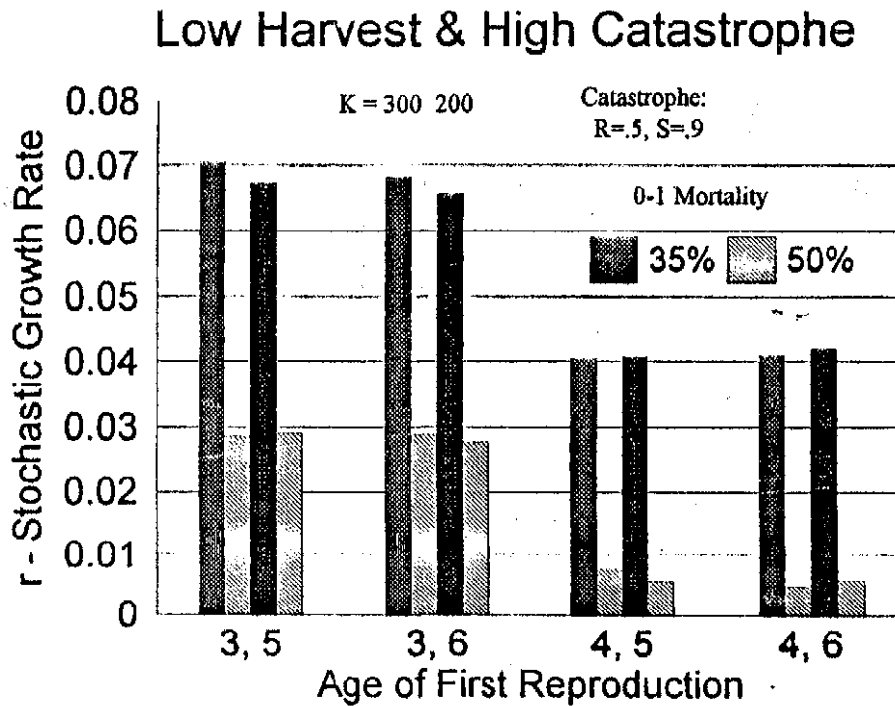


Figure 5

Low Harvest & Low Catastrophe

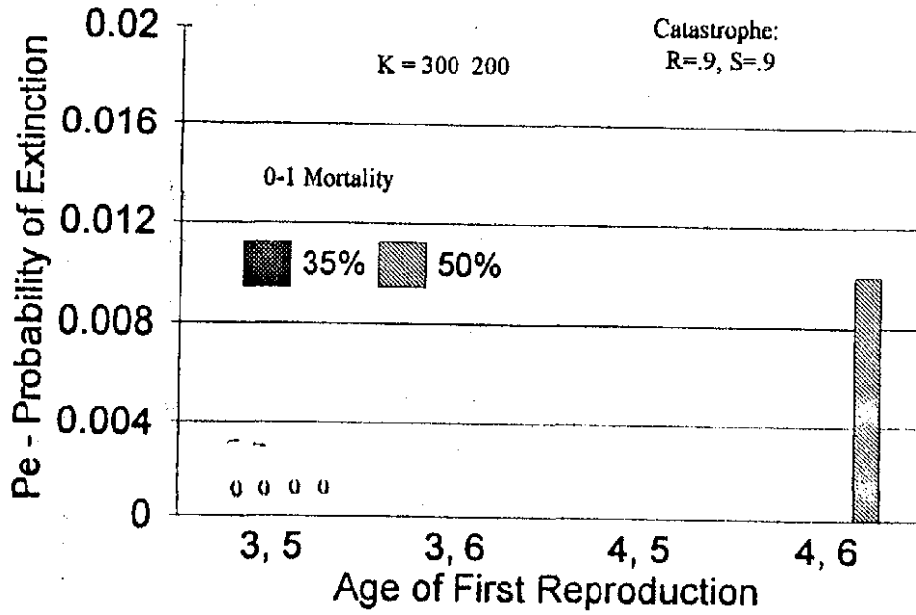


Figure 6

Low Harvest & High Catastrophe

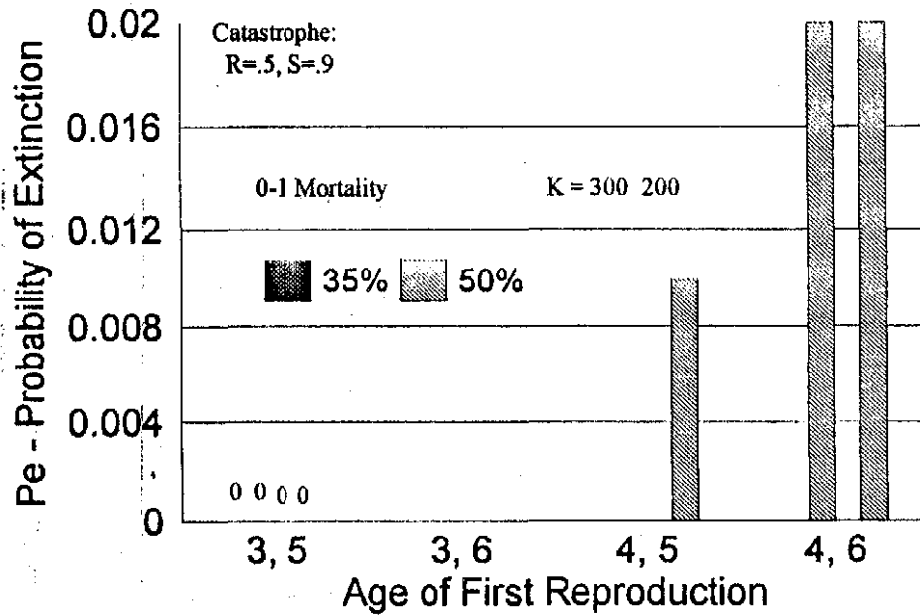


Figure 7

High Harvest & Low Catastrophe

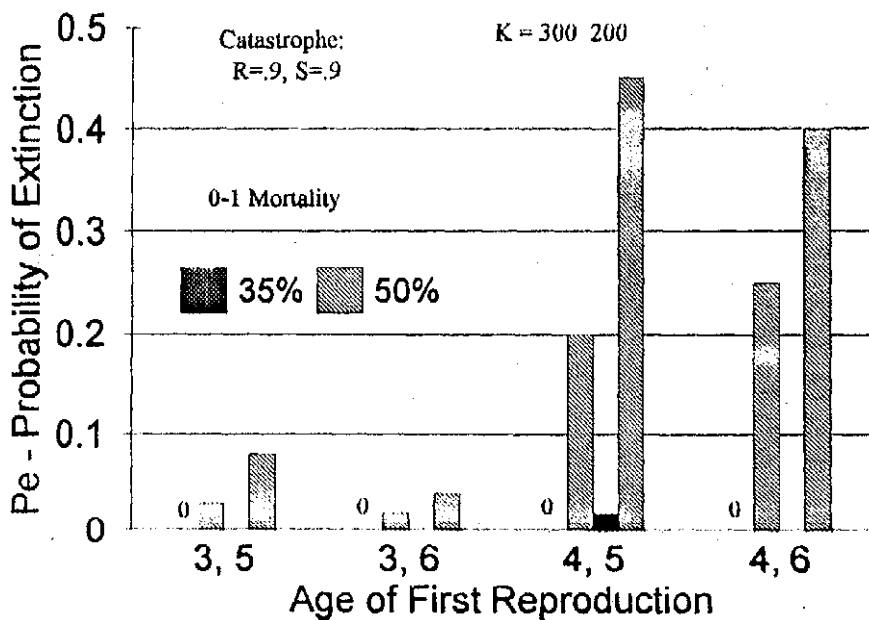


Figure 8

High Harvest & High Catastrophe

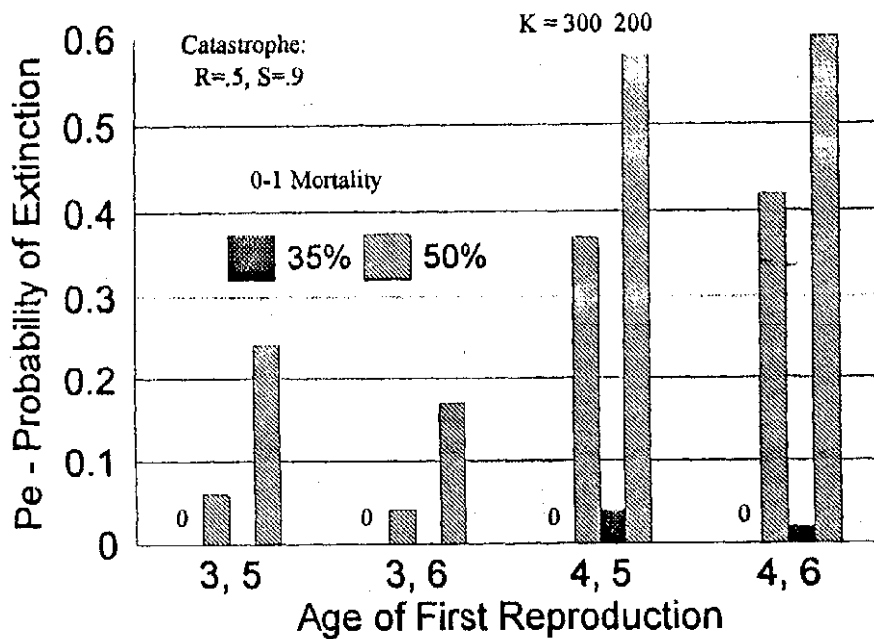


Figure 9

Low Harvest & Low Catastrophe

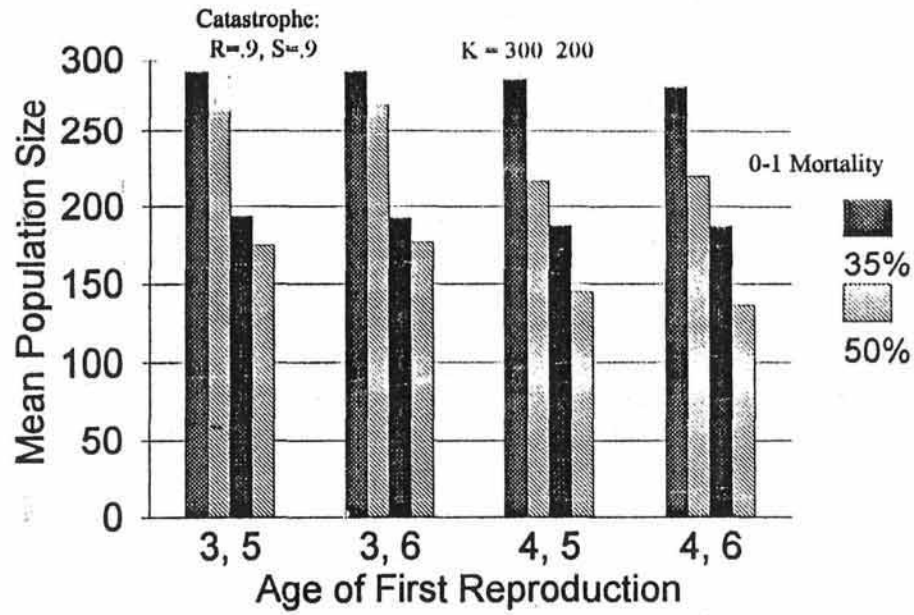


Figure 10

Low Harvest & High Catastrophe

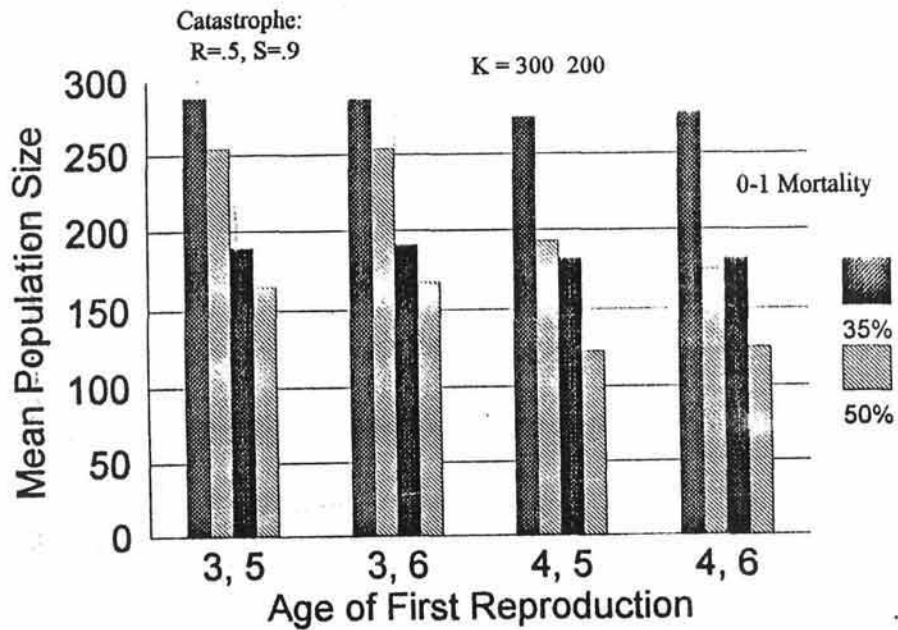


Figure 11

Low Harvest & Low Catastrophe

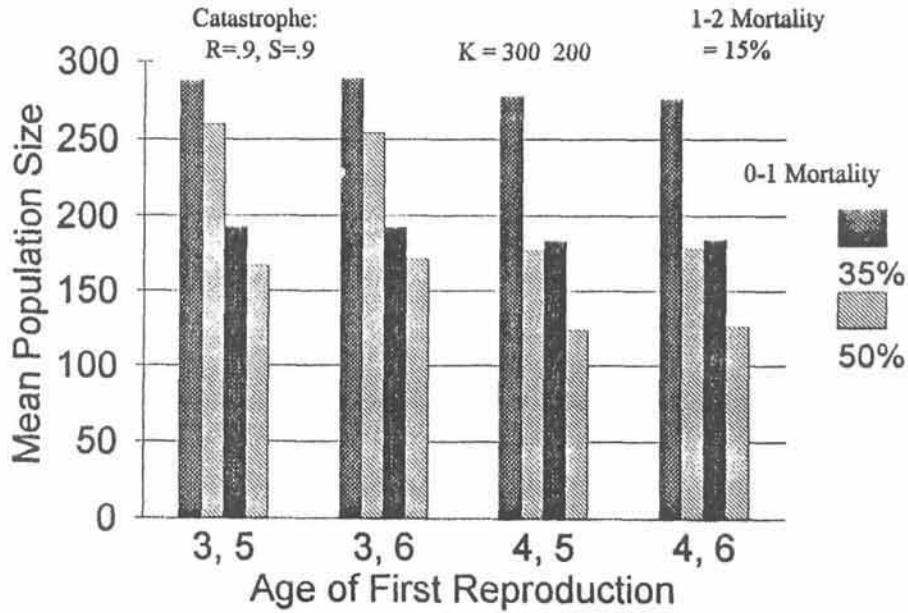


Figure 12

Low Harvest & High Catastrophe

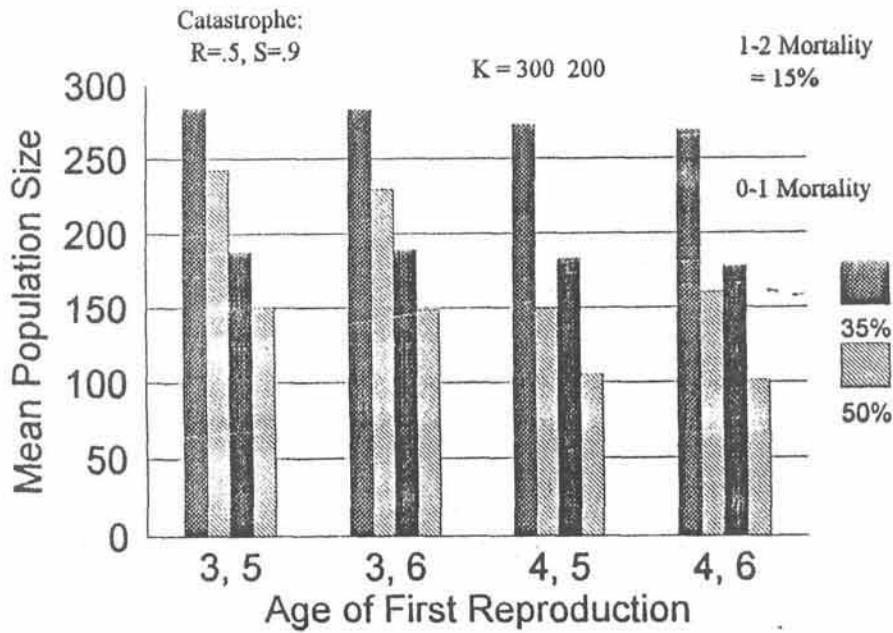


Figure 13

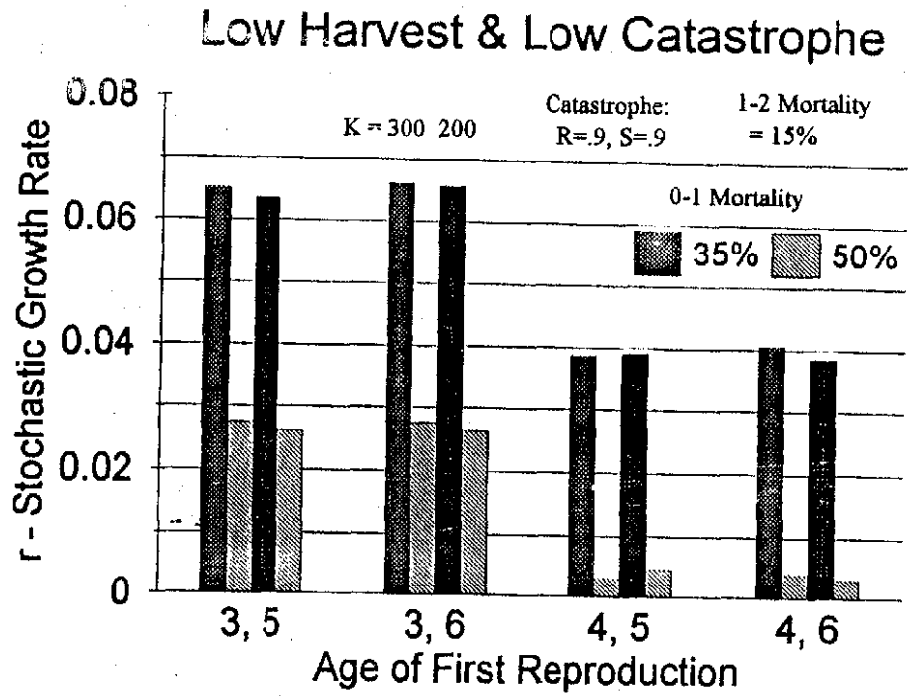


Figure 14

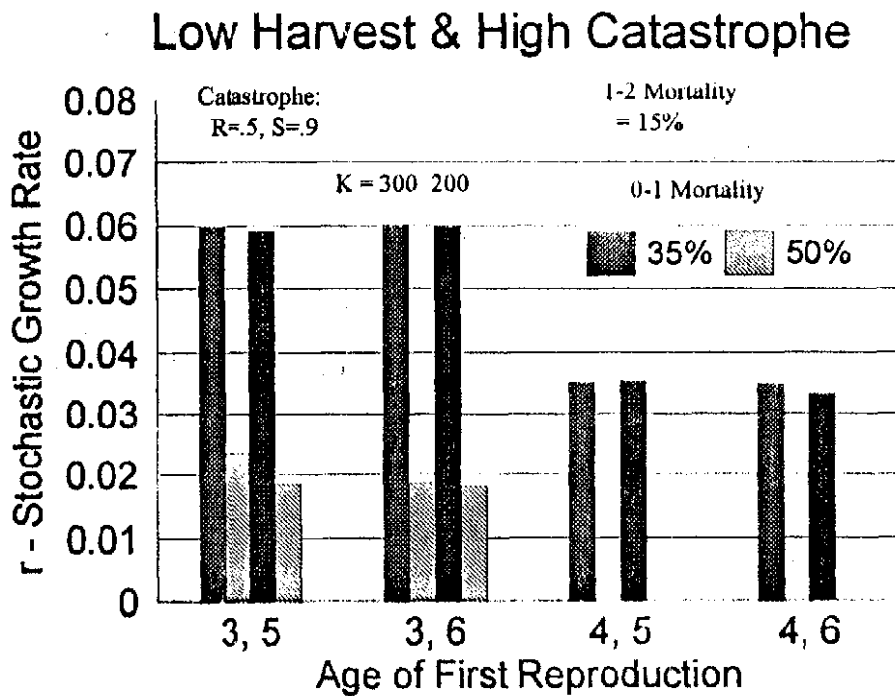


Figure 15

Low Harvest & Low Catastrophe

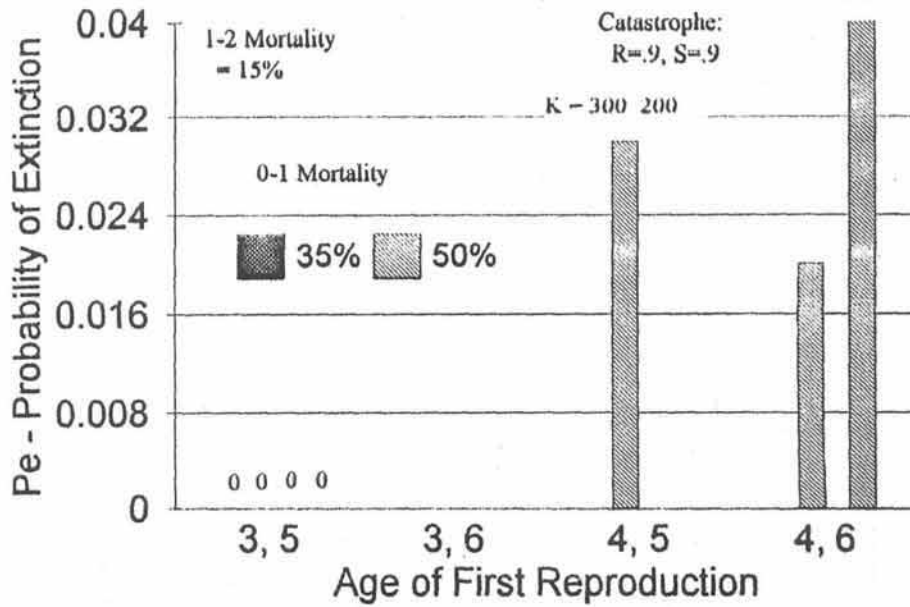


Figure 16

Low Harvest & High Catastrophe

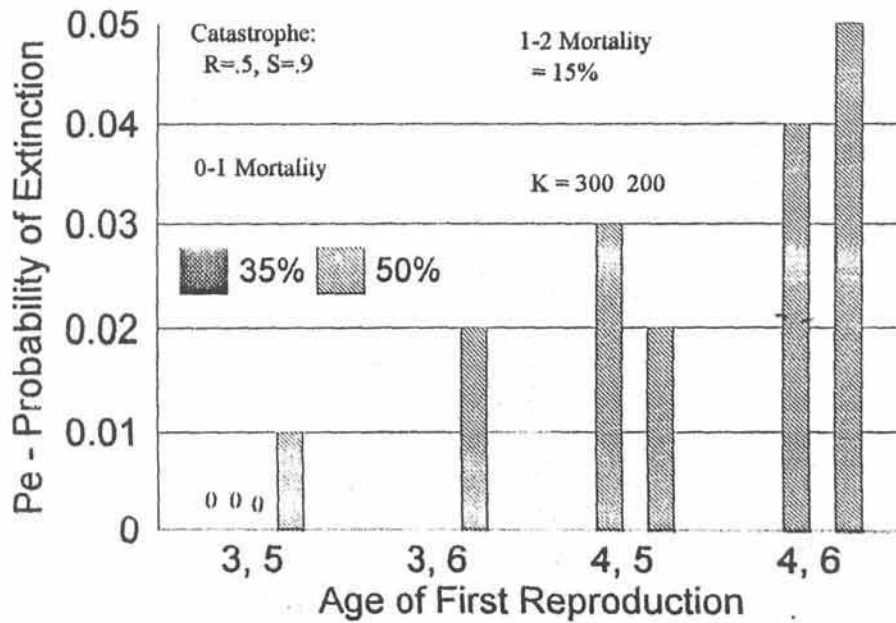


Figure 18

Low Harvest, Catastrophes 1 or 2

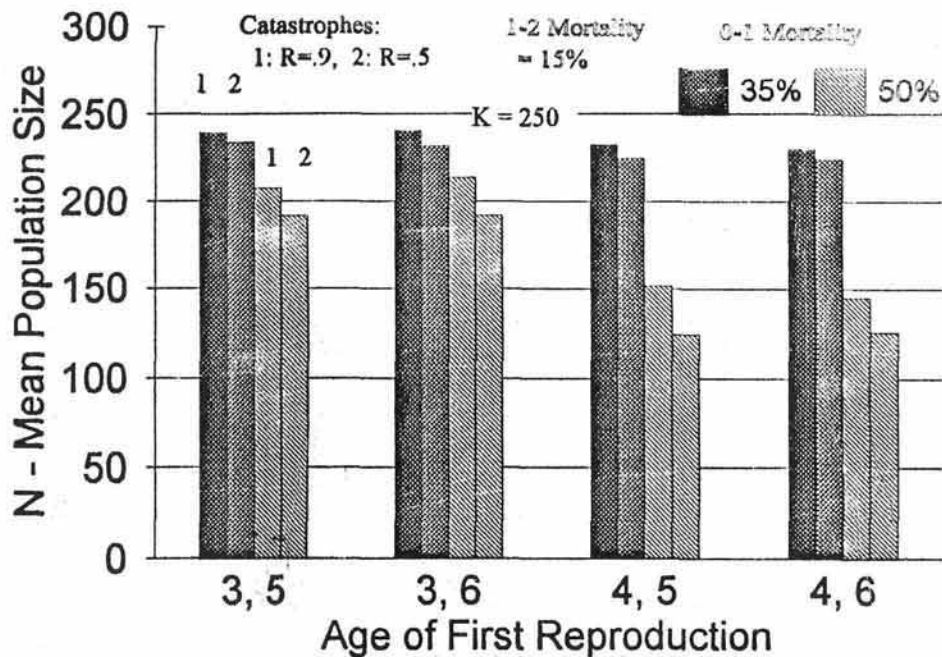
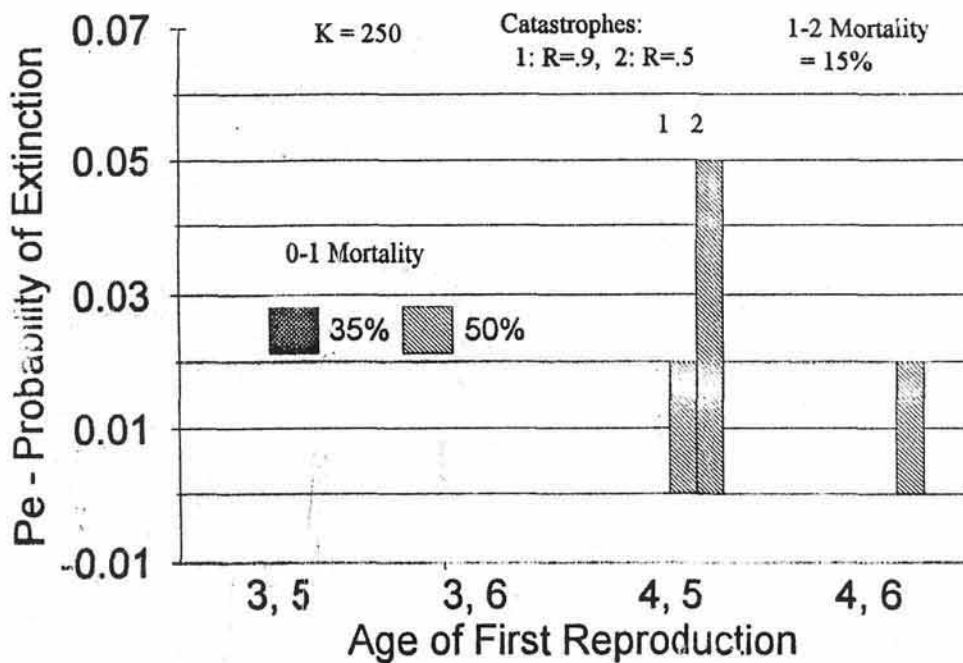


Figure 17

Low Harvest, Catastrophes 1 or 2



TRANSLOCATION

Members : Diwakar Sharma, Nita Shah, Sanat Chavan, Shivabadhrasinji, Bhuva Vimal, B.M. Parasharya, R.S. Shekhawat, J.P. Vasava, Dr. C.D. Patel, V.K. Sinha, M.K. Misra, N.S. Bundela, D.M. Richardson, Pushp Kumar

[Habitat Synthesis Group was transformed into this group since this group had more knowledge about the habitat & problems of the proposed area.]

The recommendations are made based upon the guidelines of the IUCN/SSC Reintroduction Specialist Group.

I. Pre-translocation phase

A. Feasibility studies and background research

1. Previous history of reintroduction/translocation of Asiatic lion
2. Choice and evaluation of sites
3. Identification of suitable release stock
4. Socio-economic and legal studies

B. Public Awareness and Education Phase

C. Professional training of individuals.

II. Planning, preparation and release phase

- 1 a. Governmental and funding approval and establishment of well-defined institutional support.
- 1 b. Fulfillment of pre-release habitat management obligation.
2. Transport plans.
3. Identification of short term and long term, objectively verifiable performance indicators.
4. Time-frame of release process
5. Appropriate genetic assessment and health screening of the stock to be released.

6. Design of monitoring process.
7. Appropriate Veterinary support.
8. Actual release strategy.
9. Specific policies for intervention.
10. Continuation of conservation education.
11. Planning and execution of eco development program for local people.

III. Post release phase

1. Constant post-release monitoring of the released stock.
2. Demographic, ecological, genetic and behavioral study of the released stock.
3. Undertaking restocking policy
4. Collection and investigation of mortalities.
5. Interventions when necessary
6. Periodical review of the project.
7. Habitat management where necessary
8. Continuing public relation activities
9. Continuing eco development program for local people.

IV. Specific recommendations

1. The Group considered five sites viz. Barda (Gujarat), Kuno (Madhya Pradesh), Sitamata, Kumbalgarh, and Darrah-Jawahar Sagar (all 3 in Rajasthan). Evaluation chart is appended.
2. With reference to these sites, it was considered necessary that feasibility studies should be carried out as given in Item 1 – 4 pre-release phases.

3. As a pre-release activity, the public awareness and education program should be taken up.
4. Training of individuals who are going to be involved in the project should be taken up well before the actual implementation.
5. Prior to release, the State/Central governments concerned should ensure institutional support in respect of staff, administrative machinery, infrastructural development and finance. It is recommended that a well-defined executive body for the project should be formed.
6.
 - a. A well considered protocol should be drawn up for the actual translocation/reintroduction program by the authorities and experts who are going to execute the programmes.
 - b. The program should also lay down a time-frame for the release process.
 - c. The release process shall take into account the appropriate veterinary protocol for screening the released stock.
 - d. Incidence of disease inimical for the release stock in the proposed habitat should be dealt with well in advance.
7. During this program the authorities concerned should take up eco-development works as per the Government of India guidelines.
8. The released stock should be regularly monitored for demographic, ecological, genetic and behavioral aspects.
9. All deaths of released animals should be subjected to scientific scrutiny.
10. In case of man-animal conflict, suitable compensation should be paid through the project.
11. The project should be reviewed on a regular basis.
12. Habitat management of the reintroduction site should be carried out.
13. Public relations and education activities should be continued throughout.

14. It is recommended that a back-up population of Asiatic lion be maintained at a captive breeding facility within easy access of the proposed translocation site.

15. That the translocation program must have a long term perspective of about 100 years during which further translocation sites in the states of Rajasthan, Madhya Pradesh, Uttar Pradesh, etc. be built up in a planned manner.

16. That the question of lion-tiger compatibility be examined in great detail.

Table 1.
Comparison of Habitat Characteristics Between Gir and the Proposed Sites for Lion Reintroduction

Habitat Parameters	Gir	Barda	Kuno	Sitamata	Kumbalgarh	Darrah-Jawahar Sagar
Locality Factor						
Area	1412 sq.km	190 sq.km	2000 sq.km. (345 sq.km now)	1300 sq.km (500 sq.km)	1060 sq.km (586 sq.km)	1100 sq.km (500 sq.km)
Geology	Deccan trap basalt	Deccan trap basalt	Vindhyan sandstone tract	Aravalli, granites	Aravalli sandstone	Vindhyan
Climate	900mm; 5 ⁰ -47 ⁰ C	200mm; 10 ⁰ -40 ⁰ C	602mm; 4 ⁰ -48 ⁰ C	650mm; 5 ⁰ -43 ⁰ C	600mm; 2 ⁰ -48 ⁰ C	600mm; 5 ⁰ -45 ⁰ C
Vegetation	Dry deciduous teak and <i>Anogeissus teak</i> , <i>Acacia catechu</i>	<i>Acacia</i> associations <i>Acacia nilotica</i> <i>Manilkara</i>	Dry deciduous <i>Anogeissus pendula</i> and <i>Acacia catechu</i> <i>Boswellia serrata</i> , <i>Zizyphus spp.</i> <i>Diospyros</i> , <i>Bauhinia</i>	Dry deciduous teak forest, teak – bamboo, <i>Lannea</i> <i>Diospyros</i> , <i>Anogeissus latifolia</i> , <i>Boswellia serrata</i>	Dry deciduous <i>Anogeissus pendula</i> <i>Acacia catechu</i> , <i>A. latifolia</i> , <i>Boswellia</i> <i>Lannea</i>	Dry deciduous <i>Anogeissus pendula</i> , <i>A. latifolia</i> , <i>A. catechu</i>
Shrub cover	Medium	Dense	Sparse	Dense	Sparse	Sparse
Ground cover	Dense, extensive	Poor growth	Dense good growth Extensive	Dense in 200 sq.km. area, restricted	Good growth Extensive	Good growth Extensive
Riverine Tract	Good riverine vegetation with 40% canopy cover. Long impenetrable patches of <i>Carissa</i> in shrub layer. <i>Syzygium</i> , <i>Pongamia</i> , <i>Holoptelia</i> is dominant species.	Small extent of thickets in patches	<i>Terminalia arjuna</i> , <i>Syzygium cumini</i> , <i>Mitragyna</i> , <i>Ficus</i> , <i>Carissa</i> , <i>Vitex negundo</i> Patches of bamboo of cover	50km length <i>Syzygium</i> , <i>Terminalia</i> , <i>Mitragyna</i> Continuous thickets	Fragmented patches of <i>Syzygium</i> , <i>Terminalia</i>	Similar to Kumbalgarh

Habitat Parameters	Gir	Barda	Kuno	Sitamata	Kumbalgarh	Darrah
Area & Shape	10	3	7.5	8.5	2	5
Climate	10	2	6	7	6	6
Temperature	10	9	6	6.5	6	6
Terrain	10	1	8	9	2	9
Water	10	5	7	6	2	7
Vegetation	10	2	7	8	7	7
Canopy Cover	10	3	8	6	7	7
Shrub Forest	10	2	7	4	7	7
Riverine Forest	10	1	6	4	3	3
Disturbance	04	1	6	4	3	3
Prey base	10	1	9	3	4	3
Total	104	30	77.5	66	49	63

Comparison of habitat parameters among the proposed sites for lion reintroduction.

Monitoring and Research Report of the Working Group

Members : C. Packer and Ravi Chellam

Monitoring the size and structure of the free-ranging lion population(s) is essential to understand their population dynamics. The following techniques are recommended.

1. Individual recognition:

With knowledge on the age, sex and reproductive history of each individual in the population, all population parameters could be measured with complete accuracy. It would also be possible to select specific individuals for relocation and to anticipate dispersal of sub adults into the surrounding communities. Field studies on African lions have been conducted using natural markings for establishing individual recognition. These markings include patterns of whisker spots and ear notches. However, lions in Gir can rarely be examined at sufficiently close quarters to permit inspection of such details. Nevertheless, every effort should be taken to establish identification files of as many animals as possible. Tracing the life histories of even a few individually distinctive animals could provide important information.

2. Radio telemetry:

Radio telemetry is essential for understanding the habitat requirement of the lions and can also be extremely valuable in estimating population numbers. By radio collaring one member of each group, it would be possible to record the ranging patterns of every pride in the reserve. Through repeated tracking of these individuals it would be possible to gain more regular censuses of the composition of each pride. In addition, telemetry greatly reduces the effort required to locate and observe the lions.

3. Cross-sectional censuses:

Lions in the Gir forest are traditionally censused by large teams of wildlife workers who set out baits throughout the reserve over a three day period. This is an excellent technique for gaining periodic “snapshots” of the lion population. In future, university students and personnel from wildlife research organization, NGO’s, and conservationists should be included in these censuses as part of their training and in order to share their expertise with the local field worker. While these “snapshots” have been extremely valuable in tracking the Gir population, it is essential to supplement this technique with radio telemetry and individual recognition. Only the latter can link population changes with events that occur at any time of the year. Further, it will be important to have a variety of available techniques in case baiting does not prove feasible in the newly established populations.

Suggested research program

A. Basic demographic parameters:

The workshop has emphasized the value of knowing age-specific survival and reproduction in estimating future population trends.

B. Social Organization and dispersal patterns:

The preliminary data suggests that the Gir lions may show significant differences in social organization from the African lions. Knowledge of the precise group structure will be essential to estimate local reproductive performance and the approximate timing of dispersal events and the fate of the dispersing animals.

C. Monitoring lion diet through collection of scat:

Systematic collection of lion scats should be performed on a regular basis to monitor the lion diet. Such samples could also be used for parasitological assay, which may prove important in assessing the prevalence of parasitic infections.

D. Monitoring prey populations:

Biennial censuses of prey species should be undertaken to provide a fine-grain measure of food availability for the lions, and to measure impact of any changes in livestock densities.

E. Monitoring leopards and striped hyenas:

Little is known of either species in India and monitoring should be expanded to include both. This can be achieved by the systematic collection of scat samples, and keeping records of all sightings.

F. Monitoring Vegetation:

Sample plots in the major vegetation types should be established and monitored for seed germination, recruitment and succession. This can be achieved by marking adequate sample area and collecting data repeatedly at an interval of three to five years for a period of 25 to 30 years. These vegetation data should also be set up in localities under varying intensities of anthropogenic pressures and under different management practices such as fire lines and grass harvested areas.

Lion Human Interaction

Report of the Working Group

Members : *Bharat J. Pathak, D.S. Narve, P.P. Raval, Diwakar Sharma, Shivabhadra Sinhji, Vimal Bhuva*

Lion-human interaction could be direct physical interaction or interaction of their activities and influences on each other. Depending on the location of such interactions, these can be of different types.

I. Interactions with resident human population.

In Gir there are two types of resident human populations viz.

- (a) Pastoralists locally known as Maldharis and
- (b) Villagers of forest settlement villages. The level of interaction can be assessed by their populations and need based activities.

A. Maldharis:

The Maldharis living in 60 Nesses (hamlets), formed of 360 families, 2540 people and 14,000 livestock graze their livestock in the protected area and to an extent there is a competition for resource between livestock and wild herbivores.

Maldharis take adequate care to protect their livestock from lions and other predators. Over generations, they have developed skills to minimise cattle loss due to predation and avoid human injuries and deaths by wild carnivores. However, to some extent livestock of *Maldharis* form parts of the diet of carnivores, which is generally in the form of non-valuable livestock not properly looking after. On the other hand there is free movement of livestock in and out of the protected area, increasing the chances of spread of cattle-borne diseases.

B. Human population in forest settlement villages:

Forest settlement villages within the 1412 sq.km of the Gir protected area are located mainly near the administrative boundary, barring 3 villages which are surrounded by the protected area. This comprises 556 families with a population of 4794 people and 4249 livestock.

These villages were settled in the protected area much before declaring the areas as a Sanctuary and National Park. Villagers collect forest produce and graze their livestock in the protected area. Wild herbivores and carnivores frequently visit these villages. Like *Maldharis*, these villagers have also developed skills to avoid conflicts with carnivores and to prevent attacks on their livestock. Though cared for, livestock of these villages occasionally fall prey to carnivores, but no incidents of human injuries or deaths have been reported. The

competition between livestock and wild herbivore is detrimental to the prey population and ultimately for the carnivore population. Administratively it seems to be difficult to relocate villagers of the forest settlement villages.

C. Villages Outside Gir:

There are 306 villages within the 10 km radius from the boundary, with a human population of 160,000 in 28,000 families and about 100,000 livestock. These villages have come up recently and have led to habitat loss. Most of the population comprises of farmers and these are the people most affected by lions along with the livestock graziers.

Because of the loss of natural habitats, the lions are forced to move out and sometimes they take shelter in agricultural field (sugarcane). Other factors include the hours of electricity availability. It is during the night hours that farmers are forced to work in their fields due to unavailability of electricity during the daytime. Night being the most active period for lions, the chances of interaction is increased, mostly due to surprise encounters.

Livestock graziers sometimes move inside the Protected Area for grazing their livestock and are prone to encounters with lions. Sometimes lions move inside the villages and kill domestic animals out of hunger. Villagers, because of their ignorance, and their emotional attachment to their animals, try to push a lion away from the kill and in the process get attacked by the lion. There have been rare cases of intentional attack by lions.

The other category includes people who go to the Protected Area for minor forest produce collection and failing to see a lion under a bush, approach too close and subsequently get attacked.

II. Interaction with people visiting Gir

Tourist, pilgrims, outside graziers, forest produces collectors from outside visit the Gir Protected Area.

Tourists	= 30,000
Pilgrims	= 100,000
Livestock	= 20,000

Tourists and pilgrims are generally regulated. However, unscrupulous visitors cause disturbance to wildlife during their visits. No physical interaction with carnivores has been recorded except one case when a young boy moved away from the vehicle and approached a lion hiding behind a bush.

Primate tourist companies and pilgrim places have expansion plans and pose a threat to the integrity of lion habitat, because of increased disturbance. Regulated tours serve the purpose of education. Facilities have been developed for tourist

assistance and interpretation. However, there is scope for further improvement in eco-tourism.

Physical interaction between man and lions has been recorded since historical times. Incidents of such physical harm to man have risen considerably in recent years. Though recorded, such incidents were not studied properly during the early part of the 20th century. Now, each incident of physical interaction is being studied properly. In most cases, the reasons are attributed to people getting attacked because of disturbing lions, chance encounters, and of mistaken identity.

Recommendations

1. Lion population management outside the park and appointing village wildlife watchers.
2. Public awareness programmes.
3. Relocation of *Maldharis* from Gir protected area in a phased manner.
4. Various studies on each of the interaction issues have been carried out. Recommendations of such studies should be considered for implementation. [Includes (1) Central Committee on Tourism in Protected Areas, 1984; (2) Experts Committee, appointed by Government of Gujarat, 1990-1993].
5. Implementation of recommendations of tourism plan.
6. Completion of implementation of protected plan recommendations.

Global Animal Survival Plan for Captive Population

Report of the Working Group

The reason for establishing captive population of Asiatic lions to further ensure the long term security of the subspecies. This scientifically managed breeding program is facilitated by a regional captive population coordinator or committee, whose job is to maintain a genetically pure, non-inbred population in tandem with the coordinators in other zoo regions.

It is the responsibility of the coordinating bodies to make breeding recommendations to equalize founder representation and family sizes. This would avoid individual lions producing more offspring than is required to maintain the level of heterozygosity, or would overstretch the amount of captive space that is available for the population.

Occasionally, wild caught individuals will be available to enhance the captive population. Generally these will be displaced or 'problem' animals that have to be removed from outside the proposed area. Any 'harvesting' has to be held at or below a recognized level (from the demographic simulation models) that does not compromise the long term security of the wild population.

The reason for establishing a "global" programme is for additional security of the population in the event of a catastrophe reducing numbers to untenable levels or destroying the local programme. In establishing a programme, priority goes to the range country of the species in setting up their own viable programme and then to outside regions. The figures reflected below are ideal goals which have been fixed as a result of demographic and genetic parameters necessary to maintain a healthy captive population with sufficient diversity to survive and maintain 95% genetic diversity (this percentage is predicated on potential for adaptive evolution) for 100 years. In the event that the wild populations – both the one in Gir and those which are to be established – require genetic supplementation, it should be done with biomaterial from animals which are healthy and fit. From a small captive population, this can be achieved by very carefully managed pairings. This is the objective of a breeding programme and of the Global Animal Survival Plan.

Currently the pure captive population of Asiatic Lion is estimated to be 82 specimens of which 59 (19.34.6) are in India and 23 (11.10.2) outside of India.

Recommendations

1. To develop a genetically pure, healthy captive population of between 400 to 600 animals. Regional coordinators will be established to manage the populations in the five global zoo regions with a proposed minimum distribution as follows:

India	200- 300
Southeast Asia	25-50
Australia	25-50
Europe	75-100
North America	75-100

Other regions may participate in this program should they fulfill the requirements stated in this document.

2. Hybridization has seriously affected the integrity of the captive population of the Asiatic lion in the past. Therefore the zoo community should identify the genetically pure lions and permanently identify those individuals. The use of transponders at the dorsal base of the tail and tattooing on the inner right thigh shall be done by each zoological institution housing these animals. Studbook numbers should be used for identification whenever possible.
3. Genetically pure individuals should be housed only at institutions that can prevent hybridization. It may be preferable that these animals be kept in facilities which have no hybrid lions. However, if all the hybrids cannot be removed, individual hybrid lions should be sterilized before accepting the facility as a member of the captive breeding program.
4. All of the zoological institutions participating in the program should abide by the recommendations of their Regional Species Coordinators.
5. Regional Species Studbook Keepers will be responsible for collecting and maintaining a current regional studbook. This information should be provided annually to the International Studbook Keeper.
6. Ownership of all Asiatic lions shall be retained by the Government of India. There will be no commercial transaction of Asiatic lions.

Global Animal Survival Plan

Population Management and Husbandry

Facilities

A major component for the care and management of the captive lion is the design of the facility in which the animal is housed. When designing the enclosure, husbandry needs, veterinary concerns and the biological requirement of the animal should be considered and incorporated in the facility. Important considerations when designing a breeding facility should include dimensions, barriers, substrate, shelter, transfer areas, and climate which can affect both the reproduction and health of the animal.

Design

The basic enclosure design is of utmost importance. The size must be adequate for movement and exercise to decrease boredom, stimulate activity, and give the lion a feeling of security and comfort.

1. Barriers commonly used for containment include:
 - a. Bars – metal can provide strength and requires relatively low maintenance. However these may promote trauma from biting or attacking, may trap limbs or heads due to inadequate spacing and may promote trauma from adjacent cats due to improperly designed barriers.
 - b. Wire – Wire is accepted for lion enclosures provided it is of adequate strength. This material, if not properly selected and installed, may trap limbs, heads or teeth, especially in young animals.
 - c. Glass – requires more maintenance, is expensive, and is vulnerable to fracture. The material used should be adequate strength to restrict any damage that could be caused by the animal or outside forces.
 - d. Moats and Grottos – should be constructed of sufficient area both horizontally and vertically (minimum 15' deep and 25' across) to prevent escape. Deep vertical or water filled moats, although effective should not be used to contain cub or in situations where interspecies aggression is high. Extra consideration should be given in zoos where water moats are subject to freezing over.
2. Shelter/ holding area(s) – must be provided for each animal in the enclosure. Protection from the elements should be accessible. Provision should be made to provide controlled warm environment of about 85 degrees Fahrenheit in shelters in zoos located in cold climates.
3. Indoor areas – should be illuminated by a combination of natural and artificial light. Proper ventilation is imperative for indoor facilities.

Exhibit Enclosure:

Natural exhibits – may be created by using vegetation and soil. Natural substrates for the outdoor enclosure are preferred. Grasses and other plant materials used within an enclosure

must be carefully chosen to avoid toxic species.

Dirt substrates will become contaminated over time with micro organisms and parasites, exposing the animals to potentially dangerous concentrations of pathogens. Provisions must be made so that the contaminated substrate is removed periodically and replaced with clean materials.

The topography of the exhibit should be varied with a combination of elevated areas, dead fallen trees, rocks and mounds. Logs or timber allow the natural behavior of scratching for claw wear and maintenance.

Natural hiding areas should be included in the exhibit. There should be adequate shade provided in the exhibit for the animals. Ponds, pools or streams add to the exhibit, but water sources must be drained if cubs are to be displayed in the area.

The area should be sub divided into a main exhibit/breeding area and holding yards for animals temporarily isolated separated from the main exhibit.

The aquatic component of exhibits, pools and moats, need to be designed for maintaining high water quality, ease of cleaning and sanitizing due to the tendency of some cats to defecate in water.

Off-exhibit holding:

Off-exhibit holding enclosures are essential for proper management and health care of the animals and including additional working holding and quarantine areas. Off-exhibit holding provides treatment areas out of public view and seclusion for a stressed or ill lion. Within this area, squeeze or restraint cages permit an alternative method of handling for procedures normally necessitating anesthesia.

Off-exhibit enclosure should provide access to a private outdoor area a dry, comfortable, denning space. These areas should be designed to facilitate feeding, watering, and cleaning, with as little disturbance as possible. Surfaces in these areas must provide good traction, especially when wet, but not be too abrasive as to cause foot pad trauma during normal movement or pacing.

Maternity Den:

Off-exhibit holding areas can also provide an area where a female can be isolated from cage mates prior to parturition. This is an essential component for successful birthing and survival ship of cubs.

A pregnant female should be separated prior to parturition. A separation date should be determined utilizing the date of mating, temperament of female, and past birthing history. Females with cubs should be kept separate from other adults until the cubs reach an age of 2 to 3 months. Adult lions of both sexes will tolerate cubs if adequate space is provided.

Examination of cubs after birth will depend upon the disposition of the female. Guidelines in the veterinarian section of this document should be referred too.

Social Organisation:

Lions are solitary animals for at least part of their life; therefore they may be housed singularly or in groups depending on compatibility and enclosure space. Only one male should be mixed with a female, or a group of females, at any given time to ensure actual recording of parentage.

It may be possible for a number of single sex groups to be established, particularly in safari parks. These can be groups that have already produced the required progeny in the captive populations. These groups will act as a reservoir that can be utilized should particular individual be needed in the breeding population.

Enclosure Maintenance:

Each enclosure design must provide an easy-to-clean water container that can be shut off and drained. The water container should be accessible to both lion and the keeper.

Disinfecting agents must be selected on the basis of effectiveness and low toxicity. For effective cleaning, hot water, a detergent plus physical effort to remove organic debris followed by disinfectant should be used. In all cases, chemicals must be thoroughly rinsed to prevent exposure to animal.

Animal Introduction:

Introduction methodology varies with each institution. Flexibility is the key to successful introductions. Individual personalities and animal characteristics must be considered.

For any introduction, adequate staff should be available to intervene, keeping in mind that aggression can occur. Methods for intervention and separation could include the transferring of the animal(s) to another enclosure, the use of the safety equipment, water spray, or CO2 canister. All introductions should be carefully monitored, and should be limited to hours when staffs are available.

Mate Selection:

Mate selection is an important factor. Consideration should be given to genetic representation of individuals. All transfers and breeding should be recommended/approved by the Regional or National Asiatic Lion Coordinator.

Disease and Veterinary Research

Report of the Working Group

Members : J.M. Anjaria, R.H. Sabapara, N.V.K. Ashraf, R.K. Sahu, V.K. Sarodra, Vinod Kumar Sharma, P.S. Solanki, V.K. Sony, Mitchell Bush, Lyndsay Phillips Jr.

The following derives from a collection of five years of medical data gathered from postmortems and clinical observations by the participants of the Asiatic Lion PHVA on captive Asian Lion (pure and hybrid) with limited data on several free living individuals. The largest set of data, both free living and captive, comes from the Sakkarbaug Zoo where the largest captive population of this subspecies resides. This data will be utilized to define the major medical problems encountered in the delivery of health care to this highly endangered subspecies. It is hoped that by critical analysis of this initial data we will focus on the major areas of concern so that additional data can be collected in a standardized method to facilitate specific diagnosis which can lead to rational preventive and treatment programs. This data is organized according to body systems.

1. Skin Conditions

The skin conditions observed have been limited to those of parasitic origin

- Ticks and fleas affect 70-80% of the animals seasonally. Infection is at peak during the wet season in the months of July to November. The effect decreases in severity when dry season returns.
- Sarcoptic mange infestation has been confirmed in 3-4 adults.
- Maggot infestation of any wound is a common problem, most prevalent in the wet season. An impression is that lions are more susceptible to maggot infestation of wounds than other felids. Fly species involve blue and green bottle fly, domestic fly, and horsefly.

II. Gastrointestinal system

A. Parasites.

- Roundworms of unknown species have been seen in the stomach in some animals on postmortem examination.
- Intestinal: Confirmed species include *Ancylostoma spp.* – continuous infection in adults despite present parasite treatment programs. *Taxascaris leonina* – particularly common in young animals: no disease associated. Coccidia – common, no disease associated, all ages are apparently infected. *Taenia spp.* – occasionally encountered.

B. Inflammatory conditions:

- Feline distemper has been confirmed causing enteritis in two adults
- Necrotic enteritis confirmed in one 6 day old cub
- Hemorrhagic enteritis has been confirmed in 10 cubs three to four weeks of age. *Staphylococcus albus* and *Streptococcus spp.* have been found to be associated with this condition.

- Ascites has been seen in both captive and free-ranging animals, both *chylous* and

transudative, involving two adult males, four adult females, and one juvenile female. One case was with a foreign body penetration of the small intestine and peritonitis and one case was with an abdominal liposarcoma (hemorrhagic ascites)

- Trichobezours have been seen in the stomach of two adult and one juvenile.

III. Nervous system:

- Viral meningitis, unknown agent, has been confirmed in one juvenile female.
- Rabies has been confirmed in two adult females.
- Suppurative meningitis/ encephalitis of unknown origin have been confirmed in one adult male.
- Posterior weakness/ paresis/ paralysis has accounted for death in six adult free-ranging animals. This condition is possibly associated with trauma.

Special Concern:

In Sakkarbaug Zoo, 30% of the cubs born exhibit signs of dysfunction associated with central nervous system. These include staggering gait, muscle stiffness (especially in the neck and forelimbs) and convulsions. Mortality is 100% in week old animals. Some animals retain ataxic signs for life. Postmortem examinations revealed no histopathologic lesions in the nervous tissue. This condition has been seen in some 30 animals in Sakkarbaug Zoo.

IV. Respiratory system:

A. Parasites:

Pulmonary pathology has been attributed to *Paragonimus westermani*, *Taxascaris leonina* larval migration and *Dictyocaulus viviparus* juvenile and adults have been seen on routine fecal examination.

B. Inflammatory conditions:

- Pneumonia has been confirmed histologically in four adults and eight juveniles, presumptively of bacterial etiology.
- Mycobacterial pulmonary disease has been confirmed in five adults. The species identify of the causative agent is unknown.
- Pyothorax confirmed in one adult associated with osteomyelitis
- Three instances of asphyxiation due to large meat chunks blocking the laryngeal opening.

V. Urinary system:

- One case of interstitial nephritis in a 6 day old cub, gram negative *Coccobacillus* isolated. Associated with septicemia.
- One case of toxic nephritis in a 2 day cub, *Streptococcus* and *Staphylococcus* isolated. Associated with hemorrhagic enteritis.
- One case of renal necrosis in an adult. Associated with septicemia.

VI. Hematopoietic system

-Acute splenitis, lymphoid depletion has been confirmed in two cases involved with acute multisystem involvement.

- Chronic splenitis with atrophy and fibrosis has been confirmed in four free-ranging adults associated with multi-system disease.

VII. Musculo-skeletal system

- One case of degenerative and coagulative necrosis of scapular muscle associated with generalized disease.
- One case of limb fracture leading to osteomyelitis and death.

VIII. Reproductive system

- No reports.

IX. Endocrine system

- No reports.

X. Cardiovascular system

- Two cases of myocarditis, myocardial hemorrhage associated with multi-system involvement

IX. Trauma

- Generalized trauma in captive animals, neonates – 10 cases involving nonspecific trauma
- Skull fracture; two adults; one free- ranging, one captive
- Spinal trauma: two adults, free ranging
- Bullet wound: One free – ranging adult
- Railway accident: two free- ranging adults
- Poisoning: One free – ranging adult, zinc phosphide
- Drowning: One free ranging adult.

In summary, it is evident from the available data, that there are three areas of future focus:

1. Parasitic conditions, particularly the area of myiasis
2. Nervous system conditions of:
 - a. Posterior paresis/paralysis in free-ranging animals
 - b. Disorders of central nervous system in young animals in captivity.

Prospects of Veterinary Research on Asiatic Lion

Additional report of the Disease working Group

The disease group looked into the possibilities of initiating veterinary research on captive as well as wild populations of the endangered Asiatic Lion. Research projects were set up based on the inferences drawn from the compilations of the disease aspects of this subspecies. The group feels the necessity of employing a full time researcher with veterinary background on this long term project and also establishing a complete disease diagnostic laboratory at Sakkarbaug Zoo which is located about 60 km away from Gir. This place has been chosen because of 2 reasons.

1. Much of the research is going to be on the captive population which is largely confined to the Sakkarbaug zoo and

2. The zoo itself has been involved either directly or indirectly on aspects of research on this subspecies.

It will be also appropriate to establish a small laboratory facility at Sasan itself primarily for preserving the collected specimen.

For convenience, we classify the research topics identified by the working group into those that could be conducted in the wild and those projects that could be carried out in captivity.

A. Research projects of particular attention in free ranging population at Gir

The different projects under this category will be based on the broad whelm of 'lion health monitoring and disease surveillance'. This includes parasitological, serological and clinical pathological investigations looking into the prevalence of infectious diseases.

1. Investigation on the prevalence of macroparasites.

This can be carried out by the collection of fresh scats from the wild. Collected scats will be preserved in 5-10% formalin and examined for parasite ova using standard methods. Both species composition and parasitic load will be taken into consideration, besides seasonal variation.

2. Investigation on the prevalence of antibodies against specific microbial infections

Serological investigations on the prevalence of antibodies to determine previous exposure to some of the common feline diseases needs to be initiated. This could be done first on the captive population. This analysis should include feline panleukopenia, feline infectious peritonitis, feline rhinotracheitis, papilloma virus, rabies, feline immunodeficiency virus and perhaps canine distemper also.

3. Research on posterior weakness

This fatal disease of unknown etiology has so far been the major cause of large scale deaths in free-ranging lions. Gir lions must be closely monitored for posterior weakness and diseased animal should be subjected to complete diagnostic investigation.

B. Research projects of concern in captive population

1. Establishing the normal physiological values of the Asiatic lion

Available data on the normal physiological values of this species comes almost exclusively from the African subspecies. It is essential that an elaborate study be conducted to establish the normal values of blood constituents and chemistry. This study should also include urine analysis.

2. Research on the probable causes for juvenile mortality

It was found, during the working session that about 30% of the lion cubs below 6 months of age die because of a mysterious nervous syndrome. The biological specimens that are going to be collected for this purpose should either be subjected for investigation within the country or if not possible, outside. Specimen that should be collected includes parts of brain, visceral organs and other tissues in different preservatives, whole blood and serum under various methods of preservation.

A second major cause of juvenile mortality is trauma during the first two weeks of life. Post-parturient management practices will be reviewed and possibly modified to minimize this cause of neonatal death.

3. Investigation of epidemiology and therapy of myiasis (maggot infestation)

This condition is a common finding secondary to traumatic wounds in captive and free-living lions. Various therapies need to be evaluated so that a single treatment can be curative. Anatomical and behavioral characteristics of the species should be studied to answer the question as to why Asiatic lions are more prone to this problem than other species of felids

Reproductive and Genetic Research

Report of Working Group

Group members: Ramesh Sabapara, Suresh Chug, Bharat Jethva, Budhan Pukazhenti, David E. Wildt, Terri L. Roth

Introduction:

The urgent need for maintaining biological and genetic diversity can be met using science and technology. For *ex situ* (zoo) breeding programs, assisted reproduction such as artificial insemination, embryo transfer, etc. can be used for overcoming problems associated with sexual incompatibility, cases of organic infertility and aged or under-represented founders unable to contribute to species preservation. Also important would be the ability to meet various breeding recommendations with fewer complications, for which transporting frozen semen, rather than living animals, would prove less stressful and costly.

Perhaps of greater importance, the use of reproductive biology also can help preserve species in wild habitats. The most profound impact could be achieved by developing systematic “genetic resource banks” (GRB’s) in the range state (that is, India), which are repositories which store germplasm, blood product, tissues and DNA. Organized collection and cryopreservation of spermatozoa from selected, free living lions (or any species) for example, would permit ‘snapshot’ storage of existing diversity. The effect would be to provide insurance against future human-induced or natural catastrophes capable of reducing genetic diversity further, or eliminating entire species.

Because of habitat fragmentation and the resulting isolation of populations, these germplasm reserves also could be used for transferring genetic vigor (i.e. interchanging genes by periodically capturing and artificially inseminating females from one isolated population using sperm from another). Each animal is producing surplus germplasm, and, given that this material is on reserve, there is no longer a need to supplement zoo populations with animals from the wild. In essence, reproductive technology can assist in keeping wild populations healthy and numerous. The most important by-product would be the simple existence of a wild population which, in turn, justifies and promotes habitat protection.

An additional benefit would be allowing the exchange of reproductive products between all living populations – both *in situ* and *ex situ* – especially transporting wild germplasm to invigorate captive populations that have become genetically stagnant.

Finally, sperm (and eventually embryo) banks could help resolve the major crisis now facing zoos, i.e. lack of space. Biologists generally agree that zoos have insufficient space for conserving all the species. This problem could be alleviated in part, by maintaining portions of needed genetic diversity in liquid nitrogen there by reducing the number of living animals required and freeing space for other species at high risk.

It is possible to use assisted reproductive technology and fresh or frozen-thawed germplasm to generate offspring in rare felid species. To date, offspring's have been produced in 5 felid species (leopard cat, cheetah, tiger, puma and clouded leopard) using laproscopic artificial insemination. The rate of these successes is directly proportional to the amount of fundamental (basic) knowledge available for each species of interest. It now is well established that characteristics unique to a particular species often prevent direct application of technologies developed in closely related species. However, given a sound database, all evidence suggests that basic reproductive knowledge can be translated into living young. Our ability to produce offspring in several felid species and multiple pregnancies in the cheetah support the assertion that these technical approaches hold exciting promise, including for conserving the Asiatic lion.

A Strategic Reproductive Research/ Management Plan for the Asiatic Lion:

The Research Working Group recognizes that the essence of ensuring a self-sustaining captive population of Asiatic lions depends upon developing a clear understanding of all of the factors regulating reproductive success. There are two urgent needs:

1. To begin some basic reproductive research projects in India that will generate normative data for the Asiatic lion subspecies. For example, it is necessary to determine if females or males have seasonal peaks in fertility and also the length of the estrous cycle, etc. This information is crucial for being able to distinguish fertile from infertile individuals and for developing, refining and implementing assisted reproductive techniques like artificial insemination.
2. To make possible the use of assisted techniques and genetic resource banking in the range country, that is, India, for managing Asiatic lions. Once the fundamentals of the reproductive system of the Asiatic lion are understood fully, then assisted reproduction can be used to keep the extant population healthy, genetically diverse and safe.

The Working Group took a two step approach. First, the Group identified what is known about the basic reproductive biology of the Asiatic lion. Secondly, the Group reviewed what is not known and then broadly outlined four research areas deserving high priority. Finally the working group addressed the vexed questions of pure v/s hybrid lions and genetic impoverishment.

I. Known factors in reproductive physiology of Asiatic lions

1. Electro-ejaculation can be used safely to successfully collect spermic semen samples from adult Asiatic lions.
2. General semen characteristics of ejaculates collected from wild and captive populations of Asiatic lions have been defined. Males generally produce motile sperm in the sample, and approximately half the sperm are structurally abnormal.
3. Preliminary data are available on methods for processing and cryopreserving Asiatic Lion sperms.

(*For a discussion on the significance of this finding, see the report by Wild in this document)

II. Unknown factors in reproductive physiology of the Asiatic Lion Research recommendations for four projects.

Project 1.

Basic reproductive characteristics of the female/male Asian Lion

There is a paucity of data on the basis of the estrous cycle of the Asiatic lion. Furthermore, there is no conclusive data indicating whether ovulation in the Asiatic lion is induced (i.e., occurs after mating) or is spontaneous (occurs without mating and as a result of natural hormonal surges). In particular this information is crucial to planning timed– matings (determining when to introduce a male) or for developing artificial insemination. Finally a seasonal effect on reproductive potential of the male or female lion has not been determined. Such information also will prove useful in dictating when animals should be mated or artificially inseminated. For these reasons, the group's recommended objectives for this project are to:

1. characterize the natural estrous cycle in the female lion;
2. determine if ovulation is induced (like in most cat species); and
3. define the effects of season on reproductive potential in both male and female lions.

To achieve these objectives the group recommends taking advantage of newly available technology to monitor hormonal metabolites non-invasively in excreted feces. The research project should be longitudinal with fecal samples collected from individual females at least four and as many as seven days per week for an entire year. Concurrently, behavioral observations should be recorded in an attempt to better understand the importance of behavioral cues with sexual activity (or inactivity). Fecal samples should be processed and stored frozen until analyzed for progesterone and estrogen metabolites. Similarly, fecal samples should be collected once or twice weekly from male lions that also will be electro ejaculated every 2 months (using a standard protocol) for one year. Male fecal samples should be assessed for testosterone metabolites. Ejaculate characteristics (volume, sperm motility, sperm concentration and sperm morphology) should be recorded for each male and then compared over time.

This project would provide all of the basic reproductive information necessary. Endocrine profiles would be defined for both male and female lions for the entire year. This information would allow defining the estrous cycle of the female, determining if ovulation is induced or spontaneous and identifying seasonal effects on natural estrous cycles. Additionally, the data obtained for male lions would identify and seasonal effects on testosterone levels and ejaculate quality as well as substantially increase the data base on ejaculate characteristics for male Asiatic lions. Simultaneously, there is a plan to radio collar a substantial number of free-ranging males in the Gir Forest Sanctuary. Given that this is possible, then the ideal method for assessing seasonality would be quarterly collection and evaluation of semen while the animals are undergoing routine checks of radio-collar equipment. This also would provide an excellent opportunity to begin systematically banking sperm from this collection (see Project III).

This technology would be extremely useful on-site to have in India, not only for monitoring the Asian lion, but other rare species as well. It is recommended that the Central Zoo Authority and the Wildlife Institute of India attempt to organize such facilities with appropriate institutes in India. * In preparation for establishment of an endocrine facility for wildlife in India, the group also recommends that at least one Indian scientist receive training in a laboratory experienced in felid work.

Project II

Developing technology for the cryopreservation and use of Asiatic Lion sperm and the systematic banking of Asiatic Lion sperm

Sperm cryopreservation is a prerequisite to transferring genetic material from wild to captive populations. A sperm cryopreservation protocol has been developed and successfully applied to several felid species including the cheetah, tiger, leopard cat, snow leopard, clouded leopard and African lion. However, this procedure requires using dry ice which is not available in all countries or under field conditions. This is particularly relevant to managing the Asiatic lions as one advantage of the technique would be to bank semen from wild males living in the Gir Forest Sanctuary. Remoteness would make dry ice access difficult. Therefore, the objectives of this project are to:

1. test the efficiency of the standard semen cryopreservation protocol using dry ice;
2. develop an alternative method for freezing sperm under field conditions using liquid nitrogen vapour.

Semen would be collected by electro-ejaculation from male lions in captivity, and each ejaculate would be cryopreserved using two methods. Method 1 would be the standard sperm pelleting protocol utilizing dry ice, and Method 2 would involve controlled freezing of sperm in plastic straws over liquid nitrogen vapor. Single pellets and straws from each ejaculate would be thawed rapidly and sperm evaluated for motility, longevity, structure and function. These data would be used to determine the efficacy of each semen freezing method. Results from this project would determine the efficacy of the standard (dry ice) sperm cryopreservation protocol in Asiatic lions. In addition, comparative data would be used to validate Method 2 as an alternative method of sperm cryopreservation under field conditions.

- ***At the time of this Report, the Central Zoo Authority has taken up such a project with a premier biotechnological institution in India. See Appendices for a review of the Proposal.***

The Working Group agreed, however, that until the results of Project II have been analyzed, semen collection and cryopreservation from wild Asian lions should be conducted opportunistically using the standard sperm pelleting procedure. To accomplish systematic sperm banking, there is a need to develop a Genome Resource Banking Action Plan, a written document that details how to cryopreserve germplasm, blood samples, tissues and DNA. Protocols for developing such an action plan are available through the IUCN's conservation Breeding Specialist Group (CBSG). The Working Group recommends that this approach be explored after identifying individuals and institutions in India interested in developing the plan.

A copy of the working protocol for developing a GRB Action Plan is appended in this Report.

Project III.

Developing ovulation induction techniques.

The success of assisted reproduction relies upon the ability to control ovarian function using exogenous hormones. Hormone treatments (regimens) followed by artificial insemination have allowed producing living offspring in several felid species.

There also may be occasions where (for whatever reason) a female in captivity may fail to naturally demonstrate estrus for natural breeding. Therefore, knowing how to therapeutically

stimulate ovulation could be useful in natural breeding management. However, different felid species react differently to the same kind of hormonal treatment. Therefore there is a need to systematically assess the effects of these treatments in each species/subspecies including in the Asiatic Lion. This may be particularly important and interesting because of the historical lack of genetic variability in this population. Therefore, Project III objectives are to:

1. determine optimal hormone dosages for stimulating ovarian activity resulting in ovulation;
2. define the time of ovulation following hormone treatment.

A hormone regimen already developed in the African lion would be applied to female Asiatic lions in captivity. This regimen would consist of two hormone injections, pregnant mares' serum gonadotropin (PMSG) and the human chorionic gonadotropin (HCG) administered at specific time intervals. Females would be subjected to laparoscopic examination following HCG to evaluate ovarian response (follicular development and ovulation). Based on these results, the hormone dosage and/ or timing of laparoscopy would be modified until an appropriate response for artificial insemination is achieved. The Working Group recommended this project be conducted using surplus hybrid animals. Once the optimal hormone regiment has been developed, then it would be tested and directly applied to pure Asiatic females.

Results from this study would determine the optimal ovulation induction regimen required for conducting artificial insemination in Asiatic Lions. At the same time, it would provide training opportunities in laparoscopic procedures to researchers in India. It is critical that this training occurs prior to artificial insemination attempts in valuable, pure bred Asiatic lions.

Project IV.

Developing, testing and applying artificial insemination techniques

There is great potential for artificial insemination as a management tool for maintaining genetic diversity in populations of Asiatic lions. This technique would eliminate the need for transporting animals between zoos and the removal of lions from the wild. It would be less costly and would allow safe and effective transfer of genetic material. The objective of Project IV is to integrate all of the newly learned information gained from Projects I – III to conduct successful artificial insemination in pure bred Asiatic lions.

The hormone induction regimen developed in Project III would be applied to female Asiatic lions. These females would be subjected to laparoscopic examination to determine ovarian responses to the hormone treatment. At this time, animals that exhibit the appropriate ovarian response would be artificially inseminated using an intra-uterine laparoscopic procedure already proven successful in several felid species. Initially, all inseminations would be performed using freshly collected semen from captive purebred Asiatic lions. After proving the technique by the production of offspring, subsequent artificial insemination attempts would be performed using frozen-thawed sperm.

Upon completion of Project IV, a standard AI protocol would be available for assisting in managing captive populations of Asiatic lions within India as well as in cooperative breeding programs in other regions. The completion of Project IV will end the first phase of developing assisted reproduction for the systematic management of captive Asiatic lions while complementing and ensuing the ongoing efforts to conserve lions *in situ*.

3. Issues of Pure v/s Hybrid and Genetically Impoverished Asian Lions

The Working Group discussed issues relating to identification of pure v/s hybrid lions. Current molecular techniques are unavailable for absolutely distinguishing a pure from a hybrid lion. Therefore, the working group suggests the following methodologies for addressing the problem.

Establish a Breeding Protocol * such that all future captive breeding involve only

- a. Lions now living in the Gir Forest Sanctuary which have to be brought into captivity.
- b. Lions now living in the Sakkarbaugh Zoo.
- c. Lions which have been translocated from the Sakkarbaugh Zoo/ Gir Forest Sanctuary to other zoos that have been positively and permanently identified before transfer.

Optional

- d. Animals that have been transferred from Sakkarbaugh/ Gir that have been appropriately tested using available genetic techniques in the presence of appropriate management techniques and records. For lions to meet criteria "d", the lions must have been isolated in the host zoo from other lions of questionable or unknown origin. These lions also must be genetically tested to exclude the possibility that they are hybrids. It should be stated, however that probabilities of maintaining purity decrease with the use lions in this set.
- e. Animals transferred from the Sakkarbaugh Zoo/Gir Forest Sanctuary and their first generation offspring that have been appropriately tested using available genetic techniques in the presence of appropriate management techniques and records as per criteria "d".

1. Undertake a systematic breeding program to reconstitute the genetic diversity of the Asiatic Lion.

If Asiatic lions are defined on the basis of an invariant genotype, then restrictive breeding programs will perpetuate the lack of genetic diversity in this subspecies. In African lions, polymorphism in the Transferrin locus is associated with variation in resistance to various forms of infection. Studies of the mammalian immune system show that many alleles are maintained after speciation, thus the large number of variants may in themselves be valuable to the fitness of individuals in the population.

The lack of diversity in Asiatic lions probably results from the recent bottlenecks in the wild population and be especially susceptible to diseases. It may therefore prove valuable to identify important alleles lacking in Asiatic lions and undertake careful hybridization with a select set of African lions to restore the missing alleles.

It is noted that Dr. Craig Packer conveyed a strong dissenting opinion from the PHVA at large in favor of deliberate and rational hybridization of African and Asiatic lions for improving the gene pool. Plan B presents a case for adding heterozygosity from the African lion to the Asiatic population to increase its long term viability.

****At the time of this Report, the Central Zoo Authority has prepared a Studbook and held a meeting of Chief Wildlife Warden and Species Coordinator for Asiatic Lion for preparing a plan or preserving genetic purity of the Asiatic lion. See Appendices for a Report of this meeting and for the Studbook details.***

POSSIBLE SCENARIOS FOR DEALING WITH THE ISSUE OF IDENTIFYING PURE VS. HYBRID. ASIATIC LIONS

Addl. Report of Reproduction Working Group.

Current molecular techniques are unavailable for absolutely distinguishing a pure from a hybrid lion.

Therefore, the working group decided that there were two possible scenarios to consider. In addition, two others were added for consideration.

1. All future captive breeding will involve only those lions:
 - a. now living in the Gir Forest Sanctuary and scheduled to be brought into captivity,
 - b. now living in the Sakkarbaugh Zoo and
 - c. which have been translocated from the Sakkarbaug Zoo/ Gir Forest Sanctuary that have been permanently identified before transfer.
 2. All future captive breeding will involve lions:
 - a. now living in the Gir Forest Sanctuary and scheduled to be brought into captivity
 - b. now living in the Sakkarbaug Zoo
 - c. have been translocated from the Sakkarbaug Zoo/ Gir Forest Sanctuary that have been permanently identified before transfer, and
 - d. animals that have been transferred from Sakkarbaug/ Gir that have been appropriately tested using available genetic techniques in the presence of appropriate management techniques and records. For lions to meet criteria “d” the lions must have been isolated in the host zoo from other lions of questionable or unknown origin. These lions also must be genetically tested to exclude the possibility that they are hybrids.
-
2. All future captive breeding should involve lions which:
 - a. now living in the Gir Forest Sanctuary and scheduled to be brought into captivity
 - b. now living in the Sakkarbaug Zoo
 - c. have been translocated from the Sakkarbaug Zoo/Gir Forest Sanctuary that have been permanently identified before transfer and
 - d. animals transferred from the Sakkarbaugh Zoo/Gir Forest Sanctuary and their first generation offspring that have been appropriately tested using available genetic techniques in the presence of appropriate management techniques and records. For lions to meet criteria “d” the lions must have been isolated in the host zoo from other lions of questionable or unknown origin. These lions must be genetically tested to exclude the possibility that they are hybrids.

3. Undertake a systematic breeding program to reconstitute the genetic diversity of the Asian lion. If Asiatic lions are defined on the basis of an invariant genotype, then restrictive breeding programs will perpetuate the lack of genetic diversity in this subspecies. In African lions polymorphism in the transferrin locus is associated with variation in resistance to various forms of infection. Studies of the mammalian immune system show that many alleles are maintained after speciation; thus the large number of variants may in themselves be valuable to the fitness of individuals in the population.

The lack of diversity in Asian lions probably results from the recent bottlenecks in the wild population and be especially susceptible to epidemic. It may therefore prove valuable to identify important alleles lacking in Asian lions and undertake careful hybridization with a select set of African lions to restore the missing alleles.

In Summary, scenario 1 insures genetic purity in the Asiatic lion population as it exists today in the Gir Forest. Scenarios 2 and 3 have safeguard measures included to preserve the genetic integrity of the population by, probabilities of maintaining purity decrease with each step. Scenario 4 presents case for adding heterozygosity from the African lion to the Asiatic population to increase its long term viability.

Eco development Report of the Working Group

Members : M.A. Rashid (Facilitator), Suresh Chugh, H.V. Asari, Mr. Nitin Pandya, Mr. Neel Gogate, Mr. Sandip Diwan, Mr. D.S. Narve

The eco-development program should be planned and executed with the interactions of local people. The following items have been proposed:

1. Grass fodder development:

Rain-fed plantation: 500 ha, Irrigated plantation: 200 ha. Treatment of gauchars (grazing land), wastelands, reserve forests, and unclassed forests should be carried out through soil and moisture conservation measures to increase fodder production. Stall feeding should be promoted.

2. Soil-moisture conservation measures:

Constructions of check-dams, gully plugs, village ponds and percolations tanks, gradient along contours with planting of suitable fodder and browse species on the bunds should be carried out.

3. Energy-related activities:

Bio-gas plants (5000 No.) for utilization of solid waste. Construction of improved crematoria to economize on fuel wood. Smokeless chullah in those villages where the bio-gas plant installations is not feasible. Provision of solar cookers and stand-alone solar power light. Setting up of wind-mills to make use of wind energy for lifting water, particularly along the coastal belts where wind velocity is favorable.

4. Employment generation activities:

Development of suitable cottage industries based on locally available raw material (eg. handicrafts, agarbattis (incense sticks) papad, soap and candle making, leather work, apiary, pisciculture, poultry farming, medicinal plants, etc.) Employment of local people for maintaining fire lines and for fire watching during the dry season. Lantana eradication program.

5. Regular program of immunization of livestock in and around Gir.

6. Provision of separate water troughs for domestic livestock in the peripheral areas to reduce pressure on the water supply in natural areas and hence decrease spread of cattle related diseases to wild ungulates.

7. Relocation of *Maldharis*:

The relocation of remaining 361 families residing presently in the Protected Area should be done. The relocation will include provision of 20 hectares per resettlements site, shifting of house hold effects, construction of approach roads, electrification, water supply (tube-wells) school building, dairy development (establishment of milk co-operatives, linking of each village to the Dairy Development Board centre at Talala, improvement of cattle breed through artificial insemination, phasing out of scrub and surplus cattle within a defined time frame, and propagation of improved fodder varieties). Difficulties and grievances of the *Maldhari* families already relocated should be properly attended to and resolved, as this will help to counteract any adverse publicity prevailing against the relocation project.

8. Community Development :

This scheme should involve 14 forest settlement villages and 3 other villages occupied by Siddi (Negro) community and villages within 5-6 km radius from the Protected Area. Provision of a community hall, sports facility and infrastructure, approach roads, nature education and orientation centers, organizing cultural functions, audio-visual publicity etc. so as to build trust of wildlife authorities.

9. Facilities:

Water pumping sets for water supply at resettlement sites, electric fencing for crop-protection, mobile grain shops, mobile dispensaries, publicity van.

10. Eco-tourism:

Ecologically friendly tourism should be encouraged and should be properly managed so as to encourage environmental awareness and affection towards wildlife conservation. With a view to divert the tourist traffic which at present is concentrated solely at Sasan, it is proposed to develop a few more tourist centers so that the impact of tourism is more equitably distributed over the Protected Area. Some locations suggested for this purpose outside the Protected Area are Jamwalah in the South, Tulshishyam/Dhari in the East and Satadhar in the North.

11. Local NGO's

Local NGO's (Panchayat, AKRSP, BAIF, and NNEF) should be involved in the project. The project should be initially implemented in a fixed set of villages and based on the results; new blocks of villages should be gradually integrated into the program.

12. Research monitoring and evaluation:

Research relating to eco-development could be undertaken by the wildlife division. The basic data prior to implementation will be collected with the help of Panchayats and NGO's. For monitoring and evaluation a small committee of officials and non-officials should be formed.

Recommendations

The workshop recommends the formulation of an elaborate, adequately funded eco-development covering the Gir Sanctuary/National Park and its surrounding settlements and villages and covering the following activities.

1. Grass and fodder development with special emphasis on encouraging stall feeding.
2. Soil and moisture conservation measures in the catchment areas coupled with planting aimed at increasing fodder and browse supply.
3. Energy related activities such as biogas plants, improved crematoria, smokeless Chula, solar cookers and power lights, windmills for lifting water, etc.
4. Employments generated activities such as environment friendly cottage industries based on local raw materials, running training courses for such training activities, eradication of *Lantana* weed in the fringe areas.
5. Regular programme of immunization of domestic livestock and around Gir.
6. Relocation of remaining *Maldharis* and resolving the grievances of those already relocated.
7. Community development programme covering forest settlements and siddi (negro) villages
8. Eco-tourism
9. Arrangement for proper monitoring and evaluation of the programme on a continuing basis.

Education and Public Awareness Report of the Working Group

Members : D. S. Narve, Saman, Rohit Vyas, Bharat Jethva, B. Rathinasabapathy, P. Manoharan, Zaida Jacob, Mihir Dave, Neel Gogate, Swetan Bhatt.

Nature education activities were started in Gujarat in 1978 and have been going effectively. The working group recognizes the efforts of the many nature clubs and other NGO's and NGL's who have been working in and around Gir.

Recommendations for nature education program

1. **To create awareness in the local community** of the value of the biodiversity in Gir forests, its benefits and the dangers of losing it.
2. **To educate the village leaders** about the problems of Gir so that a "Forest Protection Team" can be organized and their support and services can be utilised in organising and implementing various measures for solving the conservation problems.
3. **To recognize the persons and groups** working very well so that they will be motivated to work more effectively. These identified persons may be provided training in education and interpretation to other volunteers.
4. **To conduct nature education activities** throughout the year using appropriate methodologies according to the season and regional, annual public events.
5. **To identify the most effective media** for imparting nature education to different target groups and provides them with attractive and accurate baseline information.
6. **To bring about ecological awareness** and provide more favorable attitudes towards environment and natural resource conservation and management with active participation from the community. Input from community leaders, school students, local industrial houses, religious and political leaders and other association and staff desirable for achieving educational goals.
7. **To evaluate the effectiveness of the extensive nature education** activities which have taken place to date to find ways of enlarging and improving it further. Target groups should be selected for concentrated attention.
8. **To conduct all activities** with respect to the customs, traditions, religion and the way of living of the people of Gir.

Suggested target groups:

1. School children (between 13-17 years of age)
2. Local Community particularly targeting leaders.
3. Politicians
4. Press
5. *Maldharis* and people undergoing rehabilitations scheme

Role of NGO's:

The role of NGO's in public education should be defined. Creation of a network of all the NGO's all over the region will enhance the regular exchange of ideas and activities being taken up by them. The 125 Forest Youth Clubs, and other dedicated individuals and groups working effectively in Gujarat should be utilized fully for awareness work. The authorities should design the formula or process for identification and selection of the NGO's, evaluation of the work done by each NGO, and for suggesting further improvement in their work.

P.H.V.A. Workshop for Asiatic Lion

Panthera leo persica
18-21 October 1993, Baroda

Report

May 1995

Section III

Reference Materials and Appendices

P.H.V.A. – LIST OF PARTICIPANTS

Mr. S.C. Sharma
Member Secretary
Central Zoo Authority
Barrack No. 4, Bikaner House
Shajahan Road
New Delhi – 110 011

S.C. Dey
Addl. I.G. Forests
Ministry of Environment &
Forests, Paryavaran Bhavan
C.G.O. Complex, Lodi Road
New Delhi – 110 003

Mr. Pushp Kumar, I.F.S., Retd
Zoo Consultant
House No. 8-3 -33/97
Kamalapuri
Hyderabad – 500 879
Andhra Pradesh

R.L. Java
Chief Conservator of Forests
Social Forestry
Kothi Building
Vadodara, Gujarat

Shri. D.S. Narve
Dy. Conservator of Forests
Extension Division
Multi Story Building
Bhavnagar, Gujarat

Shri. Bharat Pathak
Dy. Conservator of Forests
Gujarat Ecology Commission
Narmada Bhavan Gerri Circle
Alkapuri, Vadodara, Gujarat

D.M. Naik
Chief Conservator of Forests
Wildlife, Gujarat State
Kothi Building, Vadodara

Dr. J.H. Desai
Wildlife Institute of India
Post Box No. 18, Dehra Dun
Uttar Pradesh – 248 001

Nicholas Ellerton
North of England Zoological
Society, Chester Zoo
Chester CH2 1LH, U.K

Dr. Sujit Chakraborty
Zoological Survey of India
'M' Block – New Alipore
Calcutta – 700 053
West Bengal

Sandip S. Diwan
The Baroda Nature Lovers Assn
24 "Amancio", Suvas Colony
Fatehganj, Baroda, Gujarat

Mike Fouraker
Intl. Studbook Keeper and
Species Coord for Asiatic Lion
Fort Worth Zoo
1989 – Colonial Parkway
Fort Worth, Texas – 76110, USA

Manoj Kumar Misra
Indian Institute of Forest Mgt.
Administrative Officer
Post Box No. 357, Nehru Nagar
Bhopal, Madhya Pradesh

Vijaya Kumar Pillai
Asst. Curator
Singapore Zool. Garden
Mandai Lake Road
Singapore – 2572

R. Raju
Zoo Authority of Karnataka
Sri Chamarajendra Zool. Garden
Mysore 570 010, Karnataka

M.A. Rashid, I.F.S., (Retired)
103, Jetan Apartments
Fatehganj CAMP
Baroda – 390 002

P.P. Raval, Director
Sakkarbaug Zoo
Junagadh, Gujarat

Douglas M. Richardson
Zoological Society of London
Regent's Park,
London NW1 4RY, England, UK

Diwakar Sharma
Wildlife Institute of India
Post Box No. 18, Chandrabani
Dehra Dun, UP. 248 001

Vinay Kumar Sinha
Minister of Env. And Forests
Regional Office, West
E-3/240, Arera Colony
Bhopal – 462 016

Ajit Kumar Bhowmik
Sr. Forest Ranger
Forest Dept. Tripura
P.O. Jatanbani
Tripura 799 104

K.N. Banerjee, Curator
Nehru Zoological Park
Bahadurpura, Hyderabad
Andhra Pradesh 560 264

R.H. Sabapara
Veterinary Officer
Sakkarbaug Zoo
Junagadh, Gujarat

Dr. N.K. Vyas, Principal
Veterinary College
Anand Dist, Khaira, Gujarat

Ravi Chellam
Wildlife Institute of India
Post Box No. 18, Dehra Dun
Uttar Pradesh – 248 001

V.C. Soni, Prof.
Bioscience faculty
Saurashtra University
Bio- Science Bhavan
Rajkot, Gujarat

Ahmed Abdul Aziz
Honorary WL Warden
Aziz Bagh, Sultanpura
Hyderabad – 500 024
Andhra Pradesh

Dr. U.S. Seal
Chairman, CBSG
12101 Johnny Cake Ridge Road
Apple Valley, Minnesota 55124,
USA

Vijayaraj Jadeja, Director
Sayajibaug Zoo
Municipal Corporation
Baroda – 390 018, Gujarat

R.K. Sahu
Asst. Zoo Superintendent
Kamala Nehru Zoo
Kankaria
Ahmedabad – 380 008

Shyamal Tikader, IFS
Forest Dept of Gujarat
DCF, Gir West Division
Sardarbaug , Junagadh
Gujarat – 362 001

Ms. Nita V. Shah
Lecturer, Dept. of Zoology
Faculty of Sciences
M.S. University
Baroda – 390 002, Gujarat

Dr. N.C. Bhuva
Veterinary Officer
Talak Dist
Junagadh, Gujarat

Jhava Bharat
Chief Conservator of Forests
Social Forestry, Kothi Building
Vadodara, Gujarat

Dr. Craig Packer
University of Minnesota
College of Biological Sciences
100 Ecology Building
1987, Opp. Byford Circle
St. Paul, MN 55108 U.S.A

Bruce Brewer, Curator
Chicago Zoological Park
3300 Golf Road
Brookfield IL 60513, USA

Raju Vyas, Zoo Inspector
Shri Sayaji Baug Zoo
Municipal Corporation
Baroda, Gujarat 390 018

Sally Walker, Secretary
Zoo Outreach Organisation
Box No. 1683,
65, Bharathi Colony,
Peelamedu, - 641 004

Sanjay Molur
Programme Officer
Zoo Outreach Organisation
Box 1683, 65 Bharathi Colony
Peelamedu – 641 004

P. Manoharan
Research Fellow
Eravikulam National Park
Moonoor
Kerala

N.V.K. Ashraf, Vety Officer
Coimbatore Zoological Park
Coimbatore – 641 004

S. Rathinasabapathy,
Research Assistant
Coimbatore Zoological Park
Coimbatore – 641 004

Dakhsha Jadeja
Shri Sayaji Baug Zoo
Municipal Corporation
Baroda, Gujarat – 390018

M.J. Kacchela
Shri Sayaji Baug Zoo
Municipal Corporation
Baroda, Gujarat – 390 018

Laxmansinh S. Jadeja
Shri Sayaji Baug Zoo
Municipal Corporation
Baroda, Gujarat – 390 018

Vinod Kumar Sharma
M.C. Zoological Park
SCO No. 839-40, Sector 22A
Chandigarh 160 022
Punjab

P.S. Solanki
Dy Director
Vety. Polyclinic
Junagadh, Gujarat

Mayur Patel
Veterinary Officer
11 – Krishnakrupa Society
Jotalpur Road, Baroda

S.J. Pandit
Kamala Nehru Hill Garden Zoo
Municipal Corporation
Kankaria,
Ahmedabad 380 008

Fateh Singh Rathore
MAA Forestry Farm
Ranthambhor Road
PO Sharpur Khichipur
Sawai Madhopur, Rajasthan

Maj. K.K. Seth
P.S. Sekhvat Raj
N.S. Bundela
Thiruvengadam
P.V. Asari
Bhuva Vimal Kumar
Prithi Nambar
Dr. Parasherya
DPS Verma
C.D. Patel
Neel Gogate
Devmangal Singh
P.J. Devashri
Mihir Dave
Vinod Sarada
All from the
Forest Department of Gujarat
C/o. Office of Dept. of Forest
Baroda, Gujarat.

VETERINARY WORKSHOPS –

LIST OF PARTICIPANTS

BARODA ZOO WORKSHOP – 20 October 93

Dr. M. Navin Kumar
Dy. Director (Vety.)
Nehru Zoo, Hyderabad

Dr. K.N. Vyas, Principal
Veterinary College, Anand

Dr. J. M. Anjaria, Professor and
H.O.D., Microbiology
As Above (a/a)

Dr. R. R. Parasania , Professor
and H.O.D., Surgery, a/a

Dr. Y.G Dugwekar
Professor of Gynaecology, a/a

Dr. K.S. Principal
Professor of Pathology, a/a

Dr. B.M. Jani
Hospital Superintendent, a/a

Dr. B.M. Arora, Director
National Zoological Park
Mathura Road, New Delhi

Dr. K. R. Dale
Research and Development
Baroda, Gujarat

Dr. P.S. Solanki
Dy. Director, Veterinary
Polyclinic
Junagadh, Gujarat

Dr. R.H.Sabapara
Vety. Officer
Sakkarbaug Zoo, Junagadh

Dr. Vinod Sharma,
Dy. Director
Chhatbir Zoo, Chandigarh

Dr. B.G. Patel
Asst. Director
I.C.D.P. Baroda, Gujarat

Dr. N.G. Vyas
Consultant Veterinary Officer
Sayyaji Baug Zoo, Baroda

Dr. Mayur N. Patel
Veterinarian, Baroda

Dr. S.V. Akolkar
Anand, Baroda

Dr. V.K. Sarodia
Disease Investigation Centre
Junagadh, Gujarat

Dr. D.V. Chaubal
Asst. Director
Cattle Breeding Farm, Mandvi,
Surat Distt.
Gujarat

Dr. V.K. Soni
Asst. Director
Animal Husbandry
Vety. Polyclinic, Vadodara

Dr. K.C. Patro, V.A.S.
Nandankanan, Orissa

Dr. R. K. Sahu,
Dy. Superintendent
Kamala Nehru Zoo, Ahmedabad

AHMEDABAD ZOO WORKSHOP, 21 OCTOBER 1993

Dr. N.M. Shah, Assoc.
Prof of Microbiology
Veterinary College
Gujarat Agricultural Univeristy

Dr. G.R. Patel, M.V.Sc,
Vety. Officer
Veterinary Hospital, Ellis Bridge,
Madalpur, Ahmedabad 6

Dr. R.C. Goswami
Veterinary Officer
Animal Health Branch
Directorate, Animal Husbandry
Krushbhavan Paldi,
Ahmedabad

Dr. P.U. Desai
Dy. Director
Animal Health, Directorate of
Animal Husbandry, Ahmedabad

Dr. N.T. Mehta, Jt. Director
(Animal Husbandry) a/a

Dr. J.M. Shah
Deputy Director (A.H.)
Epidemiology a/a

Dr. H.N. Kher
Assoc. Professor
Dept of Microbiology
Veterinary College, Anand

Dr. J.J. Hasmani
Asst . Professor
Dept. of Parasitology
Veterinary College, Anand

Dr. B.V. Joshi, Dy. Manager
Sabarmati Ashram Gaushala
Bidaj Farm, Kheda Distt.
Gujarat

Dr. A.K. Misra,
Project Manager
Sabarmati Ashram Gaushala
Bidaj Farm, Kheda Distt.
Gujarat

Dr. D.R. Barvalia
Asst. Profesor
Dept. Surgery
Veterinary College, Anand

Dr. J. V. Solanki
Assoc.Prof.
Dept. of Animal Genetics and
Breeding
Veterinary College,
Anand

Dr. J.L. Mahajan,
Retd. Dy. Director
Animal Husbandry
Department of Maharashtra
state, Dhulia

Capt. Dr. Dilip Mahajan
Kankaria Zoo, Ahmedabad

Dr. H.J. Derashri
Assoc. Prof., Gynaecology
Veterinary College, Anand

Dr. S.A. Chaubal
Dy. Manager (Embryo Transfer)
Sabaramati Ashram Gaushala
Bidaj Farm, Kheda, Gujarat

Dr. N.J. Mehta, Dy. Director
(Animal Vaccine Institute)
Sector – 28, Gandhinagar
Gujarat

Dr. M.K. Patel, Asst. Director
Animal Husbandry, Animal
Health Branch, Ahmedabad

Dr. M.H. Kipani
Asst. Director
Animal Husbandry
Disease Investigation
Ahmedabad

Mr. D.D. Makwana
Dy. Municipal Commissioner
Ahmedabad Municipal
Commission
Ahmedabad – 380 001

Dr. P.A. Mamnem, Asst. Supt.
42/12, Palhak Bhavan
Baharampura, Ahmedabad 22

**BOMBAY ZOO WORKSHOP –
26 OCTOBER 1993**

Mr. G. S. Sidhu, Naturalist
Member, Zoo Advisory Board
Arathi, Vasant Road
Santa Cruz, West, Bombay

Dr. Sangeeta Vengsarkar
137, Shivaji Park, Rd. No. 2
Bombay – 400 028

Mr. S. M. Kadam
World wide Fund for Nature
D.N. Road, Fort, Bombay

Dr. Santa Kelkar
Asst. Prof.
Dept of Gynaecology
Bombay Veterinary College,
Bombay

Vivek P. Dhum
Powai Park (Zoo) Borivilli
Bombay

Dr. S.V. Rizvi
Municipal Corporation Zoo
Aurangabad, M.P.

Dr. R.P. Barhati and
Mr. J. K. Kamble
Sanjay Gandhi National Park
Borivilli, Bombay

Dr. M.S. Kerawale
Veterinary Asst. Supt
VJB Zoo, Bombay

Dr. M.V. Wani,
Veterinary Dy. Supt.,
VJB Zoo, Bombay

Sri, P.A. Naringrekar
Dy. Supt. VJB Zoo, Bombay

Mr. K.L. Velodi
Superintendent
VJB Zoo, Bombay

**PATNA ZOO WORKSHOPS –
28 OCTOBER 1993**

Dr. Raj Narayan
Prof. and Head
Department of L.P.N.
Bihar Veterinary College, Patna

Dr. Ram Babu Singh, Head
Dept of Gynaecology, a/a

Dr. M.H. Akhthar, Assoc. Prof
Dept. of Gynaecology, a/a

Dr. I.B. Prasad, Project Officer
Indo-Danish Frozen Semen
Bank, Patna

S.M. Sulaiman Quli
Dept. of Wildlife Management
Faculty of Forestry
B.A.U. Kanke, Ranchi – 6

Dr. S. Kumar, Asst. Prof.
Surgery
B.V. College, Patna

Mr. Dinesh Kumar, Director
Biological Park, Ranchi, Bihar

Mr. Amod Kumar Gautam
Range Officer, Forests, a/a

Dr. (Sr) Doris D' Souza, A.C.
Patna Women's College

Dr. Y. Rama, Zoo Vety . Asst.
Surgeon, Il G. Zoo, Visak

Priya Ranjan Suiho
Conservator of Forests
22 C, Sri Krishnapuri
Patna – 1

Ashok Kumar Singh
Asst. Conservator of Forests
Zoo, Patna

Dr. J.G. Singh, Asst. Prof
Surgery, B.V. College, Patna

Dr. Mani Mohan
Univ. Professor
Animal Genetics and Breeding

Dr. S.B. Verma
Asso. Prof Genetics
B.V. College, Patna

Mr. P.C. Mishra, Director
Sanjay Gandhi Biological park
Patna

Dr. Trivedi, I.F.S. CF WL
Mr. M.S. Kerawalw, Vety. Asst.
Supt of Forests, WL
Forest Dept. of Bihar, Ranchi

Mr. P.R. Sinha, I.F.S.
Conservator of Forests
Forest Department of Bihar
Patna

P.H.V.A, VETERINARY & HUSBANDRY WORKSHOP – HOSTS, ORGANISERS, FACILITATORS, TRAINERS & SPONSORS

HOSTS – PHVA, BARODA

Mr. S.K. Nanda, Commissioner
Baroda Municipal Corporation
Baroda, Gujarat

PHVA & Vety. Workshops

Mr. Vijayraj Jadeja, Curator
Mrs. Daksha Jadeja, W/o.
Curator
Sayyajibaug Zoo, Baroda
Gujarat

ORGANISERS & FACILITATORS

Ms. Sally Walker, Secretary
Zoo Outreach Organisation
Convenor, C.B.S.G. India
Box, 1683, Coimbatore – 4

Mr. Sanjay Molur, Pgm Officer,
Facilitator
Z.O.O/ C.B.S.G. India
Box, 1683, Coimbatore - 4

AHMEDABAD VETY. WORKSHOPS HOSTS AND FACILITATORS

Mr. S.J. Pundit
Ahmedabad Municipal
Corporation
Kamala Nehru Zoo, Ahmedabad

Dr. R.K. Sahu, Asst. Supt
Kamala Nehru Zoo, Kankaria
Ahmedabad

BOMBAY VETERINARY WORKSHOPS HOSTS AND FACILITATORS

Mr. K.L. Velodi, Superintendent
V.J.B. Zoo
Bombay Municipal Corporation
Byculla, Bombay

Dr. M.V. Wani
Dy. Superintendent
V.J.B. Zoo, B.M.C
Byculla, Bombay

Dr. M.S. Karawali
Asst. Dy. Supt.
V.J.B. Zoo, B.M.C
Byculla, Bombay

SANJAY GANDHI BIOL. PARK, PATNA HOSTS

Mr. P.C. Mishra, Director
Sanjay Gandhi Biol. Park
Forest Dept. of Bihar, Patna

Dr. Trivedi
Conservator of Forests
Forest Dept. of Bihar
Ranchi

TECHNICAL SUPPORT – PHVA

Forest Departments of the
following states: Gujarat
Madhya Pradesh, Rajasthan

Dr. U.S. Seal, Chairman
CBSG
12101, Johnny cake Ridge Road
Apple Valley, Minnesota 55124

Dr. Ravi Chellam,
Scientist S.D
Wildlife Institute of India
Chandrabani, Dehra Dun, UP

Dr. Bruce Brewer
Population Biologist
Brookfield Zoo, Chicago, Illinois

TRAINERS – VETY/ HUSB WORKSHOPS

Dr. David Wildt
Reproductive Biologist
National Zoological Park
Washington, D.C. 20008

Dr. Mitchel Bush
Assistant Director
Animal Health
National Zoological Park
Washington, D.C.

Dr. Terri Roth
Reproductive physiologist
National Zoological Park
Washington, D.C

Dr. Budhan Pukazhenth
Research Scholar
National Zoological Park
Washington

Dr. Lindsay Phillips
Professor
University of California
Davis, California

Dr. Janice Martenson
Lab Technician
Laboratory of Viral
Carcinogenesis, National Cancer
Institute, Frederick, Maryland

Michael Fouraker
Dir. of Animal Collections
Ft. Worth Zoo
Ft. Worth, Texas, U.S.A.

Dr. Susan Malard
Researcher, CRES
San Diego Wild Animal Park
Escondido, California

Mr. Doug Richardson
Collection Manger
London Zoo, Regent's Park
London

**SPONSORS AND
CONTRIBUTORS**

Baroda Municipal Corporation

Zoo Outreach Organisation

CBSG, India

American Zoo Association
Species Survival Plan for Asiatic
Lion

Purina Big Cat Survival Fund

Conservation Endowment Fund,
A.Z.A.

**ACKNOWLEDGEMENT FOR
SERVICES PROVIDED**

Premier Instruments and
Controls, Ltd., Coimbatore

Modi Xerox, Coimbatore

British Airways Assisting
Conservation, U.K.

Indian Airlines

Pioneer Mills, Coimbatore

Le Shark, Tirpur

Lakshmi Mills, Coimbatore

Maya Kamath
Artist, Bangalore

Ft. Worth Zoo, Texas

National Zool. Park/ Smithsonian
National Cancer Institute

PHOTOGRAPHS BY

Ravi Chellam
E. Hanumantha Rao
Daksha Jadeja's Father
Janice Martenson
Sally Walker
Sanjay Molur

RESEARCH PROJET SUBMITTED
TO
CENTRAL ZOO AUTHORITY
(Ministry of Environment & Forests)
NEW DELHI

MOLECULAR CHARACTERIZATION OF WILD ANIMALS
BY DNA FINGERPRINTING FOR THEIR BETTER MANAGEMENT
IN INDIAN ZOOLOGICAL GARDENS

RESEARCH INSTITUTION
Centre for Cellular and Molecular Biology (CCMB)
Hyderabad 500 007

PRINCIPAL INVESTIGATOR
Dr. Lalji Singh, CCMB, Hyderabad
CO-INVESTIGATOR
Dr. Ramesh K. Aggarwal, CCMB, Hyderabad

Duration: April 1994 – March 1997 (3 years)

RESEARCH PROJECT

- I. **Project title: MOLECULAR CHARACTERIZATION OF WILD ANIMALS BY DNA FINGERPRINTING FOR THEIR BETTER MANAGEMENT IN INDIAN ZOOLOGICAL GARDENS**
- II. Area of Research: Molecular Genetics of Wild Animals
- III. Duration: 36 months
- IV. Total cost: Rs. 27.04 lakhs
- V. Principal Investigator

Dr. Lalji Singh
Deputy Director
Centre for Cellular and Molecular Biology
Hyderabad 500 007

Head of the Institution:

Prof. D. Balasubramanian
Director
Centre for Cellular and Molecular Biology
Hyderabad 500 007

SUMMARY OF THE PROJECT

Molecular Characterization of Wild Animals by DNA Fingerprinting for their Better Management in Indian Zoological Gardens

Genetic analysis under DNA Fingerprinting has the potential to reveal the breeding structure (lineage) of wild populations as well as to elucidate the degree of genetic relationship between underlying specific individuals social interactions. This is equally applicable to captive breeding populations of rare and/or endangered species. By using the universal probe for DNA fingerprinting (Bkm) developed at CCMB, the investigators propose to carry out DNA finger print analysis of Indian lions, tigers and their hybrids, leopard, snow leopard, leopard cat, fishing cat and Jungle cat. Clouded leopard and other rare and/or endangered species being used for captive breeding in Indian Zoological parks. In each case, where ever possible, large numbers of individuals will be fingerprinted for pedigree analysis and finding out the degree of inbreeding and overall breeding behavior. The results obtained would help in designing future breeding strategies and better management practices of these animals in our Zoological parks.

MOLECULAR CHARACTERIZATION OF WILD ANIMALS BY DNA FINGERPRINTING FOR THEIR BETTER MANAGEMENT IN INDIAN ZOOLOGICAL GARDENS

INTRODUCTION

Definition of the Problem

Due to continuing large scale global destructions of forests and natural habitats of wildlife, the existence of a very large number of plant and animal species is threatened. This had led to a greater emphasis for preservation of germ plasm of both plants and animals. Under the present circumstances our hopes for preserving endangered and rare species of animals are pinned on our Zoological parks. The success of any conservation programme depends practically on novel management techniques based largely on sound scientific data. Within the past decade, India has developed innovative programmes to protect its endangered wildlife species and their habitats. Foremost among these achievements is the remarkable effort to conserve the species of Indian mugger crocodile. The central strategy of the programme had been to offset the very high level of natural mortality on crocodilian eggs and hatchlings in nature by collecting the eggs from nests in the wild, incubating them in hatcheries, rearing the hatchlings in captivity and releasing juveniles at less vulnerable stages in the life span, back into the wild habitats. But in contrast to the case of Indian mugger, for the mammalian wildlife, we have to depend mostly on captive breeding wherein the basic information regarding the reproductive biology and genetic make up of the species is of utmost value. Continuous inbreeding, in general, leads to sterility at times making a species biologically unfit for survival in the long run. Therefore, it is of great importance that pedigree of each species, bred in captivity, is meticulously maintained and as far as possible, inbreeding is avoided. Today, using the powerful techniques of molecular biology one can accurately evaluate the degree of biological relatedness level of inbreeding and gross genetic defects in any given species. Therefore, these studies would help greatly in redesigning the breeding programmes and better management practices for the rare and endangered species being preserved in our zoological parks.

Eukaryotic genomes are densely interspersed with tandem repeats commonly termed microsatellites (Hamada *et al.*, 1982, Jeffreys *et al.*, 1985, Tautz 1989) and are known to be highly polymorphic for their length (Jeffreys *et al.*, 1986, Singha dn Jones 1986; Singh *et al.*, 1998; Traut *et al.*, 1992). Most of the simple DNA motifs are composed of one to five nucleotides and can be found in the eukaryotic genomes among both transcribed and non transcribed sequences. (GT)_n repeats seem to be the most abundant microsatellites in higher vertebrates (Stalling *et al.*, 1991). (CT)_n repeats from another abundant class of microsatellites in mammalian species (Beckman and Weber, 1992). All types of microsatellites have shown variation in the numbers of repeats and display a high level of polymorphism (review by Fries *et al.*, 1990). The long homologous stretches of repetitive sequence are prone to recombination, presumably through unequal exchange at meiosis or mitosis, or through slippage during DNA replication (Smith, 1976). These recombination events result in allelic differences in the numbers of repeated units present in a hypervariable locus, and hence in length polymorphism. Because of these tendencies, the hypervariable loci show higher degrees of heterozygosity and thus provide highly informative markers for linkage analysis (Jeffreys *et al.*, 1985, Hodgkinson *et al.*, 1987). Evidence suggests that the hypervariable loci exist as families, the members of which are related by core unit of this tandem repeats and are scattered throughout the genome (Jeffreys *et al.*, 1985, Nakamura *et al.*, 1987). The cloned microsatellites when used as probes against genomic DNA, under low stringent conditions, generate a complex pattern of hypervariable bands which are individual specific and the technique is now widely known as DNA fingerprinting (Jeffreys *et al.*, 1985, Singh, 1991; Lang *et al.*, 1993). The overall DNA fingerprint pattern also lends to its exploitation as a tool to study genetic relatedness of the individuals (Burke *et al.*, 1991) verification of pedigrees (Jeffreys *et al.*, 1991), estimation of genetic variability (Turner *et al.*, 1990, Reeve *et al.*, 1990) and establishment of phylogenetic relatedness (Aggarwal *et al.*, 1993). In addition, the technique of Restriction Fragment Length Polymorphism (RFLP) is quite efficient for the identification and isolation of genetic loci affecting important quantitative and qualitative traits of an organism. The utility of RFLP markers in breeding is based on the finding of tight linkages between DNA markers and the genes that control the traits of interests (Landry *et al.*, 1987, Young *et al.*, 1988, Paterson *et al.*, 1988, Lander and Botstein 1989). This method, along with Random Amplified Polymorphic DNA (RAPD), had emerged as powerful tool to analyse the genome of various plant and animal species (Tautz *et al.*, 1986; Tanksley *et al.*, 1989; Goldsmith, 1990).

Application of DNA fingerprinting to studies of wild populations has provided the behavioral ecologist with a powerful tool with which to estimate genetic relatedness among socially interacting individuals (Burke *et al.*, 1991). Among vertebrates, DNA fingerprinting has been widely applied to studies of fishes, birds and mammals. These include demonstrations of reduced genetic variation in clonal and colonial species (Turner *et al.*, 1990; Reeve *et al.*, 1990), genetic parentage in species with varied mating systems (Westnear 1990 Tegelstrom *et al.*, 1991), and genetic relatedness in species exhibiting social cooperation in breeding behavior (Rabenold *et al.*, 1990 Amos *et al.*, 1991; Gilbert *et al.*, 1991) In crocodilian, which are an endangered group we have used our Bkm probe for genetic fingerprinting for assessing genetic relatedness for their future management in the wild as well as in captive breeding programmes (Lang *et al.*, 1993). Unique fingerprints permitted to identify individuals, assign

parentage, and reconstruct the DNA profile of missing parent. We further demonstrated that band sharing can be used as a criterion for distinguishing relatives from non-relatives (Lang *et al.*, 1993).

Bkm (banded krait minor satellite DNA), which was originally isolated from a poisonous Indian Snake (Singh *et al.*, 1979; 1980), is highly conserved and preferentially associated with the sex chromosomes (Singh *et al.*, 1980). The major conserved components of Bkm is tetranucleotide repeat GATA (Epplen *et al.*, 1982, Singh *et al.*, 1984) Singh (1991) had demonstrated that under reduced stringency of hybridization and washing conditions these repeats detect sequences distributed in the entire genome, which show hypervariable individual specific pattern. Since Bkm sequences are present in all eukaryotes but absent in prokaryotes, this can be used as a universal probe for DNA fingerprinting (Lang *et al.*, 1993, Aggarwal *et al.*, 1993). Therefore, we propose to use this probe for DNA fingerprinting in wild animals in addition to the RAPD analysis.

The proposed research, which takes advantage of the recent development in DNA technologies, promise to bring a new dimension for future management of wild animals in captive as well as in the wild breeding programmes.

OBJECTIVES

1. Molecular characterization of wild animals by DNA fingerprinting using Bkm probe.
2. Assessment of parental genome contribution in the hybrid progeny of specific large fields (e.g., lions and tigers) by following inheritance of DNA fingerprint patterns of each parent studies by Bkm probe.
3. Pedigree analysis by DNA fingerprinting.
4. Determining relatedness and the degree of inbreeding by DNA fingerprinting using the principle of band sharing.
5. Determination of major genetic defects, if any, by DNA fingerprinting.
6. Analysis of chromosomes in cases suspected to have chromosomal abnormalities.

IMPORTANCE OF THE PROPOSED PROEJCT IN THE CONTEXT OF CURRENT STATUS

The anticipated results of the project will lead to:

1. Establishment of molecular indentify of each individual which will help in the study of its reproductive behavior.
2. Determination of relatedness and the degree of inbreeding which will help in designing future breeding programmes by selecting unrelated mating partners.
3. Establishment of parental genetic contribution in the hybrid and their progeny F1 and F2 generation, which may help in a) evaluating the genetic fitness of hybrid individuals; and b) identification of DNA/RFLP markers for traits of interest like fertility, disease susceptibility, body colour, height, gait etc
4. Identification of major genetic defects or chromosomal abnormalities will help in genetic counseling.

WORK PLAN

Basic Methods

1. Species to be used:

Indian lions, tigers and their hybrids clouded leopard, leopard cat, fishing cat, jungle cat and other rare and endangered species. Blood samples of a number of male and female individuals of each species will be collected from various zoos under sterile conditions, using heparinised syringes. For chromosome analysis fresh blood sample will be brought to the CCMB for short term blood culture. For DNA isolation, blood samples can be stored in frozen conditions for indefinite period and brought to CCMB in frozen state. In those species in which breeding record is kept, blood samples will be collected from the parents and all the progenies. In the case of hybrids between lion and tiger blood samples will be collected from the parents as well as the hybrids and also from the individuals of second generation if available. In those cases where semen is available, blood will not be required.

2. Screening strategy and DNA fingerprinting with Bkm probe:

DNA will be isolated separately from each blood sample collected as mentioned above. These DNA samples will then be used to generate individual specific fingerprint patterns using Bkm probe in order to assess the degree of intra-species and inter-species polymorphism. In addition, the genetic variability / differentiation in among individuals of a species would also be evaluated using RAPD analysis and short oligo probes.

3. Analysis of inheritance of DNA fingerprinting pattern and establishment of linkage with useful traits:

DNA fingerprint patterns of the parents, and individuals from 1st and 2nd generation progenies would be compared for respective maternal and paternal contributions to the hybrid, vis-à-vis the biological fitness of the individuals (hybrids). Also, a follow up of the fingerprint patterns and /or RAPD profiles of different individuals would help in establishing close linkage relationship between specific DNA markers and traits of interest like high fertility, disease resistance, colour, size, and gait.

4. Blood cultures would be established to prepare metaphase plates of dividing cells for individuals suspected to have gross genetic abnormalities. These will then be analysed for various chromosomal abnormalities to have an idea of genetic basis for the defects evident in the animal in question.

Laboratory procedures

A. MOLECULAR STUDIES

1. DNA isolation:

Blood samples will be diluted with lysis buffer (150mM Tris-HCl, pH8.0, 50mM NaCl, 5mM EDTA and 1% SDS) and frozen at -70°C for several hours. DNA will be extracted by freeze fracture method (Singh and Jones, 1986). Finally the pellet will be homogenized in lysis buffer and incubated at 37°C in the presence of proteinase K at a concentration of 100-125 Ug/ml. The samples will be extracted twice with phenolchloroform:isoamyl alcohol (25:24:1) and once with chloroform:isoamyl alcohol. DNA will then be precipitated with $1/30^{\text{th}}$ volume of 3 M sodium acetate. The precipitated DNA will be spooled out and thoroughly rinsed with 70% alcohol, vacuum dried and dissolved in 10mM Tris and 1mM EDTA, pH 8.0

The quality of DNA will be checked by running mini gel and will be quantified by taking O.D. readings in a spectrophotometer at 260/280 nm wave length. The high molecular weight DNA samples will be used for restriction digestion.

2. Restriction digestion:

10-15 Ug of high molecular weight genomic DNA will be used for each restriction enzymes will be used to select the ones which detects maximum variability in the fingerprint. The digestion reaction would be set as per the details provided by the manufacturers. A typical reaction mixture will contain:

20 UI DNA (10 Ug, 1 Ug/2 UI)
5 UI 10 x Reaction Buffer
5 UI Enz. (-3-5 U/ml in one or two installments)
20 UI Water

Total 50 UI

The samples will be incubated for 8-12 h at an appropriate temperature. The optimum temperature for incubation for Hin fl and Alu I is 37°C and for Bst NI and Taq I 60°C . The samples to be digested with Bst NI/Taq I will be overlaid with water equilibrated liquid paraffin oil to prevent evaporation. After checking the samples for complete digestion the reaction will be terminated either by addition of 0.5M EDTA or by heathshock.

The samples will then be loaded onto a 20-25 cm long 5-6 mm thick 0.8% horizontal agarose gel in 1 X E buffer (Trisphosphoric acid – EDTA), after adding the sample loading dye containing Ficoll and Bromophenol blue. Gel will be run for 16-18 h at 60 v at 20°C in a horizontal gel tank. Afterwards, the gel will be stained with ethidium bromide (0.5 mg/ml for 20 minutes), photographed and then transferred onto Hybond-N nylon membrane (Amersham) by Southern blotting.

3. Southern blotting:

Fractionated DNA in the gel will be transferred onto nylon membrane by vacuum blotting (30mm Hg.). Depurination using 0.25 N HCl will be carried out for 30 min followed by denaturation with 0.5M NaOH < 1.5 M NaCl for 30 min and neutralization with 1.5 M Tris, 1mM EDTA for 1.5 hr (all in situ during vacuum transfer). The blots will be baked at 80°C for 2 h under vacuum.

4. Blot hybridization:

The probe to be used for DNA finger printing will be Bkm derived multicocus, single standard probe 2(8). M 13 hybridization probe primer (16 bp) will be annealed to 5' end of 2(8) SS DNA by heating for 5 min in a boiling waterbath and slow cooling to room temperature. Klenow reaction will be carried out for 1.5 hr at 15°C using ^{32}P -labelled dATP. 3 units of enzyme will be used for labeling 300 ng of SS 2 (8) DNA. The reaction will be terminated using cDTA. 10 Ug of E.coli DNA (denatured) will be added as carrier DNA and the probe will be precipitated with $1/30^{\text{th}}$ volume of 3 M sodium acetate and equal volume of isopropanol. The precipitate will be washed with absolute alcohol (three times), dried and dissolved in 400 UI of 0.1 X SSC. The specific activity of the probe is expected to be 2×10^8 cpm.

Prehybridization:

Prehybridization of the blot will be carried out in 7% SDS, and 0.5 M phosphate buffer pH 7.5 at 60°C for 2-3 hrs. The volume of hybridization mix will be 15 ml for 12 X 20 cm blot.

6. Hybridization:

The probe will be added to fresh hybridization mix and incubated with shaking at 55-60 o C for 12 hr.

7. Washing:

The blots will be removed and washed 2-3 times to remove the unbound DNA probe at 55-60 o C in 2 x SSC + 0.1% SDS (1 + SSC is 0.15 M NaCl, 0.015 M Sodium citrate pH7.0). The washed blots will be then dried exposed to an X-ray film for 36-48 to obtain auto radiogram.

8. Fingerprint analysis:

Fingerprint patterns of each individual will be characterized with respect to (a) total number of bands / fragments present (b) respective molecular weights of different bands (c) evaluation of maternal and /or paternal bands (d) species specific bands if any etc. The information obtained, then would be analysed on computer for ascertaining co-efficient of similarities / dissimilarities among relatives and non relatives, overall degree of relatedness within and among species genera.

9. RAPD analysis:

The DNA samples collected from various individuals would be used to amplify polymorphic variable DNA sequences using different random sets of primers in a polymerase chain reaction using a thermal cycler. The amplified products from each reaction/individual would then be analysed for (a) individual / species marker (b) genetic differentiation or extent to genetic variability among given sets of individuals and also to identify if possible markers specific to traits of interest.

B. CYTOLOGICAL STUDIES:

Fresh whole blood cultures would be sent in selected cases to prepare metaphase chromosome spreads which will be analysed for both structural as well as numerical chromosomal anomalies.

REVIEW OF EXPERTISE:

The principal investigator, Dr. Lalji Singh is the discoverer of B km sequences and the Indian technique and probe for DNA finger printing. He has 20 years experience in DNA technologies such as cloning, characterization and sequencing of genes, in situ hybridization, DNA protein interaction and molecular genetic analysis of sex-determining factors. By using B km probe, he for the first time, demonstrated that only a small portion of the mouse y chromosome is necessary and sufficient to convert a female into a male. For this work as well as for on DNA fingerprinting he is internationally well known.

The co-investigator Dr. Ramesh, K. Aggarwal has considerable experience in genome analysis of vertebrates by using various modern techniques such as restriction fragment length polymorphism. He has done extensive work on DNA fingerprinting of crocodilians and published papers of this subject in international journal.

The CCMB has excellent facilities for molecular genetic research both in terms of infrastructural facilities and expertise.

TIME SCHEDULE OF ACTIVITES

S.No.	Name of milestone	Expected date
		Initiation - Completion
1	Collection of blood and or semen samples from 300 individuals of different species and isolation of DNA	May., 1994 - May.,1996
2	Restriction digestion of individual DNA samples with various restriction enzyme, Southern blotting and hybridization with B km probe	Aug., 1994 - April., 1987
3	Analysis of various individual DNA samples using RAPD markers	Mar., 1995 - April., 1997
4.	Cytological analysis of selected individuals suspected to have gross genetic defects	Aug., 1995 - Dec., 1996
5.	Linkage analysis of DNA fingerprinting pattern with traits of interest	Sep., 1996 - April., 1997

REFERENCES

- Aggarwal, R K., K. C. Majumdar, J. W. Lang and L. Singh (1993) Proc. Natl. Acad. Sci., USA (submitted).
- Amos, B., J. Barrett and G. A. Dover (1991) *Heredity* 67: 49-55.
- Beckman, J. S. and T. L. Weber (1992) *Genomics* 12:627-631.
- Burke, T., O.Hanotte, M.W.Bruford and E.Cairns (1991) *UNA fingerprinting, approaches and applications* (Burke et al, Eds), Birkhauser Verlag, Bascl, pp. 154-168.
- Epplen, J. T., J. R. McCarrey, S. Sutou and S. Ohno (1982) Proc. Nat. Acad. Sci. USA 79:3798-3802.
- Frica, R, A. Eggen and G. Stanzinger (1990) (*Genomica* 8; 403-408.
- Gilbert, D. A., C. Parker, A. E. Pusey, J. C. Stephens and S. J. O'Brien (1991) *J. Hered* 82:378-386.
- Goldsmith, M. R (1990) *Seriocologia* 31: 145-155.
- Hamada, H. M. G. Pernio and T. Kakunga (1982) Proc. Natl. Acad. Sci. USA 79: 6465-6460.
- Hodgkinson, S., R. Sherrington et al, (1987) *Science* 235: 1616.
- Jeffreys, A. J., N. J. Royle, I. Patel, J.A.L. Armour, A. MacLeod, A. Collick, I.C. Gray et al(1991)*DNA fingerprinting approaches and applications* (Burke et al. eds), Birkhauser Verlag, Basel, pp 1-19.
- Jeffreys, A.J., V. Wilson, S.L. Thein, D.J. Weatherall and BAJ. Ponder (1986) *Am. J. Hum. Genet.* 39:11-24.
- Jeffreys, J., V. Wilson and S.L. Thein (1985) *Nature* 314: 67-73/
- Lander, E.S. and D. Botstein (1989) *Genetics* 121: 185-199.
- Landry, B, S., R V. Kesselei, B. Farrara and R. W. Micheltmore (1987) *Genetics* 116:331-337.
- Lang, J.W., R.K. Aggarwal, K.G. Majumdar and L. Singh (1993) *Mol. Gen. Genet.* 238: 49-58.
- Nakamura, Y., M. Leppert, P.O' Connell et al. (1987) *Science* 235: 1616-1622.
- Paterson, A.H., E.S. Lander, J.I). Hewitt, S. Peterson, S.E. Lincoln and S.D. Tanksley (1988) *Nature* 335: 721-726.
- Rabenold, P.P., K.N. Rabenold, W.H. Piper, J. Haydackadn S.W. Zack (1990) *Nature* 348: 538-540.
- Reeve, H.K., D.F Westneat, W.A. Noon, P.W. Sherman and C.F. Aquadro (1990) Proc. Natl. Acad. Sci. USA 87: 2496-2500.
- Singh, L, (1991). *Curr. Sci.* 60: 580-585.
- Singh, L., H. Winking, K.W. Jones and A. Gropp (1988). *Mol. Gen. Genet.* 212:440-449.
- Singh, L and K.W. Jones (1986) *Hum. Genet.* 73: 304 .08.
- Singh, L., C. Phillips and K. W. Jones (1984) *Cell* 36: 111-120.
- Singh, L, LP. Purdom and K.W. Jones (1980) *Chromosoma* 79: 137-157.
- Singh, L, I.F. Purdom and K. W.Jones (1979) *Chromosoma* 71: 167-181.
- Smith, G. P. (1976) *Science* 191: 528-535.
- Stalling, RL., A.P. Ford, D. Nelson, D.C. Tomey, C.C., Hilderband and R.K. Moysis (1991) *Genomics* 10: 807-815/
- Tanksley, S.D., N.D. Young, A.H. Paterson and M.W. Bonier-bale (1989) *Biotechnology* 7: 257-264.
- Tautz, D (1989) *Nucleic Acids Research* 7: 6463-6471.
- Tautz, D. M. Trick and G. A. Dover (1986) *Nature* 322:652-656.
- Tegelstrom, H., J. Searle, J. Brookfield and S. XXX (1991) *Heredity* 66: 373-379.
- Traut, W., J.J. Epplen, D. Wichenhan and J. Rohwedel (1992) *Genome* 5:659-666.
- Turner, B.J., J.F. Elder, T.F. Laughlin and W.P. Davis. (1990) Proc. Natl. Acad. Sci. USA. 87: 5653-5657.
- Young, N.D., D. Zamir, M.W.Ganaland S. D. Tanksley (1988) *Genetics* 120: 579-585.
- Westneat, D.F (1990) *Behav. Ecol. Sociobiol.* 27: 67-76.

CCMB

Centre for Cellular
and Molecular Biology,
Hyderabad



NATIONAL PARTNER IN WILDLIFE CONSERVATION RESEARCH FOR THE INDIAN ZOO & WILDLIFE COMMUNITY

This feature is to introduce to the zoo and wildlife community a new national partner in research, the Centre for Cellular and Molecular Biology. The Central Zoo Authority has given a substantial grant to CCMB to carry out a variety of biotechnical works which would enhance the potential of zoos to contribute to the conservation effort in the country.

The Centre for Cellular and Molecular Biology (CCMB), is one of the national laboratories of CSIR, the Council of Scientific and Industrial Research, the premier multi-disciplinary research organization in India, funded by the Government of India. The objectives of CCMB are in brief : to conduct research and exploratory work in modern biology and seek potential applications for it; to provide both long term and short term training, particularly for staff from other institutions which may not have facilities for such training ; to provide centralised facilities for modern techniques in interdisciplinary biology and see that they are organized and administered to be of maximum use to researchers from other laboratories and institutions; to interact with other institutions doing related work and to collect and disseminate information relevant to biological research.

CCMB Facilities

The CCMB campus is set in a well planned and cared for garden environment covering about 15 acres. The main laboratory building covers half its area and consists of four wings each with different subject areas and groups accommodated there. The group-areas are identical with very carefully structured and planned laboratory and administrative facility to maximise the potential for creative work and minimise the aggravation which eats away intellectual energy.

There are very intricately organized departments to serve the needs of all researchers. All levels of scientists and researcher have equal access to equipment and facilities.

The DIC-Bioinformatics centre has an Apex centre with nine Distributed Information Centres (DICs) and several sub-DICs located in institutions all over India. It serves as a national network providing information in arcane biotechnical subjects. A staff of information scientists, technical assistants and programmers man the centre which keeps a 24-hour schedule, staffed to help scientists with complex data analysis needs. There is on-line access to appropriate international data bases,

CCMBs own data bases and up-to-date software packages. The CCMB Bioinformatics Centre develops its own customised software for scientific and administrative uses. Network facilities and training complete the profile of the very impressive informatics centre.

The other group-facilities are the Protein Analysis Facility, Peptide Synthesis Facility, DNA Synthesis Facility, NMR Spectroscopy Facility, Laboratory Animal Facility, Cell Culture Facility, Electron Microscope Facility.

Research resources include a very wide range of instrumentation which is maintained by a specially trained group which is responsible for installation and maintenance of the all scientific equipment at CCMB. Life-support systems of cultures and samples, etc. such as refrigeration and freezer units are checked 24 hours a day by a computerised central monitoring system. With this system there is no change that valuable animal samples will spoil in a broken refrigerator that goes unnoticed over a weekend. If the temperature drops by a few degrees in any of the cold areas in the facility, it is noted by the computer and the scientists who are working with those samples notified immediately.

There is a large and well organised library which it shares with the Indian Institute of Chemical Technology. Other facilities are Fine Chemicals Group, Photography, Experimental and Engineering Services.

The main areas of interest at CCMB are Biomedicine and Biotechnology, Genetics and Evolution, Molecular Biology, Cell Biology and Development, Biochemistry and Biophysics . . . and now, for some scientists anyway . . . Conservation Biotechnology !

There is much more to tell about CCMB but suffice it to say that the Central Zoo Authority, led by the Hon, Minister for Environment, Sri Kamal Nath, Chairman has made an astute choice in selecting CCMB as a major partner in implementing its research requirements.

CCMB will not be the only research institution to get projects from CZA. Research centres in different regions of the country should also have an opportunity to participate in the zoo conservation research activities of the country and this will also be convenient for zoos located in that region. CCMB will initiate some of the major research programmes, however, and also serve as a centre for others who do not have all of the required equipment and other facilities. This is, of course, one of the overall objectives of CCMB.



**MINUTES OF MEETING HELD ON
PLANNED BREEDING
OF ASIATIC LION
AS WELL AS INDIAN WILD ASS, RUSTY SPOTTED
CAT AND NICOBAR PIGEON**



A meeting to discuss the Planned Breeding Programme for the species was held at Sasan Gir on 4-5-1995.

The Chief Wildlife Warden, Gujarat, welcomed the participants and explained that breeding of endangered species of wild animals is now to more a "hit and miss" affair but that a minimum number of founders are required to get a genetically and demographically viable zoo population. He asked Sh. S.C. Sharma, Member Secretary, Central Zoo Authority to give a detailed account of the methodology for taking up planned breeding of the four species i.e. Asiatic lion, Indian Wild Ass., Rusty spotted cat and Nicobar pigeon.

Sh. S. C. Sharma explained that for planned breeding of any species the minimum number of founders required is 20 i.e. 10 males and 10 females. Breeding of the species is to be done in such a manner that all the founders contribute equally in every generation. For species like Asiatic lion a population of about 150 animals is considered genetically viable. IN view of the possibilities of new founders coming into the programme from time to time by way of capture of man eaters and cattle lifters even a population of even 100 animals may be good enough. We may get this target population 3 generations. However, the planned breeding of lions has severe problems because of the possibilities of several males being a spermatic or sperms not having adequate motility due to broken tails etc. Therefore detailed sperm testing will have to be carried out before taking any male lion into planned breeding programme. At present Junagadh Zoo has only 6 wild males of which 3 are too young for breeding purpose. The programme can not pick-up with three wild males. Government of India and the State Government will have to consider capturing of at least 10 to 15 males for planned breeding of lions.

The position with respect to the Wild ass is even worse. There is only one breeding male in Sakkarbaug Zoo, Junagadh. Therefore capture of at least 10 males essential in this case also. Sh. R.L. Jawa, Principal Chief Conservator of Forests, Gujarat, observed that he does not see any difficulty in capturing these animals for planned breeding. It was therefore decided to request the Government of India to allow capturing of required number of animals.

As an interim measure it was decided that the two breeding males of Asiatic lion should be retained by Junagadh Zoo. One breeding male of Asiatic lion could go to Vandulur Zoo, Madras which could also be given two wild females from Junagadh Zoo. As regards to three sub-adult Asiatic lion males, it was decided these along with 3 females could go to Ahmedabad Zoo, Kanpur Zoo and Vanvihar National Park, Bhopal, provided these zoos are in a position to provide enclosures exclusively for planned breeding programme. It was also decided before the animals go to their zoos a committee comprising of a representative from Central Zoo Authority, Forest Department of Gujarat and Sakkarbaug Zoo, Junagadh may visit the concerned zoos. Each zoo will have to make provisions for maintenance of proper records like animals history sheets, treatment cards and stud-books.

Regarding Wild Ass, it was decided that the breeding male was to continue at Junagadh and the zoos like Ahmedabad Zoo and Vandalur Zoo, Madras who have lone female may await the capture of Wild as from the wild. It was suggested that a committee comprising of Dr. Neeta Shah from M.S. University, Baroda, Sh. D.M. Naik, Chief Wildlife Warden, Gujarat, Sh. Majithia, Supdt. Wild Ass Sanctuary, Dhrangadhara and a representative of Wildlife Institute of India be formed to decide the issue of capture of Wild Ass for the breeding programme.

Regarding Rusty spotted cat, no planned breeding is possible at present because no Rusty spotted cat is available in the Zoos of India. However, it was decided that CZA may work out possibilities of getting some of the Rusty spotted cats from foreign zoos.

About Nicobar pigeon only three zoos presently have this species. The largest collection of 12 animals is with Ahmedabad Zoo. Hyderabad Zoo has five birds and Haddo Zoo has one wild male. The wild male from Haddo Zoo may be given to Ahmedabad Zoo in exchange of two pairs of birds. This will help in introducing new blood into the Ahmedabad Zoo population. The three zoos should continue frequent exchange of Nicobar pigeon also.

Sh. R.L. Jawa raised the point as to why India can't we send some of the animals to foreign zoos for breeding purpose as they have adequate expertise in the field. The Member Secretary, Central Zoo Authority explained that the Indian policy is that scientific breeding for re-introduction purposes should be done in the range state. Expertise available in foreign zoos would be used to the extent possible. We are getting such help in case of species like Red Panda.

The meeting ended with the vote of thanks to the Chair.

It was decided that Central Zoo Authority may request the concerned zoos and Chief Wildlife Wardens to make it a point to attend the meeting whenever called by the species coordinator i.e., Chief Wildlife Warden, Gujarat.

List of Attendees

1. R.L. Jawa, PCCF, Gujarat
2. D.M. Naik, CCF (WL) Gujarat
3. S.C. Sharma, Member Secretary, Central Zoo Authority, New Delhi
4. B. Arulprakasam, CCF, (WL) & CWLW, Madras
5. G.K. Sinha, Assistant (Adm) Principal CCF, Gujarat
6. R.D. Kamboj, D.C.F., Gir West Div, Junagadh
7. J.P. Vasava, D.C.F. Gir East Div. Dhari
8. P.P. Rawal, Asst. Director, Geer Foundation, Gujarat
9. Bipul Chakraborty, Scientist, Central Zoo Authority, New Delhi
10. B.R. Raval, ACF, Mobile Squad (Gir)
11. B.B. Modai, ACF, Talala
12. R.H. Sabapara, Veterinary Officer, Gir & Sakkarbaug Zoo, Junagadh
13. R.D. Katara, Zoo Officer, Sakkarbaug Zoo, Junagadh
14. R.H. Sahu, Zoo Supdt. Kamala Nehru Zoological Garden, Ahmedabad
15. H.S. Singh, Conservator of Forests, WL Circle, Junagadh.

SURVEY OF POTENTIAL SITES FOR THE RE-INTRODUCTION OF THE ASIATIC LION

A.J.T. Johnsingh, Ravi Chellam, Justus Joshua, Vidya Arthreya and A. Christy William

The Asiatic lion (*Panthera leo persica*) once ranged all across the middle east from Syria to India. They occurred throughout the central regions of India, from Pakistan to as far east Bihar and north of the river Narmada. Due to indiscriminate hunting and habitat conversion by man their distribution and numbers decreased drastically. By the end of the 19th century only a small population of about 15 individuals survived in India in the Gir forests, Gujarat. Now, Gir has about 280 lions.

A small population which is present in only one site is like having all your eggs in one basket and hence in danger of being wiped out due to disease, fire or other catastrophes. A need for another home for the Asiatic lion was perceived in 1957 and three lions (1 male and 2 female) were introduced into the Chandraprabha forests in Uttar Pradesh, India. They increased in number to 11 individuals after which they were not seen and it is believed that they were either shot or poisoned. Lack of ecological information and monitoring of the introduced population combined with insufficient reserve size (We know that only now!) probably were the reasons for the failure of that attempt to establish a second free ranging population of Asiatic lions more than three decades ago.

A WII study on the ecology of the Asiatic lions was initiated in 1986 with the objective of collecting data which would be necessary for a second reintroduction attempt to succeed (data on the lion's predation ecology, habitat use and home range size). In 1990 armed with the baseline ecological information of the lions from the above study the possibility of reintroducing the lion to another site in its former range was

considered. The areas to qualify as a potential reintroduction sites should be large (approx. 800 sq.km) and require minimal relocation to people living in and around the park. After detailed discussions with experienced wild lifers, Forest Department personnel, researchers and in the deliberations at the Population and Habitat Viability Analysis for the Asiatic lion in Baroda (October 1993), three areas; two in Rajasthan and one in Madhya Pradesh were recommended and are in the process of being surveyed as potential sites for re-introducing the Asiatic lion.

The three sites are characterized by dry deciduous vegetation and water is available throughout the year. The sanctuaries and the adjoining reserve forests are being surveyed by foot and vehicle in the winter and will be surveyed again in the summer. The habitat types, relative abundances of prey animals and human disturbance in each of the above sites are being evaluated.

Sitamata wildlife sanctuary in the state of Rajasthan is approximately 180 km from the city of Udaipur. A survey was carried out from November 20 – December 6, 1993. It is around 423 sq.km. in area with about 1000 sq.km in the surrounding reserve forests. Three rivers (Sitamata, Jakham and Karmoi) run through the sanctuary. There is great potential for this sanctuary to become the best test ride circuit for rough terrain vehicles like jeeps in India. That should have given the readers a fair idea of the road conditions here. Signs of four horned antelope, nilgai and sambar and leopard were obtained. The animals seen were common langur, jungle cat, jackal, Indian fox and 150 species of birds. This site has good bamboo and teak forests but these are

highly disturbed due to people cutting wood and bamboo. Encroachment and cattle grazing are a major problem in this sanctuary and the surrounding forest.

Kuno wildlife sanctuary is about 110 km from Gwalior city in Madhya Pradesh. The sanctuary is around 450 sq.km in area with about 1000 sq.km of reserved forest surrounding it. We assessed it from January 4 – 26, 1994. We stayed at a quaint forest rest house in the heart of the Sanctuary overlooking the river Kuno. Before we reached the rest house all of us were convinced that the organizers of the Paris to Darkar car rally would definitely include this place in their route if they ever got a chance to see it (So, please keep this under your hats. We don't want speeding cars to disturb our lions). Seven species of ungulates, the nilgai, chinkara, blackbuck, sambar, cheetal, barking deer and wild boar were recorded. The area has a wide range of predators; hyena, jackals, Indian fox, jungle cat and rusty spotted cat were sighted during our survey. Pugmarks of a tiger were seen. Other animals that were recorded are the common langur, leopard, sloth bear, palm civet and about 110

species of birds. There are many villages within the sanctuary and the surrounding forests. A large number of cattle graze within the forest. In spite of the rear axle breaking down, our work was carried out without much trouble. Lalbhai's (our driver) charming voice and also disconcerting at the same time (He always spoke with a tone which assumed that you were deaf) was still ringing in our ears when we caught the train to Delhi from Gwalior. The level of disturbance in this sanctuary is way below that of Sitamata sanctuary.

Darrah wildlife sanctuary near Kota in Rajasthan will be surveyed during the months of February '94 – March '94.

We hope that this survey will generate enough data to enable us to objectively evaluate these three sites for the possibility of reintroducing lions. We also hope to come up with a realistic action plan for site preparation, reintroduction and subsequent management of the second population of Asiatic lion in the wild before the end of this year.

*Sweet is the lore which Nature brings;
Our meddling intellect
Misshapes the beauteous forms of
Things;
We murder to dissect.*

----- Wordsworth

**PROPOSED IUCN RE
SOLUTION STATEMENT ON
ANIMAL GENOME RESOURCE BANKING FOR SPECIES CONSERVATION**

**Conservation Breeding Specialist Group Annual Meeting
Sao Paulo, August 27, 1994**

PROBLEM STATEMENT

The IUCN holds that the successful conservation of species requires integrated management efforts to sustain available genetic diversity. These efforts include programs to protect and manage animal populations within their natural, native habitat (*in situ* conservation) as well as supporting programs that manage populations, individuals, gametes and/or embryos outside of natural environments (*ex situ* conservation).

The IUCN recognizes that, although habitat protection is the most desirable, first approach for conserving biological diversity, supportive intensive management programs are essential in many cases. Such programs can deal effectively with short-term crises and with maintaining long-term potential for continuing evolution.

The IUCN further recognizes that the efficiency and efficacy of intensive conservation efforts can be increased many fold by applying recent advances in reproductive technology. These include assisted or 'artificial' breeding and the low temperature storage (banking) of viable animal germ plasm, namely spermatozoa, oocytes and embryos. Germ plasm banks (more broadly defined as genome resource banks): 1) offer a high degree of security against the loss of diversity and, therefore, entire species from unforeseen catastrophes; 2) minimize depression effects of genetic drift and inbreeding; and 3) provide a powerful method for managing the exchange of genetic diversity among populations. Ancillary conservation benefits include banks for basic and applied research including repositories of serum, DNA and cultured cell lines from germ plasm donors that permit studies on disease status, detection of microbial antibodies, pedigree determination, taxonomic status, geographical differentiation of populations and cellular physiology.

The IUCN also recognizes that the establishment of genome resource banks must be matched by developing strategies for use as a genuine and practical conservation asset for supporting natural breeding. Furthermore, genome resource banks should follow specific, scientifically-developed guidelines consistent with an international standard, thus ensuring their use as a meaningful, practical, ethical and cost-effective conservation tool.

The Conservation Breeding Specialist Group of the IUCN's Species Survival Commission is charged with exploring novel approaches to assisting in the conservation of biodiversity and genetic diversity. Since 1991, the Conservation Breeding Specialist Group has been developing and refining strategies for the practical implementation of genome resource bank. These activities have included: 1) publication of scientific manuscripts on the utility of this new conservation approach; 2) development of a comprehensive Action Planning process (with explicit guidelines) to ensure that all such repository programs have conservation

application; and 3) identification and coordination of a global network of people and resources dedicated to the systematic formation of genome resource banks.

RECOMMENDATIONS

The IUCN regards the development of genome resource banks as a valuable component of integrated conservation programs. Therefore, the IUCN recommends that the Conservation Breeding Specialist Group continue to pursue developing the framework for international coordination of this type of program based upon agreements to cooperatively manage species for demographic and genetic diversity.

To achieve this recommendation:

1. genome resource banking programs, where appropriate, should be incorporated directly into the framework of other conservation action strategies including conservation assessment and management plans (CAMP process), population and habitat viability assessments (PHVA process), global/regional collection planning and recovery plans for restoring species to natural situations.
2. genome resource banks should be developed only in the context of systematic, written and detailed Action Plans, thereby ensuring that there is a defined conservation goal associated with the collection, storage and use of animal biomaterials to support natural breeding. The development of an integrated plan with clear conservation goals is the single most important consideration prior to initiating banking activities.
3. the Conservation Breeding Specialist Group, when requested, should assist taxon Specialist Groups, propagation groups for species, regional conservation programs and others in developing genome resource banking strategies and specific Action Plans, The development of the Action Plan resides with those groups with specific responsibilities for *in situ* and *ex situ* conservation of specific taxa, species and populations. The CBSG will support these activities by interlinking global/regional groups interested in genome resource banking, providing specific information on banking strategies and by integrating information: a) reproductive and genetic histories of *ex situ* and *in situ* populations; b) efficiency of reproductive/genetic technologies; c) approaches for achieving genetic management goals; d) types of biomaterials requiring storage; e) appropriate protocols for banking and using biomaterials; f) ethical issues related to biomaterials ownership/distribution; g) concerns about disease and regulations; and h) areas requiring further research.
4. a globally-standardized, record-keeping database should be developed for cataloging, pooling and managing data and transfers of banked materials. It is highly desirable that these biomaterials are linked to individually-identifiable source animals to ensure meeting the objective of assisting in managing genetic diversity.

Revision of 1991 statement.

Factors to be considered in Developing a Genetic Resource Bank (GRB) for a Taxon Species: Guidelines for Writing a GRB Action Plan

The Cryopreservation of germ plasm and embryos combined with assisted reproduction could play a vital role in the preservation of bio- and genetic diversity. At the 1991 meeting of the Captive Breeding Specialist Group (CBSG) in Singapore, a strategy was presented and approved for beginning to establish genetic resource banks (GRBs) at the species and/or taxon specialist group level (see Rall *et al.*, 1991 CBSG Meeting Proceedings for the advantages and details associated with GRBs). In brief, the species or taxon specialist group petitions the CBSG GRB Coordination Committee for assistance in developing an Action Plan, a written document that will guide all aspects of collecting, storing and using germ plasm as well as other biological materials from threatened and endangered species/subspecies/populations. The following is the format and outline to be followed in developing such written action plans.

General Components and Specific Factors of an Action Plan for a Specific Taxon/Species (Document Structure):

- I. **Summary:** Synopsis (one page) providing brief description of justification, goals and overall conservation plan in the context of a GRB.

- II. **Justification:**
 1. Provide specific short- and long-term goals for the GRB.
 2. Describe in detail how a GRB will contribute to conservation ex situ and/or in situ of this taxon/species including, if appropriate usefulness to sustainable development.

- III. **Current knowledge of life history and natural reproduction:**
 1. Assemble information on sexual maturity, reproductive senescence, seasonality, duration of the reproductive cycle, induced versus spontaneous ovulation, time of ovulation, duration of pregnancy/incubation, post-partum estrus, clutch interval, litter size/clutch, embryonic and post-natal mortality.
 2. Indicate reproductive success as influenced by genetic, nutrition, disease and management events.
 3. Describe extent of technology available for monitoring/ managing animal health and provide any available evidence for vertical transmission of diseases.

- IV. **Current knowledge of assisted reproduction:**
 1. Indicate prior success at stimulating ovarian activity and or estrous activity using exogenous hormones and drugs.

2. Indicate prior success at monitoring hormonal status using circulating blood hormones or hormonal metabolites measured in voided urine, feces or saliva.
3. Indicate prior success at cryopreserving sperm, oocytes and embryos based upon in vitro function assays.
4. Indicate prior success at artificial insemination with fresh or frozen-thawed sperm.
5. Indicate prior success at embryo transfer using fresh or frozen-thawed embryos.
6. Indicate prior success at in vitro fertilization using fresh or frozen-thawed gametes.
7. Indicate prior success at oocyte maturation followed by in vitro fertilization and transfer using fresh or thawed embryos.
8. If no information is available on species of interest, indicate prior success in each area in a closely-related species.

V. ISIS, studbook and regional collection plan status:

1. Provide information on total number of males and females in ISIS, global and /or regional studbooks and, when appropriate, in private collections.
2. Indicate demographic distribution of populations and individuals.
3. Provide priority of the species in the context of the global/regional taxon master plan.
4. Identify founders and founder-lines.
5. Prioritize individual donors including providing location, age class and reproductive history/current status.

VI. Status in the wild:

1. Indicate known or predicted animal numbers in various geographic regions.
2. Provide relevant population and habitat viability assessment (PHVA) results including the status of in situ management programs.

3. Indicate prior success (if any) at reintroduction of captive born animals to the wild.

VII. Accessibility of existing animals for banking:

1. Identify and indicate accessibility of wild populations and individuals (in situ and ex situ).

VIII. Type and amount of germ plasm (and other biological materials) to preserve:

1. Define short and long-term management and genetic goals.
2. Describe how the banking program will meet stated management and genetic goals.
3. Using computer modeling, calculate the minimum number of available individuals (beginning with founders or founder lines) to meet plan objectives.
4. Identify materials to be stored (i.e., sperm, oocytes, embryos, cell lines, blood cells, tissues, DNA, body fluids [serum, milk, urine, saliva])
5. Using computer modeling, calculate amount of material needed from available, individual founders over a specific interval to meet plan objectives.

IX. Technical germplasm collection, storage, use and ownership: If no or limited technical information is available, proceed to conduct research to satisfy the following needs:

1. When appropriate and needed, identify generic animals available for basic research purposes.
2. Safe and effective methods for collecting germ plasm, including anesthetic procedures, gamete collection, ovulation induction and estrous synchronization.
3. Established baseline gamete/embryo norms (i.e., sperm numbers, sperm morphology, quality grades for embryos) and in vitro assays for determining biological viability.
4. Procedures for ensuring known health status of donors.

5. Comparative studies examining the impact of various cryobiological factors upon post-thaw viability of required biological materials.
 - a. cryoprotectant solutions and pre-freeze processing/equilibration.
 - b. normal microbial flora associated with collected germ plasm.
 - c. pre-freeze quality of germ plasm.
 - d. freezing method including cooling techniques.
 - e. storage conditions including temperature requirements and storage containers.
 - f. warming conditions.
 - g. post-thaw processing in preparation for use, especially cryoprotectant dilution.
6. Adequate post-thaw viability as determined by:
 - a. Gross morphology/quality
 - b. In vitro function assays (i.e., sperm longevity, oocyte penetration tests, embryo development in culture).
 - c. In vivo function assays (i.e., established conceptions in conspecifics or closely related taxa following artificial insemination, in vitro fertilization and/or embryo transfer).
7. Establish criteria for the minimum acceptable viability of germ plasm (an other biological materials) after thawing to meet management/genetic needs (i.e., minimum numbers of motile, undamaged sperm or minimum embryo quality grade capable of resulting in conception).
8. Potential for using frozen germ plasm with other reproductive biotechniques including interspecific embryo transfer, sperm microinjection, zona piercing, cloning, sexing, assisted embryo hatching and ultrasound-assisted aspiration and deposition.
9. Understanding the impact of disease(s) on the effectiveness and safety of banking and using biological materials.
10. Assembled information on research resources (i.e., current investigators/institutions and new/ongoing research findings) available in a computerized database.

If cryopreservation technology is available, proceed to formal banking:

1. Indicate that optimal technology is to be used based upon previous empirical studies.

2. Describe how health status of donors is to be determined to prevent disease transmission via movement of germ plasm or other biological.
3. Assemble the information to precisely identify all stored aliquots of each biological material in a central database system.
4. Establish and describe a labeling procedure containing key/coded information that is placed upon each stored sample container.
5. Identify primary and secondary (back-up) locations for stored materials and the database.
6. Describe quality control program to be used ensuring that the following is included:
 - a. periodic post-thaw viability checks of frozen materials.
 - b. a system for routine examinations of donors for disease.
 - c. multiple alarm systems to monitor security from unauthorized access and to ensure proper function of all low-temperature refrigerators and safety equipment.
 - d. back-up power generators to GRB act continuous of alarm systems, safety equipment and low temperature freezers.
 - e. back-up storage space on-site in the event of individual freezer failure.

For use of stored materials:

1. Describe a plan that allows frozen materials to contribute to conservation and genetic management including information on:
 - a. individuals and institutions allowed access to the biological materials.
 - b. how biological materials will be released.
 - c. how the various biological materials will be used.
 - d. the geographic region of use including the potential problems associated with import/export restrictions (i.e., disease transmission).
 - e. proper storage and handling of biological materials after release.
2. Indicate the circumstances under which stored materials will be provided.
3. Indicate the circumstances under which stored materials will be provided free-of-charge or sold.

4. Indicate the strategy, preferably a computerized data base, for assembling and disseminating follow-up information on the usefulness of the distributed materials.

For ownership of stored materials:

1. Determine ownership (individuals or partnership) of frozen biological materials (i.e., institution owning donor, the taxon specialist group or country of animal origin).
2. Determine ownership of offspring resulting from the use from stored germ plasm.
3. Define how patents resulting from research using this material will be handled.

X. Resources and funding

1. Define personnel resources and expertise available for each phase of the banking process.
 - a. Cryobiologists
 - b. Gamete biologists
 - c. Embryologists
 - d. Veterinarians
 - e. Population biologists
 - f. Molecular geneticists
 - g. Registrars (database specialist)
 - h. Captive breeding specialists
 - i. Field biologists
 - j. Representative of taxon/species coordination and management groups
2. Define facilities resources (including buildings, equipment, supplies) fir:
 - a. Personnel charged with the systematic collection of materials.
 - b. Primary storage site.
 - c. Secondary storage site(s).
 - d. The computerized database.
3. Identify sources of short- and long-term funding for:
 - a. Personnel charged with the systematic collection of materials.
 - b. Secondary storage site(s).
 - c. The computerized database.
 - d. Distributing stored materials.

4. Identify plan to assure the transferability of the stored collection if those responsibilities are unable to maintain the bank in perpetuity.

The principal authors of this report are: David E. Wildt, National Zoological Park, Smithsonian Institution, Washington, DC; Patricia Schmidt, Veterinary Resources Program, National Institutes of Health, Bethesda, MD; and William F. Rall, National Zoological Park, Smithsonian Institution, Washington, DC.

Indian Studbook for Asiatic Lion (Panthera leo persica)

WILD ASIATIC LIONS (*Panthera Leo Persica*) IN INDIAN ZOOS

SAKKARBAUGH ZOO – JUNAGADH, GUJARAT

Stud	Name	Sex	Birth date	Sire	Dam	Location	Date	Local ID	Event	Death Date
65.	KAMINI	F	-	WILD	WILD	GIR SANCT. JUNAGADH	25.5.76 25.5.76	UNK 54	CAPTURE TRANSFER	
132.	JESSICA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	18.1.84 18.1.84	UNK 111	CAPTURE TRANSFER	
229.	MANDA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	10.7.88 10.7.88	UNK 200	CAPTURE TRANSFER	
244.	HEMLATA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	31.3.89 31.3.89	UNK 215	CAPTURE TRANSFER	
255.	TINA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	8.5.89 8.5.89	UNK 226	CAPTURE TRANSFER	
259.	SUKESHI	F	-	WILD	WILD	GIR SANCT. JUNAGADH	28.2.90 28.2.90	UNK 230	CAPTURE TRANSFER	
260.	BIJLEE	F	-	WILD	WILD	GIR SANCT. JUNAGADH	28.2.90 28.2.90	UNK 231	CAPTURE TRANSFER	
261.	PRIYA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	28.2.90 28.2.90	UNK 232	CAPTURE TRANSFER	
267.	CAROL	F	-	WILD	WILD	GIR SANCT. JUNAGADH	1.3.90 1.3.90	UNK 238	CAPTURE TRANSFER	
268.	RUPA	F	-	WILD	WILD	GIR SANCT. JUNAGADH	1.3.90 1.3.90	UNK 239	CAPTURE TRANSFER	
299.	JAN	F	-	WILD	WILD	GIR SANCT. JUNAGADH	16.12.91 16.12.91	UNK 270	CAPTURE TRANSFER	
300.	PAUL	M	-	WILD	WILD	GIR SANCT. JUNAGADH	1.1.92 1.1.92	UNK 271	CAPTURE TRANSFER	
320.	ABHISEK	M	-	WILD	WILD	GIR SANCT. JUNAGADH	1993	UNK 320	CAPTURE TRANSFER	
321.	UNK	M	-	WILD	WILD	GIR SANCT. JUNAGADH	1993	UNK 321	CAPTURE TRANSFER	

SEX RATIO – 14 (3:11)

VEERMATA JIJABAI BHOSLE UDYAN – BYCULLA, BOMBAY

Stud	Name	Sex	Birth date	Sire	Dam	Location	Date	Local ID	Event	Death Date
264	UNK	M	-	WILD	WILD	GIR SANCT. JUNAGADH VEERMATA	1.3.90 1.3.90 19.3.91	UNK 235 UNK	CAPTURE TRANSFER TRANSFER	
266	UNK	F	-	WILD	WILD	GIR SANCT. JUNAGADH VEERMATA	1.3.90 1.3.90 19.3.91	UNK 237 UNK	CAPTURE TRANSFER TRANSFER	

SEX RATIO – 2 (1:1)

NATIONAL ZOOLOGICAL PARK – NEW DELHI

Stud	Name	Sex	Birth date	Sire	Dam	Location	Date	Local ID	Event	Death Date
265	RAMU	M	-	WILD	WILD	GIR SANCT. JUNAGADH NEW DELHI	1.3.90 1.3.90 28.1.91	UNK 236 UNK	CAPTURE TRANSFER TRANSFER	

SEX RATIO – 1 (1:0)

HYBRID LIONS IN INDIAN ZOOS

ANDHRA PRADESH

1. Nehru Zoological Park, Hyderabad – 11 (3:5); 3 Unsexed
2. Indira Gandhi Zoo, Vishakapatnam – 8 (4:4)

ASSAM

1. Assam State Zoo – 8 (5:3)

BIHAR

1. Bokaro Zoo – 4 (2:2)
2. Tata Zoo – 4 (1:1) 2 unsexed
3. Patna Zoo – 5 (3:2)
4. Ranchi Zoo – 2 (1:1)

DELHI

1. National Zoological Park – 6 (4:2)
New Delhi

GOA

1. Bondla Zoo, Goa – 3 (1:2)

GUJARAT

1. Ahmedabad Zoo – 14 (8:6)
2. Baroda Zoo – 9 (4:5)

HIMACHAL PRADESH

1. Renuka Lion Safari – 16 (8:8)
2. Dhauladhar Nature Park – 7 sex not known Gopalpur

KARNATAKA

1. Mysore Zoo – 4 (2:2)
2. Bannerghata N.P. – 3 (1:2)
3. Gadag Zoo, Dharwad – 2 (1:1)

KERALA

1. Trivandrum Zoo – 10 (6:4)
2. Trichur Zoo – 11 (7:4)
3. Neyyar Lion Safari – 7 (1:3) 3 unsexed

MADHYA PRADESH

1. Vanvihar National Park – 3 (1:2)

2. Indore Municipal Zoo – 7 (2:5)
3. Bhilai Zoo – 21 (12:9)
4. Gwalior Zoo – 10 (4:6)

MAHARASHTRA

1. Maharajbag Zoo, Nagpur – 8 (3:5)
2. Sangli Zoo – 26 (9:17)
3. Aurangabad Zoo – 9 (5:4)
4. Solapur Zoo – 10 (3:7)
5. Peshwe Zoo, Pune – 1 (0:1)
6. Borivilli National Park – 26 (15:11)
7. Veermata Jijabai Zoo, Byculla – 6 (3:3)
8. Nasik Zoo – 1 (0:1)

ORISSA

1. Nandankanan Zoo – 51 (26:25)

PUNJAB

1. Chatbir Zoo – 48 (26:22)

RAJASTHAN

1. Bikaner Zoo – 6 (2:4)
2. Jaipur Zoo – 6 (2:4)
3. Kota Zoo – 4 (2:2)
4. Jodhpur Zoo – 3 (1:2)
5. Udaipur Zoo – 3 (1:2)

TAMIL NADU

1. Vandalur Zoo, Madras – 15 (7:8)
2. V.O.C. Coimbatore – 4 (1:3)
3. Coimbatore Conservation Centre – 4 (2:2)

TRIPURA

1. Sephalijala Zoo, Agartala – 16 (8:8)

UTTAR PRADESH

1. Kanpur Zoo – 2 (1:1)
2. Lucknow Zoo – 4 (2:2)

WEST BENGAL

1. Alipore Zoo, Calcutta – 8 (3:5)
2. Purulia Zoo – 1 (1:0)