

# PROTOCOL

## INTEGRATED MANAGEMENT PROGRAM

### LEAR'S MACAW

*Anodorhynchus leari*



**Version 5.0**  
**December 2020**

## **1. INTRODUCTION**

This Protocol contains the minimum requirements and recommendations to be followed by the institutions participating in the Lear's Macaw's Integrated Management Program, created by the Brazilian government, through the Ordinance ICMBio n. 231/2013.

This document considered previous versions of the Captive Protocol and the contributions of Program members and collaborators. In this new version, the Management Protocol was prepared by Márcia Weinzettl, Curator of Birds of Loro Parque and Loro Parque Fundación and Program Management Consultant. The Health Protocol was updated and revised by Marcus Vinícius Romero Marques, Specialist in Veterinary Medicine of the Program, taking into account the considerations made by Tânia Raso, Professor at the University of São Paulo, and suggestions from the other Specialists.

## **2. MANAGEMENT PROTOCOL**

### **2.1. Goal**

Offer birds the highest levels of well-being and thus stimulate as much as possible their ex situ reproductive conditions.

### **2.2. Basic concepts**

Animal welfare is essential for the success of conservation projects, not only to comply with laws, standards, precepts of ethics and public opinion, but above all to achieve our greatest objective, which is the reproduction and conservation of the species.

Welfare is a gradual and continuous concept and its meaning can vary according to the individual, the stage of life and the circumstances. Therefore, the observation of the animals must be constant in order to allow the necessary adaptations.

To achieve the best levels of animal welfare, we must pay attention to three basic parameters:

- Normal operation of the organism: nutrition, hydration, physical comfort, thermal comfort and good health in general;
- Stable emotional state: absence of pain, fear or stress;
- Possibility to express typical behaviors of the species: behavioral needs.

All individuals in the Program must comply with the “principle of the five freedoms”:

- Free from thirst, hunger and / or malnutrition;
- Free from suffering thermal and physical stress;
- Free from feeling pain, injuries and / or illness;
- Free to develop the normal patterns of the species;
- Free from feeling fear or any other type of stress.

This protocol shall be applied to all individuals of the stock’s Program, including birds with permanent injury.

The concepts of welfare must be applied even in quarantines and hospitalizations.

In this protocol, two terms will be used: ADOPT and INDICATE. In the first case, it is an **obligation** and in the second, a **suggestion**. The obligations, indicated by the term ADOPT, will necessarily be implemented in all holders.

### 2.3. Management topics

#### 2.3.1. Nutrition and hydration

WE INDICATE suspended feeders and drinking fountains arranged at the front of the enclosure, with easy hygiene and external access.

WE ADOPT that feeders and drinking fountains will never be placed under perches and that they should be protected from pests, sun and rain.

WE ADOPT different diets for the reproductive and non-reproductive periods and, always, offering two meals a day. The maintenance diet (outside the reproductive period) should be replaced by the breeding diet 4 to 6 weeks before the reproductive period, in order to simulate the most favorable conditions that birds encounter in the wild in the pre-reproductive period. The diet should again be switched to maintenance diet in the week when the reproductive season ends or when the couple's reproductive activity is no longer observed.

WE INDICATE the offer of the most natural diet possible in order to facilitate the adaptation of birds in a possible selection for projects in situ (Table 1 and Figure 1).

WE ADOPT daily cleaning of feeders, drinking fountains and enclosures to prevent perishable items from spoiling. The seeds must be carefully inspected for quality and cleanliness and thus minimize the risk of aspergillosis.

Table 1: Diet indicated for a couple in each period of the year.

Outside the reproductive period	During the reproductive period
<b>Morning:</b> Assorted fruits and vegetables (choose five seasonal items) 20g of a balanced seed mix for Ara	<b>Morning:</b> Assorted fruits and vegetables (choose at least eight seasonal items) Mixture of previously cooked beans 20g of a balanced seed mix for Ara
<b>Evening:</b> 120g of a balanced seed mix for Ara 8 units of assorted nuts	<b>Evening:</b> 120g of a mixed seed mix balanced for Ara 10 units of assorted nuts

WE ADOPT the provision of calcium stones fixed in the enclosure throughout the year or an extra supply of calcium during the breeding season. Calcium stones are optional for birds and they are used whenever necessary to assist in the formation of healthy eggs or calcium deficiency in adults and chicks.

WE ADOPT the offer to birds of fresh green food (vegetables), such as dandelion, alfalfa or lettuce, which have a high content of active substances, with all the necessary vitamins and minerals, fibers and nutritional values. By increasing the supply of green food during the breeding season, we encourage even more breeding.



Figure 1. Examples of items from the recommended diet for Lear's Macaw, from left to right: varied fruits and vegetables, balanced seed mix and varied nuts (Photos: Loro Parque Fundación).

WE INDICATE the offering of licuri palm (*Syagrus coronata*) not only as a food item, but also as an environmental enrichment. Other ways of linking food to enrichment are described in item 2.3.3.

WE ADOPT for couples with chicks that the portion of food be doubled, adding more fresh corn on the cob and vegetables to the daily volume.

Water is essential for many physiological processes including essential temperature regulation. WE ADOPT the provision of fresh and potable water, changed at least twice a day, with chemical and biological control according to the criteria of local potability.

### **2.3.2. Enclosures - physical and thermal comfort**

#### Dimensions

You can choose a floor enclosure or suspended cage. The dimensions of the enclosure may vary, but we ADOPT that they have a horizontal arrangement that allows the long flight of the birds.

WE ADOPT as minimum measures for suspended cages, per couple: 3.5 m long x 1.8 m wide x 1.8 m high and at least 1 m from the ground.

WE ADOPT the minimum measures for floor enclosures, for a couple: 3.5 m long x 1.8 m wide x 2.3 m high.

WE ADOPT galvanized wire mesh, at least 2 mm thick and mesh equal to or less than 25 mm X 50 mm, in order to prevent the entry of other birds and some predators in the enclosure. The meshes must be galvanized to avoid metal poisoning.

WE INDICATE as the most comfortable measures an enclosure length above 10.0 m, with 2.0 m wide and 2.3 m high or, if possible, even larger dimensions.

For young or non-reproductive individuals, WE INDICATE that they are kept in groups, in larger enclosures, between 10 and 15 m in length.

#### Coverage and protection

WE ADOPT that at least 30% of the enclosure must be covered, providing protection against sun, rain, other weather and possible predators. The cover must protect at least the areas intended for food and nests.

WE INDICATE a visual separation between neighboring enclosures, at least 1 m wide, on both sides of the enclosure where the nests are located. This protection must cut eye contact (Figure 2) at least in the upper half of the enclosure, but the most suitable would be to the floor.

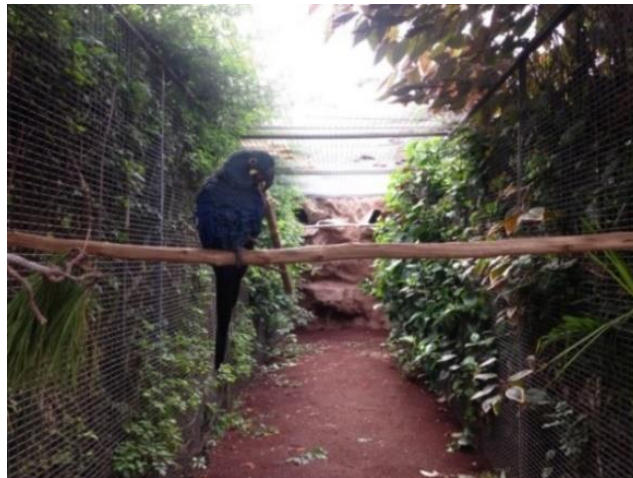


Figure 2. Visual protection with vegetation on the sides of the enclosure (Photo: Loro Parque Fundación)

#### Nests

WE INDICATE that the nests are located opposite the entrance to the enclosure.

WE ADOPT a minimum of two nests per enclosure and entrance to the nests at a minimum height of 2.0 m from the ground. The nests must be external to the enclosure, facilitating hygiene and access for handling eggs and chicks. There must be a security corridor or other structure in the place of external access to the nests, to prevent escape.

WE INDICATE nests in different formats (Figure 3) with the following measures:

- "Inclined L" presenting 90 cm long x 40 cm wide x 40 cm high at the end of the nest, and 1.2 m long in the inclined area.
- "Horizontal box", 1 m long x 35 cm wide x 35 cm high.
- "Simple L" showing 35 cm wide x 40 cm high at the end and 70 cm long x 90 cm high at the initial portion of the nest.



Figure 3. Types of nest that can be used in the enclosures. View from outside the enclosure, where the nests can be accessed for monitoring and management (Photo: Loro Parque Fundación).

Nest boxes can also be made as tunnels that lead to an incubation chamber about 70 x 70 cm. Longer tunnels that change direction are preferred by macaws. An important feature of any nest box is that it contains a dark interior. However, very important in this system is that there is an opening that allows the cleaning and disinfection of the entire set of tunnels and chambers after each reproductive season.

WE ADOPT that the location where the nests are inserted is visually isolated from other areas or from management passages. Access to the entrance to the nests must be facilitated by the adequacy of perches and / or the installation of platforms (Figure 4). The platforms are very relevant to allow the couple to be alert at the entrance to the nest and show their natural behavior. When such access is offered, it is common to observe the couple resting at the entrance to the nest.

WE INDICATE the isolation by a rocky wall (Figure 4), similar to that found in nature.



Figure 4. On the left, nest placement site, visually isolated from other enclosures by a rocky wall. Observe perches placed in order to facilitate the access of birds to the entrance of the nests. Right: couple using eaves to access and protect the nest (Photos: Loro Parque Fundación).

An option also indicated and of lower cost, which allows birds to develop their natural behavior to protect the nest, is to use the imitation of rocky wall only in the upper part of the insulation made with sheet, as shown in Figure 5.

The material used to make the artificial rock wall is shown in Figure 6.



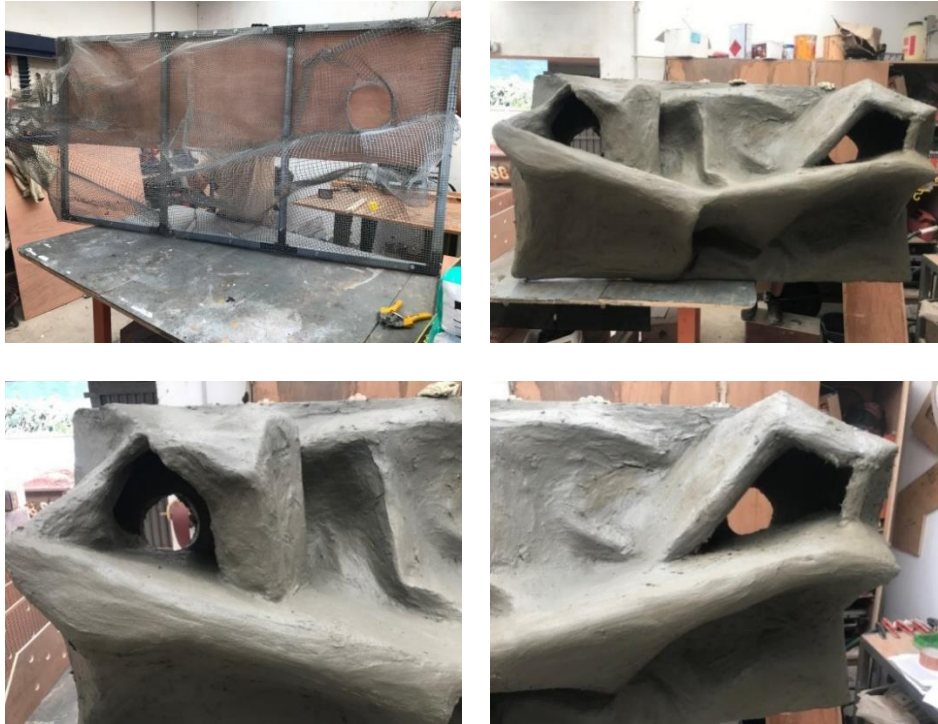


Figure 5. Manufacturing the visual insulation of nests, imitating rocky walls and providing a platform for the entrance and exit of the macaws (Photos: Loro Parque Fundación).



Figure 6. Example of material that can be used in the manufacture of artificial rock walls for Lear's Macaws. Left and center: mortar with waterproofing. Right: mortar dyes (Photos: Loro Parque Fundación).

Visual isolation of the nest area can also be found with the installation of a simple aluminum plate or aluminum plate and vegetation. If this option is used, you can provide a wooden shelf as a platform to guard the nest.

WE ADOPT the supply in any of the ways presented that represents an eave at the entrance of the nest, so that the couple can develop their natural behavior.

Lear's macaws are very sensitive to disturbances in their nests. Therefore, WE INDICATE that all nest boxes are provided with cameras for monitoring without any human intervention. In the absence of cameras, adult birds should be prevented from seeing people accessing their nest for monitoring. For this, a small sliding door can be

installed at the entrance of the nest, isolating the parents from outside and obtaining safe access to the eggs and chicks.

WE INDICATE that the substrate of the nest boxes consists of a 5-8 cm layer of washed and disinfected sand, covered with sawdust.

#### Safety hall

WE ADOPT double escape protection through the use of security doors or halls.

#### Perches and other substrates

WE ADOPT, for floor enclosures, sandy substrate or easy cleaning for daily elimination of waste and food scraps. It is also essential that the substrate has good drainage, thus preventing the formation of puddles from which birds can drink.

WE ADOPT the offer of a set of natural wooden perches, with varying thicknesses around 6 cm in diameter and forming blocks distant from each other, in order to allow and stimulate the long flight of birds (Figure 7).



Figure 7. Perches for Loro's Macaws. On the left, blocks of perches far apart. On the right, detail of the perch thickness (Photos: Loro Parque Fundación).

We recommend changing these perches at least twice a year.

WE ADOPT that in each enclosure at least two fixed perches are offered at the ends of the enclosure and a mobile perch. The mobile perch in the form of a swing (Figure 8) or one that allows the bird to climb is very important for the muscular development of birds.

For birds with permanent injury, WE ADOPT the use of well-designed perches that allow their access to feeders, drinking fountains, enrichment items, shelter and nests.





Figure 8. Perch on a swing in a Lear's Macaw enclosure (Photo: Loro Parque Fundación).

#### Exhibition Enclosure

WE ADOPT that the institutions that keep Lear's Macaws on exhibition have an educational plaque about the species, also disclosing the Integrated Management Program and its results.

WE INDICATE laminated glass as a barrier option for birds on exhibition. However, it is important to pre-apply temporary tapes to the glass until the birds are adapted to this type of barrier. The removal of the alternating half of these tapes should be after 15 days and the other half just a week later. So the birds gradually adapt.

WE INDICATE that, as a priority, non-reproductive individuals (young, elderly or unmarried animals) are taken to the exhibition. However, if the enclosure meets all the requirements of this protocol for birds in reproduction, it can also be used for this purpose.

#### Enclosures in countries with severe weather

WE ADOPT that institutions located in countries with severe winter must provide birds with an air-conditioned enclosure or indoor enclosure for greater protection. The minimum temperature indicated should be around 18° C and the humidity around 55%. These enclosures must meet all other requirements of this protocol.

WE ADOPT that institutions located in countries with temperatures above 40° C should provide cooling for birds using air conditioning or water sprinklers for daily showers in the hottest periods of the day, in addition to meeting all other requirements of this protocol.

### 2.3.3. Environmental enrichment

Well-applied enrichment allows birds to express behaviors typical of the species. Their behavioral needs must be valued to the maximum in the conservation ex situ.

WE ADOPT that, at least once a week, some new enrichment item is offered. However, WE INDICATE that this exchange is carried out twice a week.

WE INDICATE the offering of branches, trunks, pieces of palm, pine, banana leaves, pine cones and bunches of coconut, or similar. A few times a week, food can be offered in hiding places that lead birds to develop natural foraging behaviors. The location of these hiding places can be changed frequently. Food items can be offered in their natural state, such as whole fruits and vegetables, shelled nuts, etc. Calcium and mineral blocks must be offered throughout the year (Figure 9).

WE INDICATE that the enclosure has abundant lateral or posterior vegetation, in order to allow its gradual growth into the enclosure. It is important to be careful not to use toxic plants for this purpose.



Figure 9. Examples of enrichment items for Lear's Macaw. Above left, use of vegetation on the sides of the enclosure, allowing it to grow inside and continued use by birds (Photo: Loro Parque Fundación). Above, on the right, a bunch of coconuts offered in the enclosure (Photo: Loro Parque Fundación). Below, pine cones with fruit hanging from perches (Photo: Fazenda Cachoeira)

WE INDICATE that part of the enclosure contains water sprinklers to simulate rain for the birds. WE ADOPT that if artificial rain is included, the water used must be potable

and proven with a chemical and biological analysis, according to the local potability criteria.

#### **2.3.4. Breeding**

Lear's macaws are believed to be monogamous birds that form a lifelong pair.

WE ADOPT that couples should be kept together throughout the year, with continuous access to nests, which are also used as shelter. Holders who have more than one couple in their stock should maintain sound and, if possible, visual contact between the different pairs, which should encourage reproductive behaviors.

The clutch are two to four eggs. The eggs measure, on average, 23 x 43 mm and weigh 30 g.

WE INDICATE, whenever possible, that the incubation and treatment of the chicks is carried out by the parents themselves, in order to provide to the program birds least humanized than possible.

For couples less represented in the stock, WE INDICATE the withdrawal of the first posture for artificial breeding and the maintenance of the second posture with the parents.

Up to three postures in the year can be stimulated, if the first two are removed or lost. However, when the chicks are raised by their parents, they will provide only one clutch per year.

Since the couple does not raise their chicks well, we INDICATE as the first option the creation by adoptive parents, who must have a history of success in the creation of puppies. The best options for adoptive parents are species that have nutritional needs similar to the Lear's Macaw, such as *Ara chloroptera*, *Ara ambigua* and *Anodorhynchus hyacinthinus*.

WE ADOPT the premise that all the institutions in the Program have experienced professionals in artificial breeding of macaws. If any institution does not have professionals qualified for this type of management, training should be provided with another institution of the Program that already has successful experience.

In case artificial breeding is necessary, WE INDICATE that the eggs are incubated at a temperature of 37.4° C and humidity 55%. The humidity may vary according to the quality of the shell, therefore, WE INDICATE the monitoring of the egg per weight loss chart. The incubation period varies between 28 to 30 days and the egg must lose around 15% of its initial weight during this period. The eggs must be rotated 90° at least eight times a day, always changing the direction of the rotation when it reaches 360°. The rotation should be done until the baby begins to break the egg's air chamber. At this time, it should not be rotated anymore and the egg should be taken to an incubator with

a temperature of 37.1° C and humidity around 75%. The peel leaves normally complete after 48 hours.

Chicks are born weighing 17 g on average (which can range from 15 to 21 g).

For the first two weeks of life, WE INDICATE that the chicks remain in individual containers, lined with paper towels. From two weeks of age, an absorbent and clean material such as sawdust or paper should be used to line a non-slip substrate that should be placed at the bottom of the recipient. After seven weeks, a welded mesh can be used as a substrate to increase the chick's grip and prevent deformities in the feet. After 9-10 weeks the chicks are ready to roost and can be kept in a cage. WE INDICATE that, from this age on, the juvenile can live with other Lear's Macaws or other parrots of similar age, thus allowing for social interaction.

As for feeding, we INDICATE for chicks artificially raised a more protein and fatty porridge for the first 10 days of life, with about 35% protein and 19% fat. From then on, after 10 days of life, the porridge should contain around 20-22% protein and 15-20% fat. The first feeding can be offered up to 12 hours after birth, but there are variations due to the conditions of each chick and the absorption of the yolk sac, which can be observed by the skin of the bird's belly or by the color of the feces that change from yellow to brown. If no feeding is offered within 12 hours of birth, the chick should be hydrated during this period orally with a mixture of saline or 1:1 Ringer's with lactate.

The chick should be fed whenever the crop is almost empty, maintaining the frequency of porridge per day (see summary below). During the night it is INDICATED that the chick rests and no porridge is offered. On the first day, the porridge should be diluted in 6 parts of water; on the second day, in 3 parts of water; from the third day onwards until weaning in 2 parts of water for one of ration. The chick should be weighed every day in the morning and should receive between 10 and 12% of its weight in porridge with each feeding.

### **HANDLING CREATION**

#### **- AIR TEMPERATURE:**

- . First 6 hours – 37° C
- . From 0 to 5 days – 36,5° C
- . 5 days to open eyes – 35° C
- . With eyes open until the beginning of plumage – 34° C
- . Plumage already covering the body – 33° C or lower, depending on the country
- . Full plumage - Room temperature

Note: HUMIDITY must be above 40%.

#### **- FOOD:**

Frequency:

1st day - 10 times (6 parts of water X 1 of feed \*)

2nd day - 9 times (3 parts of water X 1 of feed \*)

3rd day - 8 times (2 parts of water X 1 of feed \*)  
4th day - 7 times (idem, until the end of weaning)  
5th day - 6 times  
6th day until you open your eyes - 5 times  
Eyes open until cannons start - 4 times  
With cannons - 3 times (start offering solid food)  
With feathers - 2 times  
Until weaning - 2 to 1 times a day.

NOTE: \* The dilution of the porridge may vary according to the manufacturer and it is important to always follow these indications.

For chicks raised by parents or adoptive parents, the diet is indicated in item 2.3.1 of this Protocol. After 70 days of age, the chicks begin to leave the nest. WE INDICATE that the chicks stay with their parents for at least seven months before being taken to a juvenile enclosure (Figure 10).



Figure 10. On the left, pup of macaw. In the center, Lear's Macaw created by adoptive parents. On the right, chicks are being monitored in the nest.

### 2.3.5. Etology

WE ADOPT special care when it is necessary to introduce an individual to another, thus minimizing the risk of aggression that can result in severe injury or death. For all presentations between individuals, the steps below can be used as a starting point:

- a) Place individuals in neighboring enclosures, with complete eye contact and with the possibility of establishing close contact through a double wire mesh. Alternatively, the bird to be approached can be housed in a cage (minimum 1.0 m<sup>3</sup>) placed outside (or even inside) the other bird's enclosure. For reproductive pairings, WE INDICATE that the male is taken to the female's enclosure.
- b) Observe for a few days the tolerance and compatibility of one individual with the other and assess the level of aggression (real or potential). If the birds show compatibility, without any indication of aggressiveness, you can proceed to the next step. Some birds show empathy in the first few minutes of presentation. However, if there is any sign of aggression among the birds, the approach



should not continue. This process could take up to 2 weeks to reach a conclusion.

- c) After passing the compatibility test of the previous item, the birds can be released simultaneously in a third and definitive enclosure, in the early hours of the day. The transfer of everyone to another room is preferable because it eliminates any previous occupation of territory. At this time, the team must remain attentive, monitoring the birds throughout the day, always being ready and equipped to separate the birds immediately if any severe aggression occurs. If no aggression occurs on the first day, it is unlikely to occur later.

WE ADOPT that each couple is monitored during the year (Figure 11) in order to define the level of proximity and acceptance between them.



Figure 11. Example of behavior to be monitored throughout the year, an indicator of proximity between the couple.

#### **2.4. Identification and genetic analysis of the stock**

WE ADOPT that all birds in the breeding stock, whether of nature or born in captivity, must be identified with a microchip, implanted by a qualified veterinarian. WE INDICATE that the implant is done in the bird's pectoral muscle. Individuals born in captivity must also be marked with a closed stainless steel ring 14 mm in diameter and exclusive numbering.

WE ADOPT at least one annual reading of each individual's microchip, at the time of periodic examinations, to confirm its functioning. In case it presents problems in the reading, the bird must receive another microchip immediately and the Program Coordination must be formally communicated to register the fact and inform the Genealogical Consultant, so that it updates the Studbook.

WE ADOPT that all individuals born in captivity are sexed before one year of age and that those from nature are sexed as soon as they pass through the quarantine of the institution and before entering the stock. WE INDICATE genetic determination of sex or through endoscopy, the latter of which can also provide information on the state of the gonads, maturity or diseases.

WE ADOPT that the institutions maintain individual records of the macaws of their breeding stock, so that accurate and updated information about each specimen is recorded, which can help to identify behavioral, health or reproductive problems. In addition to individual identification, the form must contain:

- a) Reproductive history: individuals with whom it was paired, number and identification of clutches and offspring, sex and destination of the offspring;
- b) Medical history: routine weighing, disease diagnoses, medical treatments, surgical procedures, etc.
- c) Changes in management: transfer to new enclosures, changes in diet, changes in environmental enrichment activities, etc.
- d) Notes about any incidents involving the individual and about any signs of abnormal behavior, stereotypes, humanization, structural problems in the enclosure etc.

WE ADOPT that participating institutions should send blood samples of all the Lear's Macaws in their breeding stock to Dr. Cristina Yumi Miyaki, a specialist of the Program in the Genetics. The collection must be carried out at the first opportunity for handling the birds, provided that there is a person qualified to do so and adequate conditions for storing the material. Dead birds, whose sample has not yet been sent for genetic analysis, must also have material collected for this purpose.

The sample for genetic analysis of live birds consists of 0.1 ml of blood stored in a sterile tube, with 0.5 ml of absolute ethanol (100%). It is necessary to use a tube and a syringe per individual, thus avoiding contamination of the material. The tube must be identified with the Studbook number corresponding to the individual sampled. If possible, additional information should be provided (e.g. microchip and ring number, if it is a bird of nature or born in captivity, etc.).

In case the sample has to be collected from a carcass, just store about 1 cm<sup>3</sup> of the bird's muscle, minced, in a sterile tube with absolute ethanol (100%), so that the ethanol has two to three times the volume of the collected muscle. The sample identification must follow the procedure informed in the previous paragraph.

The address for sending the samples is:

Departamento de Genética e Biologia Evolutiva – Instituto de Biociências  
Universidade de São Paulo  
A/C: Dr<sup>a</sup>. Cristina Miyaki  
Rua do Matão, 277, São Paulo/SP - Brasil  
05508-090

## **2.5. Containment, transport and transfers**

WE INDICATE the use of catch net as an important tool to capture Lear's Macaws safely and efficiently. The use of a quick method reduces the chance of the bird going

into stress. The capture should be made in the coolest hours of the day, avoiding thermal stress.

For transportation, periods of extreme weather should be avoided. Enclosures exchanges should be made early in the morning, so that the birds have time to familiarize themselves with the new location. Birds must be transported with adequate ventilation. Be careful with loud sounds and noises that can stress them.

WE ADOPT the cleaning and sterilization of capture instruments and transport boxes before each use.

WE ADOPT the use of IATA standard transport boxes, with individual compartments and with minimum dimensions of 50 cm long by 20 cm wide and 25 cm high (Figure 12). For transfers between holders, WE ADOPT the supply of sufficient provision of water and food.

WE ADOPT that the institution that receives a bird from another Program holder is informed in detail about the individual's medical, behavioral and reproductive history, in addition to any information deemed relevant.

WE ADOPT that the institution of origin of the bird that will be transferred must carry out, in the exit quarantine, the tests indicated in the Health Protocol and others possibly required by the authorities of the country of destination, when applicable. After reaching the destination institution, the individual will be quarantined and will be subjected to mandatory tests again. WE ADOPT that the transfer costs are paid by the destination institution, unless a different agreement has been made between the parties.

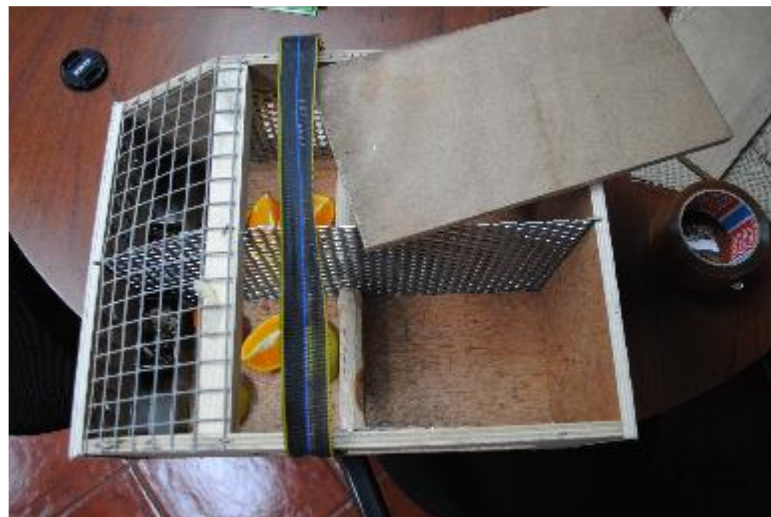


Figure 12. Example of IATA standard transport case, for two Lear's Macaws.

### **3. HEALTH PROTOCOL**

#### **3.1. Goal**

The health management should have a prophylactic approach. Its main objective is to prevent the introduction and spread of pathogens, and to diagnose, control and eliminate them before they have a negative effect on the birds. When Lear's Macaws are kept in close proximity to other birds (less than 20 m or without a physical barrier, and with the same equipment and handlers), the other birds must be tested for selected pathogens (see Appendix).

#### **3.2. Basics Concepts**

New birds and every Lear's Macaw should be fully tested prior to any transportation. Testing should be performed during the 30 days quarantine period by both holders, the one receiving and the one sending the Lear's Macaw. The institution sending the Lear's macaw should always provide to the receiving holder documentation and full clinical history of the bird to be sent including examinations performed before the transport takes place.

#### **3.3. Routine observation of behavioral manifestations of illness**

Routine assessment of health is carried out on a daily basis. All Lear's Macaws should be observed at least twice a day by an experienced avian keeper. All aviaries should be checked thoroughly once a week for potential hazards, this is over and above the daily observing of the birds. When necessary, samples should be collected to be examined in the laboratory.

Generally, observational controls are based on the following:

##### Behavior

- changes in the bird's behavior (stereotypes, the bird is quieter, sleeps, is nervous, turns away from the partner, feather plucking, aggressive);
- physical posture;
- tail wagging (may indicate respiratory diseases, cloacal problems, or egg retention);
- extremities (one or both wings are slightly hanging down, the bird obviously tries not to strain one of its feet).

##### Plumage

Ruffled plumage, first the neck feathers and then the entire plumage, regulates the body temperature of the animal; at the first stage of a disease, quite often the bird ruffles a few neck or head feathers (which may also just be a threatening posture);

### Eyes

the eyes should be carefully examined, they must be clear and fully open; at the very beginning of a disease, the birds' eyes are slightly closed and lose their shine; birds which are seriously ill have their eyes completely closed.

### Excreta

Attention is given to the quantity, color and consistency (liquidness) of excreta; both greater-than-usual and smaller-than-usual quantities might indicate an illness; although the color of the feces may strongly depend on the food ingested, it must also be carefully controlled; with regard to liquidness, the technician must distinguish between an excessive excretion of urine (urates and feces tend to be separated) and diarrhea (urates and feces tend to be mixed). Soiled under tail feathers are a sign of possible diarrhea.

### Ingestion of food and water

- the technician must assess if a bird ingests food and water in normal quantities. Changes in food intake are important to monitor.
- each bird's weight should be periodically monitored to validate that the diet they are receiving is not too high in energy. Special hidden scales could be used in order to have the birds' weight monitored on a voluntary base.

### Other

- nasal, ocular, cloacal discharges can indicate a disease or allergic reaction, e.g. to nest substrate or food;
- bill and talons: trimming, if indicated;
- leg bands: remove in case of injury.

## **3.4. Common injuries and treatment**

The most common injuries that require medical treatment result from: self-inflicted accidents in the aviary or minor aggressive incidents between partners of a breeding pair.

Feet, wings and head are the most affected body parts. Treatment is according to standard avian veterinary practice. Severe aggression is uncommon in Lear's Macaw but can lead to trauma and even death.

## **3.5. Critical infectious diseases and prophylactic management**

Most important infection diseases for macaws are listed in Table 2. Prevention is the essential guideline of this protocol.



Table 2 - Relevant infectious diseases that can affect captive psittacines.

Disease	Pathogen
Psittacine Beak and Feather Disease (PBFD)	Circovirus
Avian Polyomavirus infection (APV)	Polyomavirus
Pacheco's and Pacheco's-like Disease	$\alpha$ -Herpesvirus 1(PsHV-1)
Proventricular Dilatation Disease (PDD)	Avian Bornavirus, and possibly other infectious agents
Chlamydiosis	<i>Chlamydia psittaci</i>
Salmonellosis	<i>Salmonella</i> spp.

### 3.6. Routine medical control and sampling for tests

Biological samples of individual birds should be regularly collected for testing for infectious diseases and/or parasitological analyses.

On a regular basis, and at least once during the annual health check, each macaw must be caught and undergo complete physical examination. At the same time, weight and body condition of the bird should be checked. Average weight for adult Lear's Macaws is 867 g for males and 816 g for females (n=14 and 20 respectively, Loro Parque Fundación data). If a bird is over- or underweight it may require a special diet. Overweight might contribute to infertile eggs, and therefore this aspect should be given careful attention.

The protocol presented here can be used as a guideline when a Lear's Macaw undergoes a clinical examination. Appendix shows a list of mandatory tests for diseases detection. It is recommended to use a reliable laboratory. If it is necessary to repeat a test, this repetition must be performed in the same laboratory. In case of doubts, or in the occurrence of a positive result and another negative result, a third examination should be requested.

### 3.7. Parasite control

This control should be part of an integral parasite program including regular parasitological analysis and appropriate deworming using the most indicated drugs and dosages.

### 3.8. Necropsies

A thorough necropsy shortly after death is extremely important to determine the cause of death. It may indicate health and/or management problems that may have gone unnoticed. Necropsies must be conducted on all birds that die, and a comprehensive report with photos should be prepared afterwards. When feasible, both skin and carcass will be sent to an ornithological collection authorized by ICMBio. Immediate incineration is mandatory for carcasses of individuals positive for viral agents and for pathogenic species of *Mycobacterium* and *Chlamydia psittaci*.

### **3.9. Control of neonates**

No data could be found about specific causes of neonatal mortality in Lear's macaws. However, for similar species the main causes of neonatal and chick mortality in captivity are associated with pathogens or due to energetic imbalance (incorrect management of the parents, or incorrect hand rearing practices).

Young parrot chicks are susceptible to pathogens during their first days of life, because their immune system is not yet entirely developed. The most common infections of hand-reared chicks are bacteriological (usually Enterobacteriaceae, or other Gram negatives) and fungal (usually *Candida* spp).

Chicks must be constantly monitored for their health status. Treatment or intervention must be applied and performed as indicated.

### **3.10. Quarantine measures**

When moving birds between institutions different test may be required depending of the exporter/importer institution or country's regulations. Every Lear's macaw should be fully tested prior to any transportation. Testing should be performed during the 30 days quarantine period by both holders, the one sending and the one receiving the Lear's macaw.

Every bird that enters a given breeding facility must undergo a period of at least 30 days of strict isolated quarantine to preclude the spread of pathogens to the rest of the population/birds on site. During this period the birds should be tested for the pathogens listed in Appendix.

If the bird becomes sick within these first 30 days, complete disinfection must be carried out in quarantine and the quarantine period must start again when the bird is declared healthy again. Treatment against intestinal parasites should be provided for birds undergoing quarantine if they are positive for endoparasites eggs, cysts or oocysts. All birds have to produce at least three negative parasites faecal samples within an interval of 7 to 14 days in the quarantine. If the birds of one institution are considered free of diseases tested in Appendix it should only receive pathogen free birds or maintain positive birds separately (at least 20 m away or with a physical barrier, and equipment and handlers should be separated).

### **3.11. Reproductive health**

An investigative laparoscopy should be performed for all adult specimens, which have been paired for three years and are not reproducing.

## **4. FINAL CONSIDERATIONS**

WE ADOPT that all birds of the Program's breeding stock are the domain of the Brazilian Government.

WE ADOPT that the institutions participating in the Program must send an Annual Report to the Program Coordination, within the requested deadline. The report may contain personalized questions for each institution, if there were any doubts regarding the information provided in the report or at the previous meeting.

The requirements contained in this Protocol must be applied by the institutions participating in the Program within a maximum period of two years, counting from its divulgation.

After this period, only those institutions that comply with the obligations set forth in this Protocol will remain in the Program, especially since its preparation was participatory, with approval of the content by each institution.

The Management Consultant will present, at the annual meetings of the Program, a summary table with the scenario found in each participating institution.

Appendix – Annual mandatory tests to Lear’s Macaw<sup>1</sup>

Etiological Agent	Sample	Diagnosis Test	Management to be performed
Hemoparasites ( <i>Haemoproteus</i> , <i>Leucocytozoon</i> , <i>Plasmodium</i> , microfilarias and others)	Blood smears or Blood	Wright and Giemsa colorations or PCR	If the macaw has a high parasitism, do the treatment according to parasites
Ectoparasites (lice and mites)	Ectoparasites in isopropyl or ethyl alcohol 70°	Alpha taxonomy	Treatment according to parasites
Gastrointestinal parasites (helminthes and protozoa)	Feces (one fece per day for three consecutive days, during the early morning hours, pooling the samples, storing in the fridge and performing one analysis. Sampling on smooth disposable substrate previously disinfected)	Flotation or alternative method	Treatment according to parasites and three negatives retests post- treatment
<i>Chlamydia psittaci</i>	Required use pool samples (cloacal/oro- pharyngeal swabs), serial sampling (three consecutive days)	PCR	Isolation for treatment for clinical animals. After two negative tests post-treatment (after 2 and 4 weeks) the macaw can reincorporate to the breeding program. For birds without clinical signs treat and retest, as the stress of moving between institutions/pairing/adapting to new diets/climate could be triggering events for further shedding/manifestation of clinical signs.
<i>Salmonella</i> spp.	Cloacal swab	Culture and PCR	Isolation for treatment for clinical animals. After two negative tests post-treatment (after 2 and 4 weeks) the macaw can reincorporate to the breeding program. For birds without clinical signs treat and retest, as the stress of moving between institutions/pairing/adapting to new diets/climate could be triggering events for further shedding/manifestation of clinical signs

Etiological Agent	Sample	Diagnosis Test	Management to be performed
<i>Mycoplasma</i> spp.	Cloacal/oro-pharyngeal swabs	PCR to <i>Mycoplasma</i> spp.	Isolation for treatment for clinical animals. After two negative tests post-treatment (after 2 and 4 weeks) the macaw can reincorporate to the breeding program. For birds without clinical signs treat the decision of treatment is delegated to each institution. When decided to treat, the animal must be retest, as the stress of moving between institutions/pairing/adapting to new diets/climate could be triggering events for further shedding/manifestation of clinical signs.
<i>Mycoplasma gallisepticum</i>	Cloacal/oro-pharyngeal swabs	PCR to <i>M. gallisepticum</i>	Isolation for treatment for clinical animals. After two negative tests post-treatment (after 2 and 4 weeks) the macaw can reincorporate to the breeding program. For birds without clinical signs treat and retest, as the stress of moving between institutions/pairing/adapting to new diets/climate could be triggering events for further shedding/manifestation of clinical signs.  <u>The cases of positive animals, in Brazil, must be notified to the Ministry of Agriculture (MAPA).</u>
Cloacal and oral microbiota	Cloacal/oro-pharyngeal swabs in Stuart	Culture	Caution on interpretation whereas bacterial and fungal growth may not be related with disease. For clinical animals treatment according to bacterial and fungal isolates.
<i>Mycobacterium</i> spp.	Required use pool samples (cloacal/oro-pharyngeal swabs), serial sampling (three consecutive days)	PCR (Confirm by isolation or sequencing if it is one of the following species: <i>Mycobacterium avium</i> , <i>M. tuberculosis</i> , <i>M. bovis</i> or <i>M. genavense</i> )	All macaws that have contact with positive birds for pathogenic species of <i>Mycobacterium</i> should be isolated and tested. <u>For birds without clinical signs and considered positive should be kept in isolation for at least six months and tested for agent culture and determination of the species involved.</u> Birds confirmed as positive and with clinical signs should be euthanized. This is a screening test only for legal reasons. In confirmed positive cases further testing must be performed to investigate if it is a disease case or if it is only an environmental pathogen.



Etiological Agent	Sample	Diagnosis Test	Management to be performed
<sup>2,3</sup> Paramyxovirus – Avian Paramyxovirus type 1 (APMV-1)	Cloacal/oro-pharyngeal swabs Blood Serum	RT-PCR or Serology (haemagglutination inhibition and/or ELISA)	Only for international transfers. In Brazil, follow the orientation of Ministry of Agriculture (MAPA).
Polyomavirus (APPV)	Required use pool samples (feathers, blood and cloacal swab), serial sampling (three consecutive days for swabs; blood collection for biochemical examination on the first day and for viremia on the third day)	PCR	<p>In case of a positive result, isolate the bird and perform new monthly serial tests for six months. The individual that has three consecutive negative exams can be reincorporate to the breeding program. If there are three positive tests (consecutive or not) before the end of the six months, the bird is considered positive and no further testing is necessary.</p> <p>All birds that have contact with positive macaw should be isolated and tested (a sample pool collection, for three consecutive days). PCR have to be performed in all contact birds.</p> <p>Birds confirmed as positive but without clinical signs may be kept at the institution, isolated from the rest of the aviary. Reproduction can only occur by pairing with another positive bird or by artificial insemination. Eggs must be artificially incubated. Chicks should be raised artificially.</p> <p>If there are no conditions for isolation in the institution itself, the institution may decide euthanasia.</p> <p>Birds with clinical signs and a single positive result should be euthanized.</p>

Etiological Agent	Sample	Diagnosis Test	Management to be performed
Alpha-herpesvirus of Pacheco (PDV)	<p>Required use pool samples (blood and cloacal/oro-pharyngeal swabs), serial sampling (three consecutive days for swabs; blood collection for biochemical examination on the first day and for viremia on the third day)</p>	PCR	<p>In case of a positive result, isolate the bird and perform new monthly serial tests for six months. The individual that has three consecutive negative exams can be reincorporate to the breeding program. If there are three positive tests (consecutive or not) before the end of the six months, the bird is considered positive and no further testing is necessary.</p> <p>All birds that have contact with positive macaw should be isolated and tested (a sample pool collection, for three consecutive days). PCR have to be performed in all contact birds.</p> <p>Birds confirmed as positive but without clinical signs may be kept at the institution, isolated from the rest of the aviary, treated and retested. Reproduction can only occur by pairing with another positive bird or by artificial insemination. Eggs must be artificially incubated. Chicks should be raised artificially.</p> <p>If there are no conditions for isolation in the institution itself or if birds confirmed positive in retest, after treatment, the institution may decide euthanasia.</p> <p>Birds with clinical signs and a single positive result should be euthanized.</p>
<sup>3</sup> Circovirus (PBFD)	<p>Required use pool samples (feathers, blood and cloacal swab), serial sampling (three consecutive days for swabs; blood collection for biochemical examination on the first day and for viremia on the third day)</p>	PCR	<p>In case of a positive result, isolate the bird and perform new monthly serial tests for six months. The individual that has three consecutive negative exams can be reincorporate to the breeding program. If there are three positive tests (consecutive or not) before the end of the six months, the bird is considered positive and no further testing is necessary.</p> <p>All birds that have contact with positive macaw should be isolated and tested (a sample pool collection, for three consecutive days). PCR have to be performed in all contact birds.</p> <p>Birds with clinical signs and a single positive result must be euthanized. Birds confirmed as positive, although without clinical signs, should be euthanized.</p>

Etiological Agent	Sample	Diagnosis Test	Management to be performed
<sup>3</sup> Influenza A	Cloacal/oro-pharyngeal swabs Blood Serum	RT-PCR or Serology (haemagglutination inhibitionand/or ELISA)	Only for international transfers. In Brazil, follow the orientation of Ministry of Agriculture (MAPA).
<sup>3</sup> Avian Bornavirus (ABV)	Required use pool samples (blood and cloacal/oro-pharyngeal swabs), serial sampling (three consecutive days for swabs; blood collection for biochemical examination on the first day and for viremia on the third day) Serum	RT-PCR Serology (only in countries where it is available)	In case of a positive result, isolate the bird and perform new monthly serial tests for six months. The individual that has three consecutive negative exams can be reincorporate to the breeding program. If there are three positive tests (consecutive or not) before the end of the six months, the bird is considered positive and no further testing is necessary. All birds that have contact with positive macaw should be isolated and tested (a sample pool collection, for three consecutive days). PCR have to be performed in all contact birds. Birds with clinical signs and a single positive result should be euthanized. Birds confirmed as positive, although without clinical signs, should be in isolation for monitoring the evolution of disease and retest. Treatment is a possibility.
Aviadenovirus	Required use pool samples (blood and cloacal/oro-pharyngeal swabs), serial sampling (three consecutive days for swabs; blood collection for biochemical examination on the first day and for viremia on the third day)	PCR	Isolation of the bird. After three negative tests with a 3 months, birds can reincorporate to the breeding program. Birds confirmed as positive, should be in isolation for monitoring the evolution of disease and retest

- <sup>1</sup> The exams involving PCR can be performed every two years, instead of annually, provided that no animals have entered the institution. PCR exams are mandatory during quarantines for transfer of birds (both on departure and on arrival at institutions).
- <sup>2</sup> If Paramyxovirus is detected, it is important to identify its type (isolation and viral characterization), because not all of them are pathogenic to psittacines and only APMV-1 is notifiable. Considering Brazilian legislation, positive cases can not be imported to Brazil.
- <sup>3</sup> Birds tested positive for these viruses can not be imported to Brazil.