

included the Sabinal river mouth since the pollution and sewages it maintains seven adults animals which varies in size, ranging from 2.5 to 4.5 m of total length in no more than 100 meters.

Monthly diurnal and nocturnal visits were carried out. At the first ones, the river was reviewed as well as its shores looking for crocodiles at basking or visiting sandy areas for nesting proposes. When the nests were located, the basic measures were took such as: date, location, distance to the water, deep, circumference, incubation chamber temperature as well as environment temperature. From each detected nest at the begging of the incubation (less than three days) were collected 10 eggs (leaving the rest of the eggs in the site), were identified and measured, transported in polyurethane containers with nest material to ZOOMAT and were incubated artificially under controlled conditions. During the nocturnal visits the mayor area of crocodiles activity was ubicated and delimited. The countings begun one hour after sunset in a plane floor aluminum boat four meters long with an eight horsepower outboard motor and a pair of spotlights counting the animals eyes by reflection. Also temperature and water level were measured at the beginning and at the end of the census. The site of capture was registered at the moment; cloacal, water and air temperature were recorded with a Bravo contact thermometer. Total length, Snout - vent, head length and wide, postoccipital, nuchal, dorsal, doble crest, single crest and lateral caudal scales were measured in each crocodile. The captured animals were marked by cutting the scales of the double and single crests, and were released in the same site. Both, the hatchlings born by artificial incubation and the captured ones were transferred to ZOOMAT and were kept under stable conditions of temperature. They were marked, fed every other day and monthly measured at weight and total length. When they were one or two years old they were released in the areas with better aquatic vegetation coverin the park.

**RESULTS** 16 visits were realized since March 31st 1995 and finished on March 18th 1996, 128 observations hours were recorded from which 65 were nocturnal. Were counted since 14 to 38 animals without include 1995 neonates or crocodiles released by ZOOMAT. The dates of mayor countings were May 18th and September 6th 1995. 52.63% of the counted population is conformed by adults (2m. or more); 13.15% sub-adults (1.5-1.99 m.); 5.27% four years old (1.2-1.5 m.); 15.8% two years old and 13.15% one year old. In other samplings, five different specimens from 1992 class (three years old) were counted but they were not seen in the biggest sample from May 18th 1995 which was the one who we analyzed. 40 wild crocodiles were captured, measured, sexed and marked from the following classes: 23 (1995), 9 (1994), 6 (1993), 1 (1992) and 1 (1991), and released in the same place. Was very notorious that from the total of all captured animals 37 were males and 3 females. Some adults crocodiles permitted a good approach but we dont have experience enough to manage animals bigger than 3 meters. Territoriality was observed in adults, sub adults and some juveniles because they were found in the same places each sampling. Five active different nests were found. Three were found in 1995 and three in 1996; one of them had repetition in both years. One nesting zone was common for three females in the same year, the maxim distance among the nests was 15m. and the less 7.5m. (table 1). Laying date was from middle March to early April. Natural eclosion was from middle May to early June. In 1995, we collected 7 fertile eggs and incubated artificially at ZOOMAT; from these, 6 males hatchlings were obtained with normal characteristics and similar sizes to their brothers left at the natural nest, the artificially incubated ones, born 4-6 days later. In 1996, 28 fertile eggs were collected from 3 different nests and we expect their exclusion at early June, according to the growth of the bands, the embryos develop correctly, the temperature was maintained constantly between 29.5 to 30.5 C during the first 45 days for increasing the female proportion (Thorbjarnarson pers. comm.). Data of the collected eggs for artificial incubation and obtained hatchlings in this procedure are shown in table 2. Thirty-five wild neonates were collected: 2 m

1993, 7 in 1994 and 21 in 1995 to keep them for one or more years at ZOOMAT; all of them were males. Their growth was constant but lower in 1994 and similar in 1995 to the shown by those born in captivity (Sigler in press.). In 1995, 9 crocodiles were released, they belonged to the 93 and 94 classes and were captured at borning in the study area and in 1996 25 from 95 class will be released. From the 23 hatchling born in captivity from ZOOMAT parents, 11 were released in 1995 (Sigler 1994) and 10 will be released in 1996. From the 18 recaptured crocs, 15 of them were kept one year in captivity and released by ZOOMAT and the rest were wild animals. In March 1995, the zoo was notified about the presence of a dead crocodile in the Sabinal river, due to the moldering changes it could not be necropsied, however, her skeleton was conserved and prepared for scientific collection. This 2.94m. adult female was about to laying because were found more than 30 eggs, and in spite of our attempts to incubate them were not possible due to their softy shell. During our visits, informal interviews were made with fishermen and boatmen from the touristic cooperative, founding that the reproductive season and nesting sites were unknown, but exist knowledge about eclosion time. They also reported the presence of some small crocodiles extracted from the park and kept in captivity in some houses from the surroundings. They have not had big problems with those animals by using the illegal trammel nets. The author appreciated in the Sabinal river mouth a fisherman who crossed the river swimming in front of three adults crocodiles and surprisingly was not attacked, this man argued that he never has any problem with crocodiles and these animals are able to avoid the trammel nets when they touched them with the top of the snout and retroceded.

**DISCUSSION** It is probable that population fluctuates around 80 animals, because the river shores offer a very big vegetal floating coverture were some crocodiles could not be seen during the samplings. Some crocodiles progress in the recognition of the spotlight at posterior captures, this effect has made difficult our last samples, however is possible it can be useful avoiding human predators. When the rainy season is present (July-September) the dam is carried down to 5m. less of the normal level, this fact makes the animals be lack of hides during the samplings, but also before their predators. It is possible that some hatchlings move to places beyond the Silence cave dragged by the strong flow of the river in the rainy season and they be ubicated in the dam, area still not monitored. The nesting sites availability is reduced because the river has not adecuated beaches for the crocodile reproduction, the almost 25 km of the Sumidero Canyon are coffered by the rocky walls and because that the crocodiles came out the Canyon and are located mainly at the beginning of the park and at the end of the dam, these are places with high human pressure both the animals as well as the habitats they occupy and indispensable they need.

**CONCLUSIONS** It is important to carry out for one more year this study to evaluate the survival of the released animals which attempt to be acceptable. During 1996, the feeding habits evaluation will start. We consider important to manage the wild nests to orientated their production towards the obtention of females and in this way nivelate the sex ratio in juveniles. This National Park in considered a good place for releasing hatchlings born in captivity at ZOOMAT and due to the nearness with our facilities, we can give them a periodical monitoring. In the near future is contemplated studying the population across the Grijalva river, detecting the better populations and suggesting to the settlers of each area, measures for living together and for touristic approachment of the crocodiles.

**ACKNOWLEDGEMENTS** This study would not be possible without the enthusiastic support of IDEA WILD project 040 that donated the outboard motor and made the difference. Special thanks are given to the sapo-iguana team in collaborating with every work visit.

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# Head-starting and Translocation of Juvenile Crocodylus acutus in Lago Enriquillo, Dominican Republic

by

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## ABSTRACT

Since 1992 the Dominican Republic is carrying out a conservation project in Lago Enriquillo, a hypersaline inland lake, where the country's only Crocodylus acutus population is surviving. The population had declined from 300-600 adults in the early 1980s to some 200 in 1992, mainly due to illegal killings. The Dominican Wildlife Department (DVS), in charge of animal and plant conservation, developed an action plan to secure the crocodiles' survival and well-being. An inter-institutional executive council was formed to plan and supervise the plan's activities.

Because of the delicate state of the Crocodylus acutus population the council decided in 1993 to take eggs to the Santo Domingo Zoo for a headstart program. In April and May 178 eggs and 53 hatchlings were taken to the zoo. A total of about 130 neonates were in the zoo by the end of 1993. Most of them died in 1995 due to severe internal problems in the zoo.

In 1994 it was decided to translocate the hatchlings from the nesting beaches on the islands to freshwater habitat along mainland shores, in order to improve survival rates. 255 hatchlings were translocated, after being marked. In Los Borbollones area some 20 % were recaptured. Since post-hatching care is an important phenomenon in Lago Enriquillo crocodiles, we are not sure if mortality was reduced significantly by our translocation activities.

Growth rates in the juveniles were 30 to 35 mm per month. Mean weight gain in the first year was 63 g / month, in the second year about 200 g / month.

## RESUMEN

Desde el 1992 la República Dominicana está llevando a cabo un proyecto de conservación en el Lago Enriquillo, un lago hipersalino en el interior, donde la única población del cocodrilo americano (Crocodylus acutus) del país sobrevive. Esta población ha declinado de 300 - 600 adultos en los años 80 a unos 200 en el 1992, sobre todo por matanzas ilegales. El Departamento de Vida Silvestre, responsable para la conservación de animales y plantas, desarrolló un plan de acción para asegurar la sobrevivencia y el bienestar de los cocodrilos. Un consejo ejecutivo, de forma inter-institucional, fue formado para planificar y supervisar las actividades del plan de acción.

Por el estado delicado de la población de Crocodylus acutus en el lago, el concejo decidió en 1993 de llevar huevos al Jardín Zoológico de Santo Domingo para iniciar un programa de crianza en cautiverio. En abril y mayo 178 huevos y 53 neonatos fueron llevados al zoológico. Un total de unos 130 neonatos estaban en el zoológico a finales de 1993. La mayoría de ellos murió en el 1995, por fuertes problemas internos de la institución.

En 1994 fue decidido trasladar los neonatos desde sus playas de anidamiento en las islas hacia los sitios de agua dulce en las orillas del lago, con fines de aumentar la tasa de sobrevivencia. Unos 255 neonatos fueron trasladados, después de haber sido marcados. En el área de Los Borbollones un 20 % de ellos fueron recapturados. El cuidado que las cocodrilas les dan a sus crías parece ser un fenómeno muy importante en el Lago Enriquillo. Así no estamos seguros, si la mortalidad ha sido reducida significativamente por las actividades de traslado.

Las tasas de crecimiento de los juveniles eran 30 - 35 mm por mes. El promedio del aumento de peso era 63 g por mes en el primer y 200 g por mes en el segundo año.

## INTRODUCTION

The American crocodile (*Crocodylus acutus*) was once abundant along much of the Hispaniolan coastline (Descourtilz 1809, Moreau de St. Mery 1797). Today, in Haiti there are only small populations along the coast and in Etang (Lake) Saumatre (Thorbjarnarson 1988); and in the Dominican Republic *C. acutus* has only persisted in Lago Enriquillo (SEA/DVS 1993).

Lago Enriquillo is situated in the Neiba valley in the southwestern part of the Dominican Republic, bordered by two 2000 m mountain ranges. It has a water surface of about 200 km<sup>2</sup>, a length of 35 km, a width of 11 km and a maximum depth of 22 m (Araguás et.al, 1993). There is one big island, Isla Cabritos and two small islands, La Islita and La Barbarita. At the moment Lago Enriquillo has a salinity of 80 ppt (1996) and lies 40 m below sea level. The lake is a remnant of a marine channel that once divided Hispaniola into two paleo-islands (Mann et.al 1984, Inchaustegui et.al 1978). The climate is semi-arid: annual precipitation ranges between 470 and 780 mm and evaporation is estimated to exceed 2000 mm. Mean air and lake water temperatures are approximately 30° C. Water level, lake surface, and salinity vary significantly from year to year.

In the early 1980s the crocodile population of Lago Enriquillo was considered to be the biggest and densest for the entire species, estimating an adult population between 300 and 600 individuals (Inchaustegui and Bautista pers. comm.). Between 70 and 112 nests were found in the years 1977 through 1984 (Inchaustegui in SEA/DVS 1993).

Surveys carried out in 1990/91 by the Departamento de Vida Silvestre (Dept. of Wildlife) revealed alarmingly low numbers of nests and of crocodiles seen. In 1992 Vida Silvestre started the project "Study and Protection of the American crocodile". Surveys during the first months showed that the situation of the crocodiles was even worse than expected. Few adults were seen along the coast. Despite intense searching only four nests were located. Thereafter evidence of crocodile killings; bones, including smashed skulls, human tracks, wooden poles and crocodile traps were found in many places (SEA/ DVS 1993, Schubert and Santana 1996).

A "Surveillance Plan" was established and implemented for the whole lake. An "Action Plan for the Conservation of the American Crocodile" was worked out. It includes five programs: 1. Surveillance, 2. Investigation, 3. Reproduction, 4. Public Relations and 5. Resource Management. An interinstitutional "Executive Council" was created to plan and supervise the activities. Members of this council are representatives of the National Park Directorate (DNP), the Wildlife Department (DVS) and the Santo Domingo Zoo (ZOODOM). Grupo Jaragua (a national NGO) and the German Service for Development (DED) are included as consultants. The action plan is in its fourth year of implementation. Human impacts on the crocodiles have been reduced and public awareness improved significantly; the crocodile population is increasing slowly. More than 200 adults are now estimated for the lake area.

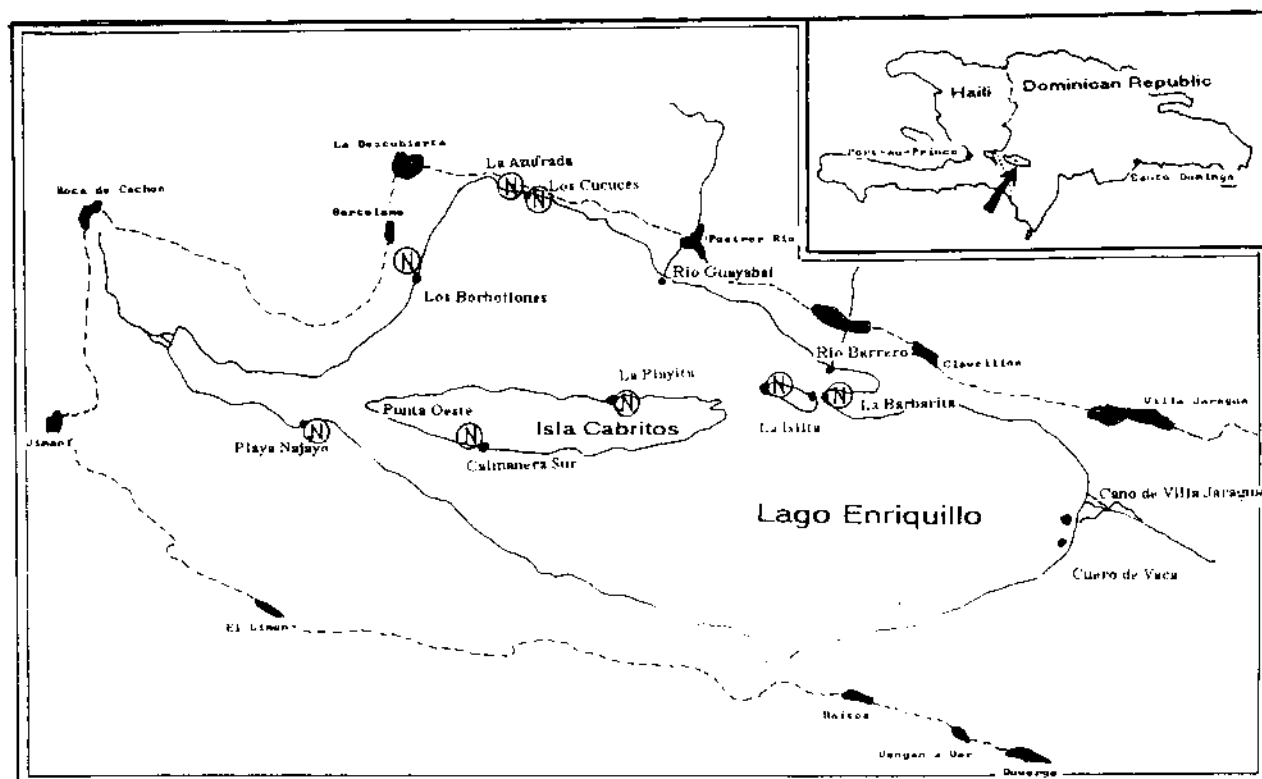


Fig. 1: Map of Lago Enriquillo. N: major nesting sites

## METHODS

Since 1993 all major nesting beaches have been patrolled every two to three days during the egg-laying period (January through March). Nests were opened, and the eggs were counted, measured and weighed, then put back in exactly the same position from where they were removed. In 1993, eight nests were reopened and the eggs were collected and taken to the Santo Domingo Zoo, after having incubated two thirds of their required time (a mean of 60 days). Once they arrived at the zoo, they were measured and weighed and checked for overall shell appearance. Then they were handed over to the zoo personnel. They were kept in an incubation room until they hatched.

Since 1993 was a good year for the crocodiles, in terms of reproduction and stabilization of the population, the "Executive Council" decided for 1994, not to take eggs or neonates to the zoo, but to evacuate them from nesting beaches, especially on the islands, to freshwater habitat on the mainland. In April and May 1994 255 neonates found on their nesting beaches were captured and taken to the mainland, where they were measured, weighed and individually marked by cutting the tail scutes according to a prearranged code. They were released in freshwater habitats along the northern and northwestern shore on the same day or a day later in four different localities.

At the same time we started a capture - recapture program with neonates and juveniles. In May and June 1994 this program focused on the neonates that hatched in the Azufrada area. They were either caught the day after hatching or some days later. After a break of three months, the capture - recapture program continued in September. Juveniles from 1993 and 1994 were caught, measured, weighed, sexed, marked and released immediately. Their position was mapped on small-scale maps (scale 1:20,000 m and 1:10,000 m).

## RESULTS AND DISCUSSION

**Nesting.** In Lago Enriquillo nesting activities generally begin in January or February. Most females frequently visit the beaches to select their future nesting site. All nests are hole nests, excavated in the sand. Mean number of eggs is 22 per nest. The smallest nest recorded had nine eggs, the biggest 36. Clutch size does not vary significantly between years or between nesting beaches.

According to Fig. 2, the number of nests varied greatly between 1990 and 1996. Due to severe human impacts only four nests were found in 1992. In 1993 and 94 the number of nests went up to 36 and 48 respectively, then dropped again in 1995 to only 14 nests (SEA/DVS 1994b and 1995b). This year we have located 30 nests so far, and we estimate a total of at least 40 nests for 1996.

### Total Number of Nests in Lago Enriquillo

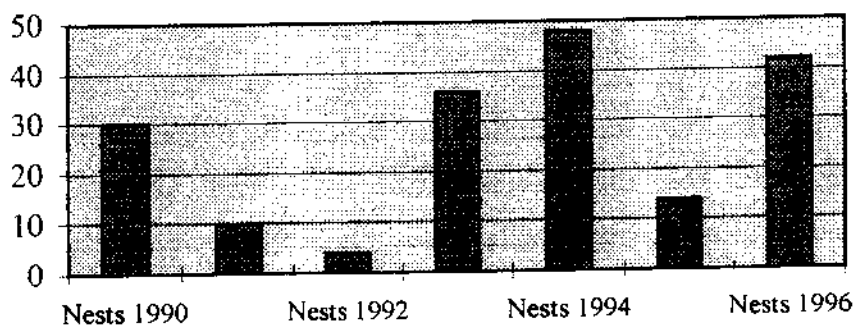


Fig. 2: Number of nest in the years 1990 through 1996. The 1996 number is estimated, based on 30 nest found after laying.



**Nesting beaches.** Some 32 km or 30% of Lago Enriquillo's coastline are sandy beaches. However less than 5 km are used for nesting. There are seven major nesting beaches in Lago Enriquillo, five of them of high importance with more than three nests per year and two of lesser importance. In two further beaches nesting was recorded on only one occasion. Four nesting beaches are situated on the islands. In the last five years a little more than half of the nests were found on island beaches. From these beaches the neonates have to cross up to ten km of the hypersaline lake to get to freshwater habitat on the mainland.

We assume that mainly human disturbances are the reason why Lago Enriquillo crocodiles tend to choose the islands for nesting. Killings of crocodiles and egg-robbing were common before conservation measures were taken. On the islands, especially on Isla Cabritos, human access is very limited.

Due to the very low reproduction success in 1992, the Executive Council decided to take eggs to the Santo Domingo Zoo for artificial incubation in a head-starting program. In April and May 1993 eight nests (22% of total) with 178 eggs plus 53 already hatched neonates were taken to the zoo. At the end of 1993 the zoo had some 130 juvenile *Crocodylus acutus* in captivity. The head-starting program failed completely. In 1995 a change of the zoo director led to severe internal problems, causing the death of many zoo animals (see also last CSG Newsletter). More than half of the crocodiles died and the rest are probably in a very delicate state of health.

In April and May 1994 255 neonates found on their nesting beaches were taken to the mainland, only 42 neonates were left on the beach where they had hatched. Visiting the same beach a day later we found that they had disappeared. In 1995 it was agreed not to evacuate any neonates, however, due to a misunderstanding by the park rangers 53 neonates were captured and taken to the mainland. Only 9 stayed on the nesting beaches. We estimate a total of 250 crocodiles hatched for 1995.

In May and June 1994 the capture and recapture rates showed that neonates were moving west towards an area with dense cattail stands and a high abundance of freshwater. They were using small freshwater ponds along the coast to hide during the daytime. On several occasions an adult crocodile, presumably the mother, was seen in the lake next to the little ponds. On one occasion the mother was even in the pond, hiding in the mud: one of us almost stepped on her.

Between September 1994 and March 1995 a stretch of about 4 km of coastline of major importance for the juveniles was patrolled monthly, ten juveniles or more were caught during each visit, all juveniles seen were registered and their position was mapped. The same activity was repeated between September 1995 and January 96. However, this time it was much more difficult to catch the juveniles, due to increased wariness. Only a mean of two animals per patrol was obtained.

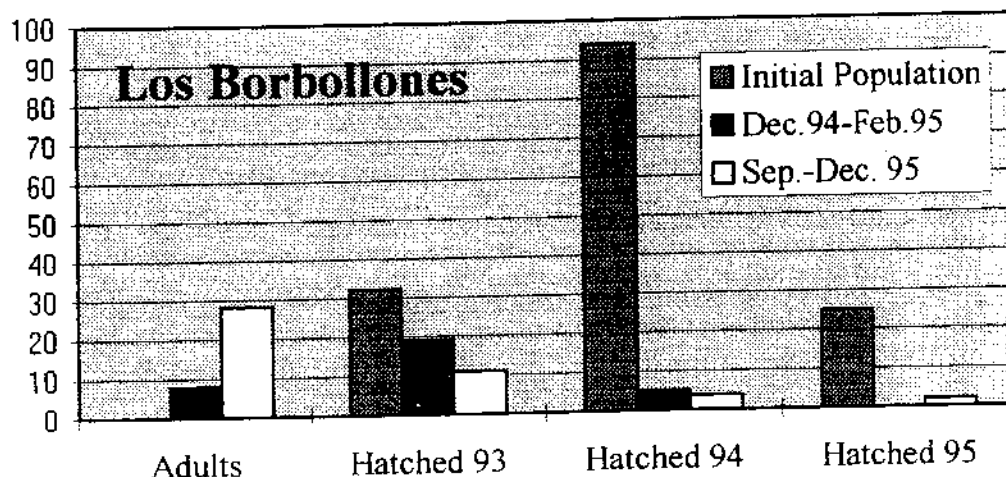


Fig. 3: Abundance of different year-classes in Los Borbollones.

Los Borbollones is one of the areas patrolled regularly. On a stretch of 1 km there are many freshwater springs that drain into the lake. Los Borbollones is an important site for adults who presumably stay here for some days to osmoregulate and, to a lesser degree, to nest (a mean of two nests per year). The streams that drain the springs and the brackish water along the shore form important habitat for neonates and juveniles. Fig. 3 shows the initial amount of neonates in Los Borbollones for each year class, as well as its abundance in Dec. '94 through Feb. '95 and for Sept. '95 through Jan. '96. While the 1993 born crocodiles diminished slowly, numbers for 1994 and 95 decreased drastically within the first months. The principal cause for this decrease is probably a very high mortality rate, rather than a high rate of migration. Even though juveniles were recorded to migrate distances of up to 3.6 km within two weeks, most of them stay in the same locality for many months. A 1993 juvenile, radio-tracked since January 1996 apparently has a rather small home range (less than 2 km of coast line).

However, it is important to point out that only a fraction of the juveniles are seen. In April and May 1994 some 82 marked neonates were released in the Borbollones area. Between September 1994 and March 1996 we captured 24 of them, corresponding to 19.5%. Another eight unmarked juveniles of the 94 age class were captured, so the relation marked : unmarked was 2 : 1. The unmarked animals came either from one of the two Borbollones nests or were brought to the area from other nesting beaches by their mother.

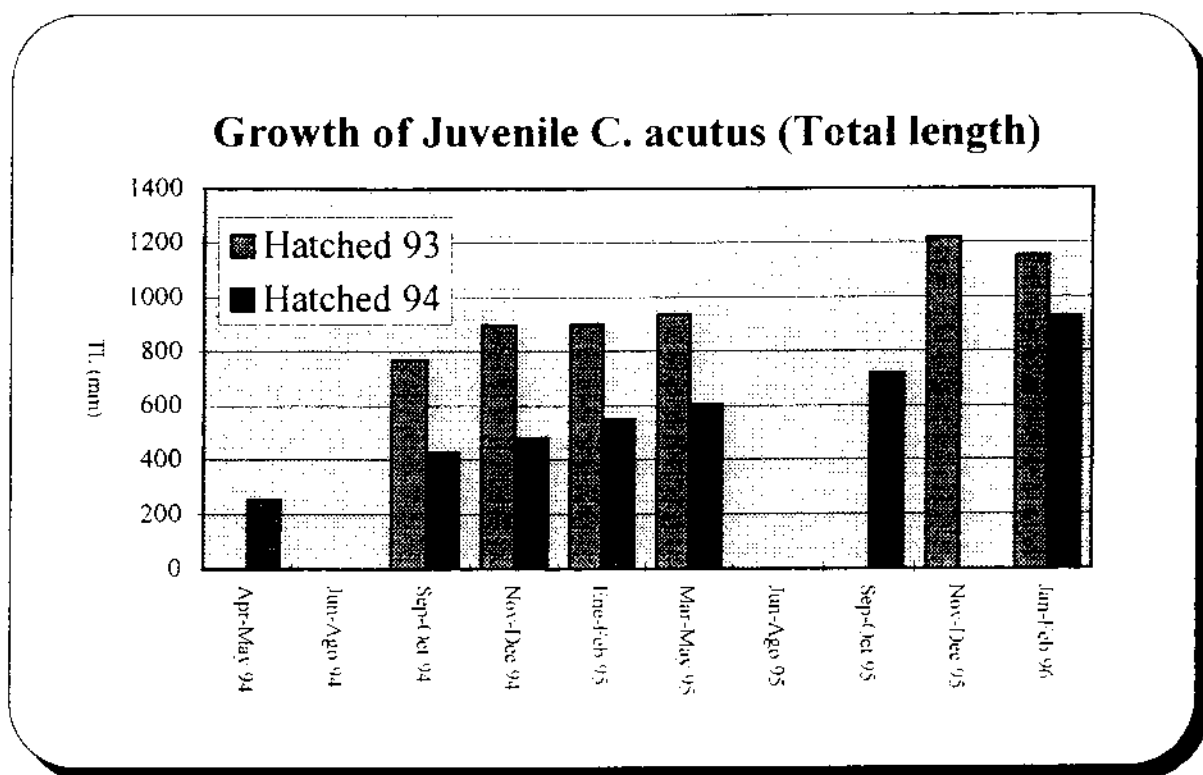


Fig. 4: Growth of *C. acutus* juveniles in mm per month (total length)

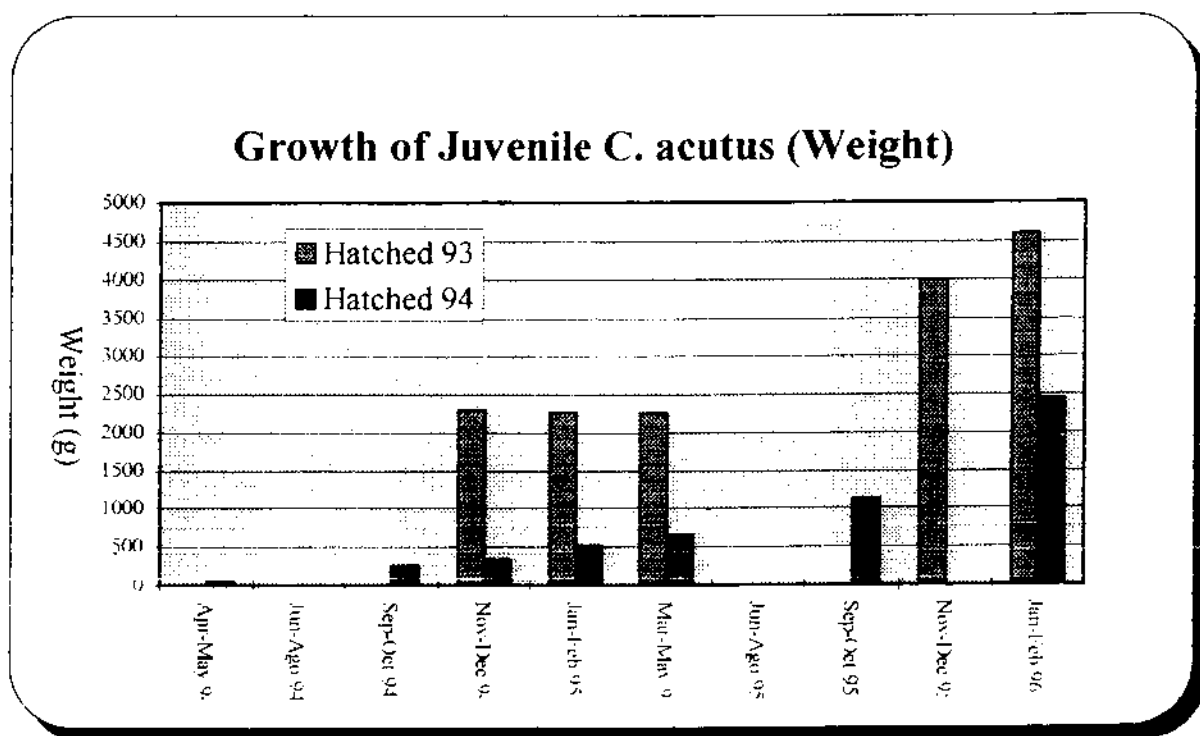


Fig. 4: Growth of *C. acutus* juveniles in grams (g) per month

**Growth.** Figures 4 and 5 show the mean total length and weight of juveniles hatched in 1993 and 94. In their first year, the total body length of both age classes increased by 30 to 35 mm per month. First-year neonates gained 63 g monthly, while second-year neonates gained almost 200 g per month. The 1993 hatchlings showed some cutback in their growth between November 94 and May 95. Their mean length and weight stagnated. Some individuals even lost weight (150 and 220 g per month). This stagnation might have been related to a rapidly lowering lake level and drying out of freshwater swamps next to the lake after August 1994. However the lake keeps shrinking but the juveniles are growing and gaining weight again.

The juveniles that were raised in the Santo Domingo Zoo (head-starting) were measured May 15 of 1995, when they were exactly two years old. Their mean total length was 532 (64) mm, corresponding to a growth rate of 11.5 mm per year or one third of the growth rate in Lago Enriquillo. They had a mean weight of 428 (171) g. This means they had gained weight by 15.5 g per month or one quarter that of Lago Enriquillo. We processed 103 juveniles, just before the severe problems in the zoo started. Originally some 130 neonates, forming part of the head-starting program were registered in the zoo at the end of 1993.

## CONCLUSIONS

The head-starting program had failed for reasons that could not have been foreseen nor was there a chance for the conservation community to influence the proceedings. Fortunately the Lago Enriquillo *Crocodylus acutus* population was not as depleted as it seemed in 1992/93.

The success of translocating hatchlings from nesting beaches to freshwater habitat has not been thoroughly evaluated yet. We are not sure if first year mortality was reduced significantly by translocating the neonates. Post-hatching care in the Lago Enriquillo *Crocodylus acutus* population might be more important than was previously credited.

On different occasions we saw crocodiles with up to six hatchling in their mouths swimming along the shore or even taking off from a nesting beach on the island. We have evidence that a mother removes neonates and hatching eggs from the nest and takes them to the lakeshore, where she hides them under rocks, in shallow pools or in the foam that is produced by wave action. Then she takes them to freshwater habitat. At least twice we saw female crocodiles staying next to their hatchlings for more than two months. This phenomenon was also recorded on video. In 1995 we observed the mother of one of the nests whose neonates were evacuated to check on its nest almost every night for over a month, before finally giving up.

## ACKNOWLEDGMENTS

We would like to thank the Departamento de Vida Silvestre of the Secretary of Agriculture and the National Park Directorate, especially their directors and all involved wildlife inspectors and park rangers for the great effort to establish and maintain the conservation activities concerning the Lago Enriquillo crocodile population. We also would like to thank the ex staff members of the Santo Domingo Zoo. The US Peace Corps and the German Service for Development (DED) provided technical help, the latter also gave very important financial support. Additional project activities are supported by the Swiss Development Agency HELVETAS and by the Wildlife Conservation Society. The IUCN Crocodile Specialist Group has always been concerned about the situation of the crocodiles in Lago Enriquillo.

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**Population Dynamics and Conservation Strategies  
for Crocodylus acutus in Bahía Portete, Colombia.**

**Gerardo Abadía**

Bahía Portete is located in the northern most part of Colombia, in the Guajira Peninsula on the Atlantic coast. Portete is a fairly small bay with consistent salinity values of 40 ppt., typified by a shallow coral reef formation on its West side, and a mangrove ecosystem South and East, which is home to a wild Crocodylus acutus population.

Since 1981, the construction of a big sea port for the exportation of coal has been under way and finally started operation in 1985. In addition to the sea port, there are two other human settlements: Puerto Nuevo and Puerto Portete both of which provide a place to unload merchandise coming from Aruba and Panama in small ships, as well as a place to live for an increasing community of fishermen.

Crocodylus acutus was formerly very abundant throughout its distribution range covering southern U.S.A., Mexico, Guatemala, El Salvador, Belize, Honduras, Nicaragua, Costa Rica, Panama, Jamaica, Haiti, Dominican Republic, Colombia, Venezuela, Ecuador and Peru (Powell, 1971). However due to the value of their hides, commercial hunting has wiped out most of the world's crocodile populations.

Crocodylus acutus was common in Colombia along the Magdalena Basin and on both the Atlantic and Pacific coasts, but due to commercial and local hunting, habitat destruction, irrigation projects for both agriculture and ranching and the fear of people, crocodile numbers have declined to a minimum (Medem, 1981). Crocodylus acutus is protected in Colombia by Resolución No. 573 of July 24th, 1969.

Crocodylus acutus in Bahía Portete is an isolated wild crocodile population with no migration or recruitment rates and therefore offers ideal conditions for the study of the population dynamics of the species. The fact that there are very few crocodiles left and

their important role within the food-chain (Fittkau, 1970; Craighead, 1967; Fittkau, 1973; Kushland, 1974) makes it worth the effort for their conservation.

## MATERIALS AND METHODS

A population estimate is not a census. Censi are very seldom carried out because they are time-consuming and expensive. Population estimates are based on direct observation of the animals during the day at their basking sites or during night counts. Usually an index as an account related to the animals presence (such as tracks, sand bank slides, nests, droppings or vocalizations) is also used (Davis and Winstead, 1987).

The Crocodylus acutus population in Bahía Portete has been monitored since November 1981.

Between 1979 and 1981 this population was subject to extensive commercial hunting. The total number of animals taken could not be determined. Two or three crocodiles were shot each night and their hides sold on the black market. By 1982 the commercial hunting ceased due to the low number of remaining crocodiles.

Between November 1992 and January 1993 a new study of the crocodile population was carried out in Bahía Portete. In total 8.8 kms. of beach and mangroves were inspected on foot and 25 hours of nocturnal surveys were completed by boat over a transect of 22 kms which was repeated six times. A Nauticol inflatable boat with an Evinrude Super 25 outboard engine was used whenever possible, and an Indian canoe was used when navigation became too difficult. An AUTOMAR Long Range Mod. 453 lamp and a 100 Watts DENJI lamp were used during night counts along with two Winchester headlights. Later, in 1996, a Frezzolini 30 Volt Spot Lite lamp and a 500,000 CP Cordless Lectronic Science Inc. lamp were both added as basic equipment during night counts. A temperature-compensated Reichert-Jung refractometer was used to determine water salinity.

## RESULTS

In 1992, night surveys revealed between 2 and 11 (mean 7) individuals in the 22 kms transect with only 2 individuals over six feet total length. Counts of tracks and sand bank slides indicated similar numbers and range of sizes in the area. Extrapolation of the survey data suggests a population of around 140 individuals with the great majority in size classes of one to six feet total length.

Ogden (1978) and Chabreck (1966) have both used the number of nests to estimate total populations of Crocodylus acutus and Alligator mississippiensis. When using their formula for six nests found in Bahía Portete, the total crocodile population ranges between 120 and 150 individuals.

In 1996, 90 hours have been accumulated in close contact with the crocodile populations of Bahía Portete and Caño Lagarto. Data collected so far suggests that the population shows stability but lacks apparent increase.

Hunt (1990), Messel and Vorlicek (1987) and Webb and Smith (1987) have all reported aggressive behavior of adults towards juveniles and sub-adults which are forced to leave the place where they were born. Adult Crocodylus acutus in Bahía Portete occupy the breeding grounds within the network of channels and lagoons deep inside the mangroves, whereas the smaller size classes are displaced to the margins of the mangroves and more open areas, where they are frequently caught in fishermen's nets. In addition to this, since 1993 smaller size crocodiles living near Puerto Portete have been forced to move further west (Gregoria Fonseca per. com.) as they are being hunted by local fishermen, all of whom carry a harpoon in their boats.

## THE HUMAN FACTOR

The construction of the sea port for the exportation of coal meant the arrival of a great number of local Wayuu Indians hoping they would be hired by the coal company. This eventually created a very poor suburb when they decided to stay despite the harsh



local conditions and a lack of work opportunities. These people have since turned to the sea as their only source of food, creating an over-exploitation of marine resources and imposing a serious threat upon the crocodile population.

In 1992, there was little interest among local people for crocodile hunting or egg consuming. However, a few hides were taken and sold in the market.

Although Wayuu Indians are not particularly fond of crocodile meat, they are eager to eat adult crocodile meat as a source of protein if other food is scarce. They find hatchling and juvenile flesh disgusting.

Since 1992, an increasing number of juveniles have been drowned when tangled in fishermen's nets. These crocodiles are never skinned but their carcasses are found minus both the skull and the tail. The meat from the tail is eaten whereas the skull is used for witch-craft. To avoid being misused in witch-craft, Indians will bury all crocodile skulls making it extremely difficult to collect skulls for scientific purposes. There is a strong belief that the strength and power of the crocodile can be obtained if the skull is ground, toasted and eaten. If buried close to the water-hole of an enemy, it will eventually render the water-hole useless. Crocodile skulls are also thought to heal wounds if their ashes are rubbed against the injured tissues. Crocodile fat is used against intestinal worms and is also considered to stimulate sexual arousal in men.

On the other hand it is of increasing concern that at present the channel leading to the breeding grounds has been well trimmed with machetes by the Indians to make access to the breeding stock very easy, whereas the channel used to be inaccessible and its navigation burdensome. Two adult crocodiles measuring 275 cms and 180 cms have already been taken away from the breeding site and slaughtered for their hides. Crocodile hides are sold for a nonsensical 25,000 pesos (US \$ 25). Within a population with only six nesting females, killing one of these means cutting reproductive potential by 17%. The total number of nests in Bahía Portete for 1996 has not yet been completed and remains unknown.

## NOTES ON REPRODUCTION

Crocodylus acutus in Bahía Portete will start nesting in late April or early May. Females will build four or five nests prior to egg laying. Clutch size will range from 19 to 41 eggs. No sand beaches in Bahía Portete are suitable for crocodile nesting, therefore females will nest in rocky beaches under desert vegetation. The roots of cactus and shrubs will keep the rocks and pebbles tightly packed and prevent the nest chamber from collapsing. The added benefit of building a nest under tight bushes is that the eggs are protected from over-heating. Females will not lay their eggs in the same spot every year but they will definitely nest in the same area and stick to their territory year round.

A couple of weeks prior to egg laying, females will come out to the breeding grounds and completely cover the beach with crocodile tracks and fake nests.

Not only is human predation of nests common in Bahía Portete, which accounts for 30% average egg loss, but so is predation by wild foxes and raccoons, Procyon lotor and Procyon cancrivorus. To cut down on such egg losses a strong smelling disinfectant solution is poured over the nests to confuse the sense of smell of the predators.

## CROCODILE BEHAVIOR

It has often been said that larger, older crocodiles are much more wary than younger animals. Robert Bustard demonstrated the crocodiles' ability to learn and avoid being recaptured (Bustard, 1968). In Bahía Portete a 3 foot long crocodile was swept away from the mangroves by the sea currents and ended up by the sea port. This animal cannot be approached whereas adult crocodiles at the breeding site have gotten used to human presence, will respond to the sound of a whistle and have been hand fed.

## CONSERVATION

A conservation strategy for Bahía Portete needs to give the live crocodile an economic value greater than a dead crocodile. It is basically two men and their families

who are well acquainted with crocodile hunting and they can easily be converted from poachers to conservationists if they are paid more for preserving the crocodile population rather than killing the animals. If any of these Indians sell a crocodile hide for US \$25, but spend US \$5 in salt and transportation, in addition to his time and effort, his net profit is US \$20 for each animal. If he were paid this US \$20 for ensuring the yearly survival of each adult crocodile under his care, he would not only save himself a lot of trouble, but he would assure a yearly income on the basis of preserving natural resources. A continued monitoring of the crocodile population is then mandatory and so is protection of vital nesting areas, not only during the nesting season but year-round since these are home for the brood stock which will remain in their territories.

#### ACKNOWLEDGMENTS

This work was supported by a contract with INTERCOR-CARBOCOL (TEC.IA.024.92) in 1992 and a grant from Fondo FEN Colombia and the McArthur Foundation in 1996. I am also indebted to Dr. Felipe Guhl and Dr. Henry Von Prah (†) from Universidad de los Andes for their earlier support to the project back in 1981.

I wish to thank Pilar Vásquez, Jorge Velásquez and Ricardo Melendro from INTERCOR as well as Angel Guarnizo and Eduardo Guerrero from Fondo FEN. Miguel Paz and Carlos Díaz have both assisted me in the field.

I wish to take this opportunity as well to thank Dr. Wayne King and Dr. James Perran Ross from the University of Florida, Hernando Zambrano from the Ministerio del Medio Ambiente and Arturo Liévano from Azoocol for their concern and support and finally, I also wish to thank Mr. Alastair Turton for his personal cooperation and tolerance in the course of my latest work.

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**Preliminary Assessment of the Status of the American Crocodile  
(Crocodylus acutus) in the Coastal Zone of Belize.**

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**ABSTRACT**

The status of the American crocodile (Crocodylus acutus) in Belize remains poorly known. Commercial over-exploitation decimated populations prior to protection in 1981, and recent survey data are lacking. Spotlight surveys were conducted in 1994 and 1995, of Turneffe Atoll, Maps Cay, and northern Ambergris Cay. A total of 187.8 km were surveyed and 86 crocodiles observed (0.45 crocodiles/km). These results indicate significant crocodile populations are present at all three localities. Evidence of nesting was found at three sites in the Turneffe Atoll and on a spoilbank near Maps Cay. Raccoons (Procyon lotor) are a major nest predator. Suitable nesting beaches are limited, and while some illegal killing is occurring, the greatest threat to the continued viability of crocodile populations appears to be development of these beaches. The distribution and status of the American crocodile on the mainland, where it occurs sympatrically with Morelet's crocodile (C. moreletii) remain largely unknown. As part of a comprehensive coastal zone management plan, a survey of the entire coastal zone is planned for 1996-97 to determine present population status and identify critical habitat.

**INTRODUCTION**

The American crocodile (Crocodylus acutus) is one of two species of crocodiles which occur in Belize. The American crocodile is a medium to large crocodile found in coastal habitats and offshore islands, while Morelet's crocodile (C. moreletii) is a somewhat smaller species inhabiting inland freshwater wetlands. Both species are currently listed as endangered under the United States Endangered Species Act and are on Appendix I of CITES (Convention on Trade in Endangered Species of Flora and Fauna) treaty (Thorbjarnarson, 1992). Surveys of the American crocodile in Belize have been accorded high priority by the IUCN/SSC Crocodile Specialist Group (Thorbjarnarson, 1992).

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Commercial crocodile hunting began in Belize during the late 1930's and 1940's, as the importance of forest products in the local economy declined (Hope and Abercrombie, 1986). Subsequent over-harvesting led to depletion of both C. acutus and C. moreletii populations (Charnock-Wilson, 1970). Crocodile skins were sold to buyers in villages and larger towns, and after progressing through a chain of middlemen, exported to Europe for processing (Abercrombie et al., 1982; Hope and Abercrombie, 1986). Unfortunately quantitative survey data from this period are lacking, and the practice of categorizing both spotted cat and crocodile skins as simply "hides and skins" in government trade statistics makes it difficult to determine past levels of exploitation (Frost, 1974; Abercrombie et al., 1982; Hope and Abercrombie, 1986). Furthermore, a considerable number of crocodiles were shot by sport hunters and members of the British garrison, and an unknown number of skins were exported illegally (Charnock-Wilson, 1970).

By the 1960's both C. acutus and C. moreletii were nearly extirpated from Belize (Charnock-Wilson, 1970). Neill and Allen (1961) reported the collection of a single juvenile C. acutus from a mangrove swamp in Belize City and considered the species rare. Powell (1971) concluded American crocodiles were present on some offshore cays (islands), but extinct on the mainland, and King et al. (1982) noted some island populations had also been eliminated. C. acutus was afforded legal protection under the Wildlife Protection Act of 1981 (Marin, 1981).

The current status of the American crocodile remains poorly known and survey data are lacking. The largest remaining population in Belize may occur in the Turneffe Atoll (Thorbjarnarson, 1992). C. acutus is also present on Cay Caulker (Meerman, 1993), and in mainland coastal lagoons of Shipstern Nature Reserve (Meerman, 1992; Ouboter, 1992). Additional reports by fishermen and others indicate American crocodiles occur widely on offshore islands throughout the coastal zone of Belize. Distributional records are summarized in Table 1. Perkins (1983) estimated a country-wide population as high as 10,000 to 20,000 animals, but these numbers must be considered somewhat speculative. The objective of this project was to obtain preliminary data on the status of the American crocodile, and provide conservation recommendations based on these findings.

## STUDY AREA

The coastal zone of Belize includes three atolls, over 200 offshore islands, and extensive mainland swamps. The Belize barrier reef extends the entire length of the 325 km coastline from Ambergris Cay in the north to the Gulf of Honduras in the south, and is separated from the mainland by a narrow stretch of shallow water known as the inner channel. Both mainland and offshore coastal habitats are dominated by red mangrove (*Rhizophora mangle*), with black mangrove (*Avicennia germinans*) swamps occurring in some hypersaline areas. Elevated beach ridges are dominated by beach thicket and cay forest associations (Minty et al., 1995). Crocodile surveys were conducted at three localities in the coastal zone: Turneffe Atoll, the Bacalar Chico region of northern Ambergris Cay, and Maps Cay (Figure 1).

1. Turneffe Atoll - This atoll, located approximately 30 km southeast of Belize City, is the largest of three coral atolls found on the barrier reef of Belize, and one of only four atolls in the western hemisphere. Turneffe Atoll is approximately 50 km long and 16 km wide with an estimated surface area of 533 km<sup>2</sup>. The atoll consists of a chain of islands partially enclosing three shallow lagoons: Southern Lagoon, Central Lagoon, and Northern or Vincent's Lagoon. A near continuous beach ridge extends along the windward side of the atoll (Figure 2). Maximum elevation is about 1.5 m above sea level (Perkins, 1983).

With the exception of three tourist resorts most of the atoll remains undeveloped. Turneffe Island Lodge is located at the southern end of the atoll on Cay Bokel, and Blackbird Cay Resort and Turneffe Flats Lodge are located at the southern and northern ends, respectively, of Blackbird Cay. Coral Cay Conservation/University College of Belize have established a research center on Calabash Cay. Small fishing camps are scattered throughout the atoll. The atoll is under increasing pressure for development and the construction of several additional tourist facilities has been proposed. A draft development plan was recently compiled to address conflicting land-use issues (Turneffe Island Development Committee, 1995).

2. Bacalar Chico National Park (proposed) - The Bacalar Chico region of northern Ambergris Cay has recently been proposed as a National Park. This area encompasses significant terrestrial, mangrove, and marine habitats (Dotherow, 1995). Elevated ridges surround a system of open water lagoons and red mangrove swamps.



Some residential development has occurred on beach ridges along the windward side of the cay (Figure 3).

3. Maps Cay - A cluster of low-lying, offshore mangrove cays within the inner channel, approximately 7 km east of Belize City. An elevated beach ridge extends along the windward shore of the cay where a small tourist lodge and several fishing camps are located. A network of deep lagoons are found in the interior of the cay, and a spoilbank extends ca. 50 m along a dredged channel on the leeward side of the cay.

## METHODS

Spotlight surveys were used as an index of crocodile densities (Bayliss, 1987). This technique is used in crocodile surveys worldwide and other methods of population estimation have confirmed its validity. Spotlight surveys were conducted from a motorboat propelled slowly along the shoreline, using a handheld Q-beam spotlight (250,000 candlepower) and auxiliary 12-volt headlights. Areas not accessible to motorboats were surveyed by canoe. Crocodiles were located by noting eyeshine reflections in lightbeams and classified according to their estimated total length (TL) as juveniles (TL < 90 cm), subadults (TL = 90 - 150 cm), or adults (TL > 150 cm). Crocodiles that could not be approached close enough to estimate total length were classified as "eyeshine only". Distance travelled in each survey was calculated with a cartometer using maps and aerial photographs provided by Coral Cay Conservation. Crocodile densities were calculated as the number of crocodiles observed per kilometer of survey route, allowing quantitative comparison with other survey data. Maps of survey routes were deposited with the Coastal Zone Management Unit, Department of Fisheries, Belize City, Belize.

Daylight reconnaissance was conducted along some proposed routes to determine feasibility, locate possible hazards, and search for nesting habitat. Potential nesting habitat was identified by noting the presence of indicator plant species such as paurotis palms (*Paurotis wrightii*) and coconut palms (*Cocos nucifera*). These plants are restricted to well-drained beach ridges, and these habitats were searched for the presence of nest mounds, excavations, eggshells and fragments, and unhatched eggs. Eggshells and unhatched eggs were counted to estimate clutch size. In areas where nests were found, physical characteristics of the site were noted. Coordinates of nesting beaches in Turneffe

Atoll were obtained with a Sony Pixis IPS-360 Global Positioning System (GPS). Fishermen were interviewed when encountered and questioned about the presence of crocodiles and possible nesting areas.

## RESULTS AND DISCUSSION

### Population Surveys

#### Turneffe Atoll

Fieldwork was conducted from 22 May to 2 June, 26 to 30 June, and 1 October to 6 October 1994, and 15 to 22 June 1995. Due to the difficulties of maneuvering boats over the reef after nightfall, most areas surveyed were on the leeward side of islands, and in sheltered creeks and lagoons. Most crocodiles were observed in shallow water along red mangrove shorelines sheltered from prevailing easterly winds. Survey results are presented in Table 2. In general, populations of low to moderate density were found in Turneffe Atoll. A total of 67 crocodiles were observed and 132.2 km were surveyed. A mean of 0.50 crocodiles/km of survey route was calculated from all surveys. However, relatively high densities of crocodiles were found at the southern and northern ends of Calabash Cay (3.0 and 2.0 crocodiles/km respectively), and along the windward shore of Blackbird Cay (2.8 crocodiles/km). Moderate densities of crocodiles were noted in Soldier Bight-Blackbird Cay (0.71 crocodiles/km), Northern Lagoon (0.70 crocodiles/km), and along the leeward side of the atoll from Northern Lagoon to Crikoozeen Creek (0.83 crocodiles/km). Low densities (0.08 - 0.46 crocodiles/km) were found in most other areas.

Crocodiles observed at the southern end of Calabash Cay were in a network of small mangrove creeks and lagoons, and in the vicinity of a fishing camp. These animals may have been attracted by fish remains discarded from the camp. An unusual congregation of subadult crocodiles was noted along an exposed beach on the windward shore of Blackbird Cay. Crocodiles usually prefer sheltered waters free from wave action (Thorbjarnarson, 1989) and the presence of these animals was surprising. The gradually sloping gradient of this beach probably offers good foraging habitat, and the barrier reef affords protection from significant wave action. Thorbjarnarson (1989) stated intermediate size classes (TL = 1.0 - 2.0 m) may be more frequently found in these

marginal habitats. Most crocodiles observed along the leeward side of the atoll were foraging in turtlegrass (*Thalassia testudinum*) which forms dense beds in shallow water and supports large schools of fish and aquatic invertebrates.

The size-class distribution of crocodiles recorded in the Turneffe Atoll is presented in Table 4. Juveniles (5.9%) are probably under-represented as they probably avoid open water and remain among flooded mangroves where they are less visible during spotlight counts. Subadults (41.7%) were the size class most frequently observed and were encountered in most areas surveyed. Adults (23.8%) and "eyeshine only" (28.3%) comprised the remainder. The largest concentration of adult crocodiles was noted in the vicinity of Blackbird and Northern Cays and may reflect the proximity of nesting habitat. Crocodiles tend to become increasingly wary with age and many of the eyeshines recorded likely represent adult crocodiles which submerged before they could be approached close enough for size-class identification. The percentage of adult crocodiles observed in the Turneffe Atoll is comparable to population data from the Everglades, Florida, USA (24.5%) and Turkey Point, Florida, USA (25.0%), but higher than Etang Saumatre, Haiti (15.7%). However, size-class limits vary somewhat making comparisons difficult (Thorbjarnarson, 1989).

#### Bacalar Chico

Fieldwork was conducted from 27 June to 1 July 1995. Survey results are presented in Table 3. A total of eight crocodiles were observed and 42.7 km surveyed. A mean of 0.18 crocodiles/km of survey route was calculated from all surveys. However, all crocodiles were encountered in mangrove swamps, creeks, and lagoons, and none were observed along the windward or leeward shoreline of Ambergris Cay. If these shorelines are excluded, the result is a mean of 0.39 crocodiles/km of survey route, comparable to low-density populations noted in some areas of Turneffe Atoll.

The size-class distribution of crocodiles recorded in Bacalar Chico is presented in Table 4. Juveniles comprised 50% of all sightings, subadults 25%, and adults and "eyeshine only" each 12.5%. Because of the small number of crocodiles observed, these results should be interpreted with caution.

### Maps Cay

A single spotlight survey was conducted on 21 July 1994. Eleven crocodiles were observed along 12.9 km of shoreline (0.85 crocodiles/km). The size-class distribution is presented in Table 4. All crocodiles were observed along an exposed shoreline on the windward side of the cay, and because of shallow inshore waters, most could not be approached closely enough to determine size.

### Coastal Mainland

Determination of the status and distribution of the American crocodile in mainland coastal habitats is complicated by the presence of C. moreletii. These two species are morphologically very similar, identification is difficult in the field, and often requires capturing crocodiles (Platt, 1996). Ouboter (1992) and Meerman (1992) found both species present in mainland lagoons in Shipstern Nature Reserve. The skull of an American crocodile that drowned in a fishing net in Southern Lagoon was examined in 1994. Drag marks and tracks of what was probably an American crocodile were also observed south of Southern Lagoon, leading from a brackish swamp, over the beach, and into the open ocean. Zisman (1989) listed American crocodiles as present in both Northern and Southern Lagoons, but the source of these records is unknown. Further surveys of mainland coastal lagoons are warranted.

Crocodiles with characteristics of both C. moreletii and C. acutus have been reported from coastal regions of Belize (Schmidt, 1924; Abercrombie et al., 1980), and an animal we collected in Kates Lagoon exhibited a C. acutus-like dorsal scale pattern. These animals typically exhibit a reduced number of dorsal scales in each transverse row, and reduced or absent caudal irregularities. Ross and Mayer (1983) suggest hybridization may occur between the two species, and Ross and Ross (1974) found C. moreletii traits in C. acutus only where the two species are sympatric. The situation regarding hybridization remains unresolved and must await application of molecular genetic techniques.

### Nesting

#### Turneffe Atoll

Three nesting areas were located during this survey. The first site is located on the leeward side of Blackbird Cay 1 km south of Turneffe Flats Resort (N 17° 25' 33.1", W

87° 49' 24.0") on an elevated sandy ridge approximately 20 m long. The vegetation corresponds to the broken palmetto thicket association which occurs on drained peat and organic sand (Minty et al., 1995). In 1994 two nests were located. One nest was a large mound of sand (ca. 30 cm high) and the other a hole nest. In 1995 a hole nest that appeared to have been opened by raccoons (*Procyon lotor*) and three eggshells were found. Additional nesting habitat was found in this vicinity, and Greg Smith (pers. comm.) reported finding a nest closer to Turneffe Flats Resort several years ago. However, an intensive search of this area failed to locate further evidence of recent nesting activity.

The second and most extensive nesting site is located on Northern Cay ca. 8.3 km north of Turneffe Flats Resort (17° 29' 35.3" N, 87° 47' 06.9"W). This site (corresponding to Site 17/26 of Minty et al., 1995) is an elevated beach ridge on the windward side of the cay, composed of well-drained organic sand, and dominated by beach thicket and coconut palms. A hypersaline black mangrove lagoon of about 1.0 ha is located behind the beach. A search in 1994 revealed five recent, and three to four pre-1994 nests along a 240 m section of beach. Both mound and hole nests were found. All recent nests appeared to have been successful (producing viable hatchlings) and opened by females. When inspected on 29 June 1994, eggs of two nests appeared to have hatched within the previous two or three days. Hatchlings (20+) were observed in the adjacent lagoon. Return visits to this site were made on 15 and 20 June 1995, but no evidence of nesting activity was noted despite an intensive search and excavation of suspected nest sites. Two crocodiles were observed in the black mangrove lagoon, one of which was a large adult (TL ca. 300 cm).

A third nesting area was located in 1995 on a peninsula on the eastern side of Deadmans Cay (17° 12' 57.4 " N, 87° 51' 49.4" W). The nest was constructed on a beach ridge with a protected cove and red mangrove swamp immediately to the rear. The nest was discovered by the property owner who reported finding 13 eggs buried in a shallow hole. All but four of the eggs were subsequently destroyed by people. The average measurements of the remaining eggs were as follows: length = 74.3 mm (SE = 2.4; n = 4), width = 45.3 mm (SE = 0.9; n = 4) and mass = 87.1 g (SE = 8.1; n = 3). This site may have been an important nesting area in the past. During our first visit (1994) the property owner described finding a clutch of hatching crocodile eggs while clearing his homesite. Since our initial visit, the peninsula has been cleared and a number of tourist cabins

constructed. This is the only nesting beach that has been located in the southern region of Turneffe Atoll, and it is doubtful that any crocodiles will utilize this site in the future.

#### Bacalar Chico

Little evidence of nesting was found within the proposed reserve, although this survey was conducted at a time when hatching occurs and nests should have been readily obvious. An excavated hole nest was found on a small (ca. 0.10 ha) island in Santa Cruz Lagoon. The hole measured 40 cm wide x 15 cm deep and was constructed in a substrate of sand and crushed shells. The island is somewhat elevated and supported a sparse growth of hammock hardwoods. Several other islands in Santa Cruz Lagoon appeared to be suitable nesting habitat, but a search revealed no nests.

A ridge, which appears to offer suitable nesting habitat, and extends for about 1.5 km along the western shore of Laguna Cantena was intensively searched, but no sign of nesting activity was found. The ridge is elevated 1.0 m above normal water level, composed of a coarse, well-drained mixture of sand and soil, and borders shallow mangrove lagoons on the leeward side, which appear to be excellent nursery habitat for hatchling crocodiles (Lutz and Dunbar-Cooper, 1984; Thorbjarnarson, 1989). Future visits should be made to this ridge to determine if crocodile nests are present. It is also probable that nesting occurs in areas outside of the proposed reserve. Based on an examination of aerial photographs, suitable nesting habitat is found on Ambergris Cay south of the reserve in Laguna de Cayo Frances and Laguna de San Pedro. Hatchling C. acutus are known to disperse widely when suitable nursery habitat is not available near nests (Ogden, 1978; Mazzotti, 1983). Protection of nesting habitat outside of the reserve may be essential if a viable population is to be maintained.

#### Maps Cay

A recently hatched nest was found on the leeward side of the cay during a spotlight survey on 21 July 1994. The nest was a hole constructed about 1.0 m above water atop a spoilbank adjacent to a red mangrove swamp, in a mixture of organic peat, shell and coral. Spoilbanks are important nesting habitat in Florida (Gaby et al., 1985). Five eggshells were found in the hole.

Based on eggshells and unhatched eggs recovered at nest sites, an average clutch size of 9.5 (SD = 3.0, n = 7) was estimated. While this is most certainly an underestimate of true clutch size, it is considerably smaller than clutch sizes of 22 to 80 previously reported for the American crocodile in other areas of its range (Thorbjarnarson, 1989), possibly indicating small female size or energetic stress. Given the 80 to 90 day incubation period reported for the American crocodile (Thorbjarnarson, 1989), nesting in Belize probably follows the general Caribbean pattern with clutch deposition in March and April.

Raccoons are responsible for significant nest losses in other regions (Mazzotti, 1983; see review in Thorbjarnarson, 1989) and are probably a major predator of *C. acutus* nests in Belize. Raccoons are abundant in mangrove habitats throughout Belize and were sighted during spotlight surveys of all three study areas. Nest losses attributable to raccoons were recorded on Blackbird Cay. Raccoons were found on isolated islands in Santa Cruz Lagoon suggesting even nests in these habitats are accessible to predators. Coatis (*Nasua narica*) are also present in many coastal habitats and represent potential nest predators.

#### Food Habits

Two juvenile crocodiles (TL = 90.8 and 65.0 cm) were captured and stomach-pumped using the method of Taylor et al. (1978). Stomach contents from both animals contained insect and crab (*Uca* spp.) remains. Fresh fish was recovered from the smaller animal, and balls of mammalian hair, probably from rodents or a small opossum, were recovered from the larger crocodile. Unidentified nematodes (David Soucek, Dept. Biological Sciences, Clemson University, pers. comm.) were found among stomach contents of the smaller crocodile. Similar prey items have been recovered from juvenile *C. acutus* elsewhere (Alvarez del Toro, 1974).

#### Crocodile-Human Interactions and Conservation

Interviews of local fishermen and resort operators suggest that major crocodile-human conflicts occur when crocodiles frequent the vicinity of resorts and take dogs at fishing camps. Crocodiles are attracted to camps where fish are cleaned and offal discarded in the water. Turneffe Flats Resort has discontinued this practice because of the presence of several large crocodiles and alternative disposal techniques seem to have

eliminated the problem. An open garbage dump on "Garbage Cay" near Cay Bokel has also been reported to attract crocodiles. Closure of this dump is recommended to reduce the possibility of crocodile-human conflicts and to address public health concerns. Several fishermen stated that crocodiles occasionally enter fish traps in the shallow waters off Ambergris Cay and consume the catch. Predation on free-ranging domestic dogs was frequently reported by residents in the cays. Keeping dogs penned or leashed, especially at night, would probably prevent predation. Several resort operators have also expressed concern over possible dangers that large crocodiles pose to swimmers and divers. While attacks on humans have been reported, American crocodiles are not normally regarded as man-eaters, and risks to swimmers are considered minimal (Pooley et al., 1989). Some illegal killing of nuisance crocodiles was reported by local fisherman, and Coral Cay Conservation Volunteers observed several crocodile skins drying at a fisherman's camp near Calabash Cay (Gail Bradley Miller, University College of Belize, pers. comm.). However, the extent of this harvest is believed to be small.

The major threat to the continued survival of American crocodile populations in the coastal zone of Belize appears to be the development of potential nesting beaches. Nesting habitat was found to be of limited extent and many of the potential sites we visited were either developed or showed signs of impending development. In the Turneffe Atoll, both Blackbird Cay Resort and Turneffe Flats Resort are constructed on suitable nesting beaches. The nesting area on Deadmans Cay is now occupied by a tourist lodge and unavailable for future nesting. The communal nesting site on Northern Cay was marked by "Keep Off" signs and much of the understory vegetation had been cleared. Greg Smith (pers. comm.) reported nesting in the vicinity of Rendezvous Point, but a search of the area revealed fishing camps on all suitable beaches. A recent draft of the Turneffe Islands Development Plan (Turneffe Islands Development Committee, 1995) included the nesting beach on Northern Cay in a list of areas suitable for development. Any forthcoming management plans must include provisions for the protection of nesting beaches. Protection of nesting habitat, whether or not located within existing reserves, is essential for the continued survival of the American crocodile in Belize. Nursery habitat where brackish water is available may also be critical for the continued survival of the American crocodile, especially on offshore cays. Hatchling and juvenile American crocodiles cannot tolerate high salinity levels for long periods and osmoregulate



by drinking freshwater (Ellis, 1981). Young crocodiles on offshore cays probably obtain freshwater from rainwater lenses (Mazzotti, 1983). Osmoregulatory failure is a significant cause of mortality among young crocodiles in some populations (Thorbjarnarson, 1989), and these brackish habitats are important for continued recruitment.

#### Future Research

A one year study of the status, distribution, and ecology of the American crocodile in Belize as part of the GEF/UNDP coastal zone management program is scheduled to begin in late spring of 1996. The specific priorities of this project are:

1. Train Belizean biologists in crocodilian research and management techniques.
2. Determine the present status of crocodile populations in coastal Belize.
3. Identify critical habitats.
4. Initiate a long-term (> 10 year) monitoring program.
5. Obtain basic ecological data on the ecology of crocodiles inhabiting barrier reef islands.
6. Resolve questions relating to the systematic status of the C. moreletii/C. acutus complex in coastal Belize.

Recommendations for a long-term crocodile management plan will be based on the results of this study. Primary consideration will be given to managing crocodile populations for non-consumptive purposes (e.g. wildlife viewing) as nature tourism is the country's largest industry, producing over \$Bz 100 million per year, and the ecotourism potential of the coastal zone is considered great.

#### ACKNOWLEDGMENTS

This project could not have been completed without the generous assistance and logistic support of University College of Belize, Coral Cay Conservation, International Tropical Conservation Foundation, United Nations Development Program/Coastal Zone Management Unit, Department's of Fisheries and Forestry, New York Zoological Society-Wildlife Conservation Society, and Clemson University. Dorian Barrow, Susan Wells, Peter Raines, Melanie Dotherow, Gail Bradley-Miller, Jonathan Ridley, and Andrew Gill provided support throughout the project. Field assistance was provided by Martin Feather, George Hansen, Marcus Marples, Domingo Ruiz, Paul Scott, Mike Somerville,

Auriol Samos, Jim Thomas, and numerous CCC volunteers. Greg Smith provided results of previous survey work. Richard Montanucci reviewed an earlier draft of this manuscript. The Belize Department of Fisheries and the Forestry Department issued the necessary permits. Boats and transportation were provided by the Belize Fisheries Department and Coral Cay Conservation.

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Table 1. Summary of distributional records for the American crocodile (*Crocodylus acutus*) in Belize. Sources in parentheses. Asterisk indicates nesting has been reported.

<u>Coastal Mainland</u>	<u>Offshore Islands</u>
Belize City (1)	Ambergris Cay (8, 12) *
Four-Mile Lagoon (2)	Cay Caulker (9) *
Mango Creek (3)	Cay Chapel (5, 8)
Northern Lagoon (4)	Hicks Cay (5)
Placencia Lagoon (3)	Lighthouse Reef (10)*
Punta Ycacos Lagoon (5)	Maps Cay (12) *
Chipstern Nature Reserve (6, 7)	Middle Long Cay (8)
Southern Lagoon (4, 12)	Moho Cay (8)
	South Long Cay (11)
	Tobacco Range (5)
	Turneffe Atoll (4, 8, 12) *

Sources: 1-Neill and Allen, 1961; 2-Bruce Cullerton, personal comment; 3-Mahler and Wotkyns, 1991; 4-Zisman, 1989; 5-Perkins, 1983; 6-Meerman, 1992; 7-Ouboter, 1992; 8-Greg Smith in Dotherow, 1995 and personal comment; 9-Meerman, 1993; 10-Colin Howell, personal comment; 11-Susan Wells, personal comment; 12- Platt and Thorbjarnarson, this study.

Table 2. Results of American crocodile (*Crocodylus acutus*) spotlight counts during 1994 and 1995 in the Turneffe Atoll, Belize.

<u>Location</u>	<u>Date</u>	<u>Distance(km)</u>	<u>#Observed</u>	<u>#/km</u>
1994				
Blackbird Cay (Soldier Bight)	30 May	4.2	3	0.71
Northern Cay (Unnamed Lagoon)	2 June	1.3	1	0.76
Blackbird Cay (leeward)	2 June	16.0	3	0.18
Calabash Cay (leeward)	26 June	10.8	5	0.46
Shag Cay (leeward)	27 June	12.6	1	0.08
Calabash Cay (South-creek/lagoon)	28 June	2.0	6	3.00
Deadmans Cay (leeward)	29 June	12.0	4	0.33
Blackbird Cay (windward)	30 June	4.2	12	2.85
Northern Lagoon	2 October	14.1	10	0.70
Northern Cay (N. Lagoon Entrance to Dog Flea Cay)	2 October	8.4	3	0.35
Crikozeen Creek-Lagoon	5 October	9.0	2	0.22
1995				
Turneffe Atoll (leeward, Cay Bokel to Creek mouth 17°14.8"N, 87°56.3"W)	18 June	11.4	5	0.43
Calabash Cay (windward)	18 June	1.0	2	2.00
Turneffe Atoll (leeward, N. Lagoon Entrance to Crikozeen Creek)	19 June	10.8	9	0.83
Northern Cay (windward)	20 June	14.4	1	0.06
Total		132.2	67	

Table 3. Results of American crocodile (*Crocodylus acutus*) spotlight counts during 1995 in the proposed Bacalar Chico National Park, Ambergris Cay, Belize.

<u>Location</u>	<u>Date</u>	<u>Distance(km)</u>	<u># Observed</u>	<u>#/km</u>
Santa Cruz Lagoon to Bacalar Chico Creek	27 June	15.0	0	0.0
Santa Cruz Lagoon	28 June	4.3	3	0.69
Bacalar Chico Creek	28 June	2.0	1	0.50
Laguna de Cantena	29 June	9.1	2	0.21
Unnamed Lagoon (see Figure 3)	30 June	2.7	1	0.37
Bacalar Chico Lagoon	30 June	2.1	1	0.47
Ambergris Cay (windward)	01 July	7.5	0	0.0
Total		42.7	8	

Figure 1. Map of Belize showing the location of three areas where American crocodile surveys were conducted in 1994 and 1995. Scale 1 cm = 25 km.

Figure 2. Map of Turneffe Atoll, Belize. Locality names in accordance with maps obtained from the Lands and Survey Department, Belmopan, Belize, or the Travellers Reference Map of Belize (International Travel Map Products, Vancouver, British Columbia, Canada, ISBN #09-21463-01-1). Scale 1:230,000.

Figure 1: Map of the proposed Bacalar Chico National Park, Ambergris Cay, Belize. Locality names in accordance with Ordnance Survey Map of Ambergris Cay, Belize (Ordnance Surveys Directorate, Southampton, S09 4DH, England). Scale ca. 1:50,000.



Table 4. Size-class distribution of American crocodiles (*Crocodylus acutus*) observed in spotlight surveys of the coastal zone of Belize. Crocodiles were classified based on estimated total length (TL) as juveniles (TL < 90 cm), subadults (TL = 90-150 cm), or adults (TL > 150 cm). Crocodiles that could not be placed in a size-class are classified as "eyeshine only" (EO). Total number of crocodiles in each size-class followed by frequency in parentheses.

<u>Location</u>	<u>Juveniles</u>	<u>Subadults</u>	<u>Adults</u>	<u>EO</u>	<u>Total</u>
Turneffe Atoll	4 (5.9)	28 (41.7)	16 (23.8)	19 (28.3)	67
Bacalar Chico	4 (50.0)	2 (25.0)	1 (12.5)	1 (12.5)	8
Maps Cay	3 (27.2)	0 (0.0)	1 (9.0)	7 (63.6)	11
<b>Total</b>	<b>11 (12.7)</b>	<b>30 (34.8)</b>	<b>18 (20.9)</b>	<b>27 (31.3)</b>	<b>86</b>

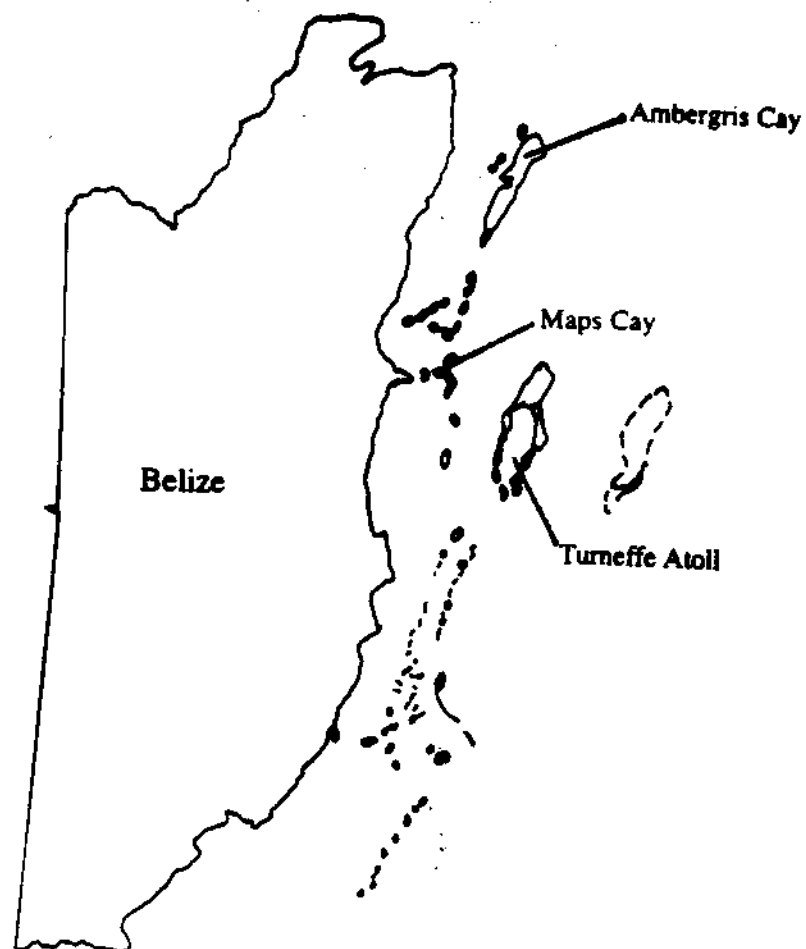


Figure 1. Map of Belize showing location of three areas where American crocodile surveys were conducted in 1994 and 1995. Scale 1 cm = 25 km.

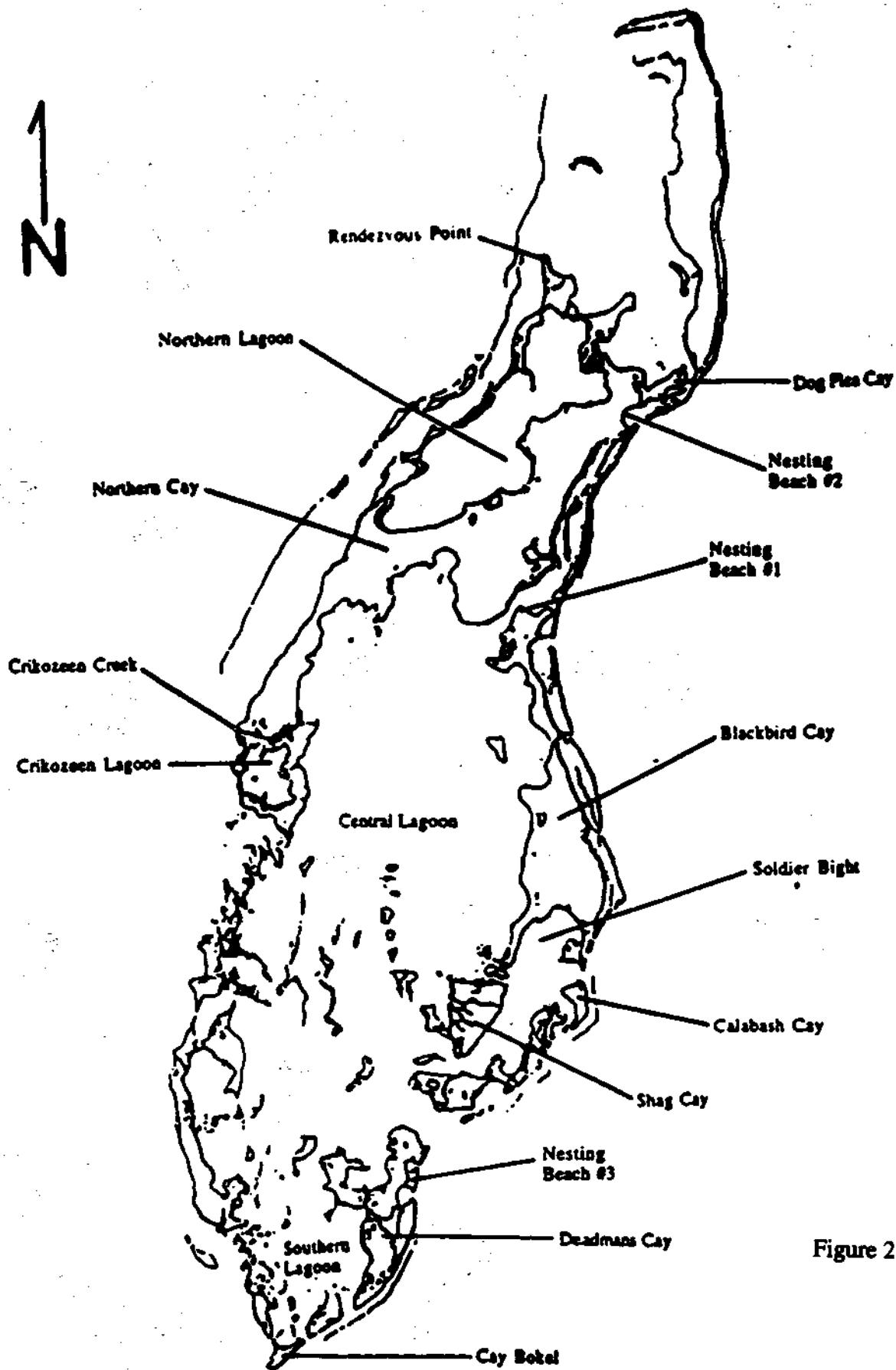


Figure 2

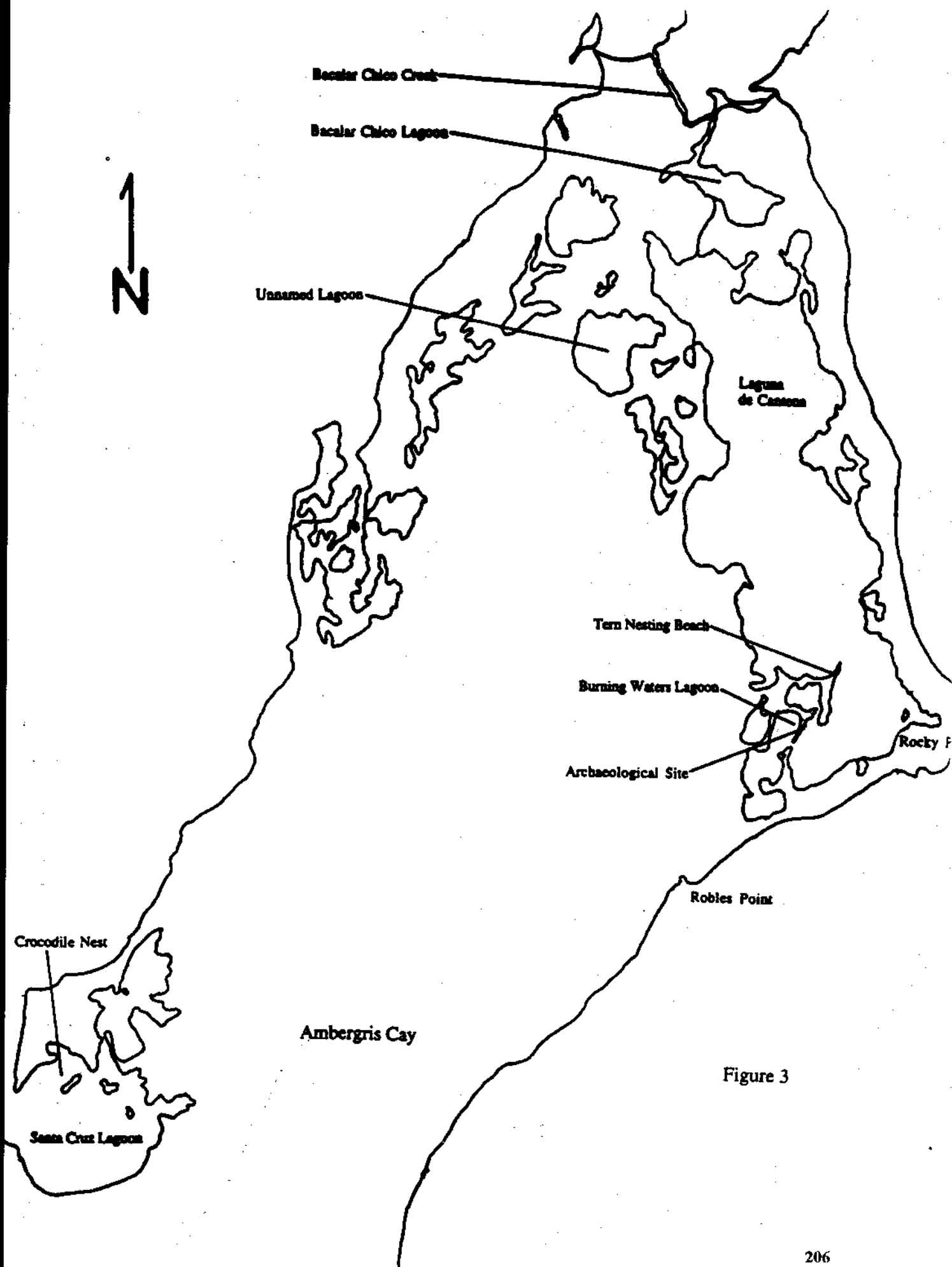


Figure 3

## Evaluation of the reintroduction of *Crocodylus intermedius* in the Caño Guaritico Wildlife Refuge (Apure State, Venezuela).

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### Summary

In the period from 1990 to 1995, were reintroduced a total of 514 Orinoco crocodiles (*Crocodylus intermedius*) in the Caño Guaritico Wildlife Refuge, in a combined effort of different national and international organizations. The crocodiles were reintroduced in three rivers: Caño Guaritico, within the boundaries of the Refuge and where 283 crocodiles (55.1%) have been released; Caño Macanillal, where the reintroduction program started and 227 crocodiles (44.2%) have been released; and Caño Mucuritas, where only 4 crocodiles have been released (0.7%). The last two localities are located inside the El Frío Ranch. Out of a total of 514 crocodiles released, 271 (42.2%) were produced in the Masaguaral Breeding Center; 114 (22.2%) came from the Estación Biológica El Frío Breeding Center; 92 (17.9%) came from the UNELLEZ Breeding Center and 41 (7.9%) came from the Puerto Miranda Breeding Center. Most of the crocodiles reintroduced were produced under a *farming* or closed cycle scheme (61.7%), while the rest (38.3%) were produced under a *ranching* or open cycle scheme. It is known the origin of most of these crocodiles, whose parents are from different geographic localities, in some cases quite distant. Out of the 514 crocodiles, there is data on length and weight for only 411 individuals (80.2%). In 1990 the average total length was 151.7 cm (SD = 22.25), in 1991 was 125.2 cm (SD = 30.9), in 1992 was 82.8 (SD = 35.1), in 1993 was 79.7 cm (SD = 13.17) and in 1994 was 86.9 cm (SD = 16.08). The decrease of the average total length is due to the fact that in the last three years were released animals of 1 year of age. The sex ratio is almost 1:2, with 264 males (64.07%), 144 female (34.95%) and 4 undetermined (0.97%). The total length and weight data were used to calculate a condition index, which shows that in general animals with TL greater than 100 cm (26%) and weights greater than 2.5 kg (46%) have indices that imply good physical condition. It is proposed the use of such indirect indices as an additional criteria to decide when the captive raised crocodiles should be released into the wild. The follow up study of the reintroduced crocodiles has not been constant, but an sporadic and partial effort, whose results show a very low mean abundance of 1.8 ind/km in the Caño Macanillal and 0.47 ind/km in the Caño Guaritico, that contrasts with a very high concentration of spectacled cayman (*Caiman crocodilus crocodilus*). A total of 16 animals have been recaptured, all of them close to the release sites, although one animal migrated a considerable distance in a year. Finally, it was determined that the mean growth rate of crocodiles recaptured in Caño Macanillal (0.5 mm/day) was lower than the growth rate of the crocodiles from Caño Guaritico (1.38 mm/day). However, further studies are necessary to confirm these results and determine the viability of this program

## Evaluación de la reintroducción de *Crocodylus intermedius* en el Refugio de Fauna Silvestre Caño Guaritico, Venezuela.

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### Resumen

En el periodo entre 1990 y 1995, se reportó la reintroducción de un total de 514 Caimanes del Orinoco (*Crocodylus intermedius*) en el Refugio de Fauna Silvestre Caño Guaritico, dentro de un esfuerzo combinado de diferentes organizaciones nacionales y extranjeras. Las reintroducciones se ejecutaron en tres cuerpos de agua: Caño Guaritico, propiamente dentro de los linderos del Refugio y donde se liberaron en dos sectores 283 caimanes (55,1%); Caño Macanillal, lugar donde se realizaron las primeras liberaciones, con total de 227 individuos (44,2%); y Caño Mucuritas, donde solo se han liberado 4 caimanes (0,7%). Estas dos últimas localidades se sitúan en tierras del Hato El Frío. De los 514 caimanes, 271 (42,2%) provinieron del Zoocriadero Masaguaral, 114 (22,2%) del Zoocriadero El Frío, 92 (17,9%) del Zoocriadero de la UNELLEZ y 41 (7,9%) del Zoocriadero Puerto Miranda. Los caimanes liberados son en su mayoría son producto de la cría en cautiverio (61,7%), mientras que el restante proviene de la recolección de juveniles producidos en vida silvestre (38,3%). Se conoce el origen de la mayoría de los caimanes, los cuales provienen de reproductores de diferentes localidades, en algunos casos bastante distantes. De los 514 animales, se tienen solo medidas y datos de 411 individuos (80,2 %). En términos de longitud total en 1990 el promedio fue de 151,7 cm (DE= 22,25), en 1991 de 125,2 cm (DE= 30,9), en 1992 de 82,8 (DE= 35,1), 1993 de 79,7 cm (DE= 13,17) y en 1994 de 86,9 cm (DE= 16,08). La disminución de la longitud total se debe a que en los últimos tres años se liberaron ejemplares de 1 año. La proporción de sexos es casi 1:2, con 264 machos (64,07%), 144 hembras (34,95%) y 4 sin determinar (0,97%). Los datos de longitud total y peso se usaron para calcular un índice de condición, con lo cual se encontró que principalmente caimanes con medidas de LT mayores de 100 cm (26 %) y pesos mayores de 2,5 kg (46 %) tienen índices que denotan buena condición física. En este sentido, se propone el uso de tales índices indirectos como criterio adicional para establecer cuando los caimanes criados están en condición de ser liberados al medio natural. El seguimiento de caimanes liberados no ha sido permanente, ejecutándose solo estudios parciales y esporádicos, cuyos resultados arrojan índices promedios de abundancia muy bajos, de 1.8 ind/km en el Caño Macanillal y 0.47 ind/km en Guaritico, que contrasta con una alta concentración de babas (*Caiman crocodilus crocodilus*). Se han practicado 16 recapturas cerca de los sitios de liberación, aunque hay un caso de un ejemplar que emigró una distancia considerable durante un año. Finalmente, se encontró que el crecimiento promedio de caimanes capturados en Macanillal fue menor (0,5 mm/día); al crecimiento de caimanes colectados en el Caño Guaritico (1,38 mm/día). Sin embargo, se requieren mas estudios para corroborar estos resultados y establecer la viabilidad de este programa.

## INTRODUCTION

*Crocodylus intermedius* is an endemic species of the Orinoco river basin, specially of the flooded savannas of the "Llanos" of Venezuela and Colombia. The commercial harvest of the Orinoco crocodile during the 1930s and 1940s, along with the habitat destruction, keep this species on the verge of extinction. The crocodile has virtually disappeared in Colombia (Lugo and Clavijo, 1991), while in Venezuela there are small isolated populations (Godshalk, 1978, 1982; Ramo y Busto, 1986; Franz *et al.*, 1985; Ayarzagüena, 1987; Thorbjarnarson and Hernández, 1992; Seijas, 1993, 1994). These relicts are located mainly in areas where the human activity is minimal, and in some cases under official protection.

In 1984 it was included by the IUCN in the list of the twelve animal species more threatened with extinction in the world. The conservation of the Orinoco crocodile have been a collaborative effort by many national and international, both non-governmental and government organizations, among which are: the Fundación para la Defensa de la Naturaleza (FUDENA), the Sociedad de Ciencias Naturales La Salle, the Universidad Nacional Experimental de los Llanos Ezequiel Zamora (UNELLEZ), the Ministerio del Ambiente y los Recursos Naturales Renovables (MARNR), the Servicio Autónomo de Fauna (PROFAUNA), the Instituto Nacional de Parques (INPARQUES), the Masaguaral, Puerto Miranda and El Frío ranches, the Agencia Española de Cooperación Internacional (AECI), the Wildlife Conservation Society (WCS), the World Wildlife Fund (WWF-USA), World Wide Fund for Nature (WWF), the National Geographic Society, and the Smithsonian Institution.

The aim of this paper is to present the results of the evaluation of the reintroduction of Orinoco crocodiles in the Caño Guaritico Wildlife Refuge, using FUDENA's database as the main source of information. This evaluation is part of the Action Plan for the Survival of this species, proposed by the Venezuelan Crocodile Specialist Group (GECV) between 1993 and 1994 (Arteaga, 1993; Seijas y Chávez, 1994).

## CAÑO GUARITICO WILDLIFE REFUGE

This Refuge was created the 11 of January of 1989, for the official protection of endangered species of the "Llanos" ecosystem, specially the Orinoco crocodile. This is the reason it was chosen as the main area for reintroduction of captive bred crocodiles. The Caño Guaritico is a tributary of the Apure river and is located between the towns of Samán de Apure and Bruzual in the Apure State, and it has a total area of 9300 has (Fig. No. 1).

## RESULTS

### Numbers and Localities

Between 1990 and 1995 were reintroduced approximately 514 captive bred crocodiles, all of them from the Masaguaral, UNELLEZ, El Frío and Puerto

Miranda Breeding Centers, as shown in Table 1. The number of crocodiles reintroduced increased every year between 1990 and 1993, mainly by the contributions of the Masaguaral Breeding Center, see Fig. 2. Between 1994 and 1995 the number decreased, and in 1995 were released only 80 animals from El Frío Breeding Center.

The firsts reintroductions were in two small tributaries: caño Macanillal and caño Mucuritas, being the first where most of the animals have been released, specifically at the Tapa de la Ramera site. As seen in Table 1, in 1992 crocodiles were reintroduced directly into the caño Guaritico, in the site of Las Ventanas in El Frío ranch, and at the site of Tres Ceibas in the Turagua ranch. To summarised, out of a total of 514 crocodiles reintroduced, 283 have been released in caño Guaritico (55.1%), 227 in caño Macanillal (44.2%) and 4 in caño Mucuritas (0.7%), see Fig. 3.

### Number per Breeding Center and Origin

Of the 514 crocodiles reintroduced, 267 (51.9%) came from the Masaguaral Breeding Center, 114 (22.2%) from El Frío, 92 (17.9%) from UNELLEZ and 41 (7.9%) from Puerto Miranda, as seen in Fig. 4. Only for 491 crocodiles there is certainty about their origins, being 303 animals (61.7%) captive bred, and 188 animals (38.3%) collected in 4 rivers and one reservoir in the Llanos. The specific origin of the 491 animals is: 238 (48.5%) Masaguaral, 131 (26.7%) Cojedes river, 60 (12.2%) UNELLEZ, 38 (7.7%) Capanaparo river, 8 (1.6%) unknown, 6 (1.2%) Camatagua reservoir, 4 (0.8%) Portuguesa river, 3 (0.6%) FONAIAP-Puerto Ayacucho and 1 (0.2%) Orinoco river, see Fig. 5.

### Sizes

Using data from only 411 (80.2%) reintroduced crocodiles, a comparison of their total length (TL) was done, which shows a decrease from 1990 to 1992, leveling till 1994, as seen in Fig. 6. In 1990 the mean TL was 151.7 cm (SD=22.25 cm), in 1991 was 125.2 cm (SD=30.9 cm), in 1992 was 82.8 cm (SD=35.1 cm), in 1993 was 79.7cm (SD=13.17 cm) and in 1994 was 86.9 cm (SD=16.08 cm). The decrease in TL is due to the fact in 1990 and 1991 animals of up to three years of age were released, and in the last three years only animals of one year of age have been released into this area.

### Sex Ratios

The sex ratio is close to 1:2, with 264 males (64.07%), 144 females (34.95%) and 4 undetermined (0.97%). In Fig. 7 are presented the sex ratios of animals released per breeding center, showing for Masaguaral (1:3), El Frío (1:4) and Puerto Miranda (1:4), indicating that these centers have produced mainly males, while on the other hand, UNELLEZ have produced mainly females, with a sex ratio of 3:1.



## Condition Index

Seijas (1993), proposed the calculation of a condition index using the total length and weight data for Orinoco crocodiles greater than 50 cm, assuming that below this size and for about six months after they hatch, the animals still have reserves, therefore not reflecting their adaptation to the new environment. According to this author the mathematical expression of this index is the following:  $CI = a \cdot W \cdot TL^{-b}$ , where  $a$  and  $b$  are the coefficients calculated in the lineal regression of  $\ln W$  on  $\ln TL$ .

According to the indicated by Seijas (1993) the CI is a measure of the relative "fatness" of the animals, so animals with a  $CI < 1$  would be relatively "skinny", and animals with a  $CI > 1$  would be relatively "fat".

Using the TL and W data for 411 crocodiles released between 1990 and 1994, a regression analysis of  $\ln W$  on  $\ln TL$  was done, resulting in the following equation:

$$\ln W = -5.376950 + 2.911783 \ln TL$$

$$(r^2 = 0.84608, p < 0.00001, 404 \text{ d.f.})$$

For the calculation of the CI of each crocodile the following equation was used:

$CI = 216.351 W TL^{-2.911783}$ . In the Graps. 8 and 9 it can be observed the tendencies of CI with respect to TL and W of the group of reintroduced crocodiles. The values of  $CI > 1$  correspond to animals in a good condition and mostly greater than 100 cm in TL and 2500 g in W.

## Night Surveys in Reintroduction Areas

A few sporadic night surveys have been conducted, most of them previous to the release of animals each year. In Table 2 are summarized the surveys completed mainly in two sectors of the caños Macanillal and Guaritico, indicating the relative water level, the number of surveyed kilometers, the number of crocodiles observed and an abundance index, presented in number of crocodiles per kilometer.

In 1991 approximately 7.8 km of the caño Macanillal were surveyed, counting 14 crocodiles and capturing 4. The number of crocodiles observed represented 25% of the total reintroduced crocodiles (62) to that date, in that caño. The survey also included the mouth of the caño Guaritico, where two adult animals were found.

Approximately 20 km of the caño Guaritico were surveyed in 1992, locating only 3 juvenile crocodiles. The low water level of that month made the study very difficult, due to the fact that there was a very high concentration of spectacled caymans in the caño. By the end of 1993 a series of surveys were conducted in the caño Guaritico, upstream and downstream of Las Ventanas site. Close to 17 km were surveyed and 8 crocodiles were observed and captured, most of them had been released in the Turagua ranch, about 50 km upstream, in June of the same year. It is important to note, besides the distance traveled by the crocodiles, that these animals grew at a higher rate than that reported for crocodiles in other areas (see table of recaptures).

Finally, in December 1994, 22 km upstream from Las Ventanas were surveyed and only one crocodile was observed amongst a high concentration of spectacled caymans, most of those between 120-180 cm and >180 cm of TL. Also were surveyed La Ramera lagoon and caño Macanillal, that were connected because of the high water level. An estimated 25 km were surveyed, observing only 3 adult crocodiles.

## Recaptures

As it can be observed in the recaptures Table, 16 animals have been caught in 5 years, by different researchers. There are evidences that some crocodiles stay in the same river where they were released: the recapture in august 1991 in caño Macanillal of a crocodile released in that river in 1990; an animal recaptured in the Tres Ceibas site of caño Guaritico in 1994, this was released in 1993 in the same area; and the 8 animals previously mentioned that moved about 50 km within six months of their release in the caño Guaritico. Although in one instance, a crocodile recaptured in December 1992 at Las Ventanas site of the caño Guaritico had been released in 1991 in caño Macanillal, might indicate the possibility that some animals would move from one river to another.

The mean growth rate of crocodiles recaptured in caño Macanillal was 0.05 cm/day, much lower than that registered for crocodiles recaptured in caño Guaritico, which was 0.138cm/day. It is necessary to conduct further studies to verify these differences, considering a much bigger sample size.

## Discussion

In order to accelerate the recovery of crocodile populations in the wild in Venezuela, in recent years have been promoted and carried out the captive breeding and release of these animals into the wild. Restocking has taken place in localities where still exist the species although in low numbers, and reintroduction has been conducted where the populations have been extirpated.

Seijas et al (1990) argue that this strategy has been tested in several parts of the world, and the ecological basis for such activity is that during the early life stages of crocodilians (eggs & hatchlings), a very high mortality occurs due to predators and other environmental factors. The collection of eggs or hatchlings in the wild, for their artificial incubation and raising, eliminates this high mortality and improves their survival probabilities. When these animals are about 1 year old and with a mean TL of 80-90 cm, are released back into the wild.

The effect of the release size of Orinoco crocodiles on their success in adapting to the conditions in the wild has not been determined. However, the results of the population surveys seem to indicate that larger animals stay close to the release sites, while juveniles and subadults tend to disperse. Thorbjarnarson (1989) suggests that the dispersion phase of juveniles and subadults, might be an integral part of the population dynamic of the American crocodile (*Crocodylus acutus*) as well as other species of crocodilians.

The authors found the intermediate size classes of this species were found frequently in marginal habitats. Mazzoti (1983) and Gaby *et al* (1981, 1985) indicate that intermediate size crocodiles tend to be found in inaccessible areas, isolated from the areas where hatchlings and adults are found, probably an effect of territoriality from adult or dominant adults. Such effect is even more significant if it is also considered the possible pressures from adult spectacled caymans, found in high densities in the release areas.

At first, the animals were released in caño Macanillal and caño Mucuritas with the idea that these would migrate to the caño Guaritico, which is the Refuge properly, while the management plan was written and facilities were built for the regulation of fishing and hunting activities. So far, none of these expectations have been fulfilled. Under these conditions, since 1992 the animals were released directly into caño Macanillal, because it was thought that the animals released in caño Macanillal would go to the Apure river in times of maximum floods, therefore outside the protected area of the Refuge.

Apparently, the caño Guaritico offers better conditions for the crocodiles, if it is considered that the growth rates are higher than those reported for the caño Macanillal. However, it is very important to conduct a more detailed study in order to confirm or deny this idea, because there are other factors involved, for instance, in caño Guaritico there is a very high density of spectacled caymans, which are certainly playing an important role, specially in regard to competition and predation. All this is important to fine tune the reintroduction program, which in the particular case of caño Guaritico seems to be the release of animals of the same size of the spectacled caymans found there, which is between 120-180 cm and > 180 cm of TL. This would mean that the animals to be release there should be more than one year old, which would increase the costs, both for operation and building of new facilities in the breeding centers.

The crocodile reintroduction program in the Refuge was initially planned *a priori* for the release of 300 animals in a 5 year period, with the aim to establish a viable population with a stock of 10% of that total, that is 30 animals. Until now, 514 animals have been released which exceeds the established goal, all this indicates the necessity of implementing detailed follow-up studies that will answer the present questions and doubts.

Finally, it can be said that one of the major problems this reintroduction program faces today is the lack of information regarding: survival and growth rates, adaptability to the wild, movements and reproduction, among others. Although the program is 6 years old, there are no specific projects aimed to establish the present situation of the species in the area. In 1992, Eddie Escalona from the Venezuelan Wildlife Service (PROFAUNA) presented a proposal to the consideration of the GECV, but the funds could not be obtained. This year, one of the authors, Gustavo Hernández, will begin a follow-up project jointly funded by FUDENA and WCS, and it is hoped that this project will receive the collaboration of national and international organizations interested in the conservation of the highly endangered Orinoco crocodile.

## ACKNOWLEDGEMENTS

The authors would like to express their gratitude to the following persons and institutions, that made this study possible: Glenda Medina and Diego Díaz, former and present Executive Directors of FUDENA; Fundo Pecuario Masaguaral and Mr. Tomás Blohm; Dr. John Thorbjarnarson, María Muñoz, Manuel Felipe Rodríguez. Also, the collaboration of the following persons was much appreciated: Tibisay Escalona, Nayibe Pérez, Carlos Chávez, Jesús Rivas, César Molina, Tito Avila, Pedro Vernet y Humberto Rivero (Kako). The data from the breeding centers was kindly provided by: Andrés Eloy Seijas and Ildemaro González (UNELLEZ), Estación Biológica El Frío (El Frío breeding center), and Manuel Denis, Pedro Azuaje and Ernesto Boede (Puerto Miranda breeding center). Finally, the authors recognize and are very grateful to VARIG Airlines and WWF-USA for their support and funding, which made possible to present this paper.

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<i>Year</i>	<i>N° crocodiles</i>	<i>Breeding Center</i>	<i>Locality</i>
1990	30	Masaguaral (16) UNELLEZ (14)	Caño Macanillal (26) C. Mucuritas (4)
1991	53	Masaguaral (39) UNELLEZ (6) El Frio (8)	C. Macanillal (53)
1992	78	Masaguaral (55) UNELLEZ (6) El Frio (17)	C. Macanillal (59) C. Guaritico (19)
1993	155	Masaguaral (98) UNELLEZ (45) El Frio (9) Puerto Miranda (3)	C. Macanillal (9) C. Guaritico (146)
1994	118	Masaguaral (59) UNELLEZ (21) Puerto Miranda (38)	C. Guaritico (118)
1995	80	El Frio (80)	C. Macanillal (80)
<b>Totals</b>	514	Masaguaral = 267* UNELLEZ = 92 El Frio = 114 Puerto Miranda = 41	RFS Caño Guaritico = 5 C. Macanillal = 227 C. Guaritico = 283 C. Mucuritas = 4

**Table No. 1:** Description of the reintroduction program of Orinoco crocodiles, from the Masaguaral, UNELLEZ, El Frio and Puerto Miranda breeding centers, in the Caño Guaritico Wildlife Refuge (Caños Macanillal, Mucuritas y Guaritico) between 1990 and 1995. Source: Database GECV-FUDENA.

<i>Date</i>	<i>Locality</i>	<i>Water level</i>	<i>Kilometers</i>	<i>Nº crocodiles</i>	<i>Croc/km</i>
August 91	Macanillal <sup>1</sup>	medium	7,8	14	1,80
May 92	Guaritico <sup>2</sup>	low	20,0	3	0,15
Dec. 93	Guaritico <sup>2</sup>	high	17,0	8	0,47
Dec. 94	Guaritico <sup>2</sup>	high	22,0	1	0,05
Dec. 94	Macanillal <sup>3</sup>	high	15,0	3	0,20

**Table 2:** Night Surveys summary of two sectors of the Caños Macanillal and Guaritico, between 1991 and 1994. <sup>1</sup> Tapas La Ramera-El Jobo, <sup>2</sup> Las Ventanas, <sup>3</sup> Laguna La Ramera-Caño Macanillal. Source: Database GECV-FUDENA.

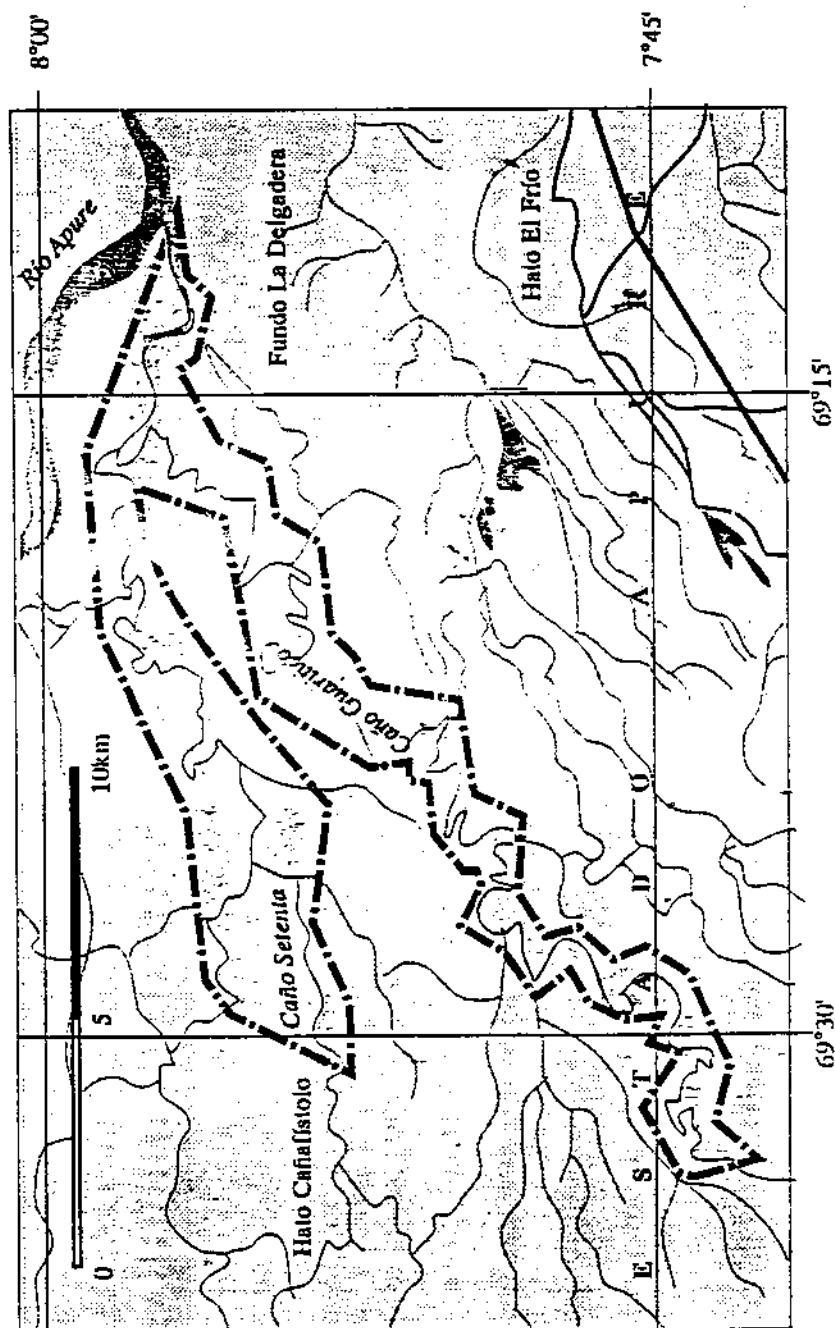


Fig. 1: Area of the Caño Guaritico Wildlife Refuge, where the Orinoco crocodiles have been reintroduced. Apure State, Venezuela. Source: MARNR/DGSP0A/ACM/01

Fig 2: Total number by Breeding Center of Orinoco crocodiles reintroduced in the Wildlife Refuge Caño Guaritico between 1990 and 1995.

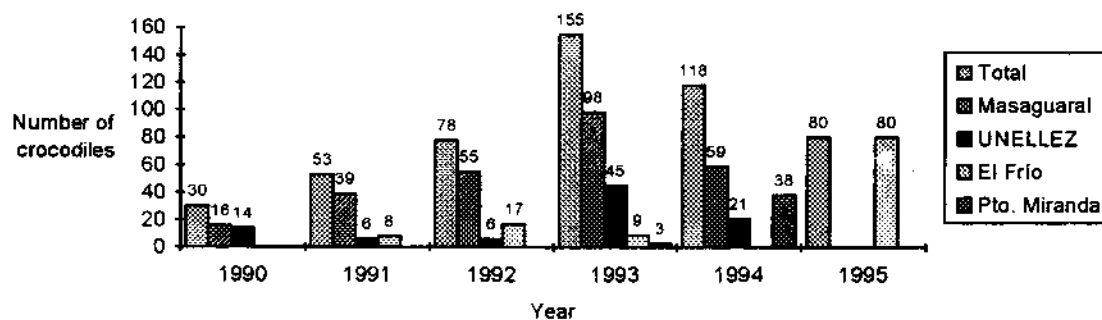


Fig 3: Orinoco crocodiles reintroduced by locality in the Wildlife Refuge Caño Guaritico between 1990 and 1995.

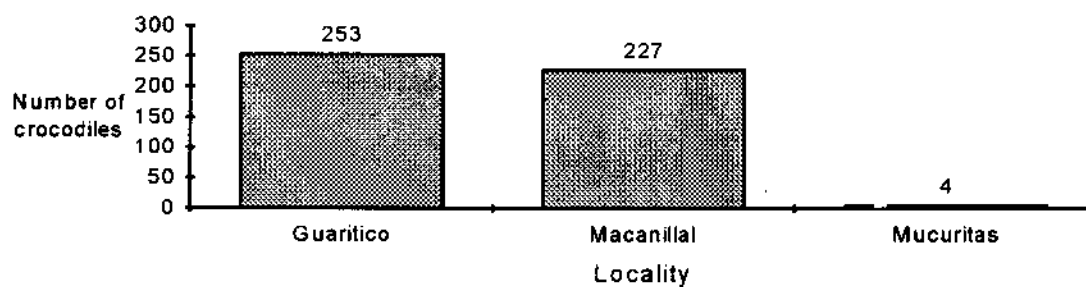


Fig 4: Number of Orinoco crocodiles in the Caño Guaritico Wildlife Refuge by breeding center between 1990 and 1995.

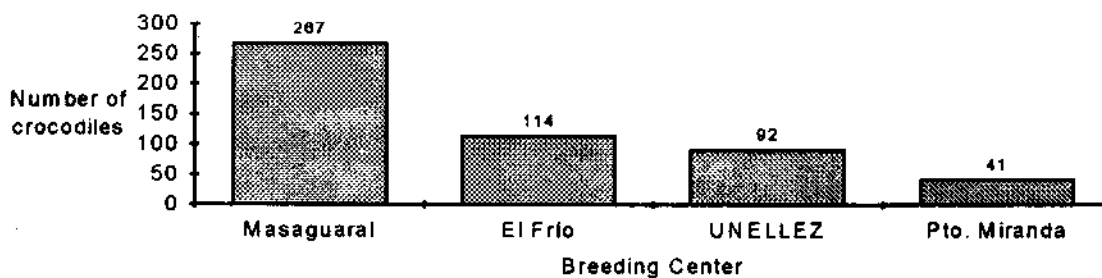




Fig. 5: Specific origins of Orinoco crocodiles reintroduced in the Caño Guaritico Wildlife Refuge between 1990 and 1995.

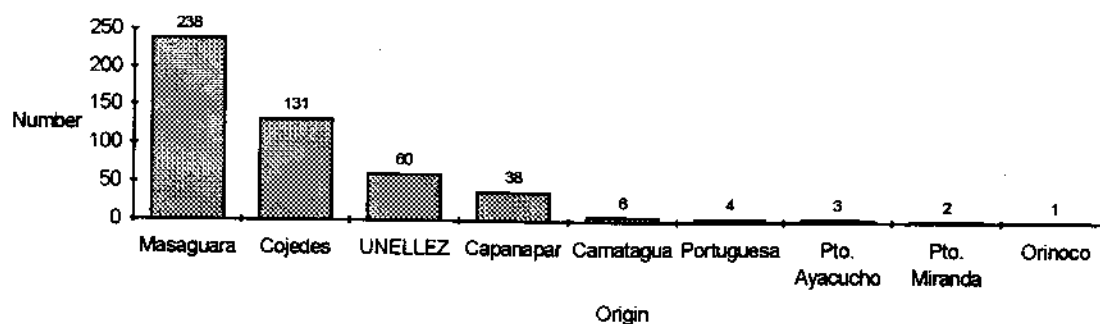


Fig. 6: Mean Total Length and Standard Deviation of Orinoco crocodiles reintroduced in the Caño Guaritico Wildlife Refuge between 1990 and 1994.

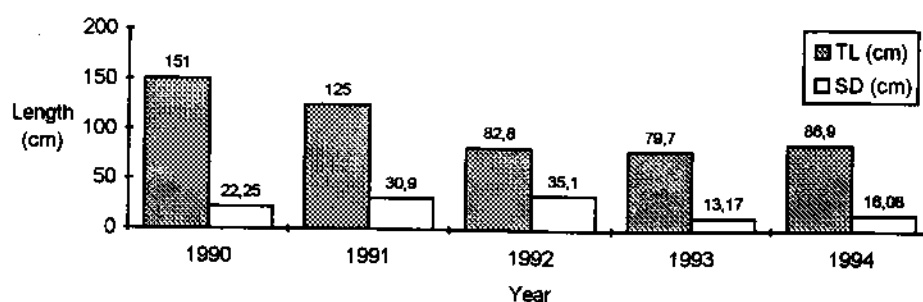


Fig. 7: Sex distribution of Orinoco crocodiles by Breeding Center reintroduced in the Caño Guaritico between 1990 and 1994.

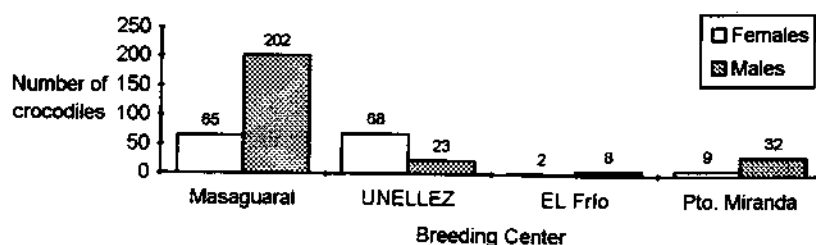
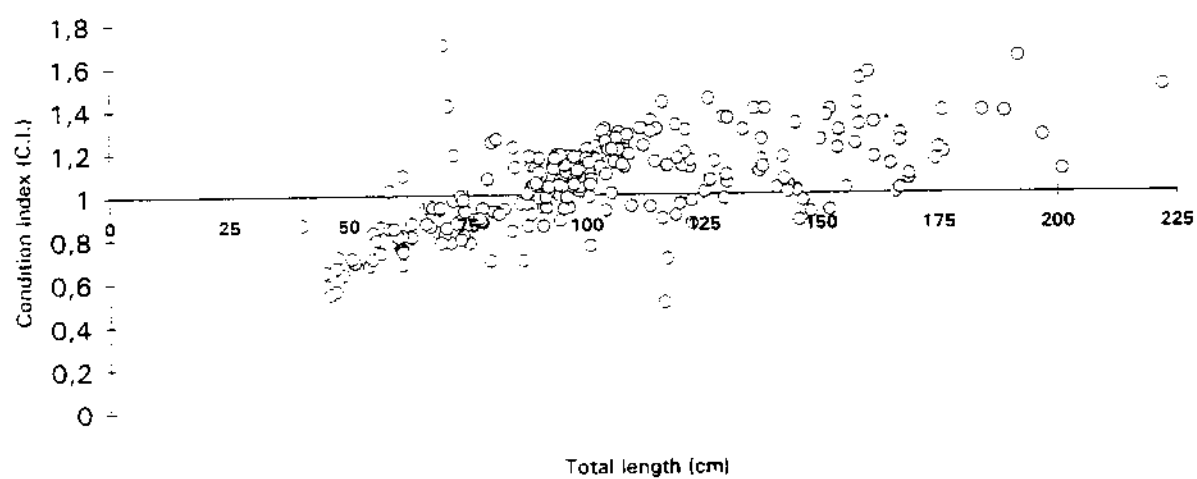


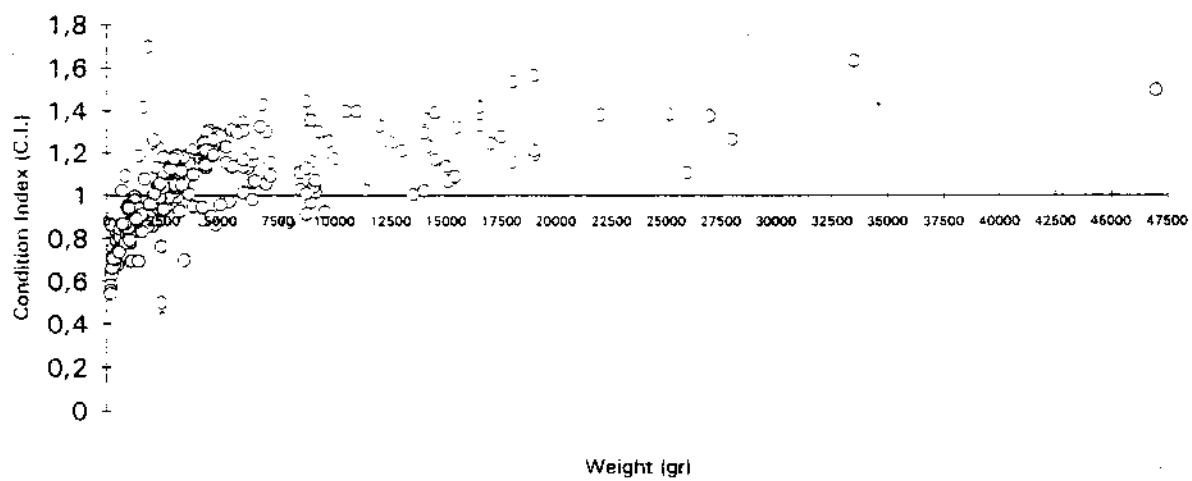
Fig. 8: Relationship between C.I. and TL of *C. intermedius* reintroduced in the Caño Guaritico Wildlife Refuge between 1990 and 1994.



# RECAPTURES OF ORINOCO CROCODILES IN CAÑO GUARITICO WILDLIFE REFUGE

DATE	AREA	RECAPTURE	INCREASE TL
2 AUG 1991	Caño Macanilla	2 Masaguaral 1991 1 UNELLEZ 1990 1 El Frio	0.04 cm/day 0.06 cm/day ND ND
21 DEC 1992	Caño Guaritico	1 Masaguaral 1991	0.05 cm/day
17 NOV 1993	Caño Guaritico	2 Masaguaral 1993	0.08 cm/day 0.15 cm/day
13 DEC 1993	Caño Guaritico	4 Masagaural 1993	0.041 cm/day 0.169 cm/day 0.142 cm/day 0.148 cm/day
14 DEC 1993	Caño Guaritico	2 Masaguaral 1993 1 Pto. Miranda 1993 1 El Frio	0.175 cm/day 0.169 cm/day 0.148 cm/day ND
3 JUN 1994	Caño Guaritico	1 Masaguaral 1993	0.134 cm/day
		16 Recaptures	
ND: no data Source: database FUDENA			

Fig. 9: Relationship between C.I. and Weight of *C. intermedius* reintroduced in the Caño Guaritico Wildlife Refuge



# Traditional methods used for hunting African dwarf crocodiles in the Congo

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## Introduction

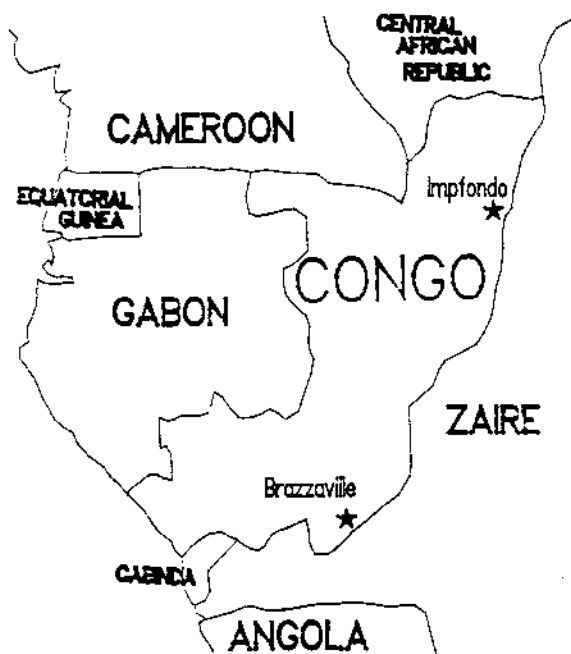
West African dwarf crocodiles *Osteolaemus tetraspis* inhabit water pools remaining in periodically flooded swamp forests (Waitkuwait, 1989). When threatened they may retreat into a burrow or hide amongst the stilt roots of mangrove trees at the edge of the pool.

Behra (1990) reported on the commercial exploitation of these crocodiles in the Congo for their meat. Rhodhain (1926) and Abercrombie (1978) described some traditional crocodile hunting methods, but the latter did not believe that the exploitation of *O. tetraspis* for food constituted an important danger to that species. Huchzermeyer & Agnagna (1994) examined wild-caught African dwarf crocodiles at markets in Brazzaville and reported on their pathology and parasites.

As the efficiency of hunting methods has an impact on the numbers of crocodiles which a hunter can collect in a given period, crocodile hunters were interviewed about the implements and strategies they employed, during a visit to the swamp forest area between Djéké and Mobenzélé, south of Impfondo during April/May 1995. This paper reports on the methods used by crocodile hunters in the visited area.

## Materials and Methods

In the villages of Dzéké and Impondi on the river Likouala-aux-Herbes and in Mobenzélé on the river Oubangui (Figures 1 & 2) villagers were asked to lead us to known crocodile hunters, who were then questioned about the different hunting methods they employed.



As the hunters first were suspicious of our motives, it was found that it was not possible to adhere to a prepared question protocol. Instead the questions were posed informally, leading from one aspect to another. However, once the suspicions were allayed, we were also allowed to photograph most of the implements said to be used in crocodile hunting.

## Results

Four crocodile hunters were found in Djéké, one in Impondi and two in Mobenzélé. In addition one of the guides accompanying the expedition demonstrated hunting methods in the swamp forest habitat of the dwarf crocodile.

The following implements and strate-

Fig 1 The Congo Republic

gies were found to be used:

The assegai is used to stab the crocodile when it is hidden in its burrow. This makes it unnecessary to subdue the animal after its capture. This method is used when the crocodile is destined for immediate home consumption.

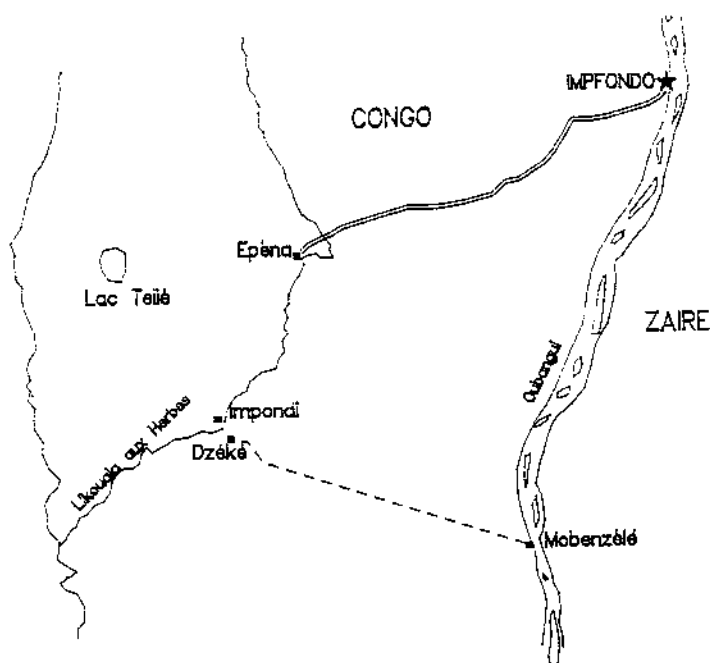


Fig 2 The research area south of Impfondo

The torch and multipronged fishing spear method is used in the rainy season when the floods connect the individual pools. At night the torch is used to pick out the reflection of the crocodile's eyes. While the crocodile is blinded, a second hunter slowly approaches the targeted animal and transfixes it with the fishing spear. The wounds caused by this weapon are not necessarily lethal.

**Calling:** By imitating the calls of adult crocodiles or of their hatchlings, the hunter induces the crocodile to come out of the water, where it is either caught with a fishing spear, with bare hands or most often with a forked stick. At night a torch may be used in addition.

**Bare hands:** During the day the crocodiles stay submerged in small open pools in the swamp forest with only the nostrils above the surface of the water. When the hunter approaches, the crocodile sinks to the bottom. The hunter notices the slight ripples caused by this movement and can make out the shape of the crocodile in the clear water, if it has not sought refuge in its burrow. The hunter then wades into the pool and grasps the crocodile's tail with his bare hands and swings it out of the water. He then places one foot on the crocodile's head, thus subduing it while he ties its snout and legs with lianas. As an alternative and more often a forked stick is used to fix the submerged crocodile at the bottom of the shallow pool.

The iron hook - gaff hook - is fastened to a long, rigid wooden shaft or to a strong but flexible length of liana and is used to pull the crocodile out of its burrow or hide, which usually is found under the roots of a tree at the edge of the pool. The flexibility of the liana allows the hunter to manoeuvre the hook even around bends in the burrow. This appeared to be a widely used implement and method.

A large iron fish hook with a dead toad or fish as bait and on a long line, fastened to a tree at the edge of the pool, is left in the water overnight. If the bait is taken and swallowed, the crocodile can be pulled out of the water the next morning.

Two long thorns are used to fashion a "hook" by tying them together at the base with the points directed away from each other. The thorns are baited and placed in the same way as the iron fish hook. This method can cause severe damage to the victim, as thorns were found imbedded in internal organs of several crocodiles with subsequent septicaemia by Huchzermeyer & Agnagna (1994). A variation of this method consisting of a single stick with two pointed ends has been described by Schmidt (1919).

A piece of cloth soaked in petrol is tied to a long stick and introduced into the (dry) burrow. The crocodile is driven out by the strong smell of the petrol and when rushing out, it is apprehended.

**Suffocation:** The entrance of the submerged burrow is blocked with soil, wood and other materials, preventing the crocodile from coming out to breathe. It suffocates (drowns) and can be pulled out a few hours later.

**Netting:** During the floods the hunter can enter the swamp in a dug-out canoe, then he calls the crocodile and when one approaches, throws a net over it.

After capture snout and legs of the crocodile are tied and the animals are carried back to the village, where they are kept, still tied, in a hut before being transported to the market or sold to a buyer on a passing river boat.

The number of crocodiles that can be caught by a single hunter on a hunting trip is limited by their population density as well as by the need to transport them back to the village, sometimes a day's march or even further through difficult terrain. Alternatively the secured crocodiles are left in the swamp forest while the hunter returns to the village for help. One of the hunters boasted that he could catch up to 30 crocodiles in one day.

#### Discussion

In the Congo African dwarf crocodiles have traditionally been consumed by the inhabitants of the villages bordering on the swamp forests. With ongoing urbanisation affluent town dwellers are prepared to pay a good price for a delicacy which they remember from the olden days. Crocodiles, like other reptiles, live for a long time without being cared for and thus can be transported even to distant markets by relatively slow river craft. Huchzermeyer & Agnagna (1994) estimated the average time lapse between capture and slaughter at a market in Brazzaville to be 30 days.

The dwarf crocodile has many bony scales, making its skin valueless (Thorbjarnarson, 1992) and it is eaten therefore with the skin. The absence of dwarf crocodile skins from the international markets led Abercrombie (1978) and Thorbjarnarson (1992) to the erroneous conclusion that this species was not exploited commercially to any serious degree.

Capture methods resulting in the death of the crocodile are used if the animal is hunted for home consumption. For the capture of live crocodiles the hunter has to develop specialized skills. This is why only few villagers become crocodile hunters.

#### Acknowledgements

This project is supported and the April-May 1995 expedition was funded by the Brazzaville Office of the Projet Nouabalé-Ndoki of the Wildlife

Conservation Society, for which we are very grateful. JR was supported by travel grants from the Carnegie Trust and the Percy Sladen Fund (administered by the Linnean Society).

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## Duodenal Morphology in African crocodiles

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Three species of crocodiles occur in Africa, the Nile crocodile *Crocodylus niloticus*, the slender-snouted crocodile *C. cataphractus* and the African dwarf crocodile *Osteolaemus tetraspis* and of the latter two subspecies *O. t. tetraspis* and *O. t. osborni*. The subspecies of the Nile crocodile are not subject of this paper.

The differences between the two subspecies of dwarf crocodiles affect the shape and length of the snout with the underlying skull bones as well as the nuchal and supracaudal scutes (Schmidt, 1919; Rodhain, 1926; Inger, 1948).

During the examination of 23 dwarf crocodiles in the Congo in 1993 and of a further 15 in 1995 as well as of a number of zoo specimens in South Africa, differences in the morphology of the duodenal loop of dwarf crocodiles emanating from different geographical regions were noted (Huchzermeyer, Penrith & Penrith, 1995).

One single juvenile specimen of *C. cataphractus* was also examined in Brazzaville in 1995, while large numbers of Nile crocodiles from South African crocodile farms were received as postmortem specimen over the last few years.

The examination of these specimens revealed that *C. cataphractus* has the longest double duodenal loop (Fig. 1). *C. niloticus* has a double duodenal loop which is shorter than that of the preceding species, but still elongate (Fig. 2). All Congolese specimens of *O. tetraspis* had a squat, almost square double duodenal loop (Fig. 3), while the West African specimens of *O. tetraspis* (from South African zoos) had a single duodenal loop (Fig. 4).



Fig. 1 Elongate double duodenal loop of *C. cataphractus*

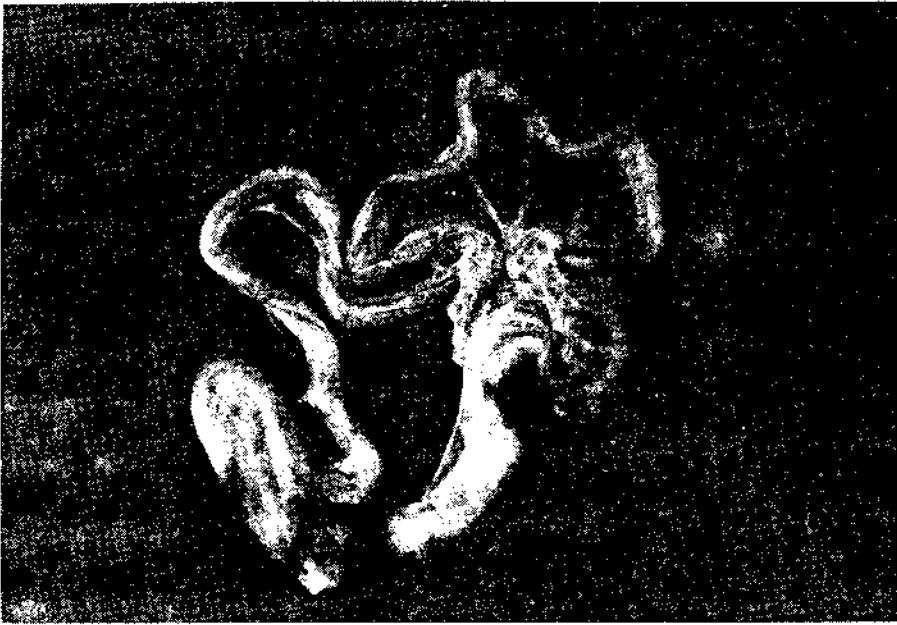


Fig. 2 Double duodenal loop of *C. niloticus*



Fig. 3 Double duodenal loop of Congolese *O. tetraspis*



Fig. 4 Single duodenal loop of a West African *O. tetraspis*

The lengthening of the duodenum is expected to increase the efficiency of duodenal digestion. It would be interesting to see whether this trait could be linked to differences in feeding habits.

It remains to be seen whether this newly discovered trait coincides with and further distinguishes between the two subspecies of *O. tetraspis*. A single duodenal loop would be regarded as the primitive form and its doubling as a specialization. If the Congolese specimens are taken as representing *O. t. osborni*, this would be in contrast with the interpretations by Inger (1948) who regarded the latter as the more primitive form.

It will be necessary to sample crocodiles at markets in other West African countries to determine the distribution of this trait in relation to the other distinctive features of the two subspecies.

Occasionally malformations of the duodenal loop have been seen in Nile crocodiles (Fig. 5) and because of the possibility that similar malformations could occur in other species as well, studies of duodenal morphology should be based on the examination of a sufficient number of specimens.



Fig. 5 Treble duodenal loop in a Nile crocodile

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Ecology of the Nile crocodile, *Crocodylus niloticus*,  
in lake St Lucia, Natal, South Africa.

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**Abstract**

This paper presents preliminary results of a project looking at the Role of the Nile crocodile, *Crocodylus niloticus*, in the Lake St Lucia ecosystem in Natal, South Africa. The project commenced in January 1994 and is progressing well. An update on the project is provided looking specifically at the feeding ecology and reproductive biology of the Nile crocodile in St Lucia.

**Introduction**

The project was designed to study the "Role of the Nile crocodile in the Lake St Lucia Ecosystem in Natal, South Africa". This broad study topic has been narrowed down to four primary objectives, as follows:

1. To investigate the diet and feeding habits of the various age/size classes of crocodiles throughout the St Lucia system
2. To determine the population status, distribution and abundance of crocodiles in the various size classes in the Lake system
3. To study various aspects of the reproductive biology of the Nile crocodile
4. To discuss conservation implications and to propose management recommendations for the Lake St Lucia ecosystem.

## Study Area:

Lake St Lucia forms part of the quarter of a million hectare Greater St Lucia Wetland Park in Natal, on the north eastern coast of South Africa. The lake body itself covers an area of 350km<sup>2</sup> and comprises approximately 80% of the estuarine area of Natal province (Begg, 1978). St Lucia has also been described as being the largest estuarine system in Africa. Although extremely large, the lakes average depth is less than one meter (Blaber, 1985). Due to its shallow nature the lake is subject to long-term salinity fluctuations and irregular inflow of fresh water (Blaber, 1980). In a normal wet year five rivers feed the lake system. Salinities vary from fresh water in the northern reaches of the lake to 35ppt at the estuary mouth and there is a net outflow of water. During periodic drought periods salinities in the northern reaches of the lake can increase to 120ppt. There is a net inflow of sea water which helps to reduce salinities. It is a dynamic and fascinating system housing approximately 1500 Nile crocodiles of various size classes. This project has focused on determining the requirements of Nile crocodiles in this system.

## Methods and Materials

### Objective #1:

To study the diet and feeding habits of the crocodile, the animals are captured in the wild and their stomachs are pumped using water, tygon tubing and a scoop. Trap sites are chosen according to crocodile distribution determined by aerial, boat and foot surveys. Baited noose traps are used to capture crocodiles and in addition more recently we began noosing juvenile and subadults from a boat at night. Once captured the animal is immobilized using a well known drug, namely flaxedil. The crocodile is moved to a safe working area and the animal is sexed and weighed, a cloacal body temperature is immediately recorded, numerous measurements are made, a blood sample is taken, the stomach pumped and it is finally tagged and released at the site of capture.

### Objective #2

To determine the population status, distribution and abundance of crocodiles, boat, foot and aerial surveys are undertaken on a regular basis. Aerial surveys were carried out on a bimonthly basis during 1994 and on a quarterly basis in 1995. Two observers and a scribe were present during each flight. Flights are carried out over a two day period, flying in the morning at approximately 200 feet and at a speed of 110km/hour.

### Objective #3:

To study certain aspects of the reproductive biology of the crocodiles in depth nesting surveys have been carried out during the past three breeding seasons - by air, by boat and on foot. Six primary nesting areas have been identified primarily on the Eastern shores of the lake. An annual survey of crocodile nests is conducted to obtain baseline data for estimating the size of, and year to year variation in the breeding population. Surveys are also conducted to monitor changes in habitat usage.

### Results and Discussion:

#### Objective #1:

To date we have caught 69 animals ranging in size from 1.4 to 4.3 meters. This figure includes 36 females, 22 males and 11 recaptures. This total also includes 3 juveniles (1.4 - 1.9m) and 2 subadults (2.2 - 2.6m).

Approximately 30% of the captured animals have had prey items of some sort in their stomach. To date prey items have included primarily fish scales and bones, fish roe, remains of ducks and other birds, and pieces of various crustaceans that occur in the system. A number of items of anthropogenic origin have also been found, such as plastic bags, fishing hooks and sinkers and even pieces of rope. Due to the stomach scooping technique which merely sub samples stomach contents, quantifying prey consumed has not been possible.

#### Objective #2:

Nine successful aerial surveys of the system have been conducted. The only disadvantage with aerial surveying is that crocodiles smaller than 1.5m are not usually seen. However, these surveys do provide good baseline data as to the number of adults in the system and to their seasonal distribution pattern.

During surveys over the past two years, the highest count was in July 1995, when 771 adult animals were counted.. Counts during the winter months were generally higher - not necessarily because there are more crocs in the system, but the animals are more concentrated around the available fresh water sources and more of them are usually basking in the early morning, which makes them more visible. During periodic boat surveys many of the tagged crocodiles have been seen. Some have been found 18km from the site of capture.

The distribution and abundance of juveniles and subadults in the system still needs to be determined. Animals in this size class are rarely seen. We have been radio tracking a 3.2m female for the

past 51 days in order to determine her home range, and we also plan to track a number of juveniles and subadults to determine their distribution and abundance in the system.

### **Objective #3:**

The six nesting areas we identified occur primarily on the eastern shores of the lake. This region receives twice the annual rainfall when compared to the western shores and it is inundated by fresh water seep lines originating from the nearby sand dunes. It is also characterized by flat, low lying sandy areas.

A total of 90 nests were counted in the 1993 season, 69 nests in 1994 and 127 in 1995. Nest counts for the 1993 and 1994 seasons were low, due to a three year drought we experienced. Constraints of both time and manpower limited previous survey efforts to areas where nesting was known to occur before. Nonetheless, some comparisons can be made between the past three years results and results of three earlier surveys carried out in the mid to late 1980's. Some minor changes in both the number and distribution of nests over the years is apparent in the system. Most notable is the southwards shift of nesting areas. This shift could have resulted from a human disturbances, varying salinities, availability of fresh water, food availability, alien plant invasion, or other factors. However, further research is required before any conclusions can be made. There is some natural recruitment into the system and we recently started a hatchling notching program catching them from a boat at night.

### **Conservation Implications:**

The most recent red data list categorizes the Nile crocodile as being at a "low risk", yet conservation dependent (Figure 1). The St Lucia Nile crocodile population is one of three major breeding nuclei of wild Nile crocodiles in South Africa, all of which are under some form of protection. Additional surveys have reflected isolated populations in most rivers and waters of the Eastern side of the country, although these crocodiles are not afforded any form of protection. Westward distribution in South Africa is restricted by climatic conditions as well as human pressure. Over the last hundred years these isolated populations have been sadly depleted by hunting, intolerance and destruction of habitat. Unless steps are taken to control poaching, which is carried out primarily for medicinal products, and to improve available habitat, crocodiles will only survive in the sanctuaries of South Africa. Unfortunately even these sanctuaries are now being threatened. During the 1995/1996 breeding season we lost an estimated twenty breeding females in one particular region of Lake St Lucia.



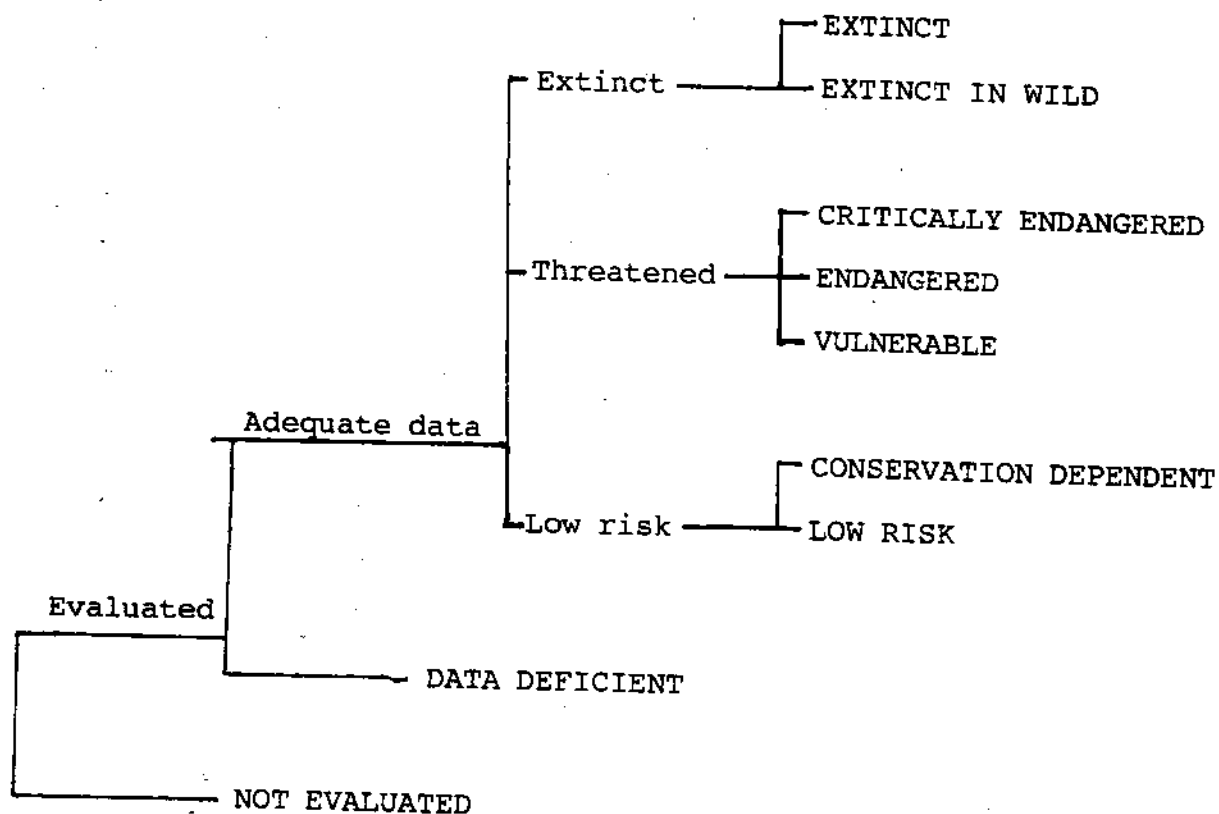


Figure 1: RED LIST CATEGORIES, VERSION 2.2

#### Future management:

If the Nile crocodile in South Africa is to survive outside of the available sanctuaries and for that matter even within the sanctuaries, a number of steps need to be taken to ensure their future:

- 1) Educational programmes and a neighbour relations policy need to be established
- 2) River systems need to be correctly managed and
- 3) a network of reserves needs to be established to adequately conserve the genetic diversity of the species and to protect populations throughout their range.

#### Future project plans:

The project will continue for an additional twelve months. By mid-June 1996, a food consumption versus growth rate experiment will be implemented. This experiment will look at osmolarity and growth rates of various age classes of crocodiles under varying salinities, various diets and ambient temperatures.

With the results of the above mentioned experiment, our additional field data, and assimilation and digestion rates from Games' (1986) study in Zimbabwe, we will be able to construct an energy budget for the various size classes of crocodiles in the Lake St Lucia ecosystem.

We plan to design and implement a management plan for the St Lucia crocodile population.

#### Acknowledgements:

The authors would like to thank the following organizations and companies for their generous financial assistance and for thus making this study possible:

Drexel University, USA

Earthwatch, Inc., USA

Natal Parks Board, South Africa

Alusaf, South Africa

Leomat, South Africa

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Conversation Starter:  
Summarizing Croc Growth Using Von Bertalanffy Curve

**Note:** This brief paper reflects a disjointed series of continuing conversations amongst Zilca Campos, Chris Hope, Paul Moler, Franklin Percival, Kenny Rice, Wayne VanDevender, and Phil Wilkinson. The summary provided here was typed up by Ab Abercrombie.

We have been concerned with (among other subjects) the von Bertalanffy growth curve. We like the curve OK for four reasons. (1) In some cases it appears to describe adequately the shape of croc length as a function of time. (2) It has been used by a number of researchers, and mathematical procedures exist to permit cross-population comparisons amongst such descriptions. (3) It at least purports to rest upon semi-reasonable biological assumptions. (4) In practical applications familiar to us, it has been statistically indistinguishable from (more complex) higher-order Richards curves.

In this meditation we want to do four things. First, we'll list the von Bertalanffy assumptions; this is just to remind you what they are. Second, we'll show in considerable detail the math by which the equations for growth in length are derived from the assumptions.<sup>1</sup> Third, we'll tell you our feelings about those underlying assumptions. We'd really like for you to think about this-- to show us where we are incorrect and to help us expand any interesting insights we might have. Fourth, we want to give you the chance to review just a little bit of the calculus-- which is, of course, the language we'll need to use (a) if there is a heaven and (b) if we get there.<sup>2</sup>

If you want to talk about growth-type issues, one possibility would be to get into electronic correspondence with Ab Abercrombie at [abercrombic1@wofford.edu](mailto:abercrombic1@wofford.edu) (note that the final "e" in "Abercrombie" is dropped). Add to or subtract from this non-paper. Tell us whether you found it at all interesting or helpful-- and whether we should complete the second-verse meditation we've planned for the next CSG meeting. You may write in English, Spanish, or Portuguese, but any reply will be in English.

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<sup>1</sup> We do this for two reasons. First, you might catch us in an error and thereby teach us both algebra and humility. Second, we used the von Bertalanffy curve for a long time without understanding why the equation looked the way it did. Perhaps we can help a few people avoid such blind application.

<sup>2</sup> We do not necessarily advise wagering the entire research budget on either (a) or (b).

## INTRODUCTION

If your entire goal is to describe your data<sup>3</sup>, then it is often appropriate to present an age-size graph without fancy summarizing lines. This can be particularly useful if you are dealing with known-age animals. Furthermore, some other croc workers will be most appreciative if you will also offer to share your raw data for their analysis.

If you believe it is important to summarize your data with lines, then you might ask yourself exactly why you want to do such a thing. Here are a few possible answers-- and some tentative suggestions on appropriate and inappropriate techniques.

1. Perhaps you simply wish to draw a representative line through a "busy" descriptive graph of size against age. Please do not fit a straight line; technically, such a model will almost always be "underspecified." Polynomial regressions can give very tight fits, but some of your readers may not be able to resist the temptation to extrapolate the line's equation beyond the data, which is intellectual suicide (please see our paper from the 1992 Zimbabwe conference)<sup>4</sup>. One appropriate technique might be to apply a (relatively) assumption-free locally weighted regression such as SYSTAT's "Lowess" curve. When using any such curve you will probably end up playing around with an inclusion "window"<sup>5</sup> that will affect how the computer program treats your

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<sup>3</sup> We recognize that many journals are unlikely to be enthusiastic about such descriptions. However, some of us in the "croc community" like to see regular old length-as-a-function-of-age descriptions of study animals, and our CSG proceedings may offer a vehicle for their publication.

<sup>4</sup> An example of a polynomial regression would be to specify croc length as a function of age, age squared, age cubed, etc. In our more lucid moments a few of us remember that any function can be expressed with any desired degree of precision (yielding an R-square arbitrarily close to 1.0) by piling higher powers into the polynomial equation. Technically this can be OK for pure description. However, if one projects a polynomial regression beyond the confines of sample data, one often derives spectacularly stupid results! We've demonstrated this by simulation.

<sup>5</sup> Lowess-type techniques work sort of like this: They choose a Y-value (a size) for Age =  $X_1$  by taking account of points within a region  $X_1 \pm W$ . W is (or is a function of) a user-selected parameter that we might call the examination window. There are no firm guidelines for choosing how wide to make this window, and choice of window-width involves a tradeoff. If you make your window wide, then you obtain a smooth curve that considers many of your observations in selecting each Y value;

observations. Because there is no widely accepted formal procedure for selecting window width, and because window width affects curve shape, the shape of each fitted Lowess curve will reflect not only the distribution of data points but also the researcher's gut-level professional judgment on how the curve should look. We don't see anything wrong with that subjective component. However, it does largely preclude formal comparisons<sup>6</sup> of growth curves from different studies.

2. You might wish to compare descriptions of two (or more) populations. One common exercise might be to compare a description of your crocs against a published report. In that case you could beg the author for the original data and re-analyze them however you wanted to. Otherwise you may be trapped by the original author's techniques-- and (for your own data set) would need to duplicate as precisely as possible those techniques applied in the published report. On the other hand, if you own all relevant data sets, then you might wish to use the von Bertalanffy curve, described below.

#### THE VON BERTALANFFY CURVE AS AN EXAMPLE OF GROWTH MODELS.

Here, as promised, are the assumptions and mathematical operationalizations of them.

Assumption 1. An organism's overall growth may be defined as the change in that organism's volume over time. This change in volume may be considered as the difference between material added through anabolic processes and material lost through catabolic processes.

Assumption 2. The addition of material to an organism is constrained by the rate at which the organism's surface can expand; or, to put it differently, gains in "stuff" must be reflected by gains surface area. I.e., von Bertalanffy's model posits a functional relationship between anabolic gain and organism surface area.

Thus, if A is a rate constant representing the combined effect of anabolic processes, then

$$V_{g,t} = AS_t \delta t, \quad \text{where}$$

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such a curve can obscure local changes in the age-size relationship but will be relatively free of "noise" (a particular animal's life-history effects, measurement-error effects, etc.). If you make the window narrow, you will obtain a relatively jagged curve. This curve should reflect how the animals' size changes with age but may also include all sorts of unwanted "noise."

<sup>6</sup> By formal comparisons we mean "comparisons on which you can do arithmetic to generate numbers that mean something."

A, a positive number<sup>7</sup>, is the anabolic rate constant,  
 $V_{g,t}$  is "material" (in the loose sense, meaning "stuff," or  
 "that which constitutes Volume"<sup>8</sup>) gained around time t  
 (due to anabolic processes),  
 $S_t$  is surface area at time t, and  
 $\delta t$  is an arbitrarily short period of time.

Assumption 3. Catabolic loss processes can operate anywhere throughout the entire volume of the organism. Therefore, catabolic loss is a function of the organism's volume. Thus, if C is a rate constant representing the combined effect of catabolic processes, then

$$V_{l,t} = CV_t \delta t, \quad \text{where}$$

C, a positive number, is the catabolic rate constant,  
 $V_{l,t}$  is "material" lost around time t (due to catabolic processes),  
 $V_t$  is volume at time t, and  
 $\delta t$  is an arbitrarily short period of time.

Note that by these three assumptions an organism's small amount of volumetric growth across a very short time is given by

$$\delta V_t = AS_t \delta t - CV_t \delta t,$$

or, as a differential equation,

$$dV/dt = AS - CV,$$

Equation 1

where we drop all the t-subscripts for convenience.

Assumption 4. Some organisms may be said to retain approximately the same shapes throughout their lives-- so that x and y (both positive numbers) may be considered shape constants in the following equations:

$$S = xL^2 \quad \text{and} \quad V = yL^3,$$

where S is surface area, V is volume, and L is a linear dimension of the organism.<sup>9</sup>

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<sup>7</sup> Note that  $V_{g,t}$  is the gross gain, not the net gain, in stuff; thus it can make sense to require that A be positive.

<sup>8</sup> It sounds a little better to talk about Volume rather than "stuff," material, or mass. Here we are assuming that any of those things may be appropriately considered to be a linear function of Volume.

<sup>9</sup> It should be easy to see what is meant by shape constants if we think about an easy, specific example. Consider, for

Now let's look at a little of the math. Substituting the relationships from assumption 4 into Equation 1, we obtain,

$$d(yL^3)/dt = AxL^2 - CyL^3.$$

If we divide both sides of this equation by  $y$ , we obtain

$$d(L^3)/dt = (x/y)AL^2 - CL^3.$$

Let's simplify the left-hand side of that equation. From our first semester of differential calculus we should recall that the first derivative of  $L^3$  with respect to time is  $3L^2 dL/dt$ ; substituting this identity into the above equation, we obtain

$$3L^2 dL/dt = (x/y)AL^2 - yCL^3.$$

Now divide<sup>10</sup> both sides of this equation by  $3L^2$  to obtain:

$$dL/dt = Ax/3y - CL/3 \quad \text{Equation 2}$$

Since all elements of Equation 2 are positive, we should note that growth-rate is now demonstrated to be a linear decreasing function of the length-dimension,  $L$ , for  $L > 0$ : the bigger they are, the slower they grow.

It is possible (see Appendix 1) to solve differential Equation 2 and show that under the 4 above assumptions,

$$L_t = L_{\max} - (L_{\max} - L_0)e^{-kt}, \quad \text{Equation 3}$$

where  $L_t$  is length<sup>11</sup> at time  $t$ ,  $L_{\max}$  is the maximum length<sup>12</sup> to which the organism can obtain,  $L_0$  is the length when  $t = 0$ ,  $e$  is the base of natural logarithms,  $k$  is the catabolic growth

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example, a cube, the surface and volume of which would be given by  $xL^2$  and  $yL^3$  respectively, where  $L$  would be the length of an edge: in this case shape-constant  $x = 6$  and shape-constant  $y = 1$ . Try that with any cube you like.

That is to say, one more time, if a growing organism does not substantially change its shape, then one may consider  $x$  and  $y$  as fixed parameters of the organism under consideration.

<sup>10</sup> We can do this only if  $L$  does not equal 0 (that is, if the organism has some length). Note that the degenerate case also makes a sort of sense: if  $L$  does equal 0,  $dV/dt$  also equals zero; organisms without size do not grow.

<sup>11</sup> Or of course  $L$  could be another linear dimension such as snout-vent length or head width.

<sup>12</sup> In Appendix 1 we show that this maximum length is equal to  $Ax/Cy$ .

constant, C, divided by 3, and t is time. For some purposes it is convenient to express this equation as:

$$L_t = L_{\max}(L_{\max} - Pe^{-kt}), \quad \text{Equation 3'}$$

where  $P = 1 - L_0/L_{\max}$ ; P may be understood as the proportion of the maximum length remaining to grow when  $t = 0$ . Anyhow, either way you write it, this function is known as the von Bertalanffy growth model<sup>13</sup>.

Perhaps you understand completely all the elements in Equations 3 and 3'. Some of us, however, have difficulty in visualizing what k might mean. We sometimes try to think of it as a shape parameter indicating when crocs grow fastest. Of course, under the von Bertalanffy model, all crocs grow faster when they are little-- but k indicates how much faster. All other things being equal, larger values of k indicate crocs that concentrate larger proportions of their growth in the early years. Appendix 2 provides three example graphs.

If the world were kinder to us, then croc researchers might always have sufficient size data from animals of known ages. In such a world researchers favoring the von Bertalanffy curve would presumably fit Equations 3 or 3'. Actually, of course, we are more usually forced to describe growth rates using data from capture-recapture studies. In this case it is still possible to estimate  $L_{\max}$  and k as used in the von Bertalanffy Equation 3 above. We usually do this as follows (see Appendix 3 for a derivation):

$$L_s = L_{\max} - (L_{\max} - L_f)e^{-k(\Delta t)} \quad \text{Equation 4}$$

where  $L_s$  is length at second capture,  $L_f$  is length at first capture, and  $\Delta t$  is the length of time between first and second captures.<sup>14</sup>

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<sup>13</sup> Unfortunately, it's also known as a number of other things. In our paper for the 1992 Zimbabwe meetings we called it "the von Bertalanffy model for linear growth." It is also commonly termed "the Monomolecular model." To make things particularly confusing, there is another model, for volumetric growth, that is also termed "the von Bertalanffy model." We'll mention this one in another footnote below.

<sup>14</sup> Occasionally animals are caught more than two times. In such cases you can, of course, employ any two captures (and the relevant inter-capture interval) for fitting this model. Most folks usually use first and last captures. There can be instances in which one might wish to use other capture-recapture pairs. In general you should not use more than one inter-capture interval per animal; otherwise your data points will not be independent. (That is, animals captured more than twice will contribute more than their "fair share" to the estimate of growth)



If you accept assumptions 1-4, then Equations 3, 3', and 4 express the way you think crocodilians grow. But the important question is, do you really want to accept the assumptions? Let's look through them all.

Assumption 1. An organism's overall growth may be defined as the change in that organism's volume over time. This change in volume may be considered as the difference between material added through anabolic processes and material lost through catabolic processes.

It is difficult to disagree with the essential idea of this assumption. Basically it states (1) that any change in size is the mathematical difference between stuff added and stuff subtracted, (2) that additions are not simply glued on and subtractions are not simply chopped off. For purposes of the mathematical model, it shouldn't really matter very much what anabolic and catabolic mean. We buy this assumption; can you find any problems in it?

Assumption 2. The addition of material to an organism is constrained by the rate at which the organism's surface can expand; or, to put it differently, gains in "stuff" must be reflected by gains surface area. I.e., von Bertalanffy's model posits a functional relationship between anabolic gain and organism surface area.

We like the verbal statement fine, but we are troubled by the assumption that the anabolic gain "constant," A, is truly constant for all values of S (surface).<sup>15</sup> We also believe that this assumption (as well as the #3, that follows) will crash if assumption 4, on conservation of shape, does not obtain. What do you think?

Assumption 3. Catabolic loss processes can operate anywhere throughout the entire volume of the organism. Therefore, catabolic loss is a function of the organism's volume.

We feel the same way about assumption 3 as we did about # 2 above. The words sound OK, but we are not convinced that (for

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parameters.)

<sup>15</sup> We can prove an analogous proposition under reduced dimensionality-- e.g., for change in area as a function of change in perimeter for 2-dimensional bacterial colonies-- given biologically reasonable assumptions about growth. We also believe this assumption might hold for growth of spherical colonies of bacteria. We are less confident about crocs.

Maybe we do need to know a bit more about the definitions of anabolic and catabolic.

crocs) the catabolic loss "constant,"  $C$ , is truly constant across all values of  $V$ .<sup>16</sup>

Assumption 4. Some organisms may be said to retain approximately the same shapes throughout their lives.... It is trivial to point out that the shape of an adult crocodilian is different from the shape of a hatchling, so assumption 4 is not precisely true. However the shapes of sundry-sized crocs are basically a lot alike, so this proposition may hold. We have tried to write some analytical models for testing this assumption, but the algebra gets pretty ugly, there are many opportunities to make small mistakes, and our output has been difficult to interpret. Nevertheless, we have scared ourselves a little because it appears that (at least toward the ends of a von Bertalanffy growth curve) relatively minor changes in shape may affect the model. As far as we know, nobody who does von Bertalanffy work seems to worry about this. If you do, let us know, and maybe together we can tease out the effect of shape.

OK, folks, that's all we want to say at this time. Pretend that this is an Internet communication, embryonic and poorly thought out. If it bores you, then neglect it. If not, let's figure out a way to deal with it together.

---

<sup>16</sup> We are not as worried about this assumption as we are about # 2 above.

## Appendix 1

$$dL/dt = Ax/3y - CL/3$$

Equation 2, from text above

factor out C/3:

$$dL/dt = C/3(Ax/Cy - L)$$

Equation A1

Recall from intro calculus that we can maximize  $L^1$  (or, put differently, determine  $L_{\max}$ ) if we set  $dL/dt = 0$ :

$$dL/dt = 0 = C/3(Ax/Cy - L_{\max});$$

$$L_{\max} = Ax/Cy$$

Thus, if we substitute and also re-name C/3 as k, we have

$$dL/dt = k(L_{\max} - L)$$

Equation A2

Now let's work on differential equation A2. We separate the variables and integrate:

$$\int \frac{dL}{(L_{\max} - L)} = \int (k dt)$$

Next we evaluate the integrals to give:

$$-\log_e(L_{\max} - L) = kt + c,$$

Equation A3

where c is the constant of integration. Now we impose the initial condition that  $L = L_0$  when  $t = 0$ , and we plug  $t = 0$  into equation A3 so that we can afterwards solve for c:

$$-\log_e(L_{\max} - L_0) = k(0) + c,$$

$$c = -\log_e(L_{\max} - L_0),$$

and we substitute this value of c into equation A3 to give:

$$-\log_e(L_{\max} - L) = kt - \log_e(L_{\max} - L_0)$$

---

<sup>1</sup> Note that this will indeed be a maximum, and not a minimum, for L. We explained in the text that when L is small, growth rate,  $dL/dt$ , is greater than zero, and as L gets larger,  $dL/dt$  gets smaller. It should be clear that small  $dL/dt$  will be associated with a big L (biologically,, big animals increase more slowly in length).

Taking exponentials of both sides of this equation, we get

$$1/(L_{\text{Max}} - L) = e^{kt}/(L_{\text{Max}} - L_0)$$

Invert both sides of that equation and you get:

$$(L_{\text{Max}} - L) = (L_{\text{Max}} - L_0)e^{-kt};$$

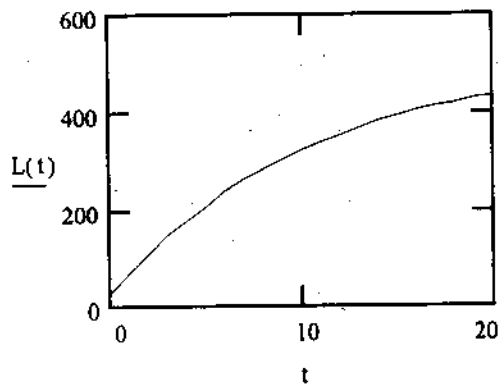
Now just solve for L, also restoring its t-subscript:

$$L_t = L_{\text{Max}} - (L_{\text{Max}} - L_0)e^{-kt} \quad \text{Equation 3 in the text above}$$

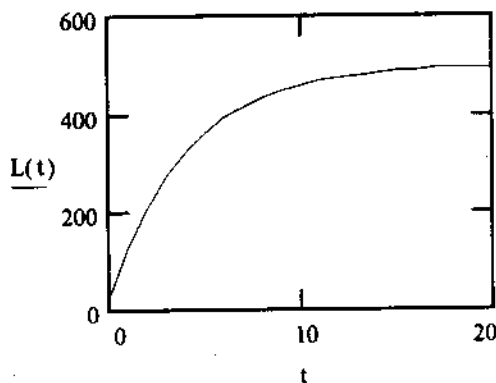
## Appendix 2: The Mysterious k Parameter

The precise numerical effect of changes in the k-parameter will depend on the hatching and maximum sizes of the varmint in question (our hypothetical example hatches at 25cm and maxes out at 500cm) as well as the units in which t is measured. However, the relative mathematical shapes of the growth curves will be as illustrated below.

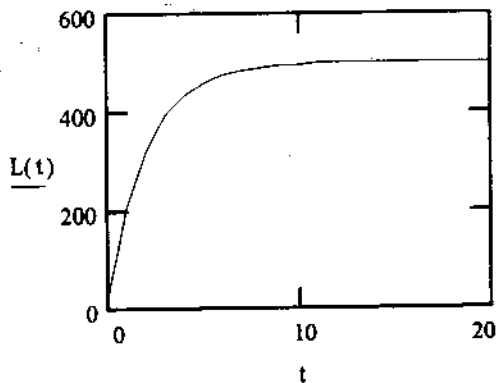
First diagram:  $k = 0.10$ :



Second diagram:  $k = 0.25$ :



Third diagram:  $k = 0.50$ :



Here's the deal: Other things being equal, larger values of  $k$  indicate faster initial growth.

## Appendix 2

Fitting the Von Bertalanffy growth model to data generated by capture-recapture studies on animals of possibly unknown age.

Look back to equation A3 in Appendix 1:

$$-\ln(L_{\max} - L) = kt + c \quad \text{Equation A3}$$

Now let us take a specific time, say the time at which  $t = F$  (eventually we may want to think of  $F$  as being age at first capture). We can represent length at  $t = F$  by  $L_f$ . Now let us enter those values into Equation A3 and solve for  $c$ :

$$-\ln(L_{\max} - L_f) = (k)(F) + c$$

$$c = -\ln(L_{\max} - L_f) - (k)(F)$$

Now substitute this value for  $c$  into Equation A3:

$$-\ln(L_{\max} - L) = kt - \ln(L_{\max} - L_f) - (k)(F) \quad \text{Equation A4}$$

Say we wish to think about length,  $L$ , at the time  $t = S$  (eventually we may want to think of  $S$  as being age at second capture). We can represent length at  $t = S$  by  $L_s$ . Now let us enter those values into Equation A4:

$$-\ln(L_{\max} - L_s) = (k)(S) - \ln(L_{\max} - L_f) - (k)(F)$$

Our next goal is to solve for  $L_s$ . First we shall collect the terms that include  $k$ :

$$-\ln(L_{\max} - L_s) = -\ln(L_{\max} - L_f) + k(S-F)$$

Next we shall rearrange terms algebraically:

$$-\ln(L_{\max} - L_f) = -\ln(L_{\max} - L_s) - k(S-F)$$

Now we take "take the antilog" of both sides of the above equation:

$$1/(L_{\max} - L_f) = (e^{-k(S-F)})/(L_{\max} - L_s)$$

Rearrange a little (multiply both sides of the equation by both denominators to get everything up on the same line):

$$L_{\max} - L_s = (e^{-k(S-F)})(L_{\max} - L_f)$$

Solve for  $L_s$ ; note that we could define the difference between times  $t = S$  and  $t = F$  as  $\Delta t$ ; rearrange just a little:

$$L_s = L_{\max} - (L_{\max} - L_f)e^{-k(\Delta t)}$$

**THE ENVIRONMENT AND ITS RELATIONSHIP WITH EGG SIZE  
CLUTCH SIZE AND HATCHING SUCCESS IN DIFFERENTS  
*Caiman latirostris* POPULATIONS AT SANTA FE, ARGENTINA**

**Alba Imhof; Ana Costa and Alejandro Larriera**

**Bv. Pellegrini 3100, Santa Fe-3000, Argentina**

**Introduction**

As was reported by Larriera (1995) in the distribution range for *Caiman latirostris* in Santa Fe province, is possible to find five different nesting environment with specific characteristics each one:

- **Artificial water bodies (ponds or channels):** Man made facilities for water reserve or transport for cattle ranch exploitation. This places are normally far from natural water bodies, but has been colonized for broad snouted caimans for a while. The preferred construction materials here are the gramineous.
- **Ridge in swampy land:** Small elevations in swampy lands with predominance of *Spartina argentiniensis*, *Typha sp* y *Scirpus sp*. No arboreal and bush vegetation could be found here.
- **Ridge in (or around) water bodies:** Small elevations around lakes, narrow rivers and brooks, periodically flooded. Nests are found relatively close from the water and are normally build by gramineous, *Spartina* and *Typha*.
- **Floating vegetation in swampy lands:** Floating formations in swampy lands, formed from death vegetation and sediments. Sometimes are colonized for bush and always by grass (gramineous) that is the nesting material used here.
- **Forest :** Environments with variable density of trees and bushes, elevated with respect to the main water bodies. Nests here could be found in some cases more than 2 kms. far from water bodies, but in general, close to a small pond within the forest. Nesting materials here are grass, soil and some sticks and branches.

The objective of this work is to evaluate if there are differences on clutch size, egg size and hatching success in the environment categories described before.

There are not previous research relating environment with *Caiman latirostris* nesting, but Campos and Magnusson () and Campos *et. al.* () report some information for *Caiman crocodilus yacare*.

At the moment to evaluate the results, must be considered that there are remarkable variations in the climatological situation at the breeding season, that certainly will incide in the final reproductive success. Moderate rain, heavy rain,

drought, and severe drought could happen in different years, so this work must be replicated in the successive years in order to improve and adjust the results.

The severe drought occurred this year could explain the fact that we found nests from just three of the five categories. No nest were found at artificial pond and channels, neither in ridges in (or around) water bodies. The exploitation at these places is cattle ranching, so the animals were frequently concentrated at these places looking for water what certainly could disturb the reproductive process.

### Methods

Eighty four nests were harvested between January and February 96' in different environments northern Santa Fe. For this work were considered 74, and from those, seven correspond to Forest, 20 to Ridge in (or around) water bodies, and 46 to Floating vegetation in swampy land.

Nests were marked for local inhabitants and for us when the helicopter was used. The harvest and transportation was made by horse, track, boat or helicopter, depending the place.

The working area was minutely studied, so we can suppose that most of the nests were found.

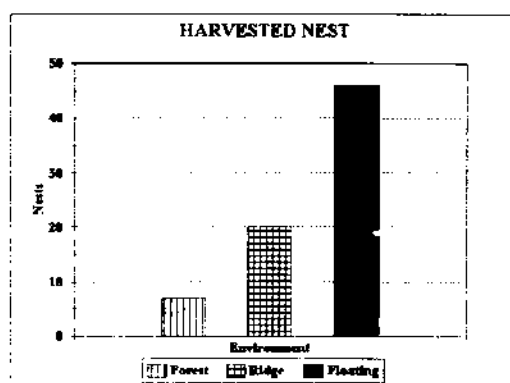
Harvested and marked eggs were transported to the incubator at the breeding station. There were measured with a caliper and the volume was calculated.

Hatch occurred since the end of February until the middle of April. The clutch size and number of hatchlings per nest was recorded, and from this information the hatching success was calculated.

### Results

1- The preferred environment for nesting this year was Floating vegetation (Embalsado). (Fig. 1)

**Figure 1**





In order to evaluate significative differences between Habitat preference a Chi square Test was made.

#### Chi - square Goodness of fit Test

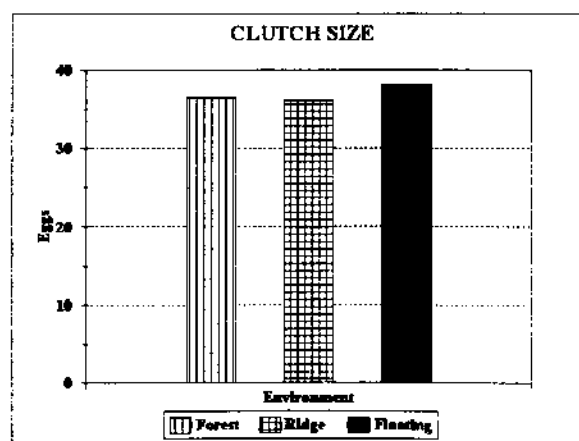
Environment	Observed Frequency	Expected Frequency	Chi Square
Monte	10	33	16,90
Albardón	27	33	1,05
Embalsado	63	33	26,49

Chi square = 44,4341 with 2 d.f.

Sig. Level = 2,22451E - 10

2- In Fig. 2 are showed the differents clutch size recorded for each environment tipe.

Figure 2



In order to evaluate if the differences were significatives, a Kruskal Wallis Test was made, showing that they are not significative differences.

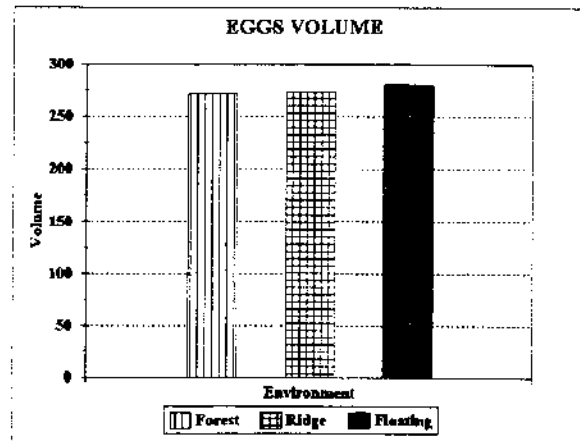
#### Kruskal - Wallis Test

Environment	Sample Size	Average Rank
Monte	7	39,5
Albardón	20	34,1
Embalsado	46	37,8

Test statistic = ,551908 p-value = ,758848

3- In Fig. 3 is showed the different eggs volume for each envaironment tipe.

**Figure 3**



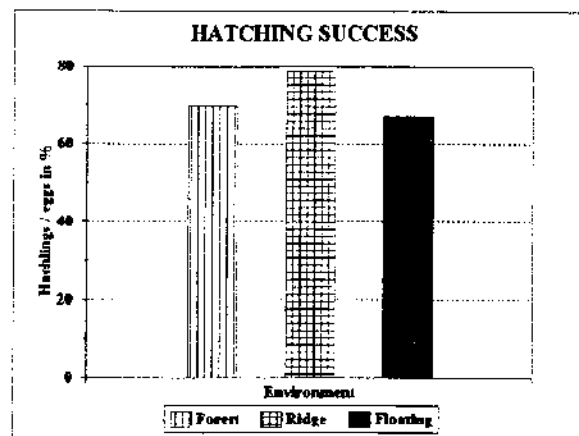
In order to evaluate the different eggs volume between environments, an Analysis of Variance was carried out.

**ANOVA Table - Analysis of Variance**

Source	Sun of Squares	Df	Mean square	F - Ratio	p - value
Between groups	1595,2	2	797,601	1,33	,2725
Within groups	40927	68	601,868		
Total (Corr.)	4522,2	70			

4- In Fig. 4 are displayed the different Hatching success recorded for each environment type.

**Figure 4**



In order to evaluate differences, a Kruskal Wallis Test was made, showing that there are significative differences between environments.

#### Kruskal - Wallis Test

Environment	Sample Size	Average Rank
Monte	7	35,1429
Albardón	20	47,375
Embalsado	46	32,7717

Test statistic = 6,63523 p-value = ,0356997

#### Conclusions

Taking into account the fact that most of the nests were found in the working area, the floating vegetation in swampy lands appear as the preferred nesting environment for this year., followed for the ridge in (or around) water bodies, and finally for the forest.

Despite the fact that the preferred environment was the floating vegetation in swampy lands, the hatching success there was lower than in the other places. The question is, caimans nesting there because preference or because they have no choice? Further studies could respond it.

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# EGG SIZE ON *Caiman latirostris* AND ITS RELATIONSHIP WITH CLUTCH SIZE, HATCHING SUCCESS, SURVIVOR AND GROWTH

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## Introduction:

Broad snouted caiman (*Caiman latirostris*) is one of the two species occurring in Argentina, it is widely distributed in northern Santa Fe and certainly is the most abundant in Santa Fe province.

The experimental ranching program is currently developing different researches about various issues. One of the most important is to evaluate reproductive parameters relating clutch size, hatching success, survivorship and growth with the egg size, and by this mean eventually, with the female size.

The objective of this particular paper is to give some information about the recorded differences on hatching success from wild harvested *Caiman latirostris* eggs and its relationship with egg volume and clutch size. We investigate too, the relationship between the egg volume and the hatchlings size.

## Materials and Methods

Eggs come from wild nests harvested in different places, (San Cristóbal, and San Javier States, where the "Proyecto yacaré INTA \ MAGIC \ MUPCN", develops its program of recovering and monitoring wild populations of "Broad snouted caiman" (*Caiman latirostris*).

The 365 eggs of *C. latirostris* were measured with a caliper (0.1 mm). The volume of the eggs was calculated according the ellipsoid equation;  $\frac{3}{4}\pi \cdot A \cdot B \cdot C$ , being *A* the larger diameter and, *B* and *C* the smaller ones.

The weight of the clutch (only hatchlings) was measured with a pesola of 10 gr. precision at the hatch moment and with a 100 g precision when they were released.

The results are presented in  $\text{mm} \pm \text{SDT}$ , the weights are the averages recorded in each nest.

## Results

Table N°1: Maximum medium and minimum values with its SDT, of 365 *Caiman latirostris* eggs

n=365	Big axis	Small axis
max. value	76,6	47
midium	66,53±3,31	40,71±1,85
min. value	56,65	31

Table N°2: Big and small axis midium values from 11 *C. latirostris* nests eggs with its clutch size.

Nest	Big axis	Small axis	Clutch size
1	71,38±2,52	44,96±0,58	41
2	68,53±1,43	41,16±0,75	43
3	60,46±3,95	40,82±1,57	29
4	68±2,32	40,23±0,75	33
5	68,58±2,58	37,14±1,42	35
6	72,18±1,77	44,99±0,69	39
7	67,09±2,44	40,57±0,35	38
8	66,96±2,9	40,05±0,75	37
9	65,47±1,76	42,40±0,42	49
10	65,45±1,84	40,19±0,68	41
11	63,27±1,27	40,08±0,81	43

Table N° 3: Averange weight of the nests at moment of hatching and releasing.

Nest	Weight during hatching*	Weight during releasing
1	40	140
2	40,24	177,5
3	40	200
4	36,84	192,86
5	33,33	104
6	40	153,33
7	39,29	111,53
8	44,12	173,53
9	45,65	250

10	40,26	202,5
11	36,25	315,15

\*Weights are the averanges of the clutch

The average weight of all animals is: 40,23 g.

Table N° 4: Clutch size, hatching success and survival of the 11 nests in relation to the eggs volume.

Nest	Volume (cm³)	Clutch	Hatching success	Survival
1	339,96	41	0,98	1
2	273,55	43	0,95	0,98
3	237,36	29	0,35	0,8
4	259,3	33	0,58	0,74
5	222,88	35	0,94	0,46
6	344,23	39	0,41	0,94
7	260,18	38	0,74	0,93
8	253,06	37	0,92	1
9	277,31	49	0,92	0,98
10	249,08	41	0,95	1
11	239,47	43	0,93	0,83
Average	268,76	38,91	0,81	0,89

## Discussion

Verdade (1995, in São Paulo, working with 17 nests, collected in 6 years, obtained an average of 33 eggs per nest, being its range from 18 to 49; the hatching success was 7,33 hatchlings per nest (average); the average size of the eggs was 67,3 mm (big axis) and 42,8 mm (small axis), being its range 60 mm to 75 mm and 34 mm to 45 mm.

The values recorded by other authors range from 57,1 to 75,5mm and 37,2 and 46,6mm, big and small axis respectively (Achaval and González, 1983). This values are included within our range recorded in this paper (Table N° 1).

Ferguson (1984) and Ferguson and Joanen (1983), described a relationship between the female age and the size of the eggs and the clutch size; they conclude that young females (younger than 15 years) laid few and little eggs, the females between 15 to 20 years laid big clutch and the eggs are big; and the females older than 30 years laid few but the biggest eggs.

The picture N° 1, shows the relationship between clutch size and the volume average of the eggs; an increase can be appreciated of the clutch size when the volume raises, until eggs bigger come from clutch smaller. In South Carolina, Wilkinson (1985), report the relationship between the eggs size, clutch size and the female size. Casas Andreu and Rogel (1986), did not found correlation between the female size and the eggs size, not either between the female weight and the number of laid eggs. When they try to make a correlation between the female weight and the clutch weight, they did not found any significative result. Others works in species such as *Alligator mississippiensis* (Deitz and Hines, 1980; Joanen, 1969; Joanen and Mc Nease, 1975; 1989) and *Crocodylus porosus* (Webb *et al.*, 1977; 1983), did not found relationship neither. The clutch size (average  $n=11$ ) found in this work was higher than the described by Larriera (1991) (37,1 eggs/nest  $n=12$ ). Values for other specie (*C. yacare*), range between 55,8 and 75,5mm for the larger axis and 33,5 to 46,6mm for the smaller (Tables N° 6 and 7); this ranges are narrower than the range found in this work to *C. latirostris* (Table N° 1).

In the relationship between the eggs volume with the animals weight at hatch (Fig N° 2), appears a weight raise of the hatchlings when the eggs volume increase. The regression of this relationship shows a low  $R^2$  value ( $R^2=0,42$ ) suggesting that exist a lots of other factors influencing on the hatchlings weight, but the correlation between the same relationship shows a significant  $r$  value ( $r = 0,65$ ;  $P<0,05$ ), this means that the relationship between the eggs volume and the weight of the hatchlings at hatch is significative. Schulte and Chabreck (1990), found that the hatchlings size was affected by the nesting environment and the incubation temperature, but certainly were more influenciaded by the nest itself. In the same paper, Schulte and Chabreck (1990), found that the hatchling weight was strongly influenciaded by the eggs volume. In others species of crocodilians, the same relationship was found too (Deitz and Hines, 1980; Staton and Dixon, 1977; Webb *et al.*, 1983). Casas Andreu *et al.* (1993), observed a significative relationship ( $r = 0,72$ ;  $P<0,05$ ) between the weight of the eggs and the hatchlings at hatch; they foun that possibly the hatchlings of the big eggs, have a larger survive rate.

After 9 month, when the animals were released, they were weighted (Fig N° 3), the result of this correlation ( $R^2=0,048$ ;  $b=-0,34$ ) shows that the eggs volume do not act on the following growth of the animals.

Speculating about the relationship between the regression value and the information displayed on figures 4 and 5, seems that from nests with small eggs, hatch and survive few (sometime large) animals, because the trend show that hatching success and survivorship are increased as larger are the eggs and the fact of its larger weight could be explained for errors calculating average in few animals.

## Figures

Figure N° 1: Relation between clutch size and the volume average of the eggs.

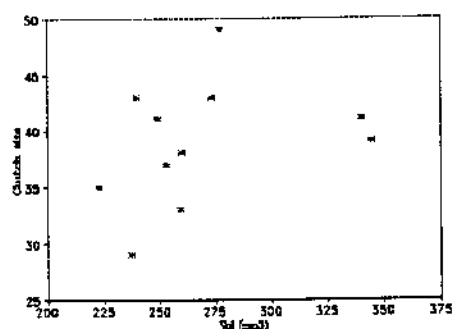


Figure N° 2: Relation between eggs volume and the animals weight at hacht.

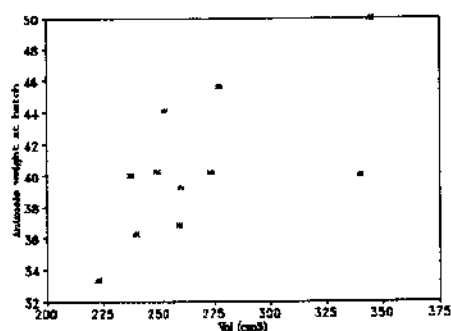


Figure N° 3: Relation between eggs volume and the animals weight at releasing time.

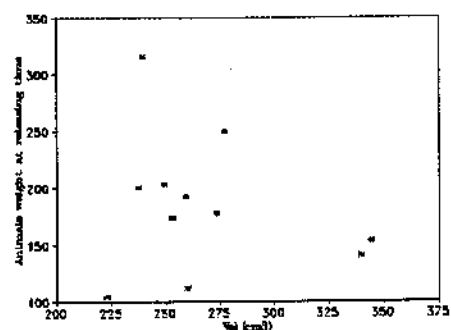


Figure N° 4: Relation between eggs volume and the hatchling-success.

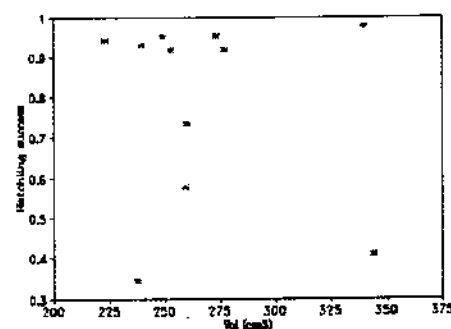
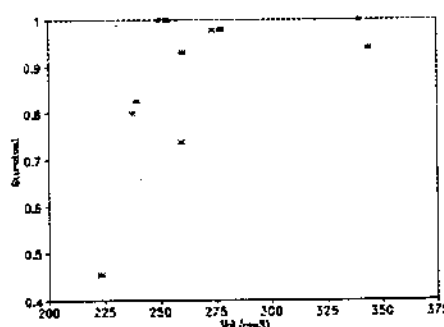




Figure N° 5: Relation between eggs volume and survival.



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CALCIFICATION BAND AND EMBRYO DEVELOPMENT OF *CAIMAN LATIROSTRIS*  
EGGS INCUBATED AT THREE DIFFERENT TEMPERATURES

Larriera, A., P. Donayo, A. Imhof & C. Piña

## INTRODUCTION

The opaque band we see in eggs changes as the egg content reorganizes itself, during the development of the embryo (Webb et al.1987).

Such opacity would be caused by structural changes and changes in the optical properties of the shell, caused by dissolution of calcium carbonate crystals, as the albumen dehydrates and the chorioallantois expands or the embryo membranes (Ferguson.1982 and Webb et al.1987). This way, until the egg become completely opaque, it is possible to calculate the embryo age, according to extent of the band.

These facts, as well as the total period of embryo development (from the laying up to the eclosion) and the influence of temperature during the development time have been reported from different crocodilians species, being the study in *Alligator mississippiensis*, *Cocodrylus porosus* and *Cocodrylus jobstoni* more detailed.

For this species, *Caimán latirostris*, only total average time has been calculated from the laying up to the eclosion in the wild (Larriera.1992-1993).

## OBJETIVES

- To develop a cheap and fast technique that allows to know the degree of embryo development from eggs collected in the wild to determine the date of the laying.
- To determine the temperature effects on the embryo development time.

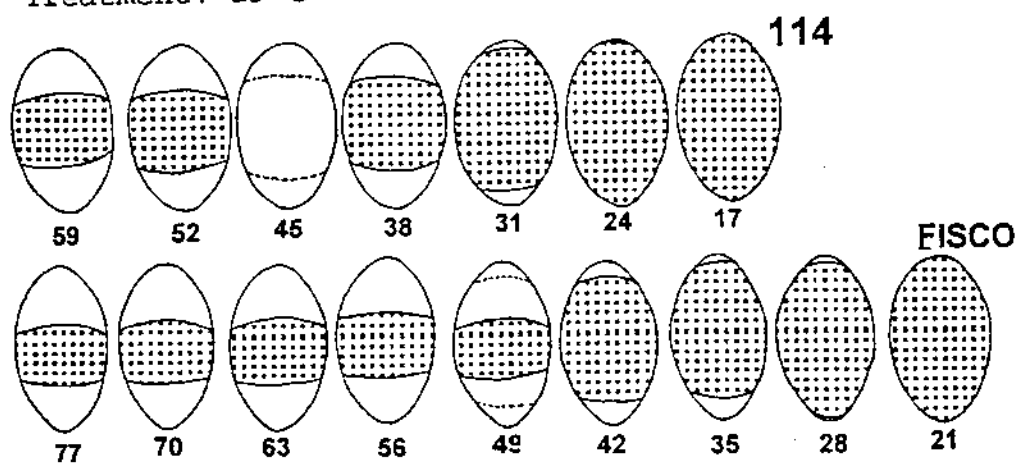
## MATERIALS AND METHODS

Eggs coming from two nests of the harvesting of wild eggs or experimental Ranching collected through the ordinary routine were used (Larriera.1990-1992-1994), being the date of laying estimated according to what Larriera had previously recorded. In one of the nests (Town 114) the development time (from the laying) has been estimated in 14±2 days, and for the other (Fisco), in 5±2 at the moment the artificial incubation started.

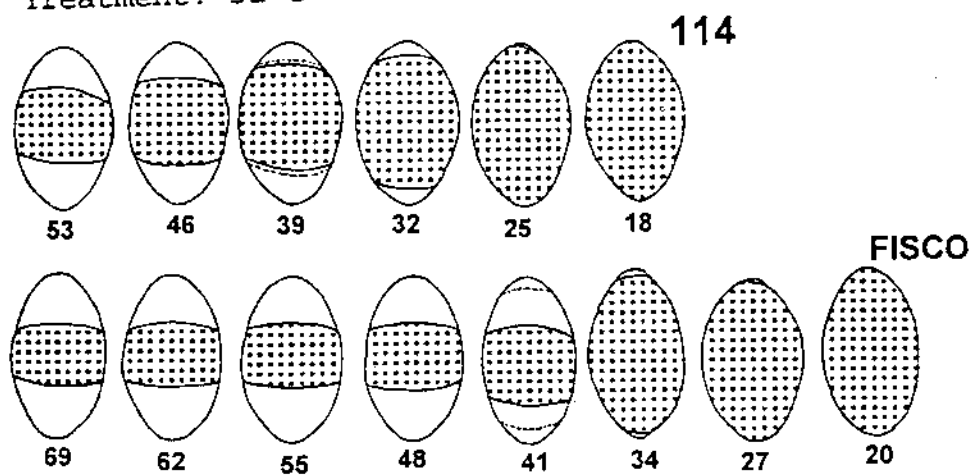
Three hand-made incubators were used. They consisted of plastic punts of 47 cms tall and a diameter of 50 cms, 2 cms thick tergopol lids, and a -30°C to +30°C thermostat. Each of which was filled with 10 cms water to keep humidity and ten eggs of each harvested nest altogether with nest material. The eggs were 8 cms above the water with a plastic grille that supported them.

Fig. 1: OPAQUE BAND DEVELOPMENT DURING ARTIFICIAL INCUBATION  
Days prior to eclosion

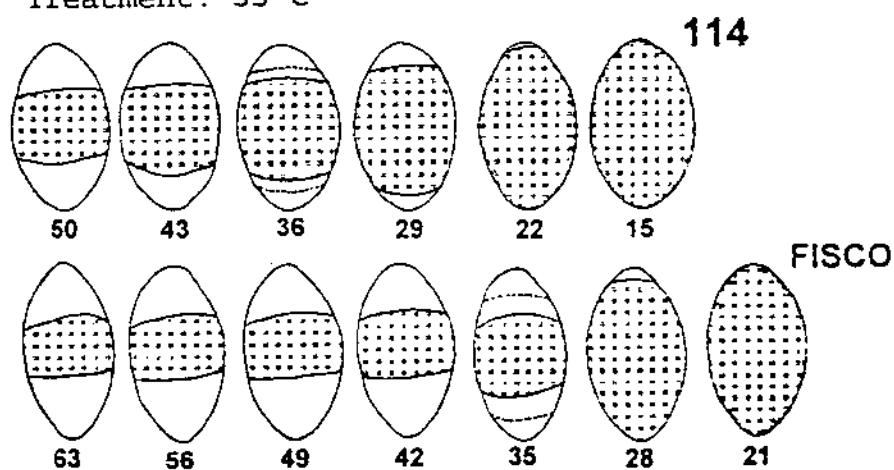
Treatment: 29°C



Treatment: 31°C



Treatment: 33°C



Incubators were kept at three different temperatures: 29°C, 31°C and 33°C. The only condition that changed during the incubation period was temperature.

The calcification band that we observed with the "ovóscopo" (directed powerful light fountain which acts by contrast), was individually measured with a caliper every seven days in the greater axis of the egg, without variation in the position they had in the nest from the first day of artificial incubation until they become completely opaque. The average value was calculated in each nest.

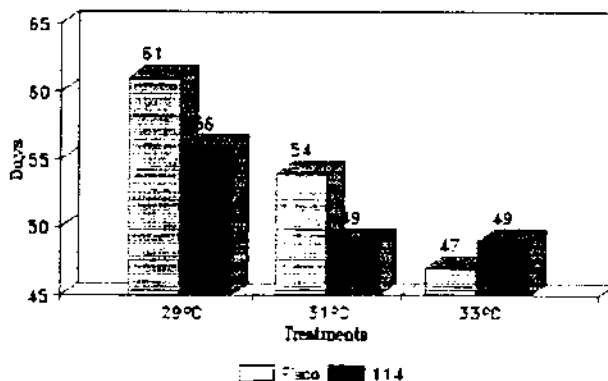
## RESULTS

It has been observed that the extent of the calcification band increases simultaneously with the development time, the same as for the other crocodilians already studied (Webb et al.1987 for *C. porosus* and *C. johnstoni*; Joanen and McNease.1991 for *A. mississippiensis*). (Fig.1)

Total opacity has been reached between the second and third week prior to the eclosion (15-21 days) without markedly differences in the different temperature treatments. But if we take into account the two nests, *Fiscons* ended its opacity in every temperature long before its eclosion.

If the estimated laying date is considered, total opacity is reached between 47 and 61 days after the laying, increasing time as temperature diminishes (Fig. 2). Approximately 65 days have been estimated for *C. porosus* eggs incubated at 30°C (Webb et al.1987), and 52 days for *A. mississippiensis* eggs incubated between 29.4°C and 32.7°C (Joanen and McNease.1991).

Fig. 2: ESTIMATED TIME UP TO TOTAL OPACITY (from the laying)



It is also observed a period in the two nests and in the three temperature treatments where the band limits are diffuse, with a generally darker zone in the center that coincide with a fast and great extension of the band. It has to be considered that as the band was weekly measured, in many cases we may be talking about a process that has just begun or may be ending. (Fig.1)

The time in which the diffuse band takes place since artificial incubation started is the same for the three treatment of the same nest, although they are different if we compare the two harvested nests. The period required for the Fisco nest eggs is larger in all temperatures in relation with Town 114 nest eggs. (Fig.3)

The same happens if it is considered when the diffuse band is reached from the estimated laying date. (Fig.4)

In *C. porosus* eggs, there is an evident band expansion approximately to the 40-45 days from the time of laying (Webb et al.1987); the estimated total incubation time is an average of 100 days (Magnuson and Taylor.1980). Also a *C. porosus* study in which embryos are slaughtered to assess morphological changes that occurs altogether with development shows that about 38 days after the laying a noticeable development is observed in embryos. It can be seen, for instance, in the increase of forelimb length and hindlimb length as well as in the head (Magnuson and Taylor.1980).

Therefore, the band diffuse limits in *C. latirostris* could be due to continuous and quick changes in the eggshell components and in the egg membrane due to vascularization and reorganization of its contents during its development. This would be the reason why they cannot be delimited.

On the other hand, the time that lasts to the eclosion varies starting from this situation in the different treatments and in the two nests, being this greater as temperature diminishes. (Fig.1)

Fig. 3: TIME UP TO DIFUSE BAND  
(from artificial incubation)

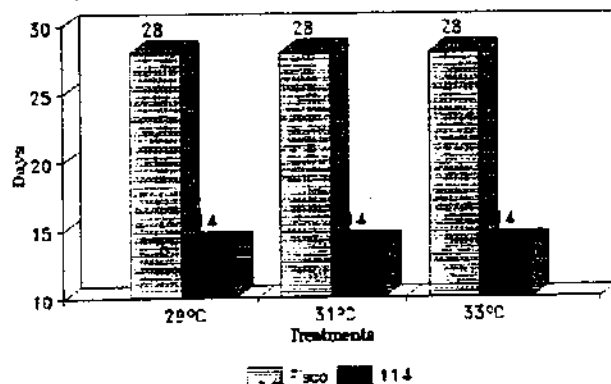
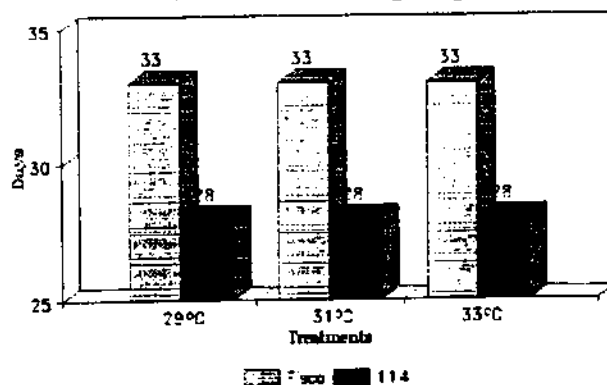


Fig. 4: ESTIMATE TIME UP TO DIFUSE BAND  
(from the laying)



If the artificial incubation time in the higher temperature treatment (33°C) is analyzed, it is less. On the contrary, in lower temperature treatment (29°C), it is larger. (Fig.5)

The same happens if total development time is considered (from laying estimated date up to eclosion) which varies between 64-82 days; in the wild, this period had been estimated between 71-79 days (Larriera.1992-1993). In the higher temperature treatment, time is less; and in lower temperature treatment it is larger (Fig. 6), which can also be seen in *C. porosus* and *C. johnstoni* (Webb et al.1987).

Fig. 5: ARTIFICIAL INCUBATION TIME

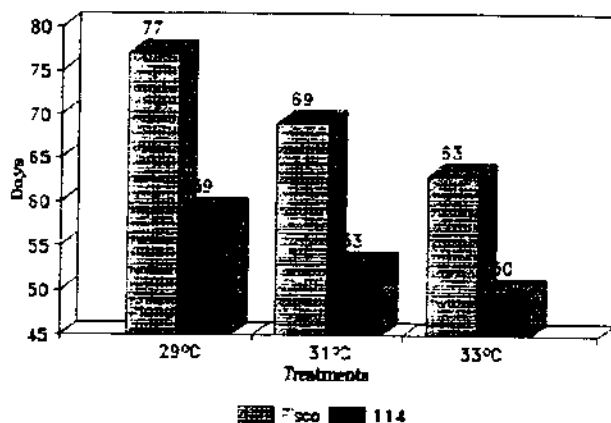
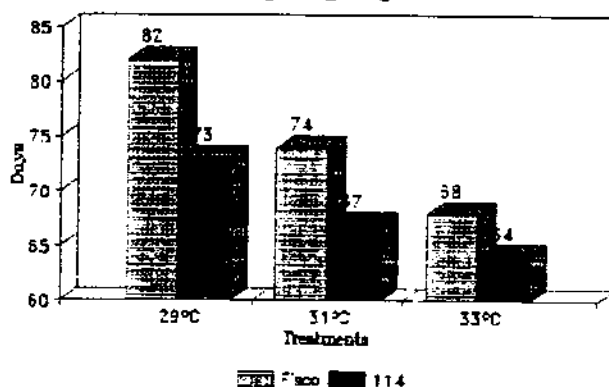


Fig. 6: DEVELOPMENT ESTIMATED TIME (from the laying up to eclosion)



In embryos of the same nest and whatever treatment is being considered, it can be seen that there are not markedly differences in the early stages of embryo development (approximately up to the diffuse band, close to half the total development time). From now onwards it is harder to estimate the embryo age. These results are similar to those of *C. porosus*, in which most differences take place after 60 days development (Magnuson and Taylor.1980). (Fig.1)

At the same time, in the two nests harvested, and taking as reference the medium temperature treatment (31°C), there are always a wider difference (greater amount of days) between 31°C and 29°C treatment (lower temperature) than between 31°C and 33°C treatment

(higher temperature) (Table 1). Temperature influenced a lot delaying the development.

In addition, more markedly differences can be observed among Fisco nest eggs three treatments (regarding difference in days between the three treatments) taking into account the estimated total development time and the artificial incubation time (Table 1). This nest was the one that had less development time at the moment of artificial incubation. This would suggests that as soon as posible the treatment stars, temperature will influence much more in the development. Complementary studies should be carried out since this difference could be due to individual variations among the nests.

Table 1: DEVELOPMENT ESTIMATED TIME  
(from the laying up to eclosion)

	29°C	31°C	33°C
Fisco	82±2	74±2	68±2
114	77±2	67±2	64±2

#### CONCLUSIONS AND DISCUSSION

- The opaque band observation is an useful method to estimate the embryo age in the early development stages.
- Temperature has an inverse effect on the embryo development time.
- Temperature would influence much more delaying the development rather than speeding it up.
- Temperature effect on development time would be larger as soon as posible the treatment starts.



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## **EFFECTOS DE LA TEMPERATURA DE INCUBACION SOBRE EL CRECIMIENTO Y DESARROLLO EMBRIONARIO DE *Caiman crocodilus fuscus*.**

**C. L. SIERRA D., M.A. RODRIGUEZ M., G. A. ULLOA D., P. M. RUIZ-CARRANZA & G.GALVIS.**

Durante el desarrollo embrionario se suceden en forma secuencial cambios morfológicos y funcionales. Al considerar estos cambios surgen tres aspectos importantes del desarrollo como son: diferenciación celular, morfogénesis y crecimiento. Estos tres aspectos en los Crocodylia y los Testudinata están regulados por la temperatura de incubación y por esta asociadas la humedad relativa y el ambiente gaseoso del medio en que se desarrolla el proceso.

Packard y colaboradores (1987) afirman que las variaciones temporales y espaciales en la humedad y la temperatura dentro y entre nidos naturales probablemente sean las responsables de variaciones ecológicas importantes en el tamaño y sexo de neonatos, de ahí la importancia de determinar experimentalmente los efectos de la temperatura de incubación sobre el desarrollo.

Se incubaron entonces 815 huevos embrionados de *Caiman* provenientes de 405 nidos obtenidos por reproducción en condiciones controladas del núcleo de parentales mantenidos en la granja Monterrey Forestal de Pizano S.A. De estos 310 fueron incubados a 29.5 °C, 265 a 31.5 °C y 240 a 33 °C, manteniéndose constantes las variables humedad relativa (98%) y tensión de oxígeno (20.5%).

Se estableció un patrón de colección, sacrificio y fijación de los huevos que permitió obtener cinco embriones en cada uno de los estadios de desarrollo y para las cuales se conocía exactamente la edad a partir del día de postura. Para la fijación y obtención de los embriones se siguió la metodología propuesta por Ferguson (1985).

Una vez cada embrión fue separado de sus membranas extraembrionarias se procedió a tipificar su estadio de desarrollo, para posteriormente tomar 14 dimensiones morfométricas: Longitud total, Longitud de la cola, Longitud cefálica, Longitud cefálica 1, Longitud del tronco, Longitud del brazo,

longitud del antebrazo, longitud del muslo, Longitud de la pierna, Longitud del pie, Distancia axila- ingle, Longitud ojo- narina, Diámetro orbital, Altura cefálica y Anchura máxima de la constricción interorbital.

**Periodo de Incubación:** El periodo de incubación de *Caiman crocodilus fuscus* varió con la temperatura; así, tomó 85 días cuando el proceso se realizó a 29.5 °C, 75 días a 31.5 y 70 días a 33.0 °C.

No obstante lo anterior se presentan algunas variaciones en cuanto la duración total del periodo entre 4 y 6 días.

El incremento continuo en la temperatura de incubación, no produce un descenso lineal en la duración del periodo de incubación. Así, el incremento de 29, 5 a 31,5 disminuye el periodo en 10 días, mientras que el incremento de 31.5 a 33 solo lo reduce en 5 días.

**Cronología del desarrollo:** para la caracterización de los estadios embrionarios se siguieron tanto el método morfológico como el morfométrico. El primero no sigue una secuencia estrictamente continua de organogénesis por lo cual no produce relaciones respecto al tiempo de incubación, mientras que el segundo si contempla los cambios en las dimensiones del embrión durante el periodo de incubación, cuantificando la variación en forma cuantitativa y secuencial, lo que permite establecer relaciones con variables como el tiempo y la temperatura.

La relación entre los estadios morfológicos de desarrollo descritos y la edad del embrión en secuencias logradas bajo regimenes de temperatura constante : En los primeros estadios, cuando los cambios son evidentes y rápidos la relación tiende a ser lineal, no así a partir del estadio 21, cuando los cambios morfológicos son apenas perceptibles, lentos y priman las variaciones morfométricas las cuales no son tenidas en cuenta durante la descripción morfológica, pues simplemente hay un incremento rápido en el tamaño del embrión.

De otro lado, un incremento en la temperatura de incubación como también se observa no produce un descenso lineal en la duración de los estadios en particular, pasándose por alto las diferencias específicas en el desarrollo originadas por la temperatura.

Para la evaluación de las tasas de desarrollo se compararon las 19 variables morfológicas y su comportamiento a través del tiempo de incubación mediante gráficas cartesianas, con las cuales se probó el ajuste de los datos a diferentes modelos de crecimiento:

El modelo logístico de crecimiento explica el comportamiento de las variables longitud total, longitud de la cola, longitud del tronco, longitud de las extremidades, longitud axila- ingle, longitud ojo narina, longitud interorbital, longitud internarinas y máxima anchura de la cabeza.

De otro lado, el incremento de peso a través del tiempo se ajusta al modelo de Gompertz, mientras que el comportamiento de la altura cefálica y la longitud cefálica se representan con modelos potenciales. Aunque los modelos de crecimiento de los seres vivos tradicionalmente se ajustan a la ecuación de Von Bertalanffy, este no se ajusta a los datos de crecimiento obtenidos de los embriones y fetos en desarrollo y crecimiento, encontrándose básicamente la diferencia en el valor del máximo tamaño calculado del observado (388.4 vs 270.5) en el caso de la longitud total.

Un caso interesante y para el cual no fue posible calcular ningún modelo de crecimiento es el avance del diámetro ocular.

Con base en los modelos de crecimiento registrados para la variable longitud total, peso, altura cefálica y longitud cefálica se calcularon las ecuaciones que permiten predecir la edad del embrión o del feto cuando se conoce la temperatura a la cual se desarrolla el proceso de incubación.

Variable	Temperat	ECUACION	r <sup>2</sup>
Long Total	29.5	$264 / 1 + e^{\exp(-(0.08343 * t - 3.3105))}$	0.9695
Long Total	31.5	$264 / 1 + e^{\exp(-(0.12060 * t - 3.5272))}$	0.9885
Long Total	33.0	$264 / 1 + e^{\exp(-(0.12196 * t - 3.4095))}$	0.9802
Peso	29.5	$10 \exp(-1.934 + 3.3065 * (\log t)) / 1000$	0.9689
Peso	31.5	$10 \exp(-1.290 + 3.1005 * (\log t)) / 1000$	0.9931
Peso	33.0	$10 \exp(-1.143 + 3.0532 * (\log t)) / 1000$	0.9597
Alt. cefal	29.5	$10 \exp 10 \exp(0.5453 + 0.2847 * (\log(\log t)))$	0.9932
Alt cefal	31.5	$10 \exp 10 \exp(0.5579 + 0.2587 * (\log(\log t)))$	0.9584
Alt cefal	33.0	$10 \exp 10 \exp(0.5585 + 0.2561 * \log(\log t)))$	0.9446
Long cef	29.5	$e^{\exp(-0.8257 + 0.9868 * (\ln t))}$	0.9510
Long cef	31.5	$e^{\exp(-0.5777 + 0.9601 * (\ln t))}$	0.9619

Long cef      33.0       $e \exp (-0.6816 + 1.0164 * (\ln t))$       0.8591

Con base en las ecuaciones calculadas se graficò la primera derivada de la funciòn correspondiente a cada modelo como valor representativo del cambio diario de las dimensiones del embriòn.

Se alcanza la mayor tasa de incremento en la longitud total en embriones incubados a 33 °C en el día 28 de la incubaciòn (8.04 mm/día) y que corresponde al estadio morfològico 22. Dicha tasa es 0.11 y 0.45 veces mayor que la tasa calculada para embriones incubados a 31.5 y 29.5 para el mismo día de incubaciòn.

Al aumentar la temperatura de incubaciòn 3.5 grados, la tasa diaria de crecimiento longitudinal se incrementa en un 69.52%, para el estadio 22. Hasta este mismo estadio, los procesos de organogènesis va acompañados de un incremento gradual de las tasas de crecimiento las cuales en este estadio alcanzan su máximo valor, a partir de este punto cuando la organogènesis cesa y se continua una etapa de crecimiento las tasas disminuyen gradualmente hasta la eclosiòn.

En lo que respecta, se obtienen las máximas tasa de incremento en masa embrionaria en fetos incubados a 33 °C ( 0.96 g/día) a partir del día 60 de incubaciòn, dicha tasa es 0.13 y 0.57 veces mayor que la tasa de incremento observada en embriones incubados a 31.5 y 29.5 respectivamente, manteniéndose la proporcionalidad hasta el final del desarrollo

Las tasas diarias de incremento de la variable altura cefálica , son mayores durante los primeros 10 días de incubaciòn. En embriones incubados a 33 °C la tasa diaria de incremento es 0.026 y 0.29 veces mayor que a 31.5 y 29.5 respectivamente.

En lo que respecta a la tasa de crecimiento longitudinal de la cabeza se aprecia un leve aumento pero continuo con valores máximos al final del desarrollo (70 días) en embriones incubados a 33 °C. La tasa diaria de cambio de la longitud cefálica es 0.87 y 0.23 veces mayor en fetos incubados a 33 °C que en individuos incubados a 31.5 y 29.5.

Se puede concluir entonces que los embriones de *Caiman crocodilus fuscus*

incrementan su masa y su longitud día tras día a diferentes tasas según la temperatura. A altas temperaturas las tasas diarias de incremento son mayores durante todo el periodo embrionario, a temperaturas medias (31.5) dichas tasas son mayores que las registradas a baja temperatura.

Finalmente indicar la edad de un embrión en campo, resulta fácil con base en las ecuaciones desarrolladas, siempre y cuando se conozca la temperatura de incubación, lo que resulta mas fácil que el realizar un examen detallado de la morfología embrionaria.

Temperature Dependent Sex Determination in St Lucia Nile  
Crocodiles in Natal, South Africa.

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**Abstract**

The factors controlling sexual differentiation in crocodilians are unknown, but heteromorphic sex chromosomes are absent from all species. The sex of *Crocodylus niloticus* was shown to be determined by the temperature of egg incubation in constant temperature and in shift temperature laboratory experiments. Temperature shifts from 32.0°C to 33.0°C and visa versa were conducted at varying times during the incubation period in order to define the thermosensitive period (TSP). The duration of the incubation period varied with temperature and was 74 days at 33°C, increasing to 87 days at 31.0°C. Preliminary results indicate that the TSD pattern is female-male-female; females are produced at low and high incubation temperatures, while males are produced at intermediate temperatures.

The shading effect of *Chromolaena odorata*, an alien plant found to be invading crocodile nesting sites in the Lake St Lucia area, may reduce nest temperatures thereby altering the sex ratio of crocodile hatchlings entering the lake ecosystem.

**Introduction**

Lake St Lucia forms part of the Greater St Lucia Wetland Park which is located in the north eastern corner of South Africa. The lake body covers an area of 350km<sup>2</sup> and comprises approximately 80% of the estuarine area of Natal Province (Begg, 1978). It has been described as being the largest estuarine system in Africa. This dynamic and fascinating system is home to approximately 1500 crocodiles of various size classes.

Temperature dependent sex determination (TSD) is a phenomenon



nearly restricted to certain taxonomic groups of reptiles (Bull, 1980 and 1983; Ewert and Nelson, 1991; Janzen and Paukstris, 1991). However, the majority of animals possess genotypic sex determination in which the sex of the individual is determined at or before conception. During TSD the sex of the individual is determined by the temperatures experienced during embryonic development. Other environmental factors may also affect sex determination and for this reason the phenomenon may also be called environmental sex determination or ESD. Throughout this paper we use the term TSD.

Among crocodilians, the universal absence of heteromorphic chromosomes (Cohen and Gans, 1970) points to the likelihood that all living crocodilia exhibit TSD (Ferguson, 1985). Lang and Andrews (1994) state that at the present time, half of the 22 species of extant crocodilians show evidence of TSD. However, the TSD patterns in a few studied representatives differ substantially and therefore Additional species have to be studied before valid generalizations may emerge. The presence of TSD in reptiles has important implications for their sex ratios, habitat requirements and reproductive success. From a management perspective the relevant information for conservation and/or utilization programmes must therefore be species specific.

Since the discovery of this sex determining mechanism by Charnier in 1966, the effort of most researchers has been directed toward laboratory studies of TSD with relatively little attention being paid to the operation of TSD in nature. This study however, was designed to include both laboratory and field experiments.

#### Methods and Materials

##### a) Laboratory experiment:

During the 1994/95 and the 1995/96 nesting seasons, four clutches of wild crocodile eggs were collected from three different nesting areas in the Lake St Lucia area. The eggs were randomly placed onto one of seven styrofoam incubators which were maintained at the following temperatures: 31.0°C, 31.5°C, 32.0°C, 32.5°C, 33.0°C, 34.0°C and 35.0°C. An "open" method of egg incubation was used. In the 1994/1995 nesting season a shift experiment was carried out with an additional 40 eggs in order to determine the thermosensitive period or TSP, which is the time phase during incubation that the sex of the individual is determined. For this experiment, at various stages during incubation, 10 eggs at any one time were shifted from a higher to a lower temperature and visa versa.

#### b) Field Experiment:

In 1994 we hypothesized that an alien plant, namely *Chromolaena odorata* (commonly known as parrafen bush) was negatively impacting breeding success by: 1) invading crocodile nesting areas and preventing nesting and 2) reducing incubation temperatures by creating shade. The alien plants invasion of nesting sites is a particular problem due to the fact that crocodilians have been shown to demonstrate nest site fidelity. In order to test this hypothesis, during August and September of the same year we cleared experimental areas on the banks of the Mpate river, one of the nesting habitats on the western shores of the lake. Some areas were previously utilized nesting sites that had since been invaded by *C. odorata*. We also created a number of totally new experimental sites within the dense stands of *C. odorata*.

We established two thermal and moisture transects, 12m in length, perpendicular to the river bank. One transect was in full sun and the other in full shade, shaded primarily by *C. odorata*. Thermocouples connected to a CR10 data logger, which was buried and powered by a solar panel, recorded soil temperatures on the hour at 4 depths at the 8 transect stations for the duration of the incubation period. Soil tensiometers measured soil tension at average nest depth at all stations.

To complete the entire picture, it was of vital importance to collect data on incubation temperatures in wild crocodile nests. "Hobo" thermal recorders were placed within a number of wild nests, programmed to record temperatures every one hour and twenty minutes throughout the incubation period.

#### Results and Discussion:

##### a) Laboratory experiment:

Hatching commenced at 74 days of incubation at the warmer temperatures and continued for an additional 10 days for both nesting seasons. We attempted to sex the individuals macroscopically at 3 months of age and then again at 6 months and 8 months of age according to criteria described by Hutton, 1987. However, we had difficulties doing this. We developed a technique for sexing the hatchlings using a laparoscope. Although time consuming this technique is an important one as most studies to date have had to sacrifice the hatchlings in order to verify their sex. Blood samples have been taken from the 1994 hatchlings and in collaboration with Dr. Val Lance we will be looking at hormone concentrations in order to verify their sexes. Preliminary results indicate a pivotal temperature between 31.5°C and 32.0°C. Although lower than the 32.5°C pivotal temperature for Zimbabwean

Nile crocodiles, it is not surprising due to the fact that Lake St Lucia is at the southern most extent of the Nile crocodiles breeding range and they would therefore be more cold adapted in this region.

b) Field experiment:

When comparing daily mean temperatures between the sunny versus the shaded sites at a depth of 25cm (average crocodile nest depth) we find that soil temperatures in the shaded sites are on average 5-6°C cooler than those in the sunny sites (Figure 1).

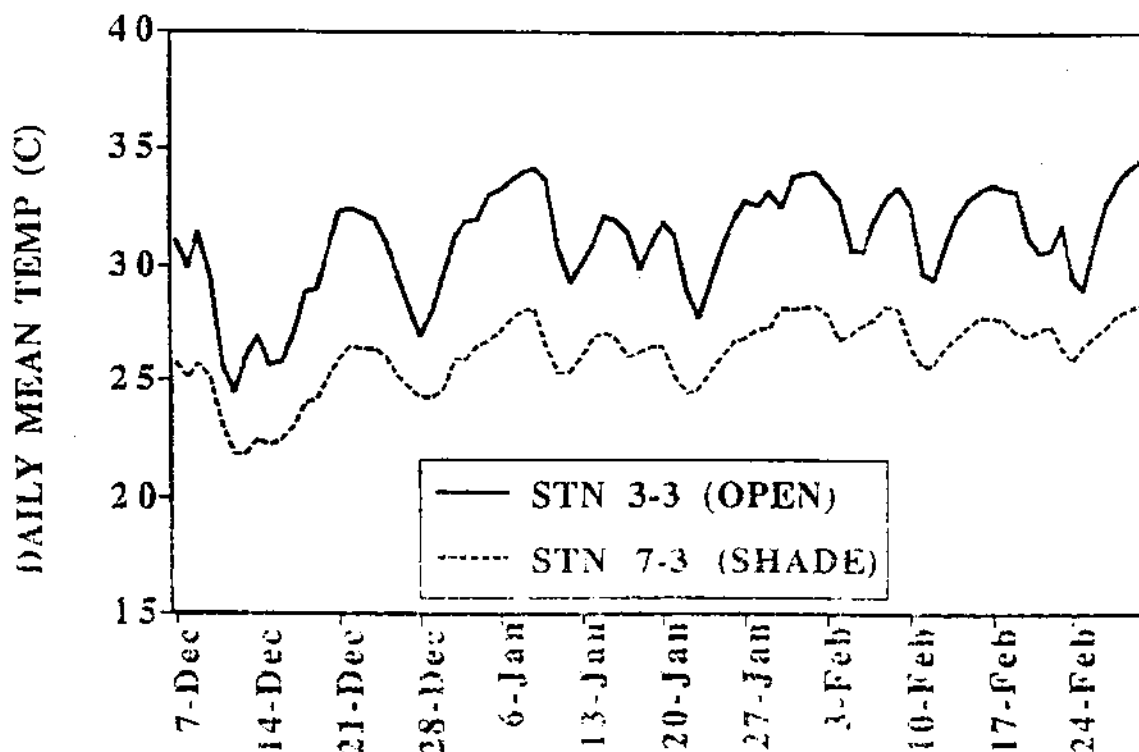


Figure 1: 25cm daily mean temperatures - Mpate River

The 32.5°C dotted line seen in figure 1, represents the predicted pivotal temperature for Nile crocodiles based on research by Hutton (1987). Hutton's pivotal temperature was used because the results from both seasons of this TSD experiment have not yet been verified. Therefore even if we consider metabolic heat production within the nest, shaded site temperatures may still be too low for

the development of Nile crocodile embryos. Lang and Andrews (1994) found that at 28°C most American alligator eggs complete development but fail to hatch; Chen (1990) found that survivorship of Chinese alligator eggs was poor at temperatures below 27°C and according to Magnusson et. al. (1990) survivorship of Dwarf caiman eggs was also reduced below 27°C. Hutton (1987) states that Nile crocodile eggs incubated at 27°C never hatch. This alien plant is therefore not only altering the sex ratio of partly shaded nests, but it may very well prevent embryonic development in a totally shaded or partially shaded site.

We obtained some very valuable incubation temperature data. Figure 2 shows data from a nesting site on the eastern shores of lake St Lucia. Control temperatures were recorded at the same nest site, approximately 0.5m from the eggs themselves and at the average nest depth.

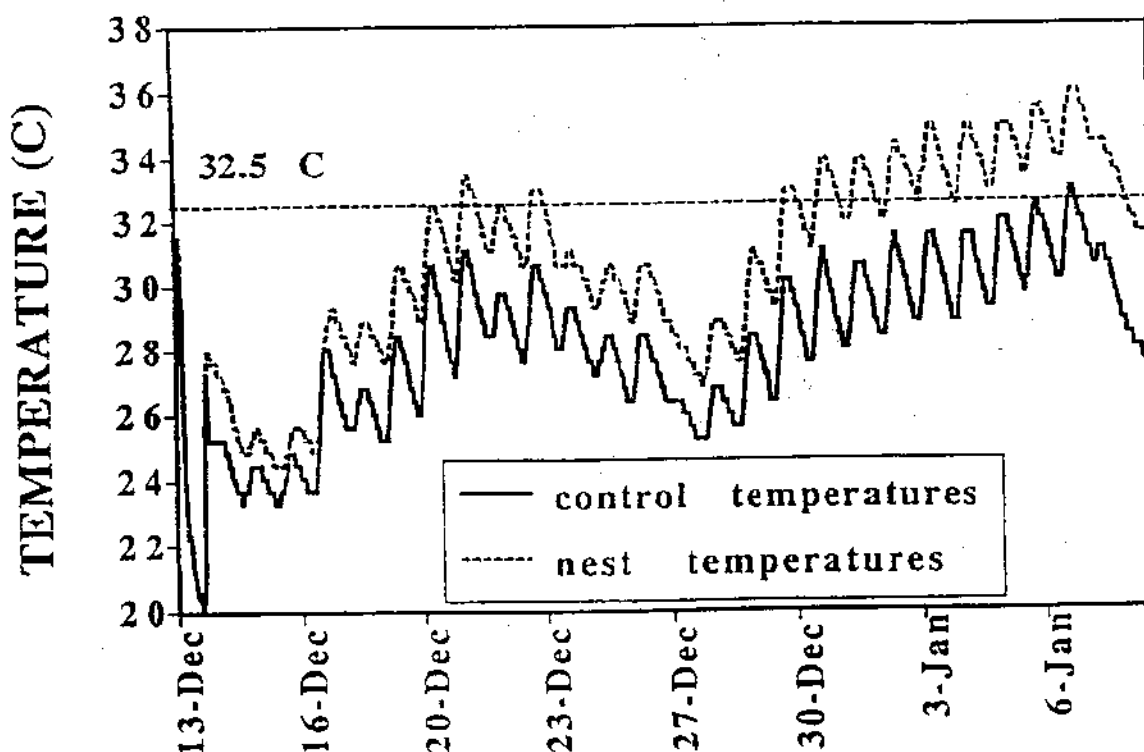


Figure 2: Nile crocodile nest vs control temperatures - Nkazana Stream

The upper line on Figure 2 represents metabolic heat production within the nest which ranges from 1 - 2° C. As development continues, metabolic heat production increases. The predicted thermosensitive period for the Nile crocodile, based on work by Hutton (1987), is between day 30 and day 50. It is approximately around this time that metabolic heat production increases and incubation temperatures reach the predicted pivotal temperature of 32.5°C.

When comparing results from nesting sites on both the eastern and the western shores of the lake - we found a substantial difference in soil temperature between the two areas (Figure 3). These temperatures were all measured in the sun transects. The Mpate river area is characterized by reeds, shrubs, high sand banks and it is generally a more open area. Nkazana stream, on the other hand, is a 1km long woody riverine area bordering a grassland. We would thus expect, and as the graph in Figure 3 confirms, soil temperatures at the average nest depth in the Mpate area to be higher than those recorded in the Nkazana stream area. Figure 3 shows a comparison in soil temperatures in both the two nesting areas.

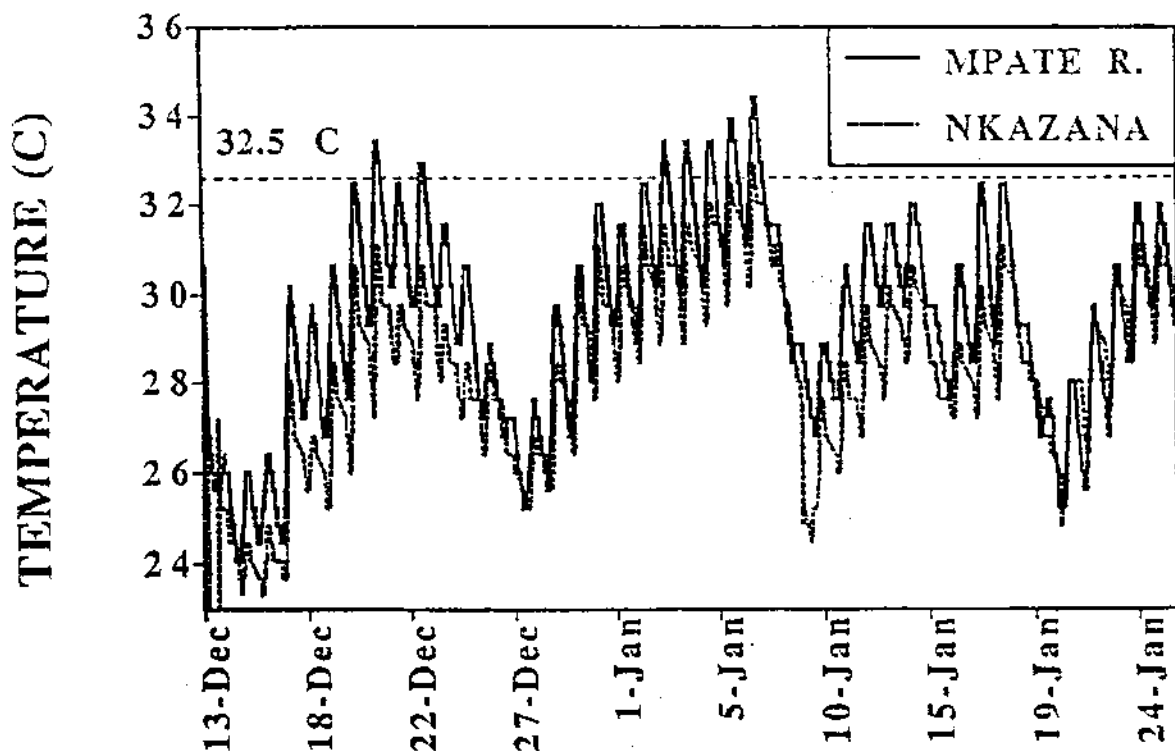


Figure 3: Nesting area soil temperatures - Nkazana Stream vs Mpate River

If we add the predicted 2.0°C metabolic heat production to the sun transect temperatures (as seen in figure 3) in the Mpate area, the 25cm depth temperatures are more centered around the 32.5°C pivotal temperature and actually go a little higher into the primarily male producing temperatures. Bear in mind that this finding excludes the influence of *C. odorata*, which is not true for the 25cm depth shade transect profiles. When metabolic heating is added to the Nkazana stream temperatures, incubation temperatures approach the pivotal temperature, resulting in a balanced sex ratio. This nesting area is still unaffected by *C. odorata*.

#### Conclusions:

1. Nesting areas of Nile crocodiles in the Lake St Lucia ecosystem are thermally marginal for the production of males.
2. Without the combination of full sun and metabolic heating it is unlikely that croc nests will ever achieve enough heat to produce male crocodiles
3. Continued alteration of nesting habitat by *C. odorata* in the Lake St Lucia system, will result in a female - biased sex ratio and eventual extinction of the species.

In Late May we once again attempted to macroscopically sex my yearlings. Preliminary results show that at 31.0°C and 31.5°C we obtained 100% female hatchlings; at 32.0°C over 80% male hatchlings were produced and at 33.0°C, we obtained 100% male hatchlings. However, even with the possibility of a lower pivotal temperature for Nile crocodiles in the Lake St Lucia region, *C. odorata* is still encroaching upon nesting sites and in so doing affecting the hatchling success and sex ratio balance of hatchlings.

#### Acknowledgements:

A special thanks to Drexel University, the Natal Parks Board, Earthwatch, Inc. and numerous others who have helped make this project a success.

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## A PRACTICAL METHOD FOR SEXING NEONATES CROCODILIANS

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**INTRODUCTION.** Like many of reptiles, crocodilians do not have external sexual organs neither visible sexual characteristics, even when the hatchlings are neonates. In the cloacal cavity three systems crowds, the digestive, the urinary and the reproductive. Also two musk glands are found inside. The common techniques for sexing are based on the appreciation by touch or at sight of the male sexual organ called penis. The female of course is lack of this, however it posses an erectil structure known as clitoris but smaller in size and with remarkable differences in its color and structure. During the males life, the penis will be loccated inside of the cloacal cavity and only will be exposed in the natural way during mating, or when the position and applied management over the animal makes that the visceras make pressure over the penis exposing it. In case of cloacal tissues inflammation, the penis will be exposed as well.

In order of knowing the sex of the crocodilians at birth, several methods has been employed:

- A) Euthanasized animals to identify their gonads.
- B) Abdominal pressure and dorso-ventral flexion of the lumbosacral region.
- C) Cloacal digital inspection.
- D) Cloacal endoscopy using the otoscopy.

The objective of this paper is to present a practical and economic technique for sexing crocodilian neonates by using the rinoscope.

**MATERIAL AND METHOD.** The employed rinoscope (Medicon Germany) number 25 stainless steel sizes 13.5 cm, the inserting edge measures 2.5 mm wide, 6 mm length without applying pressure. It can be inserted to a depth 25 mm inside the cloaca and opened to 30 mm. The technique for sexing by the use of the rinoscopy requires that the hatchling can be placed in dorsal recumbency and introducing kindly the inspection edge of it. Pressure is applied over the forceps and the cloaca opened laterally. If the crocodilians is a male will be observed the penis with a characteristic shape and a darker color than the rest of the cloacal epithelium. Generally the stimulation with the rinoscope produces the urine excretion through the penis.

**RESULTS** With this instrument were sexed 7.0.0. *Caiman crocodilus fuscus*, 31.0.0. *Crocodylus acutus* and 73.0.0. *C. moreletii* obtained by artificial incubation at Miguel Alvarez del Toro Zoo. Also have been sexed 57.7.0. mexican crocodilians of the three species captured in the wild.

**DISCUSSION.** Since two years ago the rinoscope has been employed with very good results at Miguel Alvarez del Toro Zoo, since the crocodilians could be sexed few days after hatch, all were males and we could restructure our incubation techniques. Our project is focused to the crocodilians reintroduction and obtaining females is our principal goal now. The euthanasia technique for gonads identification is out of practical objectives of breeding centers, zoos and conservation sites. The abdominal pressure and the dorso- ventral flexion of the lumbosacral region requires that the crocodilians be bigger than 600 mm and their body condition be adecuate so that the penis be exteriorized; if the animal is thinner, the internal pressure will not be enough. The



cloacal digital inspection is an accurate method but is needed that whom makes the inspection possess a thin finger able to be introduced through the cloacal vent without injuring the tissues. By introducing the otoscopy through the cloacal vent it can stimulate the liquid secretion that make difficult the sight through the instrument cone.

**CONCLUSION** The use of the rinoscopy is recommended for sexing neonates crocodilians with no error margin .

# Grow rates of *Caiman latirostris* under two different diets

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## Introduction

Breeding animals with the aim of production, feeding is one of the priorities to be taken into account to their optimum development. Monitoring and recovering program comes from an arrangement between INTA\MAGIC\MUPCN, and its aim is to determine if the Ranching technique (CITES Res. Conf. 3.15), used successfully in other countries, is adapted to our species conditions and environmental situation.

Broad snouted caiman (*C. latirostris*), is one of the two species that lives in Argentina, it has a wide distribution in the North of the country, covering almost Buenos Aires province (YANOSKY, 1990).

There is a lot of bibliography about crocodilians diets (ALVAREZ del TORO, 1974; JOANEN *et al.*, 1988; KERCHEVAL, 1990; LARRIERA y AGUINAGA, 1990; McNEASE *et al.*, 1983; MORPURGO *et al.*, 1990; POLANCO, 1991; SEIJAS y RAMOS, 1980; VERDADE *et al.*, 1990; VERDI *et al.*, 1980; WILDHOZER *et al.*, 1986), where costs, quality, availability in market, and the possibility of stocking are examined although we don't have many papers referred to *C. latirostris* (LARRIERA y AGUINAGA, 1990; VERDADE *et al.*, 1990).

ALVAREZ del TORO (1974) and VERDI (1980) have determined that in the wild *C. crocodilus fuscus* diet for animals shorter than a meter is:

Insects:	67.7%	Crustaceans:	2.9%
Arachnids:	5.8%	Fish:	1.4%
Amphibious:	14.5%	Reptiles:	1.4%

Little mammals: 1.4%

LARRIERA and AGUINAGA (1990), published a paper where two diets for *C. latirostris*, are compared, in which those animals fed on red meat develop much more than those fed on fish.

The objective of this paper is to give information about the development of broad snouted caiman hatchlings under two different diets, comparing the grow between them.

## Materials and methods

Two treatments were made from different diets with animals coming from 23 nests with a total of 570 animals (288 fed on chicken and 282 fed on fish). The basis of one of the diets consisted in chicken, the other was based on "sábalo" (*Prochilodus lineatus*), both with sawdust and a vitaminic-mineral complex.

The characteristics of each diet were:

		Chicken	Fish
Humidity (Heater 105-110°C)	%	65.1	60.1
Ethereal extract (Fat)	%•	22.8	32.5
Total protein	%	17.3	19.2
Total fiber	%•	8	5.3
Ashes	%•	12	12.1
Phosphorus (expressed as P)	%•	0.8	1
Calcium (expressed as Ca)	%•	3.6	3
•Dried weight			

The experiment started in May 1995, when animals were distributed in six pools. During 15 days they were fed on a mixture of the two diets, with the intention that both treatments change the food composition. Animals were fed three times a week *ad libitum*, from 8 AM to midday, when leftovers were taken away. Animals were kept in concrete pools, which were 50% filled, and where they were fed. During the cold season the environment was heated with a combustion turbine of 100,000 BTU.

Animals were weighed and measured individually at the beginning and the end of the experiment (after six months). They were weighed with an analytical balance of 1 gr., and measured with a 1 mm precision meter. Data were analyzed through Kruskal-Wallis test (SOKAL and ROHLF 1979).

## Results

Table N°1: Animals growing in average weight during the experiment

Days	Chicken	Fish
1	82.51	97.63
49	108.83	103.7
172	159.5	143.96

Table N°2: Average length of animals at the beginnings and at the end of the experiment

Day	Chicken	Fish
1	28.18	29.67
172	37.63	36.79

Table N°3: Cruskal-Wallis test results

Beginning		H	H Vs $\chi^2$
	Weight	75.28	>*
	Length	59.26	>*
End			
	Weight	102.3	>*
	Length	4.99	>

Value of  $\chi^2$  (0.05; 1) = 3.841

\* Significant values up to  $\alpha = 0.005$

Table N°4: Maximum and minimum values recorded on both treatments

	Maximum		Minimum	
	Chicken	Fish	Chicken	Fish
Length	32.7	34.1	23.2	24.1
Weight	127	157	38	58

### Discussion and conclusions

Our results are different from the MORPURGO *et al.*'s (1990) results, working with Nile crocodile, where they tested 3 diets, fish (live and dead), red meat and chicken breeding. Animals preferred fish, without significant differences in growing among the 3 treatments. In this paper, we have found significant differences as regard growing, as much in length as in weight (Table N°: 1, 2 and 3). Animals from chicken treatment were heavier and longer than those fed on fish (159.5 gr Vs 143.96 gr and 37.63 Vs 36.79 average values) (Tables N°: 1 and 2). These differences can be attributed to the fact that animals preferred chicken. In the pools where animals were fed on fish, the food that remained was larger than in others pools. Although it is supposed that the diet based on fish (pag. 2) has a better nourishing quality, those animals fed on chicken grew much more.

Though animals fed on chicken grew much more, red meat seems to be better for animals growing. GARNETT *et al.*, (1986); McNEASE *et al.*, (1983); LARRIERA and AGUINAGA, (1990) papers agree that animals fed on red meat have a better development than those fed on fish, not because of the amount of food that they ate (dry weight) but for the conversion rate is lower (GARNETT *et al.*, 1986).

LARRIERA and AGUINAGA (1990), reached an average daily growth in weight on fish diet about 0.21 gr, our treatment based on chicken reported 0.06 cm/day growing and 0.45 gr daily increase. LARRIERA and AGUINAGA (*op. cit.*) benefited notoriously from the diet based on red meat (0.80 gr/day); these results are only useful to show us different growing rates since you can't compare them because they worked with the animals in a different way. Although red meat diet is more expensive it is more profitable from the growing point of view, which coincide with McNEASE and JOANEN, (1983) and GARNETT *et al.*, (1986) results.

When PACHON was working with animals fed on fish he found that the *C. Crocodilus* growing curve agreed with the equation:  $\ln y = 3.11 + 0.057x$ , where  $y$ =length in cm and  $x$ =time in months.

LIEBERMAN and HILDEBRAND (1979) when feeding animals on *Iguana iguana* and *Hydrochaeris hydrochaeris* they found that the growing of young *Caiman crocodilus* was equivalent to the equation  $\ln y = 3.15 + 0.60x$ . Fifty animals bred in Monterey Farm, fed on inlaid made of bones and meat fish, cattle visor, red meat and vitaminic complexes, allowed to establish as a developing pattern the following equation  $\ln y = 3.15 + 0.068x$ , RODRIGUEZ (1988). According to this equation young animals show an increment of about 0.66mm/day, almost the same we reached in this work with animals fed on chicken (0.6 mm/day).

McNEASE *et al.* (1983), report a difference of 20% in weight and 3% in length, in this work the difference in weight isn't too big, just 11%, however the difference in length is approximately the same as the above mentioned 2.3%, what suggests the same length grow rate in animals fed on chicken and red meat regarding those fed on fish. LARRIERA and AGUINAGA (1990), report a difference of 49% (those fed on red meat grew much more) between red meat diet and fish diet, that make us suppose, that a diet based on chicken and red meat could be better to one based on chicken only. When we observe the big range in weight and length of the two treatments (Table N°. 4) we could deduce that we are far away to reach the optimum potential rate of *Caiman latirostris*. RODRIGUEZ (1988), report the same after finding that the growing equation of *Caiman crocodilus fuscus* in the wild is  $Ln y = 3.15 + 0.12x$  and in captivity is  $Ln y = 3.15 + 0.064x$ , being  $y$  = length in cm, and  $x$  = time in months. PIÑA (*in press*), quotes that the growth of *C. latirostris*, fed on chicken, bran and a mineral-vitaminic complex is  $Ln y = 3.771 + 0.0061x$  in length and  $Ln y = 3.168 + 0.002x$  in weight, being  $x$  = time in days and " $y$ " is gr. or cm.

The regression (weight and length) values of the two treatment show a good relation between them, the  $R^2$  value for animals fed on fish was 92.1%, and the other 93% (Fig.: 1 and 2).

Survival during the experiment was similar in both treatments, 94.4% and 97.16% for the animals fed on chicken and fish respectively.

## Figures

Figure 1: Regresion values and dotted line for chicken treatment.

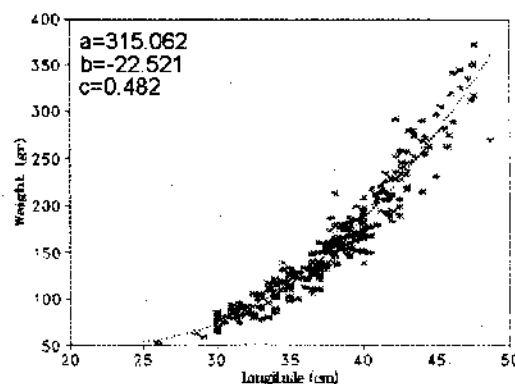
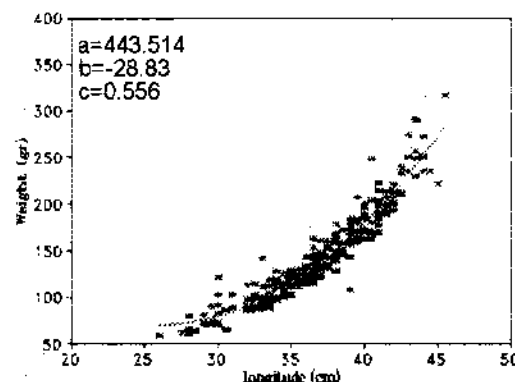


Figure 2: Regresion values and dotted line for fish treatment



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**Abnormalities in Eggs, Embryos, and Juvenile  
Farmed Estuarine Crocodiles, *Crocodylus porosus*.**

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Poster paper presented at:

The 13th Working Meeting of the IUCN Crocodile Specialist Group,  
Santa Fe, Argentina, 13-17 May 1996.

**Abstract**

A survey of abnormalities in eggs, embryos and juvenile *Crocodylus porosus* bred in captivity at a commercial crocodile farm was carried out during 1992 - 1994. A photographic record was made of the deformities found and preserved examples of some of the more interesting cases were kept for display at the farm.

Eggs with the following anomalies were observed:- soft shells; partial shell formation; no shell; additional calcareous protrusions on the external shell surface; under- or over-sized eggs; eggs of deformed shape with incomplete sealing.

Recorded abnormalities in embryos or hatchlings were:- unabsorbed yolk sac; dizygotic twins; malformation or absence of tail; kyphoscoliosis; shortened body; misaligned and/or reduced upper or lower jaw; anophthalmia; microphthalmia; exophthalmia; meningoencephalocele.



## **Aim**

To record abnormalities in eggs, embryos and juvenile estuarine crocodiles, *Crocodylus porosus* for future scientific reference.

## **Introduction**

Research into fungal disease in farmed *C. porosus* commenced in 1990 as a part-time Master of Applied Science research program (Hibberd, 1996). During 1990 and 1991 various abnormalities in eggs, embryos and hatchlings were observed in passing however no record was made of these. From 1992 to 1994 inclusive, photographs were taken of any abnormalities observed, and preserved specimens were kept as a reference collection.

## **Methods and materials**

Animals, facilities and staff support were provided by Koorana Crocodile Farm.

During breeding seasons clutches of eggs were routinely collected for artificial incubation. The condition of any abnormal clutch or individual egg was recorded. The eggs were then cleaned and incubated with apparently normal eggs. During incubation, eggs which failed to develop were opened and the contents examined. After the expected hatching date, those eggs which failed to hatch were also opened and the condition of the embryo recorded. All live hatchlings were also checked for abnormalities.

Hatchlings with only minor defects were housed with the rest of their clutch. Live hatchlings with gross malformations and little chance of survival were euthanased. Specimens to be kept for future reference were injected with and then stored in 10% formalin. These were placed on display in the farm's tourist facility.

Photographs were taken by the Author using a Canon AE1 Program camera with a variety of lenses and microscope attachments.

## **Results**

**Eggs** with the following anomalies were observed:-

soft shells; partial shell formation; no shell (ie. membrane only); additional calcareous protrusions on the external surface of the shell (ie. pimpling); under- or over-sized eggs; eggs of deformed shape with incomplete sealing (Plate 1).

**Embryos and hatchlings** with the following deformities were observed:-

- abnormally large yolk sac (Plate 2)
- dizygotic twins (Plates 3 and 4)
- malformed tails (Plates 5 to 10)

- congenital kyphoscoliosis (Plates 11 and 12)
- absence of tail (Plates 13 to 16)
- shortened body, misaligned jaws, and bilateral anophthalmia (complete absence of eyes) (Plates 17 and 18)
- microphthalmia (undersized eyes) (Plate 19)
- misaligned and/or reduced lower jaw (Plates 20 and 25)
- multiple abnormalities, no tail, reduced and/or misaligned upper jaw, anophthalmia, exophthalmia (abnormal protrusion of the eyeball), meningoencephalocele (protrusion of brain through defect in the skull), (Plates 21 to 24, and 26 to 28).

## Discussion

Twinning was reported in *C. niloticus* (Blomberg, 1979 cited in Frye, 1991). Suggested causes were sudden changes in incubation temperature, abnormal incubation temperature, and occasional period of anoxia (oxygen deprivation) (Frye, 1991). Various congenital deformities were reported in *C. niloticus* (Foggin, 1987) and were thought to be directly related to extreme incubation temperatures (Ferguson, 1985 cited in Foggin, 1987).

Birth defects in *A. mississippiensis* were recorded by Ferguson (1989) and included scoliosis, microphthalmia, anophthalmia, hydrocephalus (accumulation of cerebrospinal fluid on the brain) and incomplete twinning. Causes of the defects were breeding age of female (very old or very young), maternal diet, egg incubation at the extreme limits of the temperature range, and egg dehydration.

Kyphoscoliosis was recorded in *C. siamensis* by Frye (1991) however it was not known whether the deformities were genetic or acquired through environmental or nutritional disturbances.

A wide range of congenital anomalies in *Crocodylus* sp. (*C. siamensis*, *C. porosus*, and hybrids) were reported by Youngprapakorn *et al* (1994). These included maxillo-facial malformations, meningoencephalocele, exophthalmia, microphthalmia, anophthalmia, twinning, and eggshell abnormalities. No causes of the defects in the hatchlings were reported. However, lack of dietary calcium and/or oviduct abnormality in the laying adult was suggested as the possible causes of poor eggshell formation.

Egg, embryo and hatchling deformities in both *C. porosus* and *C. johnstoni* were observed by Webb and Manolis (1989). High incubation temperature was cited as the cause of spinal abnormalities, strongly coiled tails, skull malformations, and protruding lower jaws.

In this study, low levels of calcium in the breeding females' diets were thought to have caused the faulty egg formation reported here. Addition of calcium di-phosphate and shell grit to the meat rations of the females prior to the subsequent breeding season almost eliminated the production of malformed eggs.

Unscheduled high incubation temperatures for the 1990 and 1991 breeding seasons were assumed to be the cause of the embryo and hatchling deformities for those years. However, in subsequent years different temperature and humidity monitoring and control methods were employed with fewer fluctuations in 1992 and none in 1993 or 1994. What then caused the deformities in embryos and hatchlings for those years? Were they due to the breeders' ages? Was there a genetic factor? Was overheating or desiccation of the eggs in the nests prior to collection the cause? Since not all eggs from individual clutches were affected, perhaps none of these questions will give the answers. A proposed cause may be microbial contamination of the egg immediately after laying thus compromising the developing embryo and causing abnormalities as depicted in the following plates.

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### Acknowledgments

This survey of abnormalities in farmed crocodiles was carried out in conjunction with a Master of Applied Science research program by the Author at Central Queensland University. The work had the approval of the CQU Animal Experimentation Ethics Committee, and operated under the relevant Queensland National Parks and Wildlife Service Permits. The survey was jointly sponsored by the Biology Department at

CQU and Koorana Crocodile Farm and was supported by a CQU Research Grant in 1994. Funding for attendance at the 13th Working Meeting of the CSG was provided by the Biology Department, the Faculty of Applied Science, and the Pro-Vice Chancellor Research, all of CQU, and Koorana Crocodile Farm. Thanks are extended to the CSG for providing student accommodation for the duration of the meeting.

**List of photographs used on poster.**

**Plate 1** Infertile malformed eggs, incompletely sealed and leaking albumen, highly susceptible to microbial attack.

**Plate 2** Abnormally large yolk sacs in both hatchlings, anophthalmia (complete absence of eyes) and meningoencephalocele (protrusion of brain through defect in the skull) in the hatchling to the left.

**Plate 3** Dizygotic twins at hatching (separate yolk sacs).

**Plate 4** Dizygotic twins from Plate 3 shortly after hatching, the larger survived for three months, the smaller survived for ten days.

**Plate 5** Permanently coiled tail, live hatchling.

**Plate 6** Permanently curled tail, live hatchling.

**Plate 7** Permanently curled tail, live hatchling.

**Plate 8** Permanently kinked tail, live hatchling (scale in cm).

**Plate 9** Shortened and reduced thickness tail, at hatching.

**Plate 10** Hatchling from Plate 9, 2 months later with normal hatchling.

**Plate 11** Congenital kyphoscoliosis (curvature of the spine with accompanying hump), live hatchling.

**Plate 12** Hatchling from Plate 11, several months later.

**Plate 13** Absence of tail.

**Plate 14** Absence of tail, live hatch.

**Plate 15** Absence of tail.

**Plate 16** Absence of tail, ventral view of hatchling in Plate 15.

- Plate 17** Shortened body, misaligned jaws, and bilateral anophthalmia, live hatch.
- Plate 18** Hatchling from Plate 17, survived for 5 months.
- Plate 19** Microphthalmia (undersized eyes) and shortened lower jaw, live hatch.
- Plate 20** Reduced lower jaw (scale in mm).
- Plate 21** Multiple abnormalities, no tail, reduced and misaligned upper jaw, anophthalmia, meningoencephalocele (scale in cm).
- Plate 22** Close-up of hatchling from Plate 21.
- Plate 23** Ventral view of hatchling from Plate 21 and 22.
- Plate 24** Multiple abnormalities, reduced upper jaw, anophthalmia, meningoencephalocele, abnormally large yolk sac.
- Plate 25** Misaligned and reduced lower jaw, exophthalmia (abnormal protrusion of the eyeball).
- Plate 26** Incompletely sealed skull (scale in cm).
- Plate 27** Multiple abnormalities, reduced and misaligned upper jaw, anophthalmia and exophthalmia, meningoencephalocele (scale in cm).
- Plate 28** Close-up of hatchling from Plate 27, left and right views.

## Winter sores, a dermatitis in farmed Nil Crocodiles kept at suboptimal temperatures

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### Introduction

Although they are poikilothermic, crocodiles try to maintain a constant core temperature of approximately +32°C. They achieve this by thermoregulatory behaviour making use of temperature gradients in their habitat. In the wild the components of these temperature gradients are sunshine and shade, warm and cold air, warm and cold substrates, warm and cold water as well as burrows. From these the crocodiles can choose the appropriate spot where to warm up, to cool down or to maintain the achieved temperature. It is believed that blood circulation from the skin to the internal organs is used to enhance the flow of thermal energy to the core and that the flow of blood to the skin is reduced when the crocodile is trying to conserve heat.

Farmed crocodiles are either kept at a near-constant temperature of around +32°C or are exposed to temperature fluctuations in an environment with limited scope for behavioural thermoregulation. In the latter case they are kept either in outdoor enclosures with shallow water and little cover or in a poorly insulated building with widely fluctuating temperatures.

The smaller the crocodile, the poorer is its surface to mass ratio. Therefore, smaller or younger crocodiles are more prone to be affected by adverse thermic conditions than adult ones.

On most farms the hatchlings are pampered. Thus it is the juveniles, which also require more space and which are believed to be quite hardy, that are likely to be kept under less than ideal conditions and consequently to suffer from temperature problems.

### Winter sores

On several farms where juvenile Nile crocodiles were kept under such adverse conditions, between 10 and 50% of the animals developed a dermatitis towards the end of winter. This dermatitis affected mainly the ventral skin and was characterized by crusty lesions between the scales which were eating into the scales. The crusts were cream to light brown in colour. Because of the brownish colour of the crusts the farmers usually suspected them to be pox lesions.

Histopathologically there was a destruction of the epidermis, the crusts being formed from necrotic cells and many bacteria. There was an accumulation of round cells in the dermis. Inclusion bodies were never seen in the epidermis cells.

Bacterial culture produced a very large variety of nonpathogenic, potentially pathogenic and known pathogenic bacteria, typically in mixed culture from single specimens.

After correction of the thermic environmental conditions the lesions healed slowly without the need to resort to an antibacterial treatment. However, there was a tendency for scars to remain visible.

### Discussion

Under crowded conditions which are typical for all crocodile farms the ventral skin is often subjected to minor wounds such as scratches or small bite wounds. Under the same crowded conditions there is also a likelihood of a massive bacterial build-up. These bacteria contaminate and try to

infect the minor wounds.

At constant warm temperatures the defenses of crocodiles easily overcome such infections, but when in cold or repeatedly cold (fluctuating) conditions the blood supply to the skin is chronically reduced and the immune system is thermically compromised as well, the bacteria can gain a foothold and chronic, scarring lesions result. However, the crusts on these lesions are much lighter in colour and larger or more confluent than those found in pox in Nile crocodiles (Huchzermeyer, Huchzermeyer & Putterill, 1991).

As this condition is typically seen at the end of winter on farms with poor temperature control, the term "winter sores" was chosen for this type of dermatitis.

Even in well insulated and fully environmentally controlled houses a thermogradient can be offered to the crocodiles by heating the air to +34C or even +35C and bringing in the water at a lower temperature of between +25C and +30C.

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**Poliartritis asociada a micoplasma en cocodrilos de criadero (*Crocodylus nilocotus*).  
Se transmite verticalmente?**

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**Resume:..**

Por primera vez hemos informado acerca de brotes de poliartritis asociada a micoplasma en cocodrilos en cautiverio de diversos criaderos en zimbabwe. Reprodujimos la enfermedad experimentalmente en cocodrilos de 1 a 3 años. Sin embargo la fuente y modo natural de transmision no pudieron ser identificados (Mohan et.al., 1995). Al principio, los criadores pensaron que la fuente de la enfermedad eran las aves de corral y los menudos con que se alimentaba a los cocodrilos; pero en los cultivos no parecia estar entre los micoplasmas conocidos de las aves de corral y los menudos. Una caracterizacion mas amplia de los animales aislados en el instituto para microbiologia de la escuela veterinaria de hanover ha confirmado que los mismos pertenecen a una nueva especie (Kirchhoff,1995). Y lo que es aun mas importante, recientemente se han registrado brotes similares en Israel. Los cultivos portaban similitudes en los fenotipos con las cadenas de Zimbabwe (Levisohn, 1995).

Para poder identificar el modo natural de transmision, intentamos la transmision experimental de la enfermedad a traves un contacto cercano entre los cocodrilos infectados experimentalmente (de 1 a 3 años) y los aparentemente sanos (de 1 a 3 años). Nuestros intentos no tuvieron exito, a pesar de que tanto a los cocodrilos sanos como a los enfermos se los mantuvo durante 12 semanas en una misma pileta, la posibilidad de que esta enfermedad se transmita verticalmente como la infeccion *M.gallisepticum* y *meleagridis* en aves de corral se discutieron en esta presentacion.



Is *Mycoplasma*-associated Polyarthrititis in Farmed Crocodiles (*Crocodylus niloticus*) a vertically Transmitted Disease?

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Introduction

Outbreaks of polyarthrititis from several crocodile farms affecting only the rearing stock with *Mycoplasma* sp. as its etiology were reported for the first time from Zimbabwe. The disease was later experimentally reproduced in crocodiles of the same age groups (Mohan et al., 1995). The source of infection and natural mode of transmission could not be identified. With a view to understand the natural mode of transmission we attempted to transmit the disease through close contact between experimentally infected and apparently healthy crocodiles. Results of this study and the possibilities that this disease might be vertically transmitted are being reported in this paper.

Materials and Methods

Details of affected farms, disease manifestations, pathology, technique of isolation and characterisation of the isolates have already been reported (Mohan et al., 1995). Six crocodiles (1-3yrs old) were selected from a particular farm which had not reported the *Mycoplasma*-associated disease. These animals were left under observation in the Crocodile Unit of the Veterinary Research Laboratory for four weeks. Nasal swabs from all the six were cultured for *Mycoplasma* and other pathogenic aerobic bacteria at intervals of two weeks, following the procedure described earlier (Mohan et al., 1995). These animals were also observed for clinical signs of arthritis or any other disease. After having been found free for *Mycoplasma* and any potentially pathogenic bacteria, four of these were infected intra-peritoneally each with 2.5ml of 48 hrs. old broth-grown culture comprising  $6 \times 10^8$  CFU/ml of a field isolate in its second subculture. The four inoculated animals and the two healthy ones shared the same especially designed rearing ponds (Foggin et al., 1988) for 16 weeks under observation. Weekly examinations for lameness and signs of arthritis were carried out. Animals which showed lameness and/or swollen joints were considered to have picked up the infection. Synovial exudate from affected joints was cultured for *Mycoplasma* and aerobic pathogenic bacteria (vide supra). Giemsa stained smears of the exudate were also examined for *Chlamydia* and *Rickettsia*. One of the infected animals with swollen joints was euthanised twelve weeks post-infection and attempts made to reisolate *Mycoplasma* from heart-blood, spleen, lungs and synovial exudate.

## Results

Neither Mycoplasma nor any potentially pathogenic bacteria could be cultured from the nasal swabs of the six animals prior to infection. Two of the infected animals developed lameness three weeks post-infection, movement of these was characterised with dragging of the limbs when made to move on the ground outside the pond water. The joints of the affected limbs gradually developed swelling, more pronounced in the rear ankles. The third one discerned lameness and swelling of right shoulder joint six weeks post-infection while the fourth infected crocodile developed similar manifestations nine weeks post-infection. Mycoplasma but no other bacteria was reisolated from the swollen joints of all the four animals. The disease manifestations gradually tapered-off but swelling in some of the affected joints persisted despite absence of lameness. The two uninfected incontacts showed no signs of lameness or any other disease throughout 16 weeks' period of observation.

When the fourth infected animal was sacrificed 14 weeks post-infection Mycoplasma could be reisolated only from the affected joints but the culture from heart-blood, spleen and lungs could yield no bacteria. The smears of the synovial exudate were also found to be negative for Chlamydia and Rickettsia.

## Discussion

Several aspects of the new disease, Mycoplasma - associated polyarthrititis have been investigated; the etiology has been confirmed and the disease experimentally reproduced (Mohan et al., 1995). Experimental trials with an alum precipitated vaccine have given encouraging results (Mohan et. al., 1996). Nevertheless, the source of infection and mode of transmission in nature remained elusive. The isolates have been characterised in detail and assigned to a new species yet to be named, ecology of which is not known (Kirchhoff, 1996). These information have brought out evidence that the disease might be native to crocodiles and the earlier notion of the farmers to incriminate the poultry and goat offals fed to the crocodiles as the source of infection lacked scientific evidence.

Knowing that any infection spreads either vertically or laterally (contact) or both, we mounted this study to establish the lateral mode of transmission by close contact. The incubation period among the experimentally infected animals ranged from 4-9 weeks. We therefore exposed the healthy with the diseased crocodiles in the ratio of 1:2 for a period of 16 weeks to ensure a sustained and intensive contact. Despite this long and sustained exposure the disease could not be transmitted. Mycoplasma remained confined to the joint sacs following infection; neither the visceral organs nor the heart blood was found to be infected. Consequently, the possibilities of dissemination of the infectious agent to outside environ, a prerequisite to lateral spread appear remote. Experimental evidence thus rules out the possibility of the disease being transmitted through close contact.

Vertical mode of transmission now remains to be investigated as the other method of spread of this disease in nature.

Wide spread prevalence of polyarthrititis in Zimbabwe and no such record in South Africa (Huchzermeyer, 1995) is difficult to explain. The farmers in Zimbabwe have emulated the management and breeding practices of the South Africans; the species being commercially exploited and the disease spectrum happens to be the same (Foggin, 1987, 1992). Nevertheless, the Zimbabwean farmers, besides raising hatchlings from their breeding stock also collect eggs from wildlife to raise hatchlings (Foggin, 1992), whereas the South Africans do not do so (Huchzermeyer, 1995). Having collected the eggs from wildlife the farmers do not use a marker to identify the hatchlings originating from wildlife and those from inbred stock. Could the eggs collected from wildlife be infected? We have already commenced investigations in this direction.

A new dimension to the epidemiology of this disease has been added with the recent report from Israel of isolation of Mycoplasma similar to the Zimbabwean strains and Chlamydia from cases of polyarthrititis in farmed crocodile (Levisohn, 1995). Chlamydia has been incriminated in South Africa and Zimbabwe in causing hepatitis in crocodiles hatchlings and yearlings (Huchzermeyer, et al. 1994).

Although we have not been able to confirm that this disease is egg-transmitted but the preliminary results of investigation suggest that this Mycoplasma infection might be transmitting in nature like some of the avian Mycoplasma sp. which are known for transovarian transmission and the possibilities that eggs collected from wildlife might be the source of its spread to commercial farms cannot be ruled out.

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**Diseases of Juvenile Farmed Estuarine Crocodiles,  
*Crocodylus porosus*.**

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The 13th Working Meeting of the IUCN Crocodile Specialist Group,  
Santa Fe, Argentina, 13-17 May 1996.

**ABSTRACT**

A summary of necropsy examinations, histological findings and bacterial and fungal isolations from 62 juvenile, farmed crocodiles (*Crocodylus porosus*) is described. Ill-thrift, anorexia, gum degeneration and skin ulcerations were the most frequently observed clinical signs. Necropsy findings included granulomatous masses in the visceral organs and fibrinous exudates associated with the pericardium and pleura. The bacterial pathogens most commonly isolated from were *Salmonella* spp. and *Providencia rettgeri*. Fungal isolates included *Fusarium* sp, *Penicillium* spp. and *Aspergillus* spp. Superficial and deep mycoses, salmonellosis, pericarditis, hepatitis and suspected stress related mortality were the most common diagnoses.

**AIMS**

- To identify the causal agents of diseases in juvenile *Crocodylus porosus* at a sub-tropical commercial crocodile farm. The information obtained would enable the farmer to modify farm husbandry methods to reduce disease morbidity and mortality, thereby increasing the numbers of healthy animals for skin and meat production.
- To add to the crocodilian disease database those causal pathogens which may have not been previously reported.

## INTRODUCTION

This applied research carries an economic significance to what is still a fledgling Australian primary industry. Major losses of juveniles due to disease cause financial problems for crocodile farmers (Lever, pers. comm). This report represents the preliminary investigation of mortalities in a high latitude crocodile farm.

In 1980 Koorana Crocodile Farm was established as a commercial enterprise near Rockhampton on the Tropic of Capricorn. It consists of 113 hectares of melaleuca and eucalyptus open woodland, mangroves, mud flats and salt pans. The farm is bordered by tidal creeks on three sides, becoming an island with causeway access at 4 metre tides. Salt water used for adult and sub-adult lakes was pumped from the creeks at high tide. Koorana is the most southerly crocodile farm in Australia and is on the southern extremity of the estuarine crocodiles' natural east coast range. Because of cold winters (range 5°C to 25°C) hatchlings and juveniles must be kept indoors in a heated environment. The farm was officially opened in 1981 with nine crocodiles. Captive breeding on the farm commenced by 1985 with some mortality of hatchlings in 1986. From then until 1990, hatchling and yearling losses continued to be a problem. The major cause of mortality and morbidity was found to be systemic mycoses, predominantly caused by the fungus *Fusarium solani*, an ubiquitous fungus normally only pathogenic to plants. Losses of hatchlings up to age one year had been in excess of fifty percent (J. Lever, pers. comm.).

This survey of diseases of juvenile farmed crocodiles was carried out during 1992, 1993, and 1994 in conjunction with a Master of Applied Science research program (Hibberd, 1996) by the primary author at Central Queensland University. Information on mycotic disease has been reported previously (Hibberd and Harrower, 1991, 1993; Hibberd, 1994a, 1994b, 1994c). A survey of abnormalities in eggs, embryos and juvenile farmed *C. porosus* is the subject of a poster at this meeting.

### Literature review

Reports of diseases of crocodilian species have been published by several authors. Foggin (1987, 1992) reported a range of bacterial, fungal, viral and parasitological infections in farmed *C. niloticus*. Huchzermeyer and Agnagna (1992) described parasites and pathology of *Osteolaemus tetraspis* which had been recently captured from the wild. Jacobson (1989) reviewed viral, bacterial and fungal diseases as well as parasitic infections of a range of crocodilian species, including American crocodiles and alligators, gharials and muggers, caiman, and several *Crocodylus* species. Frye (1991) also reviewed crocodilian diseases (viral, bacterial, fungal and others) in a comprehensive text on captive reptilian husbandry.

Ratanakorn (1993) reported the health problems of captive crocodiles in Thailand, including congenital defects, non-infectious disease (metabolic disease and toxicosis), and infectious bacterial and fungal disease. Youngprapakorn *et al* (1994) illustrated

and described a comprehensive range of diseases and abnormalities of crocodilian species (*C. porosus*, *C. siamensis* and hybrids of these two species) in their colour atlas.

Ladds and Sims (1990) investigated diseases of captive *C. porosus* and *C. novaeguineae* in Papua New Guinea. The animals had been caught in the wild as hatchlings then held locally until transported to a commercial farm. Bacterial septicaemia and coccidiosis were common as were parasitic infections. Adaptation failure was also suggested to be a contributing factor to mortality. Ladds *et al* (1995) reported the major diseases of captive *C. porosus* and *C. novaeguineae* in Irian Jaya to be coccidiosis, pentastomiasis, and bacterial pneumonia and septicaemia. Fungal pneumonia was also reported. Multiple parasitism was the major cause of mortality in the crocodiles which had been collected from the wild as hatchlings.

Melville (1993) reported parasitic, infectious (viral, bacterial, fungal) and nutritional and other diseases diagnosed in farmed *C. porosus* and *C. johnstoni* in the Northern Territory of Australia. The most commonly identified diseases were bacterial hepatitis and septicaemia, and superficial and deep mycoses.

Buenviaje *et al* (1994) investigated disease-husbandry associations in *C. porosus* and *C. johnstoni* from four farms in the Northern Territory and three farms in Queensland, Australia. Major disease findings were similar to those reported by Melville (1993). Minor diseases were probable adaptation failure and chronic infections of unknown cause. Highest mortality and disease occurrence were highest during winter months and in farms at greater latitude. Parasitic infections were reported to be relatively infrequent. Bacterial septicaemia and mycoses were becoming less common where artificial heating was provided.

A comprehensive literature survey of fungal disease in all crocodilian species was carried out by Hibberd (1996) as part of an epidemiological investigation into a long term and major fungal disease problem in *C. porosus* at a Queensland crocodile farm.

## METHODS AND MATERIALS

### Clinical history and gross examination

Sixty two moribund or dead crocodiles with age range from newly hatched to two years old were presented for post-mortem examination and disease diagnosis. Representative specimens had been selected from groups of animals exhibiting similar clinical signs. Carcasses had usually been refrigerated from the time of collection at the farm which was approximately 40 km from the laboratory. Live crocodiles were euthanased at the laboratory. Where possible carcasses were weighed and the snout-vent length measured. Year and clutch of origin were recorded from the identifying

pattern of removed tail scutes. Prior history and time of death, if known, was recorded. Any external signs of disease were noted. Observations of internal pathological changes were also recorded during the post-mortem examinations.

### **Histology**

Tissue for histological examination was fixed in 10% buffered neutral formalin, processed to paraffin embedding and sectioned at 5 $\mu$ . Sections were routinely stained with haematoxylin and eosin. Grocott's Methenamine Silver (GMS) stain was performed for the detection of fungi, and bacteria were detected using either the modified Brown and Brenn or Churukian and Schenk procedures.

### **Bacteriology**

Bacteriological cultures were incubated at 37°C on Sheep Blood Agar. Bacteria were identified using standard techniques and confirmed with the Microbact computer identification system. *Salmonella* species were cultured at 37°C on a variety of media (Blood Agar, MacConkey's Agar, Xylose Lysine Desoxycholate Agar and Tetrathionate Enrichment Broth) and were serotyped at the Institute of Medical and Veterinary Science, Adelaide.

### **Mycology**

Mycological cultures were incubated at 28°C on Sabauroud's Dextrose Agar for two weeks and identified at the Animal Research Institute, Yeerongpilly.

### **Other investigations**

Viral and parasitological infections were not investigated in this study. Biochemical and haematological studies were specifically carried out for a separate research program and results have been reported elsewhere (Hibberd, 1996).

## **RESULTS**

Sixty-two animals in the following age groups were examined:- newly hatched (3), one to six months (26), seven to twelve months (12), thirteen to eighteen months (18), nineteen to twenty four months (1) and twenty-four to thirty months (2).



## **Clinical history**

Moribund animals presented for clinical and post mortem examination had one or more of the following clinical signs:- lethargy, depression, ataxia, poor response to stimuli, partial paralysis of the limbs, body tremors, abnormal head inclination and ulceration of the gums, limbs and ventral abdominal skin.

## **Gross examination**

The most frequently observed external abnormality was ulceration of the skin between the scales of the ventral abdomen. Abrasions and ulcerations of the digits was common in more severe cases with extensive changes occurring between the leg and body wall.

Some animals had no significant gross abnormalities. The severity of internal abnormalities varied from a mild excess of turbid fluid in the pericardium and/or abdominal cavity to severely affected cases with up to ten, white caseous masses (1.5cm diameter) scattered throughout the liver, kidney, spleen, lung and other tissues. Post-mortem decomposition was common in carcasses delivered more than six hours after being found dead. The actual post-mortem interval was most likely considerably longer as most deaths occurred during the night.

## **Histological examination**

Severely affected livers with large granulomas had extensive necrosis with large numbers of branching, septate, fungal hyphae surrounded by a granulomatous response. Similar changes were found in the spleen, kidney and lung.

The least severe changes in the skin appeared between the scale margins as crusted abrasions and ulcerations. Fungal hyphae were located between layers of partially exfoliated epithelium. Severe ulceration of the gum mucosa around the teeth extended to deeper necrosis and granulomatous inflammation. Branching, septate, fungal, hyphae were common throughout the necrotic debris and the adjacent epidermal layers.

Pleuritis and a moderate to severe epicarditis was seen in some younger animals (four to six months). Fibrinous pericarditis was common in more severely affected cases.

## Bacteriology

All bacterial isolations associated with a suspected or confirmed diagnosis are listed in Table 1. Bacteria were most commonly isolated from the liver (8), lung (6) and heart (5). Bacterial isolations occurred most frequently in the age groups one to six months (18), and thirteen to eighteen months (10).

Species	No. of isolates
<i>Salmonella typhimurium</i>	6
<i>Salmonella</i> spp.	4
<i>Salmonella johannesburg</i>	1
<i>Providencia rettgeri</i>	10
<i>Edwardsiella</i> sp.	2
<i>Morganella morganii</i>	2
<i>Enterobacter agglomerans</i>	1
<i>Pasteurella multocida</i>	1
<i>Pseudomonas</i> sp.	1

Table 1: Frequency and species of bacteria isolated from juvenile crocodiles.

## Mycology

All fungal isolations associated with suspected or confirmed diagnoses are listed in Table 2. Fungi were most commonly isolated from skin (17), gum and tooth (10) and liver (10). Fungal isolates occurred most frequently in the age groups thirteen to eighteen months (25), and seven to twelve months (15). However, fungi were isolated from at least one animal in each age group from newly hatched to two year old.

Species	No. of isolates
<i>Fusarium</i> sp.	24
<i>Penicillium</i> sp.	9
<i>Paecilomyces</i> sp.	5
<i>Aspergillus niger</i>	4
<i>Aspergillus flavus</i>	2
<i>Aspergillus versicolor</i>	2
<i>Geotrichum candidum</i>	4
<i>Geotrichum</i> sp.	2

Table 2: Frequency and species of fungi isolated from juvenile crocodiles.

## Diseases diagnosed

Table 3 lists the range of conditions diagnosed in the 62 animals. Each animal may have more than one diagnosis.

Diagnosis	Number
Congenital Abnormality	5
Conjunctivitis	2
Dermatitis	20
Epicarditis	6
Fungal Granuloma	31
Hatching Abnormality	3
Hepatitis	8
Hypoglycaemia	2
Nephritis	3
Pericarditis	6
Pleurisy	2
Pleuropneumonia	1
Pneumonia	5
Polyserositis	1
Retained Yolk Sac	1
Salmonellosis	5
Septicaemia	1
Skeletal Muscle Myositis	1
Splenitis	3
Stress Related Mortality	9

Table 3: Frequency of all diagnoses made on juvenile crocodiles.

## DISCUSSION

Animal deaths usually occurred during the night. Carcasses collected during the 6 am daily inspection often showed most severe autolysis when presented for necropsy during the afternoon. Prolonged exposure to the heated environment (32°C) exacerbated the post mortem changes. Severely autolysed carcasses were unsuitable for histological or bacterial examination.

On this farm all crocodiles up to the age of two years were housed in heated, indoor pens with heated water supply. Indoor air and water temperatures were normally regulated to 32°C. Due to this heated environment, temperature stress was not a normal occurrence. However, two major episodes of high stress-related mortality/morbidity were suspected in cases where no bacterial or fungal disease could be identified. One episode corresponded to a short period of heating malfunctions in one indoor facility when air and water heaters failed concurrently. For several days

indoor air temperature dropped to overnight minima of ~26°C (external air temperature overnight minima in the range 4-13°C). Ambient temperature of unheated water at entry to the pens dropped to ~13°C. Forty animals died in the subsequent week, all without external signs of disease. Nine representative animals from that group were necropsied. A similar situation occurred to a lesser extent during and subsequent to a heat wave when indoor air temperatures reached approximately forty degrees without use of heating devices.

Temperature and its effect on disease in farmed *C. niloticus* has been demonstrated by Foggin *et al* (1988) and indicated as being a contributing factor to disease by Buenviaje *et al* (1994) and Melville (1993).

Other stresses on the animals were reduced over the survey period during concurrent fungal disease investigations (Hibberd, 1996). Changes which markedly reduced the incidence of fungal disease included:- sourcing a more consistent food supply and the regular addition of food supplements to eliminate nutritional stress; a reduction in animal numbers in each pen to reduce possible density stress; provision of additional covered retreats in each pen and regular grading of animals according to size to avoid social stresses.

Bacterial infection in eggs at the time of deposition may be one of the sources of hatchling infections, as are environmental contaminants after hatching. Washing and disinfecting eggs prior to artificial incubation has reduced the incidence of fungal infections in both embryos and hatchlings on this farm (Hibberd, 1996).

Environmental sampling of pen water, food preparation implements, storage and work areas during a period of low hygiene isolated five *Salmonella* species. Increased standards of hygiene and changes to the cleaning regime were put into effect to reduce or eliminate possible infections from these sources.

This report represents the preliminary investigation of mortalities in a high latitude crocodile farm. Further research is required to confirm the association of fungal and bacterial isolates with histological changes where the post-mortem interval is reduced.

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## ACKNOWLEDGMENTS

This survey of diseases in juvenile farmed crocodiles was carried out in conjunction with a Master of Applied Science research program by the primary author at Central Queensland University. The work had the approval of the CQU Animal Experimentation Ethics Committee and operated under the relevant Queensland National Parks and Wildlife Service Permits. The survey was jointly sponsored by the Biology Department at CQU, the Queensland Department of Primary Industries and Koorana Crocodile Farm and was supported by a CQU Research Grant in 1994. Funding for attendance at the 13th Working Meeting of the CSG was provided by the Biology Department, the Faculty of Applied Science, and the Pro-Vice Chancellor Research, all of CQU, and Koorana Crocodile Farm. Thanks are extended to the CSG for providing student accommodation for the presenting author for the duration of the meeting.

Our thanks are also extended to the staff of Koorana Crocodile Farm, the staff of Rockhampton Veterinary Laboratory (Department of Primary Industries) for their technical assistance, Grant Campbell (Department of Primary Industries, Toowoomba) for pathology examinations, the Institute of Medicine and Veterinary Science, Adelaide for *Salmonella* identification, and Dr K M Harrower for editorial advice.

## Research on Juvenile Farmed Saltwater Crocodiles

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The Queensland Department of Primary Industries (QDPI) established a research facility in Townsville in late 1993 to investigate the effects of environment and nutrition on farmed saltwater crocodiles from hatching to one year of age. Plans are currently underway to construct a second facility in which research on 'grower' crocodiles (one year to slaughter size) can be conducted. The aim of this paper is to present an abbreviated outline of major research projects carried out to date. Each year QDPI will publish a "Crocodile Research Bulletin" containing full scientific details of the research carried out at Townsville. Please contact the authors at the above address if you would like to be put on the mailing list for this publication. It should be noted that the thrust of our research relates to commercial farming applications for *C. porosus*.

### INTRODUCTION

Tours of commercial crocodile farms in the 2 main farming areas in Australia, Queensland and the Northern Territory, indicated that farming practices varied considerably in terms of environmental conditions provided for the animals, management practices, and diets used. About the only commonly adopted practices were providing 32°C water in juvenile crocodile growing tanks and use of mixtures of red meat, chicken heads and vitamin/mineral supplements for diets. Many farms have access to meat from wild animals (horse, pig, kangaroo, donkey) but some of these sources are dwindling or are becoming more expensive to buy. Industry recognises the need to develop a dry food pellet for *C. porosus* to

- reduce direct feed costs
- eliminate the need for costly freezer storage of meat
- enable precise dietary requirements of the animals to be met.

For juvenile crocodiles there were a whole range of different environmental factors being used on farms, especially relating to light, type of tank (material, design, with or without lids), water quality and depth, type of hide-board, use of radios to provide noise. Farms also employed different management strategies and feeding regimes (morning or afternoon, use of "fasting" periods), animal density per tank, and grading on size or clutch. This signified the need for quantitative research to be carried out on these issues, and results to be extended to the industry. Consequently it was decided, with the aid of an Australian Government grant (through the Rural Industries Research and Development Corporation) to build a special purpose research facility for juvenile crocodiles with flexibility to cater for a wide range of different environmental, management and nutritional issues. Although most of the crocodile farms in Queensland are located in the far north around Cairns and on Cape York, the necessary infrastructure

in QDPI was located in Townsville at the Oonoonba Veterinary Laboratory site. There was ample land available and specific scientific skills of staff at the laboratory as well as research laboratory equipment and facilities. Members of the current research team on crocodiles all work on other research programs as well in a multi-disciplinary regional role. In all there are currently 10 team members contributing the equivalent of 3.7 full time positions on crocodile research, development and extension.

## **METHODS**

### **Research Animals**

Each year some 250-300 hatchlings are provided by a commercial farm in Queensland (the Edward River farm, now called the Cairns farm). These are hatched at the Edward River Aboriginal Community enterprise on western Cape York Peninsula. At the end of the year's experiments these animals are returned to the farm. Crocodiles are tagged individually (using small stainless steel web tags) and number series on the tags are linked to clutch of origin.

### **Research Facility for Juvenile Crocodiles**

Crocodiles are housed in a fully insulated, environmentally controlled building. The building measures 26 metres long by 4.6 metres wide, with a 5 degree sloping roof 2.4 to 2.1 metres high. It is divided into six equal sized rooms. The building is constructed of insulated polystyrene sandwich panelling, incorporating 0.6mm white colorbond skins internally and externally (coldroom panelling). The roof is 100mm thick, external walls 75mm and internal walls 50mm thick. Each room contains two fibreglass tanks, one radio speaker and a wall mounted air conditioner. The floor of the shed is concrete. An additional 'hospital' tank has been set up in a separate building at the site and can cater for 30 animals in 3 compartments.

Hot water is supplied to the building through 3 x 315L hot water systems (one per 2 rooms). Hot water runs through copper piping and cold water through PVC piping. Incoming water comes into the shed at three points only, along the internal walls between rooms 1 and 2, 3 and 4, 5 and 6. Each tank has its own set of taps and a hose for hosing out and refilling. When filled, water temperature in the tanks is controlled through solenoid valves, which pulse in hot or cold water when required.

Control panels for water, air temperatures, lighting and radio are situated on the outside of the shed so that servicing and data recording does not have to be done in the rooms. Only repairs to plumbing and sensors are done in the rooms.

Water temperature is controlled by Brainchild microprocessor based auto tuning controllers. A stainless steel sensor is placed in the middle of each tank. A diffuser tube made of copper pipe runs along the bottom of each tank and six small equally spaced holes along the tube disperse the incoming water. When the sensor detects the preset



temperature the controller closes the solenoid valve. Water temperature control range is  $\pm 1^{\circ}\text{C}$  of the set point. Digital faceplate display gives current temperature and set point.

Air temperature is controlled by reverse cycle room air conditioners. A sensor on the wall of each room controls the air temperature. A digital display unit gives set point temperature and actual room temperature. Control range is  $\pm 1.5^{\circ}\text{C}$  set point.

Lights are on timers and dimmers, so that light duration and intensity can be controlled. Each room is illuminated by two 100 watt light bulbs in clear covers located over the two tanks.

Tanks are made of fibreglass, grey in colour and are 3 metres long x 1.3 metres wide 650mm high on water side 500mm on land side. The dryland area is flat and slopes down to the water area. To provide an equal area of land and water the water depth is maintained at 160mm. A stand pipe at one end of the tank controls water depth. A laminex hideboard, 1200mm long x 600mm wide, is suspended by an aluminium frame across each tank 60mm above the dry land, increasing over the water due to the sloping of the tanks. The height of the hideboards above the tank bottom can be adjusted to allow for growth of the animals.

### **Food Preparation Room**

A feed storage and preparation room has been provided close to the rearing unit. This room houses a refrigerator and freezers for food supplies, mincer, bench space for preparation and washing facilities for equipment used in food preparation and feeding. It also contains television monitors connected to video viewing equipment which can be rotated among the rooms in the research building.

### **Egg Incubation Room**

A crocodile egg incubator room has recently been added to the complex. The room contains 6 hatching incubators each capable of holding 50 eggs. This will permit laboratory studies on microbial contamination of eggs, hatchability and cleaning and disinfection of eggs.

### **DNA Laboratory**

This is currently being set up in the veterinary laboratory for fruit and vegetable breeding programs and beef cattle research. We hope to develop the technique for use on crocodiles as well. There has been interest shown by conservation groups in Queensland in using DNA to test whether there are differences between animals from different geographic locations (eg. one location seems to contain crocodiles with just 4 prominent nuchal scales, compared with the more common 6, and there is a theory that crocodiles in another area may be naturally smaller or 'dwarfed').

## Statistical Design of Experiments

Researchers have shown that clutch differences in crocodiles are very important, so statistical designs of experiments must take this into account. Where different types of treatments are to be trialed, such as food types, at least two replicates (statistical repeats) of each treatment are used and, in every case, the experimental unit is a tank of animals. For each replicate, animals are allocated to tanks on the basis of their original clutch and then by body size, so that each group is as similar as possible before the different treatments are applied.

Different sets of clutches can be used to form different replicates, since clutch differences are thereby taken in account. Farms tend to avoid mixing clutches as much as possible because clutches vary in temperament. The only problem with this research strategy occurs if clutches respond differently to the experimental treatments. For experiments when the animals are older, there is a natural wide spread of sizes, even within clutch, so replication is based on different size classes: this generally necessitates mixing animals from all clutches together in tanks, but is regarded as a better strategy than mixing animals of very different sizes.

For experiments with treatments consisting of a range of levels, like water temperature, the emphasis is on estimating response trends, and replication is reduced, or more commonly, sacrificed to achieve a greater number of levels of the treatment. Again, animals are allocated to tanks by the methods described above.

There is a settling down period of at least two weeks between successive experiments, during which all tanks are kept at the same conditions. A grading strategy is used for the hospital tank, where smaller animals are transferred in and out between experiments. Special extra attention is sometimes given to runts to try to get some of them up to trial useable size.

In any temperature trial, water and/or air temperatures are gradually increased or decreased by 1°C each day.

## Collecting Research Data

Amounts of food given to each tank, and any uneaten residue left on food trays are recorded at each feeding. Any food scattered in the water is not measured. Because of the stress caused by measuring, animals are not fed on measuring days. Measurements are generally done on a Monday morning, following the weekend fasting. This results in fasted liveweights, which are more accurate measures of actual size. An experienced catcher, wearing a chain-mail butcher's glove, catches an animal firmly behind its neck. Usually, he catches the animals in the water, where they generally display less aggression.

Measurements are taken only at the commencement and the conclusion of each experiment, and then again after the 'settling down' period. Scientifically it would be useful to collect data several times during the trial but

- the extra stress that this causes can affect comparison between treatments, and also invalidate direct application to commercial applications
- it is only the final response difference that is really useful.

Data recorded include liveweight, total length, snout-vent length, head length, skin colour, temperament exhibited, and any skin disease or blemish. At one weighing, the belly scale pattern of each animal is recorded by photo-copy. Scute material from the 1995 research animals has been stored for possible future DNA investigation.

## RESULTS

### Temperature Trials

In experiments using *C. porosus* between hatching and one month of age, Webb *et al.* (1990) showed that when temperatures of 30°C to 34°C were imposed, maximum body weight was achieved at 32°C. Hutton *et al.* (1993) claimed a marked improvement in growth and survival of crocodile species reared at temperatures between 30°C and 32°C over animals reared at temperatures only a degree or two cooler, but hypothesised that the optimal rearing temperature for individual animals could vary depending on clutch origin and incubation temperature, among other things.

An experiment using 140 *C. porosus* hatchlings from five clutches was conducted, subjecting balanced groups of animals to a range of temperature regimes over a 10 week period (Turton *et al.* 1994). The stress levels in animals were assessed by measuring their levels of plasma corticosterone - the hormone which is released into the bloodstream in response to stress. Results indicated that at the highest temperature of 36°C, animals were more stressed than at 32°C but that there was no difference between 28°C and 32°C. Over the period of 10 weeks, body weight changes in the animals were not significantly affected by water temperature differences.

Historical research indicates that water temperature is perhaps the most important environmental factor influencing the growth and well-being of young crocodiles. So, it was essential that the best rearing temperature for different ages of research animal be established for the specific nature of the Oonoomba rooms/tanks, taking account of factors such as design of tank, depth of water, type of hide-board, humidity, air flow, level of noise and lighting.

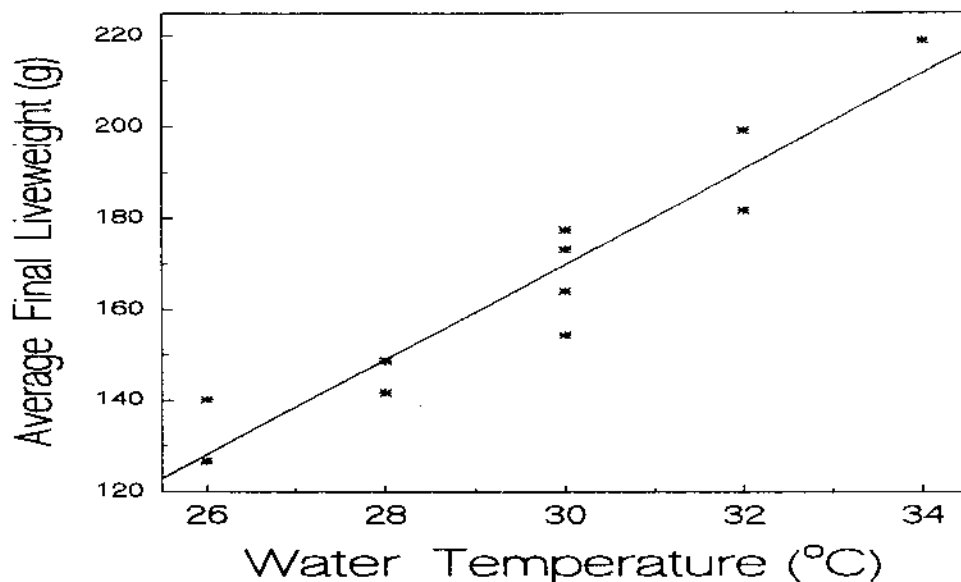
#### *'Experiment on 2 - month old hatchlings'*

Since water temperatures in native crocodile habitats are generally between 25°C and 28°C, and some crocodile farms in Australia have been set up in sub-tropical areas such as Rockhampton and Fremantle, it was decided to research the effects of water temperatures as low as 26°C. These temperatures are directly relevant to the many zoos

and wildlife sanctuaries in southern Australia which keep crocodiles. There is a school of thought that farmed animals should be given the chance to adjust their temperature by farms providing a thermal gradient in the animals' rearing environment (Lang 1987). The Oonoonba facility consists of just six rooms, each of which can be maintained at a certain air temperature, so there is a limit to the degree of thermal gradation that can be imposed as part of a research treatment. From published literature, an upper limit of 34°C water temperature was chosen for this initial experiment, carried out when animals were two months of age.

A serious fungal disease occurred in room 1, tank A (the coldest treatment) requiring animals to be caught and treated with Betodine. There were seven deaths in this tank so it was decided to terminate this experiment at the six week stage and return all tanks to a water temperature of 32°C.

The experiment consisted of imposing different air/water temperature combinations and recording growth over the 6 week period. For the 6 rooms, air temperatures were set at 28°, 30° and 32°C in a randomised block design, with 2 replicates. Within each room, water temperatures in the 2 tanks were set at 2°C above and 2°C below the particular air temperature, resulting in a range from 26° to 34°C. Three different clutches of animals constituted each replicate and animal density was either 18 or 20 per tank (0.20 or 0.22 square metres per animal). Animals were fed daily on the standard diet of chicken heads, kangaroo and beef, fortified with a special vitamin/mineral supplement powder, and a fasting period of 2 consecutive days each week to aid complete digestion of food. The average liveweight of the hatchlings at the start of the trial was 106g (range from 40g to 200g). Figure 1 shows the average final weights for animals reared at the different water temperatures. There was no significantly detected additional effect of air temperature to this response trend.

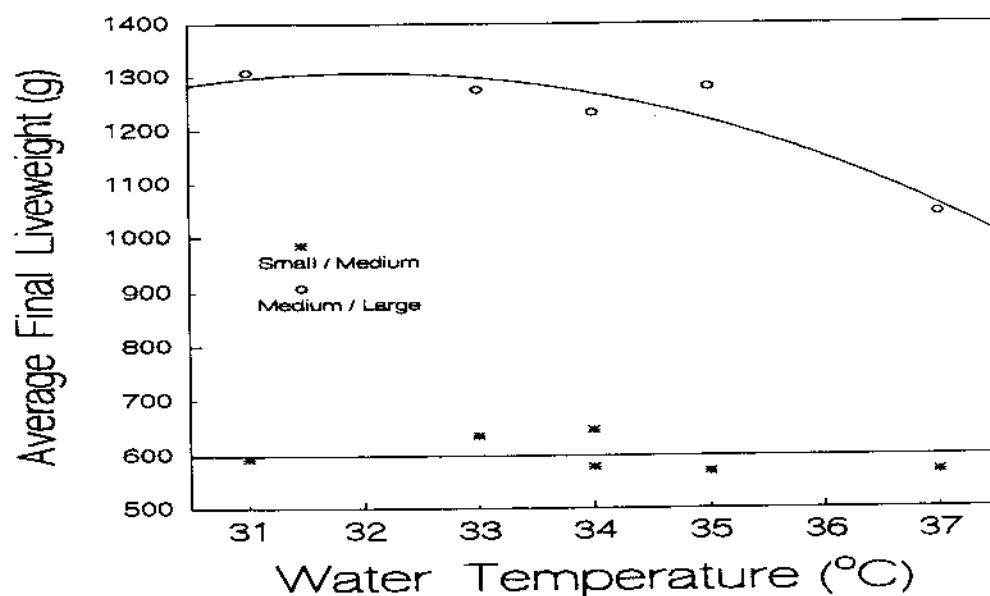


**Figure 1: Liveweight response of 2-month old hatchlings to water temperature**

Because of the nature of the tanks with their high, solid walls, it is not surprising that room air temperature differences of 2°C around a particular water temperature did not have any significant effect on animal growth. For air temperature warmer than the water, there would be a cooler layer of air sitting just above the water, and spilling on to the land area. Conversely for air temperature cooler than the water, the water would be constantly warming the air inside the tanks with the air rising. Hence, the actual air temperature in the immediate crocodile land environment would be closer to the water temperature than the air in the rest of the room.

*'Experiment on 7-month old juveniles'*

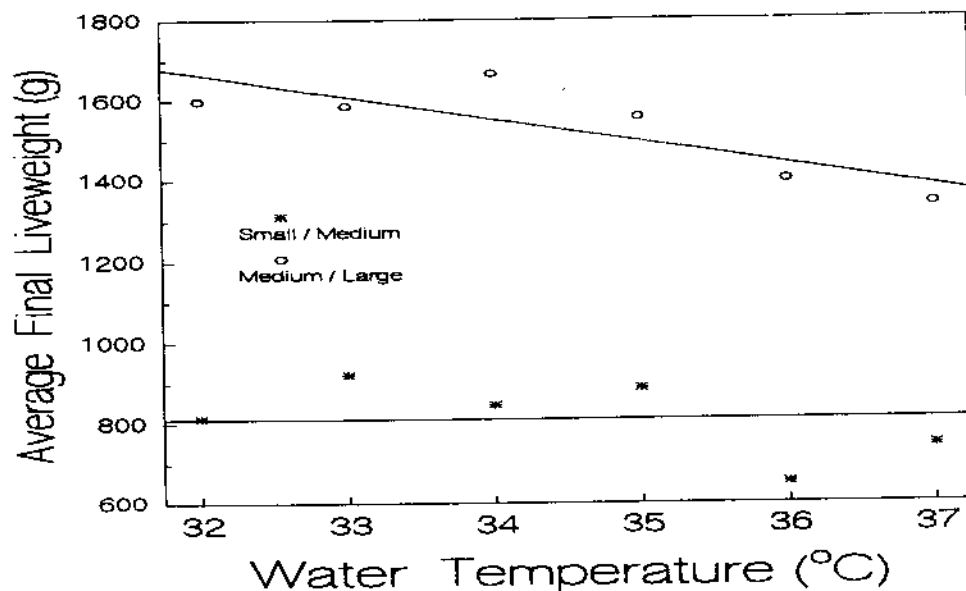
At this age there were large differences in size of the animals (200g to 1900g) so it was decided to divide the animals into 2 size classes, called 'small/medium' (av. weight 460g) and 'medium/large' (av. weight 910g), and investigate the effect of different water temperatures from 31° to 37°C on each group over a period of 9 weeks. Air temperatures were maintained within 1°C of water temperatures in each tank. Densities were 17 and 14 per tank for the 2 size classes, and the diet was the same as for the initial experiment on temperature, but minced more coarsely. There was no significant response to water temperature over this range for the small/medium group, but a detrimental affect at higher temperatures for the medium/large group is indicated by the final average liveweights as shown in Figure 2.



**Figure 2:** Liveweight response of 7-month old juveniles to water temperature

*'Experiment on 10-month old juveniles'*

This was a repeat of the experiment on 7-month animals, with a lower limit of 32° instead of 31°C, to see whether the same responses would occur in slightly older animals. It was run over a 7 week period, and there were again 17 small/medium or 14 medium/large animals per tank. The average initial weights were 650g and 1250g for the 2 size groups. Figure 3 shows the final average liveweights for each temperature treatment and the estimated response trends. These trends reinforced those observed in 7-month animals.



**Figure 3: Liveweight response of 10-month old juveniles to water temperatures**  
 The results from the experiments on 7 and 10 month old animals indicate that the smaller specimens at these ages seem to be quite tolerant to air/water temperatures anywhere between 31° and 37°C. However their heavier relations suffer a slow down in growth when temperatures climb to more than 34° or 35°C.

### Pellet Development and Evaluation

A feed milling company has been cooperating on this project, and has manufactured 5 prototypes of pellets for evaluation. The first four were distributed to commercial farms for them to try on their different aged animals. At the same time we were trying a range of pet foods (both dry and wet) on some of our runt animals. The only success we had was with one high priced, fish based tinned cat food. Following is a summary of industry's reaction to the prototypes.

#### *'Prototype 1'*

This was a small, hard base-shaped extruded pellet, and was released to farmers in January 1993. None of the farmers recorded any success, although one did get animals to eat pellets when mixed with meat and another crushed the pellets and added it to the diet, which was eaten. Suggested improvements were

- a softer, rubbery texture needed
- a different aroma (suggest prawn)
- a pellet that would float, yet not breakdown readily in water.

#### *'Prototype 2'*

This was manufactured in May 1993, and was smaller and softer, with a prawn flavour. Again, the only success was when mixed with minced meat. Farmers suggested that an even softer, moister pellet was needed.

#### *'Prototype 3'*

This pellet had a higher moisture content of 21%, and so required the addition of an anti-spillage agent. Protein content was 30% and protein sources consisted of cotton seed, soy bean and fish meal. Juvenile crocodiles would still not eat pellets by themselves at any of the farms.

#### *'Prototype 4'*

Moisture content was raised to 25%, protein to 40%, and a prawn flavour was added. As well as being distributed to commercial farms for evaluation, a proper scientific experiment was set up using our research facility. Animals were 3 months old at the start of the investigation, and were fed 3 different treatments consisting of mince:pellet mixtures (by weight).

Treatment	Diet for week			
	1	2	3	4
A	100 : 0	100 : 0	100 : 0	100 : 0
B	100 : 0	75 : 25	50 : 50	50 : 50
C	100 : 0	75 : 25	50 : 50	0 : 100

Thus, treatments B and C were attempts to 'wean' the crocodiles from their standard meat diet onto pellets. Unfortunately, at the 50:50 rate food consumption dropped off with the animals picking out the pieces of mince and for the week when treatment C animals were fed only pellets, they did not eat anything.

A second experiment was run, in which different quantities of pellet were actually put through the mincing process, and so thoroughly incorporated into the mince (so animals could not pick out pieces of pure mince). The treatments were imposed as follows:

Treatment	Diet for week					
	1	2	3	4	5	6
D	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0	100 : 0
E	100 : 0	85 : 15	70 : 30	55 : 45	50 : 50	60 : 40
F	100 : 0	85 : 15	70 : 30	55 : 45	40 : 60	50 : 50

It had been planned to take treatment F right up to 100% minced pellets, but the results on weeks 4 and 5 showed little of the mixed diets of E, F were being consumed, so on week 6, the mince content was increased. This experiment was aimed more at evaluating the nutritional aspects of a pellet/mince diet, rather than trying to get animals to eat pellets by themselves.



#### *'Prototype 5'*

We have just received the new products which consist of

- small, soft, ball-shaped pellets of different colour (red and brown)
- the same basic ingredients but in a 'mash' form.

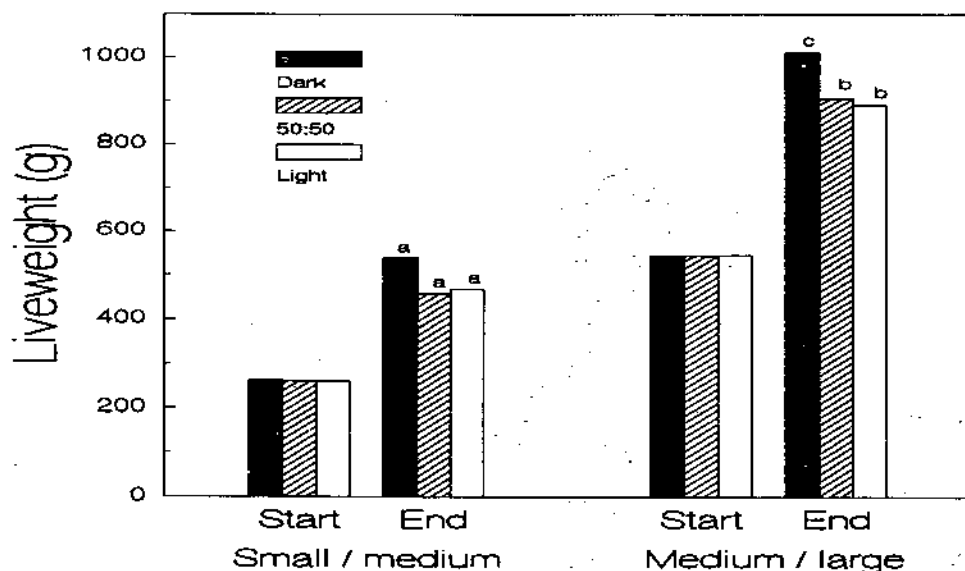
We are trialing these on hatchlings which have not been exposed to any other food since hatching.

#### **Light Trial**

This experiment was conducted on 6½ month old juveniles and ran for 8 weeks. Animals had been reared under previous experiments on a diurnal light cycle, with artificial light provided in the enclosed rooms from 6.30am and until 5pm each day. For this experiment animals were divided into 2 size groups, with average weights 262g and 544g, and stocked at 19 and 16 per tank respectively for each group. Three light treatments were imposed:

- artificial light kept on continuously
- normal diurnal cycle
- rooms kept continually as dark as possible (very dim lights put on during feeding and cleaning operations).

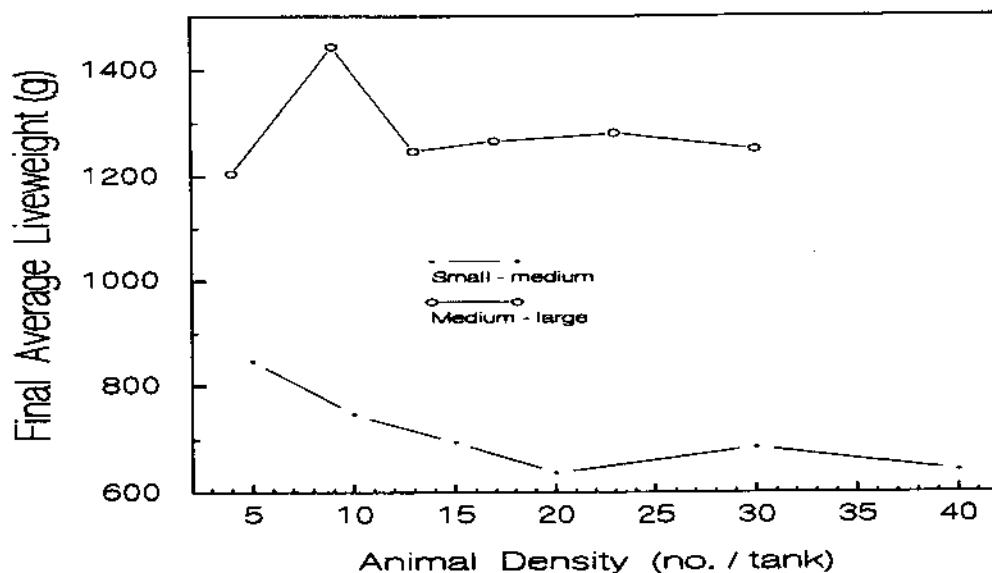
The design used was 2 room replicates of each light treatment in a randomised block array, and within each room the 2 tanks contained the 2 different size groups resulting in a split-plot design. Responses, in terms of liveweight change for the 2 size groups, and shown in Figure 4.



**Figure 4:** Average initial and final liveweight for the 2 size groups reared under 3 different light regimes. Columns headed by similar letters do not differ significantly ( $p > 0.05$ ).

#### Density Trial

This was the final experiment conducted on the 1995 animals, which were 8½ months old. The animals ranged in weight between 200g and 1710g, and were split into 2 groups about a division point of 690g, resulting in average weights of 470g and 945g for the 2 size groups. The smaller group were allocated to 6 randomly selected research tanks at densities of 5, 10, 15, 20, 30 and 40 per tank (area 3.9 square metres), and the large group to 6 other tanks at densities of 4, 9, 13, 17, 23 and 30 per tank. The trial was run for an 8 week period, on a dimmed light diurnal cycle. Figure 5 shows the response patterns of final average liveweight to density for each size group.



**Figure 5:** Final average liveweight for the 2 size groups reared at different densities

This shows that for the smaller animals there was a general decrease in growth rate with increasing density from 5 to 20 per tank (or 0.8 sqm to 0.2 sqm per animal) but no further decrease up to 40 per tank (0.1 sqm per animal). For the larger animals, apart from the one tank which produced an extraordinary response, there seemed to be little effect on density (in the range 4 - 30 per tank) on growth rates.

The odd result occurred at a density of 9 medium/large animals per tank. Compared with an overall average weight gain of 35% among all the large sized animals (945g to 1277g increase), 6 of these 9 animals recorded weight gains of more than 50%. This cannot be explained by clutch differences, since, as part of the standard design allocation procedure, animals at the different densities were selected from the available clutches in the same proportions: for this group of 9, there were single representatives from 5 clutches and pairs from another 2 clutches. This highlights some of the difficulties in having to deal with crocodiles as research material. From this single experiment there is no way of knowing whether a density of 9 per tank for larger animals is some magical number or whether it was a result of putting a particular mix of animals together which may have interacted with each other in some beneficial manner (eg. more tolerance, especially during feeding).

### FUTURE RESEARCH

QDPI is committed to pursuing research into further aspects of nutrition, environmental effects and management practices as applied to farmed *C. porosus* from eggs to slaughter. Research to date has necessarily focussed on single, specific issues. However

it is recognised that employing optimal individual practices does not necessarily translate into an optimal combined strategy (eg. response to different diets may change depending on environment or animal density). Thus, research will naturally lead to experiments looking at selected combinations of near optimal individual factors.

The research team recognises its strengths and weaknesses and has established links with the farming industry and with other crocodile researchers around Australia to draw on their expertise and ensure that research is focussed on key issues. It has drawn together the required skills of veterinarian, bacteriologist, biometrician, geneticist, animal attendant, industry manager, extension officer and computer programmer from QDPI staff.

### ACKNOWLEDGEMENTS

The authors would like to acknowledge the invaluable contributions made by other members of the research team: Dr Steve Johnson, Dr Annette Thomas, Dr Cam McPhee, Bernie Davis, Rob Jack, Rod Bloomfield, Honor Stephenson and Scott Blyth. For providing us with research animals each year we would like to thank Vic Onions and Don Morris (Edward River Farm) and Keith Cook (Cairns Farm). The product research unit at the feed milling operations of Ridley Agriproducts, Paul Mason and Andrew Stallman, have supplied a range of prototype feed pellets, and Rhône-Poulenc Animal Nutrition has supplied the crocodile vitamin/mineral supplement used in our diets. Individual members of the Queensland crocodile industry have been very supportive of our activities and we would especially like to thank Harry and Peter Freeman (Hartley's Creek Farm), Vic Onions and Keith Cook, and John Lever (Koorana Farm) for the many times they have shown team members and visiting scientists around their farms, and time spent discussing R & D issues. We have established useful dialogue with researchers at the universities of Queensland (Brisbane), Central Queensland (Rockhampton) and James Cook (Townsville), and also with Steve Irwin (Qld. Reptile and Fauna Park), and we acknowledge their input and support. Dr Peter Mullaney (Primary Tasks) is assisting the team in its extension program.

The Rural Industries Research and Development Corporation has supported our research effort through several research funding grants, and the program coordinator Dr Peter McInnes has maintained a close personal interest in our work and provided invaluable support.

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## **DISEASES ENCOUNTERED IN GENUS *CAIMAN* INTENSIVE BREEDING**

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### **SUMMARY**

The different pathologies and diseases found in five years of work in captive crocodilians of the genus *Caiman*, are presented. For practical purposes the diseases are listed as follows;

- a. Diseases caused by inappropriate management.
- b. Alimentary Diseases.
- c. Parasitic Diseases.
- d. Infectious Diseases.
- e. Miscellaneous Diseases.

For each group of pathologies; symptoms, etiology, lesions, treatments and prevention measures are presented. We conclude that the principal diseases affecting *Caiman* in captive condition are the palmar and plantar abrasion caused by rough and abrasive soil, wounds caused by interspecific aggressive behaviour. Alimentary diseases caused by an inappropriate Calcium: Phosphorous ratio cause Metabolic Bone Diseases in growth animals feed with raw beef meat.

On the other hand, the parasitic and infectious diseases are less common and not cause diseases and death as describe some authors and only few number of parasites has been described to be dangerous for the crocodilians. Finally, some miscellaneous diseases as development abnormalities are described.

**Key Words:** Pathologies - Intensive Breeding - *Caiman* - Parasitic Diseases - Infectious Diseases- Alimentary Diseases.

### **INTRODUCTION**

Compared with the knowledge and amount of scientific papers in upper vertebrates pathologies, the amount of information in reptilian, and specially, in crocodilian pathologies and diseases is poor. Only few reports are found in scientific publication and only in the last five years, some publication are available. This fact is more notorious in Latin American country, in which the management of captive crocodilians, have importance, due mainly to the decreased natural population or habitat destruction.

Is the aim of this report to communicate the experience of the authors in five years of working in this field of investigation and secondary a revision of the available literature in Argentina about this topics.

### **MATERIALS AND METHODS**

The work was carried out under different captive populations of both genus of *Caiman* that live in Argentina (*Caiman latirostris* and *Caiman crocodylus yacare*) maintained in different locations in Northern Argentina (Corrientes and, Chaco province) and, Buenos Aires Zoological Garden. The animals are feed in chopped beef, chicken meat, and fish meat, entire fish, chicken carcasses, etc., depending the location and the disponibility

of the food. The animals was from different ages, sexes and sizes .of both *Caiman* species .

## RESULTS

### a. Diseases caused by inapropiatte management

In this pathologies ,two mainly problems was found. The interespecific agresive behaviour and the palmar and plantar skin surface abration..

In the first case , overcrowding , different sizes of animals in the same pool , lack of amount of food, different sizes of animals , was the predisponenet factor to this behaviour .The type and deep iof the wound appears to be dependent of the size of the animals .You can see in the photo 1. a severe wound caused by other animal . This wound was able to cut the rostral bone of the atacked animal, but it survive the attack , cicatrize the wound and live., with some difficult to eat and breath.

Other slides show different wounds, varying from a simple wounds to phalanx and other bones severe loss . The treatment of this wounds is not difference with the same wounds seen in other animasl as mammals and the treatment ibclude the gently cleaning , dessinfectaMt aplicattions (Povidone Iode ), bandagge if it is necesary and in some severe cases broad spectrum parenteral antibiotics as Enrofloxacin ( 10 mg/kg/24 hs s.i.d) or another commercial available quinolones

The palmar and plantar skin surface abration caused buy rough and abrasive concrete was a cronic problemas for years in many breeders.as mentioned some authors (Jacobson, 1984 - Bolton, 1989) The construction of new pools , with polystyrene coberture in the soil, painting the bottom of the pools with sintetic pinture, were some of the solution . In this cases , the animals were treated with a dairy gently clean of the abration with iodine, bandage with parafinned band and aplication of parenteral broad spectrum, antibiotics as quinolones . In some cases te wounds of the abrated surfaces were contaminated with bacterial and/or fungal agents. In this cases it was necessary to remove the necrotic tissue , and keep moisture of the bandagge ., in order to avoid further complications to cause death.

### b. Alimentary Diseases

The bibliography mentioned a variety of pathologies and diseases caused by lack or deficit of minerals and/or vitamins (Jacobson, 1984- Bolton, 1989 - Kuhen, 1990). In our exprience , we only found one of this pathologies and we think the most important of the alimentary diseases found in all vertebrates , specially when this animals are bred in captive condition. with artificial alimentation.

The photo 2 show a little *Caiman crocodylus yacare* , wich are affected by a disease so called "Metabolic Bone Diseases " or in the classic veterinary book test "Ricket", "Osteomalacia", Fibrous Osteodystrophia", "Secondary Hipeparatyroidims Nutritional Disease". The basic patagenic mechanism is the lack in the Calcium aported by food, an excess of Phosporous, when the animal fed in meats with high muscle content, lack of Vitamine D caused by inapropiatte sun exposition and another situation as lack of proteins in the diet, excess of Phtalic acid in diet ,etc. In this cases above mentioned, the animal is not able to maintain the blood Calcium levels and develop the diseases wich are characterized by inflammation of the ribs and limbs, curvature of the dorsal spine, paresia or in some cases complete paralysis.and softness of some bones as the jaw and limbs bones with visible deformations.and spontaneous fractures

The treatment is based in the adequate aport of calcium salts in form of Carbonate by oral route or Gluconate or Lactate if the parenteral route is preferred. In the first case, the addition of Calcium Carbonate in the food at a dosage between 10-20 mg/kg of food is needed to prevent and in some cases revert the paralysis, the softness of the bones.

If the second option is selected, the dosage of the drug (Calcium Lactate or Gluconate) is 10 mg/kg of body weight, by intramuscular route, using a syringe. The adequate prevention of this pathology is the best practice. The control of the blood calcium level in suspected animals (Normal values 7 - 10 mg%) is an adequate practice or a periodical evaluation of the adequate balance in Calcium -Phosphorous relation (normal values 2:1 or 1.5:1). This technique has a low cost, is available in the majority of the diagnostic laboratories and needs few blood for the determination.

The supplies of Calcium Carbonate is necessary in growth young animals and the easy manner to supply this salt is add crushed egg-shell in the meat or supply powdered Calcium Carbonate.

Another pathologies related to inadequate nutritional balance, as deficit of vitamin B or vitamin C and the so called Steatitis caused by lack of vitamin E is still not found in our work.

### c. Parasitic Diseases

The description of all parasites is not the aim of this report, because at this date, more than 450 species of parasites affecting crocodiles was described (Jacobson, 1984- Frye, 1990- Reichembach -Kilne &Elkan, 1964 - Bolton, 1989 - Liegh, 1978 - Lombardero-Apostol, 1953, Medem, 1988) including Cestodes, Trematodes, Nematodes, Acanthocephales or thorny-head worm, Pentastomides and some Arthropoda as Insects of Diptera order (Tabanidae family); we only described two of our findings in *Caiman* species. One of them, is a filaroid nematode which was found free in the coelomic cavity of sudden dead animals. The parasite was classified as *Micropleura vazii*. Some authors consulted found this worms in the stomach and gut of the same definitive host (*Caiman crocodylus yacare*) but we only found in coelomic cavity. Interestingly, microfilariae was not observed in peripheral blood.

The second of our findings is a pentastomid, *Alofia platycephala*, first described in our country by Lombardero and Apostol and now redescribed. This Pentastomid was found in the lungs of animals and we think that is not a principal cause of death, however a high number of this parasites was found in affected animals.

In the periodical routine fecal and blood examination of the animals was possible to see some eggs of unclassified parasites and in blood, intraerythrocytic parasites resembling *Hepatozoon* or *Hemogregarina*.

### d. Infectious Diseases

This infectious agent was not found in the variety and quantity described by others report found in the bibliography. In our country, the use of electronic microscopic technique is still unapproachable, because of its high cost; so, the viral agent which causes pathologies and diseases in crocodiles are, at the moment, impossible to determine.

The fungal and bacterial agents were isolated from mouth, skin, stomach, liver, lungs and other organs and systems. The most common isolated fungal agent is *Aspergillus fumigatus* and *Cladysporium spp* from skin lesions.



*Aeromonas hydrophyla*, *A. liquefaciens*, *Pseudomonas putrida*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Proteus retgerii*, *Proteus mirabilis*, *Sphaerophorus necrophorus* were the most common bacterial agents isolated from the lesions in the above mentioned organs and systems.

#### e. Miscellaneous Diseases

In this topic we include different types of congenital abnormalities, one of which are shown in the photo 3. The most common findings were anomalous tail, lack of tip of the tail, bicephalic animals, lack of limbs and one case of albinism.

#### CONCLUSIONS

Based on our findings and the available bibliography in our country, the first conclusion was that the inappropriate care and management of captive condition was the major source of diseases and pathologies in captive *Caiman* species. The aggressive behaviour and the palmar and plantar skin abrasion due to the rough soil or floor of the ponds and pools in which the animals live are proof of this bad condition.

Infectious and parasitic diseases appear not to be an important impact in the appearance of illness and death of *Caiman* in captivity, but some parasites as *Alofia* and *Micropleura* can be found and some unclassified eggs and intraerythrocytic parasites required further studies in order to elucidate their exact pathological role.

Finally, the Metabolic Bone Disease appears to be an important pathology in young growth animals, that fed on meat with high muscle content and their prevention is most important than the treatment.

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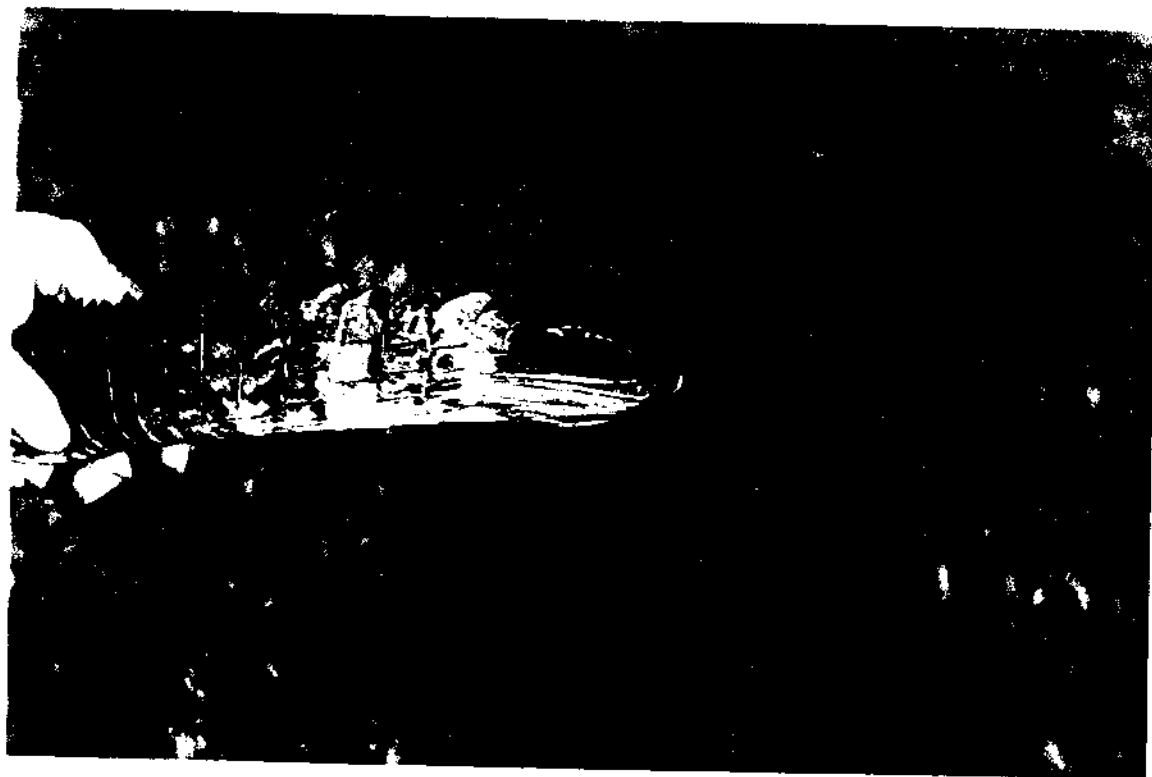
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**PHOTO 1 - CAIMAN CROCODYLUS YACARE  
LOOS OF ROSTRAL BONE. AS CONSEQUENCE  
OF AGRESIVE BEHAVIOUR**



**PHOTO 2 - CAIMAN CROCODYLUS YACARE  
METABOLIC BONE DISEASE**



**PHOTO 3 - CAIMAN LATIROSTRIS  
ANOMALOUS TAIL**

## **ALOFIA PLATYCEPHALA; PENTASTOMID WORM IN LUNG OF CAIMAN CROCODYLUS YACARE**

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### **SUMMARY**

A finding of tongue worms or Pentastomida in lungs of *Caiman crocodylus yacare* in Northern Argentina is reported. The morphology of the larval stage and the adult is described, also the localization, the definitive host and the lack of macroscopic lesion in the lung parenchima.

Based in the morphology and the localization in the host, was posible to stablish that the tongue worm is *Alofia platycephala* (Lorhman, 1889) Giglioni, 1922.

**Key Words:** Tongue Worms - *Alofia platycephala* - Lungs - *Caiman crocodylus yacare* - Northern Argentina

### **INTRODUCTION**

Poorly know, the pentastomid or tongue worms are internal parasites, when adults, of the respiratory system of various vertebrates, specially reptiles. Since its first description by Leuckart in 1860, its exact systematic position remain still in discussion, for some authors it is included in the Phylum Artropoda, because the perforatory apparatus in the larval stage resembling the mouth pieces of Insecta. On the other hands, another described closely analogy to the Anelidae due to the internal and external body segmentation<sup>13,14,15</sup> or recent investigations found that the morphology of the spermatozoa is similar at wich of the Crustacea, specially the Argulidae<sup>16</sup>. Finally, a fourth opinion give a range of Phylum(Pentastomida).

Into this Phylum are two Orders. The **Cephalobaenida** order are the most primitive and their biological cycle include insects, fish, amphibians and reptiles. Their biological cycle is related to be direct, without intermediates host.<sup>13,14,15</sup>

The **Porocephalida** order required the utilization of mammals as intermediate host and the reptiles as definitive host and are in a superior evolutionary scale of the another order.

This last order include some families, as adult, parasites of reptiles and are characterized by have a quitinous mouth surrounded by five hooks and the vulva in the posterior end of the body.

**Sebekidae** family include the tongue worms or pentastomids of respiratory tract of crocodilians, with a variety of genus, one of the most important is *Alofia*; malayan, african and southamerican crocodiles internal parasite.

Genus *Alofia*, with the species *A.ginae*, *A.merki*, *A.indica*, *A.adriatica* y *A.platycephala*, is the genus described in this report. In Argentina, Lombardero y Apóstol were the authors of the first description and finding in 1951<sup>1</sup>, but the definite host was described in a unclear form.

The objective of the present paper is a revision of the definitive host originally cited by Lombardero y Apóstol in their original paper and the redescription of the larval and adult stages morphology as well as the localization in the respiratory system of the affected animals.

## MATERIALS Y METHODS

During four years of works, necropsies was made in specimens of e *Caiman crocodylus yacare* and *Caiman latirostris*, that die suffering a variety of diseases and derived for the diagnostic at the Laboratorio de Identificación de Parásitos de los Animales Silvestres, of the Cátedra de Zoología y Recursos Faunicos, U.N.N.E., Corrientes city, Argentina. The procedence of the animals was Jardín Zoológico de Buenos Aires, Zoológico de Roque Sáenz Peña (Chaco province), Zoológico de Corrientes, Zoológico de Goya (Corrientes province).

The necropsy techniques was carried out following the decription of E.Jacobson,1984.

The parasites found in the lungs of the affected animals was fixed by Raillet's solution for further observation under stereocopic microscope, previuos diafanization and clear the cuticule by means of the glicerine technique.

The parasite eggs was obtained by means of the pression lungs washing with saline solution and a syringe, whereas the gravid females and observed under optic microscope.

## RESULTS

From the lungs of the death animals was taken 12 to 15, white, litle parasites measuring 2 cmts in large, round body, with a sharp end. The media of the parasites found was  $14 \pm 3$  by animal, and already 80% of *Caiman crocodylus yacare* examined have this parasites in their lungs.

The microscopic observation of the parasite, show a dorventrally aplaned, segmentd body with thorny tegument extended side to side of the parasite. The cefhalic or pregenitaly region named Prosoma is wide, measuring 0.6 to 1 mm, with males and females significative variation ( $p < 0.01$ ).

In this area the mouth was encountered, surrounding with a quitinous frame and four retractile hooks, conforming the fixing system. Each hooks have  $86,4 - 97,2 \mu$  in long and  $12,9 - 21,6 \mu$  in width.

The abdominal portion or Metasoma is narrow, segmented and their measuring are 1 to 1,8 mm. In the female is 10 mm in width and the male is litle, 1 mm in width.

The entire genital system was observed in females, with a uterous filled with eggs, and opening in the posterior end of the body through a genital porus.

The eggs was obtained by bronquial and lungs washing, they are oval, double membranous yellowish, measuring  $99,3$  to  $64,8 \mu$ , with a develop larvae in difrent growth stage. In the first larval stage, the larvae are amorfous, but is posible to see the three further body segmentation (see figure). Also was posible to observe a eggs containing a well develop second larval stage measuring  $83 \mu$ , with four legs and claws measuring  $13,6$  to  $16 \mu$ .

Macroscopic evidence of bronquial epithelium and lungs parenchima lesions was not observed.

## CONCLUSION Y DISCUSSION

In accordance with the bibliography available was posible determinate that the parasite bellow describe is *Alofia platycephala* (Lorhman 1889) Giglioni, 1922. Likewise, the description is in accordance with the parasite originaly describe in our country by Lombardero and Apóstol in 1952. The definitive host of the parasite is mainly *Caiman crocodyllus yacare* or "black yacaré" "yacare negro" or "yacaré de hocico largo" in our report. This fact is in dissapointing with the Lombardero and Apóstol description, which cites *Caiman latirostris* as definitive host in their