Using Captive Propagation to Help Save the

Puerto Rican Parrot[1]

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The Puerto Rican parrot *(Amazona vittata)* is a small (28 cm, 250-350 g) amazon parrot with emerald green plumage that provides perfect camouflage for the bird in the lush tropical foliage of its rain forest home. In flight its brilliant blue wing feathers appear jewel-like as the parrot manuevers over the forest canopy. The parrot's raucous calls once filled the forests of the West Indian island of Puerto Rico. Tragically, it is now one of the many *Amazona* parrot species in dire jeopardy and is listed as endangered along with the red-necked *(A. arausiaca)*, red-tailed *(A. brasiliensis)*, St. Vincent *(A. guildingii)*, imperial *(A. imperialis)*, and St. Lucia *(A. versicolor)* parrots (King 1978). In fact, most of the Caribbean parrots are in peril and many, such as the Cuban macaw *(Ara tricolor)* and the Puerto Rican parakeet *(Aratinga maugei)*, are already extinct (see Clark 1905a, 1905b, 1905c; Greenway 1958).

Parrot's Decline

Although the Puerto Rican parrot was found throughout Puerto Rico when Columbus discovered the island in 1493, the population steadily declined with European man's colonization of the island. Much of the original forest was cleared to make room for farms and grazing land for domestic animals. In the early 1900's there were still several thousand parrots, but a drastic decrease in the parrot's range had occurred; by about 1940 the species was found only in the Luquillo Forest of eastern Puerto Rico where it exists today in the 11,330 hectare (28,000 acre) Caribbean National Forest.

The U.S. Forest Service recognized the Puerto Rican parrots' vulnerability in the mid-1940's and began programs to protect the bird and its habitat. However, nothing had been done to understand the species' ecology or reasons for its decline and little was done for the next 20 years. Despite the Forest Service's efforts, people continued to harvest the young birds for pets. The Service, not realizing its importance, even encouraged the removal of the parrots' favorite nesting tree, the palo colorado *(Cyrilla racemiflora)*, then considered a weed species. A study conducted by biologists of the Commonwealth of Puerto Rico Department of Agriculture in the early 1950's revealed that the parrot population had declined from the estimated 2,000 in the 1930's (Wadsworth 1949) to about 200 birds (Figure 1) and the species was experiencing extremely low reproductive success resulting from a number of factors (Rodriguez-Vidal 1959).

Research

The first full-time parrot biologist was hired in 1968 by the U.S. Fish and Wildlife Service's Endangered Wildlife Research Program and the U.S. Forest Service, with financial incentive from the World Wildlife Fund. The precipitous decline of the parrot was further verified when the Service biologist found fewer than 15 percent of the population (24 of 2,000 parrots) recorded only 14 years before. The possibility for the species' survival looked extremely poor and a crash captive breeding program appeared essential to save the species. In captivity the parrots would be provided security against such natural disasters as tropical storms and disease that might wipe out the remnant Luquillo population.

The potential for captive propagation of the Puerto Rican parrot appeared to be good because of the species' behavioral characteristics and nutritional requirements, and the successful captive propagation of other *Amazona* species (about 60% of the species have been bred in captivity; Nichols 1978, Noegel 1979). Captive propagation has been a useful tool in the recovery of other species in the U.S. Fish and Wildlife Service's Endangered Wildlife Research Program (e.g., masked bobwhite, *Colinus virginianus ridgwayi*; whooping crane, *Grus americana*; and Aleutian Canada goose, *Branta canadensis leucopareia*), but none of these other efforts have been as closely integrated into the management program to save the species as has been for the Puerto Rican parrot. The Endangered Wildlife Research Program began a propagation effort for this species at the Patuxent Wildlife Research Center in 1970 with the construction of facilities and testing of them with the more abundant Hispaniolan parrot *(A. ventralis)*. The use of a surrogate (captive population of a closely-related, nonendangered species) is part of

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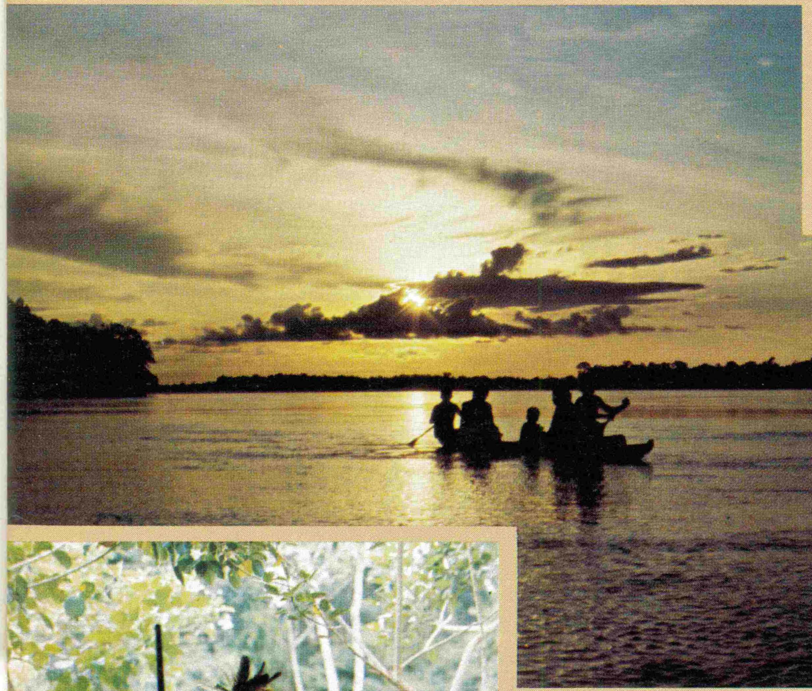

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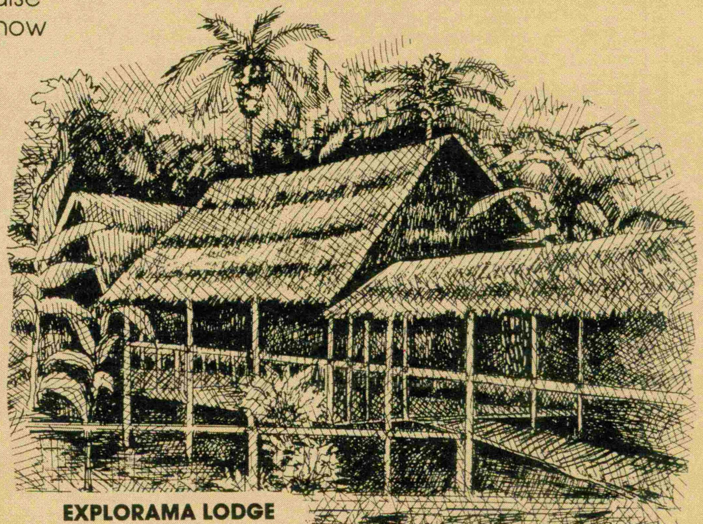

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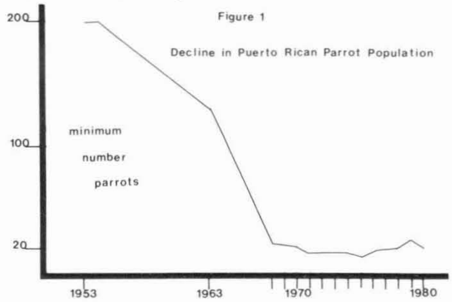
all captive studies at Patuxent. Treatments, techniques, and risks of any kind are tested with the surrogate species before using them on the rarer species (Erickson 1968).

Luquillo Aviary

In 1971 the Puerto Rico Zoo at Mayaguez donated the first 2 Puerto Rican parrots to the Patuxent program. These birds had been taken as nestlings from the Luquillo Forest in 1956, but the zoo did not attempt serious captive propagation until the late 1960's. In 1972, 1 of 2 wild birds recently caught in the Luquillo Forest for transfer to Patuxent died while in the U.S. Department of Agriculture Animal and Plant Health Inspection Station quarantine in Florida. The survivor was added to the Patuxent flock after an additional quarantine at Patuxent. An outbreak of Asiatic Newcastle disease in Puerto Rico dictated that these and all other parrots transferred from Puerto Rico would undergo a similar rigorous and potentially hazardous quarantine. The decision to establish a 2nd aviary in Puerto Rico was made shortly after the loss of the quarantined bird, as such risks were unacceptable, and a captive facility was needed to support other aspects of the field studies in Puerto Rico. The Luquillo aviary would furnish a place to care for eggs, and to rehabilitate sick and injured birds from the wild, as well as a site for captive breeding. Construction of the Luquillo aviary was completed in late 1973. Facility design, medical care, nutrition, and other management programs were provided by Patuxent and successful psittacine aviculturists.

In 1973, 5 Puerto Rican parrots were taken as eggs or chicks from the forest and placed in the new aviary. Since 1973 all additions from the wild to the Luquillo aviary have been either as eggs or nestlings. Meanwhile, captive propagation studies continued at Patuxent; in 1973, the first Hispaniolan chick was hatched and reared there. However, it was decided that the parrot captive propagation effort should be centralized at the field aviary, so by the end of 1978, the captive Puerto Rican and Hispaniolan parrot populations were transferred from Patuxent to the Luquillo aviary.

During the development of the captive program, the team of Fish and Wildlife Service and Forest Service biologists and technicians found a myriad of problems threatening the parrots' survival in the wild. It was immediately evident that the parrot is an extremely difficult species to study in the field, living in dense tropical forests and being exceedingly wary and inconspicuous. However, through the use of treetop observation lookouts and blinds placed near the nests, the habits of the birds were slowly revealed.



Reproduction

The parrot's reproductive season normally extends from February through June (egg laying to fledging); most eggs are laid in late February and March. The bird lays 3 eggs per clutch (observed range 2-4), and double-clutching is known at nests where the first clutch has failed. Incubation, done by the female without help from the male, lasts 25 to 27 days. Normally the male feeds the female near the nest during the incubation and early nesting periods. The young leave the nest when they are 60 to 70 days old, but the birds continue as a family unit for the remainder of the year, and the chicks may remain closely associated with the adults through the early part of the next breeding season. The birds eat fruit and seeds in season, especially fruit of the sierra palm *(Prestoea montana)* and tabonuco *(Dacryodes excelsa)*, leaves of the magnolia *(Magnolia splendens)*, and flowers like those of the *Piptocarpha tebrantha* vine.

Threats to Survival

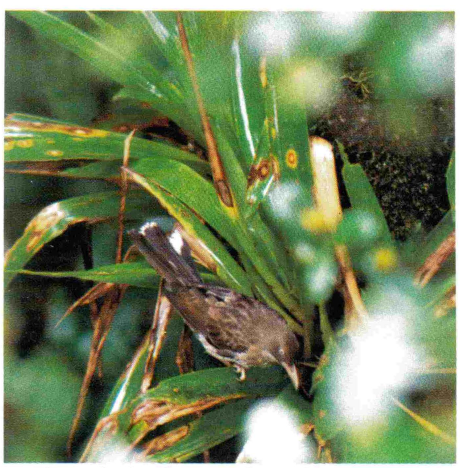
Threats to the adults, although few, include probably 2 raptors (red-tailed hawk, *Buteo jamaicensis*, and broad-winged hawk, *B. platypterus*; fighting among themselves for nesting sites; tropical storms; disease; and man. Threats to the eggs and young include severe weather, pearly-eyed thrashers *(Margarops fuscatus)*, rats *(Rattus rattus)*, honeybees *(Apis mellifera)*, and the parasitic larvae of the warble fly *(Philornis pici)*. Man has been a historical threat through habitat destruction and harvesting chicks for pets and food. Honeybees pose a moderate threat to chicks in the late nesting season, but are a more serious threat after the chicks have fledged (the normal honeybee swarming period) when they may take over a vacant nesting cavity for their hive and render it unavailable to parrots for the next season. Rats are normally not a problem at parrot nests unless the eggs or young chicks are left unguarded by the adults overnight, an unusual occurrence unless something has happened to the adult female parrot.

Warble fly larvae burrow under the

nestling's skin and may seriously debilitate a chick by destroying muscle tissue or causing infection, which could lead to the chick's death. Removal of the maggots has saved the lives of 3 badly infested parrot chicks. The warble fly also parasitizes thrasher nestlings, so that species has been the subject of tests to determine the effectiveness of several preventative treatments. Pyrethrin powder dusting at 2-day intervals and a commercial "pest strip" attached to the roof of the thrasher nest boxes controlled parasitism and subsequently improved thrasher nesting success. We will soon attempt some biological control experiments (garlic, tobacco). As with all other procedures used in the project, the warble fly control methods will be thoroughly tested on surrogate species before incorporation into the Puerto Rican parrot management program.

Pearly-eyed Thrashers

Like parrots, pearly-eyed thrashers also nest in cavities and in prospecting for nest sites may discover and destroy the uncovered eggs or young parrot chicks while the adult female parrot is off the nest. During the early years of the study (1968-1971) parrot nesting sites were thought to be plentiful due to the size of the forest and the number of large trees. However, it was later discovered that the number of suitable cavities and the fierce competition for these sites from cavity-nesting thrashers severely limited the availability of adequate nesting chambers (Snyder 1978, Snyder and Taapken 1978). To determine thrasher cavity depth and design preferences, we tested a variety of nest box styles and dimensions in several experiments. Although they showed a wide tolerance



Pearly-eyed thrasher searching for prey among bromeliad bracts.

for nest cavities, thrashers choose shallower nest sites than the parrots. In fact, the parrots were apparently using shallower cavities than they preferred because only shallow cavities were available. Special parrot boxes were designed or natural cavities modified (deepened and

Artificial nest site used by pair of Puerto Rican parrots in Luquillo Forest. Polyvinyl chloride tube replaced the natural palo colorado tree cavity which had rotted away.

made darker within) to take advantage of differences between the parrot and the thrasher nest site preferences. The parrots have responded positively toward these deepened and protected nests so that now all wild pairs are using such modified sites.

By encouraging a pair of thrashers to nest in the parrot's nesting territory, we have actually used the thrasher to protect the parrot nest site. A thrasher nest box is placed distant enough from the parrot's nest to reduce conflict between the 2 species, but close enough to contain the parrot nest site in the thrasher's territory. The nesting thrasher pair defends its territory from other intruding thrashers, thereby protecting the parrot nest from incidental thrasher nest prospecting and predation (Snyder and Taapken 1978). Since incorporating these measures into the program, the parrots have experienced no serious problems with thrashers.

All parrot nests have now been modified with plastic, fiberglass, and other materials to exclude rain and discourage predators from entering. A trap door near the bottom of the cavity allows access to the chicks and eggs. Doors in the bottom of the nesting cavities are essential if young are to be regularly inspected for general health, proper growth, and to detect infestations by warble fly larvae.

Propagation

Our objective in both the wild and captive parrot projects has been to get every bird we can back into the wild to bolster the wild population. However, when sufficient wild nests are not available for fostering captive-produced chicks or when such wild nests are out of synchrony with the captive production, we have had to retain chicks in the aviary. We are anxious to work on another alternative: the release of captive-held chicks into the wild as free-flying birds. This will require pre-conditioning so the birds will be able to cope with the environment of the forest without the succor of adults to teach them such survival essentials as predator avoidance and which foods to eat.

Many captive Puerto Rican parrot eggs and most of those rescued from jeopardized wild nests are incubated in an incubator room separate from the rest of the aviary. A Petersime[2,3] (Model 1) and several Lyons[2,4] cabinets are used for incubation and hatching. The incubation and hatching environments are similar to those used for the domestic fowl. The hatchery and its equipment are monitored constantly during the incubation season. The hatchery has a stand-by generator to supply electricity to the facility when the municipal supply is interrupted, a situation which seems to occur at the remote field aviary with amazing regularity during the hatching period.

Sexing

Sexing the Puerto Rican parrot was one of the most frustrating tasks in the propagation effort. The Puerto Rican parrot is not sexually dimorphic, and although behavior can be a reliable method of sexing heterosexual pairs, homosexual bonds do form. The Puerto Rico Zoo pair, mentioned early, was one of these homosexual pairs. Pair bonds formed by the homosexual pairs are strong and almost impossible to break once established.

Since it is important to know the sex of our captives to avoid homosexual pairings, we sought techniques that carried as little risk as possible to the bird, but could be used to accurately determine sex. We tried 3 sexing techniques based on dif-

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[1]Research in collaboration with Institute of Tropical Forestry, Southern Forest Experiment Station, USDA, Forest Service.

[2]Use of trade name does not imply endorsement of commercial products by the Federal Government.

[3]Petersime Incubator Co., Gettysburg, Oh 45328.

[4]Lyon Rural Electric Co., San Diego, Ca 92112.

ferences in the genetic material found in the cell: karyotyping, nuclear density, and sex chromatin. At the Houston Zoo, Greg Mengden attempted to use a karyotyping procedure early in the program (1975) to determine the sex of 8 captive Puerto Rican parrots. The chromosome technique is a safe procedure, as it only requires cells from the pulp of a rapidly growing feather (Mengden and Stock 1975). Even after some repeated samplings, good preparations had not been obtained from several birds. At least 2 of the birds were found to be improperly sexed a few years later. Sexing errors may be attributed to the poor samples rather than the technique. Although this is basically a very good procedure, it is not a practical technique for sexing large numbers of birds or for sexing birds within a short time.

Adult male Puerto Rican parrot at entrance to artificial nest cavity in Luquillo Forest.

Nuclear density, a technique to measure DNA (genetic material) in the nucleus of the cell (red blood cell in birds), can be used to detect the small difference in DNA between sexes. The difference exists because the ZZ sex chromosome of the male contains more DNA than the ZW chromosome of the female. However, Dr. Ellen Rasch (East Tennessee State University Medical School) found that while the differences in nuclear density are sufficient to determine sex between male and female cranes, they are insufficient to determine sex in Puerto Rican parrots with the present sensitivity of the method.

Sex chromatin is a reliable way to determine sex in most mammals and fish, and in some birds. Sex can be determined by locating the sex chromatin (W-body) in the non-dividing cell. In birds, the tissue sample is obtained by pulling a growing feather and examining the feather pulp cells (Bloom and Macera 1974). Dr. Steven Bloom (Cornell University) examined the feather pulp from our Puerto Rican parrots in 1977 but was unable to locate the sex chromatin. The number of pigment granules in the feather pulp interferred with the procedure. It may be possible to locate sex chromatin in cells from a different source. It is unfortunate that these techniques (karyotyping, nuclear density, and sex chromatin) are not yet usable since these methods are equally effective for any age birds. Sexing at an early age has the advantage in that if the sex ratio of the captives is skewed toward one sex, the sexes of chicks can be determined at the wild nest and the desired individual selected.

Laparotomy (surgical observation of the gonads) is a quick and very effective sexing technique, especially with the new optic fiber scopes. But since it does require surgical procedures, including restraint and an abdominal incision, the more conservative sexing techniques, such as those previously described, are generally preferred. Eight laparomtomies were performed on Hispaniolan parrots, but 2 of the animals died the night following the operations. Postmortem examinations revealed no hemorrhage or other apparent complications associated with the operation. Stress of the operation combined with a possible mild nutritional imbalance may have contributed to the deaths. No laparotomies have been performed on Puerto Rican parrots.

Our sexing method of choice is the fecal steroid analysis conducted at the San Diego Zoo by Dr. Arden Bercovitz, Nancy Czekala, and Lyndall Erb (Bercovitz, *et. al.* 1978) because it does not traumatize or stress the captives and it has proven to be reliable. The technique involves the collection of fecal material from which sex hormones are analyzed to determine relative estrogen:testosterone levels. Although fecal steroid analysis has been an effective method for determining sex in our adult parrots, as yet sex cannot be determined for birds less than a year old. We need a reliable sexing method for our young birds too, and the biologists at San Diego are now refining their technique for use with the young parrots.

Artificial Insemination

The captive Puerto Rican parrot flock contains more females than males (8 females and 6 males), 1 pair of homosexual females, and 2 heterosexual pairs that produce infertile eggs. The reason for the infertility appears to be a reluctance of the males to copulate with the females. Female Puerto Rican parrots have shown no reluctance to lay eggs, and even unmated Puerto Rican and Hispaniolan parrots have laid eggs. Obviously, artificial insemination of the mateless females and the homosexual pair will be necessary to obtain fertile eggs. Artificial insemination would also be useful in fertilizing eggs of females whose mates have some behavioral problems and are not copulating (Gee and Temple 1978).

In 1978 a week was spent in Puerto Rico developing methods of semen collection and insemination. Although 3 fertile eggs were produced by an inseminated bird, the fertility was probably due more to subsequent copulations by the male than to artificial insemination. Semen was successfully taken from a second male in 1979, but in such small quantities that it was not used to inseminate a female. The time available for insemination-related activities has been limited by demands from wild parrot management and nest protection.

Some day we may be able to collect semen, freeze and store it in a sperm bank. Cryogenic (low temperature) semen storage has been possible in cranes, geese, and ducks at Patuxent. Fertility rates are not adequate for general insemination (about 30% compared with 85% with fresh semen), but are satisfactory for introducing the genetic traits of the semen donor into the population inseminated. It is imperative that the genetic diversity of captive endangered species be maintained because that diversity enables an animal population to adjust to a changing environment and retain vigor. Although captive propagation or protection of a small remnant population in its natural habitat ensures survival of the species for many generations, it generally leads to a loss of the genetic diversity that has proven essential to the survival of the species (Mettler and Gregg 1969).

Captive Gene Pool

The captive flock contains birds with genetic representation of all recent wild pairs of Puerto Rican parrots, including some birds that have since died. As such there are 8 different or partially different family stocks represented among the 14 captives. Because the captive flock contains offspring from wild pairs that are no longer alive, we will be able to increase genetic diversity within the wild population by introducing captive-produced descendants into the wild flock. As the captive flock contains this wide genetic representation of the wild population, our current policy is to take additional birds into captivity only when the chick clearly would not be able to survive at the nest.

Double Clutching

Double clutching is a technique whereby removal of a pair's first set of eggs results in the female laying a replacement clutch. By artificially incubating or placing the first clutch under another female and allowing the pair to raise it second clutch, production of offspring is increased considerably. This technique has been very valuable in the recovery efforts for the endangered peregrine falcon, *Falco peregri-*

nus (Fyfe *et. al.* 1978), and whooping crane (Kepler 1978). We have experimentally double-clutched captive and wild Puerto Rican parrots, which has resulted in 2 pairs producing second clutches (1 out of 3 clutches was fertile) in the aviary and in 1980, 1 wild pair producing a fertile replacement clutch. Future management for the Puerto Rican parrot will take advantage of their clutch replacement characteristics and could greatly increase production within the aviary and in the wild.

Aviary Use

Captive Hispaniolan parrots, which are housed on a different floor of the Luquillo aviary, have proven useful in a number of ways in the conservation program for the Puerto Rican parrot. Productive Hispaniolans are used to incubate Puerto Rican parrot eggs, and, most importantly, to raise their young. Puerto Rican young brooded under Hispaniolans show more normal growth that those hand-raised by us and are better conditioned for placement into foster Puerto Rican nests in the forest. Also, fostering chicks under Hispaniolan parents greatly reduces the need for human labor and avoids possible imprinting difficulties associated with hand-raising. These Puerto Rican parrot chicks, if of the right age, are used to increase the number of birds fledged per family in the wild or to replace eggs or young in an unsuccessful wild nest.

In areas of heavy predator pressure, eggs have been taken from the wild nest (exchanged with substitute dummy eggs) for safe keeping at the aviary. The eggs hatch in the hatcher or under Hispaniolans, and all young Puerto Rican parrots are reared by Hispaniolan parrots for several days before returning them to the nest. The dummy eggs are removed from the wild nest when the young parrots are substituted. In the past, plaster eggs have been full of holes where thrashers have tried to destroy the eggs in the nest.

Hispaniolan young of a similar size are used to replace injured or diseased Puerto Rican parrot young while the latter are treated at the aviary. The Puerto Rican parrots are replaced in the nest when they are healthy and the Hispaniolan returned to the aviary. Hispaniolan parrot young are also substituted into nests in place of Puerto Rican parrot chicks when the chicks are in any kind of danger (e.g., desertion, predation, defective nest). At 1 nest in 1979 the female deserted the nest just as the chicks were hatching; only the male was left to care for the chicks. One chick survived, but rather than risk losing it, we substituted a Hispaniolan chick during this period of uncertainty.

Complicating the situation, another pair of Puerto Rican parrots tried to take over the nest. The Hispaniolan suffered through the ordeal of not being brooded during the nights, poor feeding rates (we supplementally fed the chick when the male was off foraging), and a wet nest. We could not have taken such risks with a Puerto Rican parrot chick, but use of the Hispaniolan chick let us keep the nest active until the chick was ready to fledge. The situation did not improve enough to allow the Puerto Rican chick to be replaced and fledge from this nest, but through the use of the surrogate, the nest was kept active to the very end and the pair returned in 1980 and successfully raised 5 chicks. These types of manipulations expand the reproductive potential of the wild and captive Puerto Rican flocks.

Hope for Survival

The conservation program has halted the decline in the Puerto Rican parrot population, but much needs to be done to re-establish the parrot as a healthy population in the Luquillo Forest. The species still lingers on the brink of extinction due to the small number of animals in the population, its restricted range, and small, relatively young captive population. Furthermore, the security of the captive population is compromised due to its location within the same forest as the wild population and the maintenance of all captives in one building. It is intended that other captive flocks will be established in several geographically isolated locations on Puerto Rico and elsewhere as insurance against natural catastrophes. Also, once the wild population has shown clear stabilization in the Luquillo Forest, new populations will be re-established in Puerto Rican forests within the species' former range.

The captive program has played a prominent part of the recovery effort, and its role will take on additional importance with expanding production. In 1979 a milestone was achieved in the captive program when the first captive-produced Puerto Rican parrot chick was raised. That chick was fostered into a wild nest which it shared with a wild foster sibling before fledging in June. Two captive-produced Puerto Rican parrots were raised in 1980 and these were also fostered into a wild nest from which they successfully fledged. The 1980 family of 4 was still together in February 1981 when we observed them foraging in their nest valley. If we can improve the survival of young parrots after fledging and increase captive production so more chicks can be fostered into wild nests or released, we are optimistic that the wild population may yet survive despite the tremendous odds it faced when the program was begun 12 years ago. •

For literature cited write editor.