

CHAPTER 5 DETERMINANTS OF THE REPRODUCTIVE OUTPUT IN GREEN ANACONDAS.

5.1 INTRODUCTION

Reproductive value or lifetime reproductive success (LRS) is the number of potential offspring that an individual can leave in the population over its lifetime. Natural selection should maximize it since it is a true measure of fitness (Williams 1966; Daan and Tinbergen 1997). In animals with undetermined growth, clutch size increases with female size due to an increase in the coelomic cavity. An adult female that has enough fat reserves can either breed in that year and secure some offspring or skip reproduction and breed the following year when she can produce a larger clutch. The decision to breed, including the age of first reproduction, depends largely on the increase of fertility of the animals due to the growth attained by delaying reproduction an extra-year, and the likelihood of dying during that year. If the female can increase significantly the number of offspring by increasing her size during one year, selection should favor skipping reproduction that year. If the increase in fertility is not large and the female faces a high mortality during that year, selection should favor breeding and securing some offspring in the current year (Bell 1980; Kozłowski and Weigert 1986).

Once the female makes the decision to breed another question arises: How to invest her breeding resources? She can produce a large number of neonates of small size or a few offspring of very large size. Large offspring may reach larger size more quickly and should suffer less predation. Larger individuals can also kill and subdue larger and more diverse prey than small ones, an additional benefit of having larger offspring size (Arnold 1993; Shine 1978*a*, 1989). On the other hand, it would also benefit a female to have as many offspring as possible. Thus a trade-off appears between the size of the neonates and the number of them in every clutch (Ford and Seigel 1989*a*; Sinervo and Licht 1991; Stearns 1992). Generally, if the animals have a high mortality early in life it would benefit the mother to have many offspring (*r*-selected strategy); on the other hand if there is a relatively secure environment it is best for her to have few very well endowed offspring (Wiewandt 1982). Thus, the female should optimize clutch size in order to have the largest number of offspring that have a good chance of survival (Kozłowski and Weigert 1986; Lack 1968).

Williams (1966) established the relationship between growth and reproduction. In animals that grow throughout life the amount energy that is invested in reproduction is at the expense of future growth, since the energy used in reproduction will not be available for growth. Any increment in size of the animal will eventually be reflected in larger clutches; thus, any investment in reproduction at any given time is at the expenses of future reproduction. Williams also introduced the idea of the Residual Reproductive Success (RRS), which is the reproductive value of the animal left at any given time of its life. Thus animals with a long life-span, that can produce many clutches in the future should invest proportionally less per individual reproduction. This model predicts changes in the reproductive investment: younger individuals are expected to have a smaller reproductive investment per clutch as a consequence of their small size and their allocation of the energy surplus into growth and reproduction. Older individuals, on the other hand, have less to lose, since they do not forfeit much growth by breeding, and are expected to incur in larger reproductive investments.

For snakes, this topic has been studied both in the field (King 1993; Madsen and Shine 1993a, 1996; Plummer 1992) as well as in captivity (Ford and Killebrew 1983; Ford and Seigel 1994; Seigel and Ford 1992). The available information deals with only a few taxa and limited geographic distributions (Slip and Shine 1988; Shine & Fitzgerald 1995; Seigel & Ford 1987; Madsen and Shine 1996; Shine et al. 1998a, 1998b). This may be the first reproductive study in any mainland neotropical snake where selection pressures leading to size at first reproduction, clutch size, reproductive investment, and the ontogenetic changes are evaluated by using data collected from reproduction of wild animals.

5.2 METHODS

Data collection

Forty-two live animals were collected in the savanna either during the dry season immediately after the conclusion of mating or during the wet season when the animals frequently bask at the river bank and elevated areas (Chapter 3; Figure 5-1). When the animals were not seen mating, the pregnancy was determined by the condition indexes of the animal (Chapter 4). The animals were put in outdoor enclosures each of which contained a pool with enough water so they could dive in and be covered. Smaller animals were put in circular cages, 4-m in diameter with a pool in the middle, and larger ones were placed in 5 m x 4 m cages. Aquatic vegetation (*Eichhornia* spp.) was provided as well a refuge on the land where the snake could hide from the sun and still stay dry. All the cages were leftover from a caiman farming program and were located in the same area where the anacondas were caught. Parallel to this work, a mark and recaptures study of 780 individuals was being carried out. The measurements of 234 adult females from the population were used to calculate the breeding frequency of the population at discrete

Measurements

I took the following measures of all females at the time of the capture: total length (TL), tail length (to calculate the snout-vent length, SVL), and mass (M). After parturition, I weighed the animal again before the next meal. A sub-sample of 5 animals was weighed three times during the pregnancy in order to monitor mass changes during gestation. The sample of animals from which I took weigh throughout the pregnancy was kept small due to the stress and disturbance that the handling meant for the female. Within the first 48 hours of birth I collected the same data from the neonates and determined sex by cloacal analysis. Due to the large number of neonates and time constraints, from 5 of the females I processed only a representative sample (10 individuals).

Parturition and gestation

The date of the delivery was converted to the day of the year in order to have an ordinal variable. I calculated the gestation time by subtracting the date of mating from the date of parturition in a sub sample of 14 individuals from which I had the exact date of both, mating and birth.

Reproductive investment

I calculated the Relative Clutch Mass (RCM) of the animals by dividing the mass after birth by the total mass of the clutch. This measure estimates the proportional amount of energy that the female invests in reproduction. The Relative Investment per Offspring

(RIO) was calculated by dividing the mean neonate mass by the female's mass and multiplying it by 100 to estimate the percentage of her mass that she invests per individual offspring.

Some of the females had many infertile eggs. This was perhaps influenced by the handling of some animals during pregnancy, although the occurrence of some infertile eggs is not uncommon (see below). I consider that the infertile egg size may be used as an estimator of the size of the eggs at the time that they are fertilized. Thus the difference in mass of the average baby size and the average infertile egg size can be used as an estimate of the increase in size of the embryo during the gestation. In this calculation I labeled "embryo" both actual embryo and the yolk associated to it; no further attempt to separate the development of any of these units is made. I estimated the growth rate of the embryo within the womb by subtracting the average egg mass from the average neonate mass of every clutch, and dividing it by the gestation period.

Path analysis

Path analysis is a method for partitioning the correlation among the variables in an attempt to identify the relationships among them. It assumes a causal relationship among the variables based on an "*a priori*" knowledge of the system (Kingsolver and Schemske 1991). For instance, if it is possible to identify a temporal relationship among the variables and label them as "earlier" or "later"; and thus draw predictions among them. This way it is possible to represent them in a diagram with arrows that shows their relationship. Furthermore, it is possible to include in the diagram the standardized partial-regression coefficient on the arrows to show quantitatively the relationship among them. The correlation among the variables can be due to direct effect or to indirect effects. An indirect effect is the product of the path coefficients representing multiple direct effects. The total effect of one variable on another is the sum of the direct and indirect effects of the different paths. The unexplained variance of each regression is calculated by the square root of $1-R^2$, where R^2 represents the proportion of the variance of the dependent variable that is explained by the regressors included in the model, and varies from 0 to 1. It is represented as U in the diagram.

The relationships in the path coefficients were derived from multiple regression models that included all the variables that had a significant contribution to the dependent variable predicted in each model. To build each model I used all possible regressions among the variables that were relevant, and selected the best model based on the one that produced a higher R^2 and used fewer variables. I made sure to include all the regressors whose contribution was significant to the calculation of the regression coefficient and that did not have important multicollinearity effects with the other regressors (Neter et al. 1996).

Breeding frequency

During the first six years of the study I collected females of all sizes and by calculating the condition index I could determine the breeding status of the animals. Two methods were used to assess the breeding frequency. One was by determining the condition index of all the animals caught during each year and then calculating the proportion of animals that bred in that year. This method offers a large sample size but is sensitive to detection bias. Breeding females were more conspicuous than non-breeding ones and thus the estimation of frequency based on it is likely to be biased towards breeding females (Bonnet and Naulleau 1996b). The other estimator was based in those animals that I caught in

consecutive years so I could estimate a breeding frequency of each animal by dividing the number of years that it was breeding by the total number of years that I caught the animal. In the collection of data during 1994 I relied heavily on Rafael Ascanio; some of these data will be published in collaboration with him.

5.3 RESULTS

Measurements

The sample was a representative of all the sizes of the population (Figure 5-2). Notice, however, that there seem to be a pattern that does not match the shape of the size distribution of the total population (Chapter 3; Figure 3-3). A Kolmorow-Smirnov test comparing the distribution of pregnant females ($n = 43$) with the adult females from the population ($n = 167$) show significant difference ($Z = 2.14$; $p = 0.000$) suggesting that the pattern found in the Figure 5-2 involves a legitimate biological trend.

Analyzing the CI of the breeding females I found another inconsistency with my former finding. The CI of the pregnant females before delivery tends to decrease on larger sizes (Figure 5-3; $r = -0.42$ $p = 0.007$; $n = 41$; $CI = 1.04 - 2.75 \times 10^{-4} \times SVL$) but the CI after delivery does not show such tendency (Figure 5-7; $r = -0.12$ $p = 0.54$; $n = 28$). The CI of the females after birth was strongly correlated to the CI before birth ($r = 0.66$; $p < 0.000$; $n = 27$).

Pregnant females moved very little using only 0.001 ha during pregnancy (Rivas unpublished). They spent approximately 70% of their time basking next to the water or inside the refuges that were provided. Initially I provided several species of fowl for feeding the animals, but they refused to eat. Once the females gave birth, however, they resumed feeding immediately, ingesting in one event as many as 8 chickens in a row. Surprisingly, despite the long fast female anacondas lose little mass during pregnancy (Figure 5-4).

Parturition

Parturition occurred at the end of the wet season from October to late December (Figure 5-5). Gestation lasted on average 202.6 days (sd 14.71), however, the time that the female was fertilized is uncertain due to the long time that the mating lasts (Chapter 6). I could collect information of the place where 19 births occurred either by witnessing them or by the place where the remains of the births were found; they were both on the land (12) and on the water (7). For 16 animals I managed to record the time of birth. Most births were in the evening after the peak of heat had passed (Figure 5-6). Most births lasted between 20 to 40 minutes, but some animals took much longer (min = 10; max = 145). In 3 cases that lasted a long time, the females expelled some neonates, or feces and then gave clear signs of distress, such as moving restlessly throughout the enclosure and spinning their bodies on the land or water. On two occasions when this happened there was a large number of stillborns, and I found that the females had some stillborns stuck in the duct. The time of a normal birth is probably between 20 and 30 minutes.

Some of the females had a large number of stillborn young that could be related to having been handled during pregnancy and mating. Some animals were very shy in the enclosures and remained in the water most of the time without coming out to the land to bask like most of the others. These individuals had a very poor breeding success and large numbers of stillborn offspring. In one case, the skin of the female developed a surprising

level of fragility, getting to the point of breaking at the very contact with the hand during handling. This animal died after having a completely unsuccessful clutch.

Relation between the reproductive variables

The path analysis shows the relationship among the variables (Figure 5-8). Notice that SVL and condition, followed by the clutch size, are the most important predictors of the rest of the variables. To understand the interrelationship of the other variables the effect of size and condition must be considered in order to eliminate the influence of these variables and possible trade-offs.

Clutch size

Clutch size was calculated as the sum of all the live and stillborn neonates plus the infertile eggs. The average clutch size of the anacondas was 29.4 but it was strongly correlated with the mass of the female (Figure 5-9; $r = 0.83$ $p < 0.000$; $n = 36$). The single variable that best predicts clutch size is the mass ($Y = 8.98 \times 10^{-4}(\text{Mass}) + 3.14$; $R^2 = 0.82$; $F = 168.8$; $p < 0.000$), but it can be predicted well also by the combination of SVL and condition ($Y = 0.26(\text{SVL}) + 113.4(\text{CI}) - 160.6$; $R^2 = 0.81$; $F = 70.2$ $p < 0.000$).

Relative Clutch Mass

The RCM mass was fairly high of average (Figure 5-10) but it showed high variation in the population (Table 5-1). The RCM can be predicted by including clutch size, SVL and CI in the model ($Y = -2.8 \times 10^{-3}(\text{SVL}) + 0.6(\text{condition}) + 1.19 \times 10^{-2}(\text{Clutch size}) + 1.48$; $F = 7,66$; $p = 0.001$). There was no clear relationship between RCM and SVL other than the fact that RCM was more variable in the smaller females, than in larger ones. However, if we remove the effect of the other variables, it is clear that the RCM decreases with the SVL of the female (Figure 5-11; $r = -0.71$; $p < 0.000$; $n = 20$) and with the female condition (Figure 5-12; -0.27 ; $p = 0.23$; $n = 20$), but this trend is not significant. Not surprisingly, the RCM increases with the clutch size (Figure 5-12; $r = 0.7$; $p < 0,000$; $n = 20$).

Relative Investment per Offspring

The neonates are on average 1 % the mass of the female (Table 5-1); although there is considerable variation depending on the female size. The snout-vent length of the female was strongly correlated to the RIO ($r = -0.9$ $p < 0.000$; $n = 30$; Figure 5-14) and was best predicted in the model by using the clutch size and the relative clutch mass ($Y = -2.48 \times 10^{-2} \times (\text{clutch}) + 2.03 \times (\text{RCM}) + 0.95$; $R^2 = 0.77$; $F = 38.5$ $p < 0.000$).

Predictors on neonates size

Neonates are relatively large at birth compared with the size of other snakes (Table 5-1). There was no significant relationship between the females length and the length of her neonates ($r = 0.25$ $p = 0.20$; $n = 27$) but the mean mass of the neonates was strongly correlated with the females SVL ($r = 0.60$ $p < 0.000$; $n = 30$; Figure 5-15). The neonate mass can be predicted by using RCM, SVL, clutch size and condition in the model ($Y = 0.94 \times (\text{svl}) - 2.95 \times (\text{Clutch}) + 222.6 \times (\text{condition}) + 202.2 \times (\text{RCM}) - 285.3$; $R^2 = 0.64$; $F = 8.9$ $p < 0.000$). The sex ratio at birth calculated from all the newborns obtained in the study was even (1.12 females: males; $n = 437$; two-tailed Binomial $p = 0.25$). However, the average sex ratio from individual females is 1.25 (SD 0.59; $n = 21$) which is a greater difference from 1 (t-test = 1.89; $df = 20$) than it would be expected by chance (Figure

5-16). So, while the total sex ratio of the newborns that are entering the population is close to 1.0, the average sex ratio of the individual clutches is not (Table 5-1). Smaller females seem to have female-biased sex ratios while larger females show a more even distribution. However, this trend is not apparent in Figure 5-16, perhaps due to the small sample size.

Neonates anacondas did not show any sexual size dimorphism (two-tailed t-tests performed on mass; $t = -0.28$; $p = 0.20$; $df = 331$) or snout-vent length ($t = -0.23$ $p = 0.82$; $df = 315$) comparing all the newborns. I also did a Mann-Whitney U test comparing the mass of the males and the females within the same clutch and found no significant difference among them ($Z = -1.32$; $p = 0.189$; $n = 22$). The tail length seems to be different at birth between males and females ($t = -1.98$; $p = 0.05$; $df = 277$). However this difference is not confirmed in a Mann-Whitney U test comparing the tail length of the males and the females within the same clutch ($Z = -1.23$; $p = 0.22$; $n = 20$). Due to the small size of the spurs in neonates, I did not measure them regularly because it was too time consuming to do it accurately.

Stillborn animals and infertile eggs

Normal clutches seem to have from none to two stillborn or infertile eggs. Most births (27 out of 34) had some stillborn and some a few infertile eggs (11 out of 34). Some eggs did not show any development, but during dissection some structures (resembling a small embryo) were identified suggesting that some of the eggs were fertile but did not develop for some reason (see Chapter 6).

Some females showed a high number of stillborns, but this might be related to my interfering with her mating activities or handling during pregnancy. Stillborn mass (avg 214.68) was not significantly different than that of live ones (217.39; $t = 0.76$; $p = 0.51$; $df = 575$), they looked fully formed physically, and did not show any deformity or physical problem other than being dead. It was possible to observe in some individuals infiltration of the bladder content out side of it, perhaps evidence that the animals died a few hours before delivery. The largest animals seem to have slightly higher likelihood of having proportionally lower reproductive success (Figure 5-17), but this trend is not clear. One female was not seen mating at all and she had the most unfertilized eggs (See Chapter 6).

At least 8 of the females studied ate or attempted to eat either stillborn or infertile eggs. Right after birth the female pushes her snout across the mass of neonates, which encourages movement in those animals that did not crawl away right after birth. As she does it, she grabs and eats both stillborn and eggs. Several times a female grabbed a live neonate that was not moving, releasing it hastily when it moved.

Growth of the embryos

Surprisingly, the average size of the eggs (124.6 g) had no relationship to the female's mass ($r = 0.14$; $p = 0.69$; $n = 11$; Figure 5-18) but the growth rate of the eggs (mean 0.44 g/day) was correlated with female's mass ($r = 0.67$ $p = 0.023$; $n = 11$; Figure 5-19).

Breeding frequency

The proportion of captured females that were pregnant during the mark and recapture efforts was 0.57. It seems to be lower in longer animals (Figure 5-20) but a similar pattern than that present on Figure 5-2 seems to appear. The proportion of females pregnant in every year varied across the years and was probably influenced by the areas where the

sampling was concentrated in a given year. For instance, in 1997 much of the sampling was done in the upper part of the módulos looking for breeding balls, investing less time in searching other areas where the non-breeding females are more abundant (Figure 5-21; Chapter 5). The proportion of females that breed in a given year may be related to the precipitation of the area in the two former years (Figure 5-22; $r = 0.66$; $p = 0.16$; $n = 6$).

The ratio (0.57) suggests that more than half of the female anacondas breed in a given year but the detection of animals is probably biased toward pregnant females. Thus it is possible that it overestimates the actual number of pregnant females in the population. I calculated the frequency in which each animal bred, from a sample of 28 females that were recaptured in successive years (17, 6, 3, 1, and 1 in 2, 3, 4, 5 and 6 consecutive years Table 5-2). The average of all the animals turned out to be 0.379. This seems to be a combination of some animals in a biannual cycle and some that breed at a lower frequency (Table 5-2).

I used the animals that were recaptured in more than three consecutive years and bred in at least one of them (to rule out animals that did not breed for some particular reason such as wounds or disease) to calculate the relationship between breeding frequency and size. I found a declining tendency when I graphed the breeding frequency against the SVL (Figure 5-23; $r = -0.81$; $p = 0.005$; $n = 10$). The breeding frequency can be predicted by $\text{Frequency} = 0.863 - 1.34 \times 10^{-3} \text{ SVL}$.

5.4 DISCUSSION

Up to the present all the studies of reproductive biology with large snakes have been using data from harvesting of wild animals (Shine et al 1998*a, b*; 1999*a, b*). This is the first study of the reproductive biology of any giant snake in the wild using naturalistic methods which allows followup of animals over several years.

The lack of activity by the pregnant females is probably due to the large bulk of the animal and the fact that, thanks to their fat reserves, the females do not seem to need to forage. This is consistent with the behavior found in some radio-implanted individuals (Chapter 3). It is surprising, though, that anacondas do not eat during pregnancy despite the expected metabolic investment that gestation involves. A possible explanation for this is the fact that anacondas often are wounded by their prey while subduing them (Chapter 3); and the body and womb of the snake is exposed during the process. To receive a wound in the body cavity holding the embryos might be very dangerous and perhaps compromise the health and success of the clutch. Indeed, long term captive animals at Bronx Zoo, that are regularly fed with euthanized animals, ate throughout pregnancy until just one or to months before delivery (Holmstrom, personal communication) and lost only 22 to 30 % of their mass (Holmstrom and Behler 1981). This supports the idea that wild females are “playing it safe” when they stop feeding. Despite a very large breeding investment, female anacondas still have fat reserves after reproduction as assessed in a few animals found road-killed after the delivery season, and the relatively high CI of some females after birth. Thus, the risk of a wound compromising the survival of the clutch is probably too high considering that the anacondas have enough reserves to survive. A similar behavior of little movement and no foraging during pregnancy has been reported in one other instance in a wild anaconda (Belluomini and Hoge 1957/8) and also in other viviparous (Sazima 1992; Martin 1992) and oviparous snakes (Harlow and Grigg 1984; Hutchinson et al. 1966).

It is surprising that the mass of the anacondas does not change during the pregnancy. There must be losses of energy due to respiration and the metabolism of the female and her embryos. A possible explanation for this is that the losses in metabolism are balanced out by adding water in the conversion of fat tissue and yolk into flesh and live tissue of the neonates.

Is it possible that anacondas reduce their basal metabolism to some sort of lethargy during pregnancy (hence the lack of activity and foraging) and, thus, behave like living incubation chambers for their brood? I do not have any data on the physiological state of the female, but the lethargic behavior of the animal suggests that it is a plausible hypothesis. The closest related species that has been studied are pythons and they use a completely different strategy. They provide heat to the eggs by producing continuous muscular contractions in a type of twitching that generates the heat needed for the incubation of the eggs (Ellis and Chappel 1987; Harlow and Grigg 1984; Hutchinson et al. 1966). Future studies on the physiology of the pregnant anacondas should address this question.

Parturition

Duration of pregnancy was relatively variable, perhaps due to uncertainty in assessing the actual time that animals were inseminated and the uncertainty in assessing when embryonic development begins. Anacondas mate for a whole month, in some cases, (Chapter 6) and it is unlikely that the development of the embryos starts as soon as the mating occurs, since it would mean a developmental difference of several weeks within the same clutch. It is likely that the sperm is stored during the mating and it is used after the mating finishes. Short-term sperm storage seems to be relatively common among snakes; some pitvipers regularly exhibit short-term sperm storage from fall to spring (Schuett 1982, 1992).

Females gave birth mostly in the evening or afternoon (Figure 5-6). This might be driven by two non-mutually exclusive reasons. One, a proximate explanation is that the females need the high temperature of the day to trigger the energetic demands involved in the delivery, and another, ultimate, reason that the neonates have a better chance of survival at night time when flying predators are not as abundant and the odds of dispersing safely are higher.

The feeding of the females on the stillborn and unfertilized eggs has been reported before (Holmstrom and Behler 1981). A likely explanation for the observed phenomena is that the females are recovering part of the energy invested in the offspring after the long fast and energy expenditure that being pregnant entails.

Relatively similar number of animals gave birth on land and in water. Neonates tend to go into the water and very seldom use dry land (Rivas, unpublished data). The most likely scenario is that the females give birth in shallow water or at the water's edge from which the neonates disperse.

Pregnancy tends to be a very critical moment for the health of the female. Four of the animals I had in captivity died during pregnancy without any apparent reason other than minor wounds present at the moment of the capture, wounds which would have not killed a healthy non-breeding animal. After parturition, the females often look very weak and thin, and may be more likely to be predated (Chapter 3; Rivas et al. 1999). Animals in this

condition may attack larger, more dangerous prey in order to overcome their energetic deficit, taking the risk of being injured or even killed by their prey (Chapter 3; Rivas 1998).

Anacondas give birth at the end of the wet season. This seasonality is not surprising given the great influence that the two extreme seasons (wet and dry) have in the life of most organisms that occur in the llanos. Just after birth, the neonates have little time to grow before the onset of the dry season. The peak of the dry season is the least favorable for the survival of the neonates, since there is less water and the newborns have fewer refuges to hide from aerial predators. Furthermore, other predatory animals such as caimans, foxes, storks, herons, and tegus concentrate around the bodies of water. It seems that it would be more convenient for the anacondas to give birth at the beginning of the wet season so that the newborns would have a longer time to grow and reach a size such that they more easily can fight off predators. On the other hand, due to the low growth rate of the neonates (Chapter 3), it is possible that any increase in survival derived from being born a few months early is not enough to produce an important selection pressure. Thus, the survival of the newborn may have less influence on it, as compared to the survival of the female. After a large reproductive investment it is on her best interest to find food and recover from the breeding investment. The beginning of the dry season is a very good time for the female to find food due to the above mentioned concentration of animals around the water bodies. Predators of newborn anacondas are potential prey for adult females.

Anacondas are not the only large reptile of the llanos whose offspring are born at this time of the year. Spectacled caimans have a similar reproductive timing. In fact, it is possible that the timing of the newborn caimans might be a selection pressure leading to the reported timing for anacondas. Newborn caimans use a habitat very similar to that of newborn anacondas and do not overlap in trophic niche. A newborn caiman (from 40 to 50 g) represents between 20 to 25% of the mass of a newborn anaconda, and this is a very common prey size relationship for young snakes. I have not found any newborn caiman among the diet of baby anacondas; however, the representation of this size class of snakes in my sampling is very small. Newborn anacondas seemed very interested in the cotton swabs rubbed on newborn caimans in the predation trials done in captivity (Chapter 3).

Another possible explanation for the seemingly inconvenient time for being born from the offspring's perspective, is that it is simply a consequence of the timing when the mating season occurs and the incubation time. Considering this level of analysis mating season occurs in the dry season as a consequence of the high encounter rate between males and females (Chapter 6) and the neonates are born at the end of the wet season as a consequence of the gestation time needed to develop the embryos.

Clutch size

Not surprisingly, clutch size is strongly correlated with female's size (Figure 5-9). This trend is expected in animals with indeterminate growth and has been previously reported in other snakes (Ford and Karges 1987; Ford and Siegel 1989*a*; Madsen and Shine 1996; Siegel and Ford 1987; Shine 1994*a*, Shine et al 1998*a*, 1999*a, b*). Larger animals have larger coelomic cavities and more room for the production and development of larger clutches (Shine 1992). Not only the size but also the condition index is important in determining clutch size. This is expected since animals with more fat reserves are expected to have a larger surplus of energy to invest in reproduction. A similar trend where the condition is important in the clutch size has been found to occur in studies from the wild (Madsen and Shine 1996) and in captivity (Ford and Siegel 1989*a*, 1992).

Relative Clutch Mass

The relative clutch mass was lower, but close, to the average value (0.57) found for other species of snake (Seigel and Fitch 1984), and within the value found for other large-sized snakes (Slip and Shine 1988; Shine et al. 1998a). The RCM is expected to be lower in viviparous aquatic species due to physical limitations for swimming if the rear is bulky and heavy; as happens with a large clutch (Siegel and Fitch 1984; Shine 1988). Furthermore, animals that have a longer life span are expected to have lower reproductive investments per clutch since the animals have a larger residual reproductive success (RRS) after every single breeding event (Williams 1966).

The average RCM in anacondas seems to decrease with the size of the snake (Figure 5-11). This does not support the idea that young individuals partition their energy investment into one part for maintenance and growth, and another part for reproduction; such a strategy should have resulted in an increase in the RCM with the size. If the larger animals do not forfeit future growth or breeding they are expected to have a relatively larger reproductive output (Williams 1966). The reproductive investment in young (smaller) animals is more variable than that of larger animals (above approximately 330 cm SVL). This is not expected for animals that do not have much benefit of saving energy for growth. On the other hand, if smaller anacondas have a larger risk of being predated (Chapter 3; Rivas et al. 1999) then saving energy for future growth would not be adaptive and the best strategy would be to make very large breeding efforts when possible. The CI of the animals is inversely correlated with the RCM (Figure 5-12). This is counterintuitive since animals with higher condition are supposed to have surplus energy to make a larger investment than animals in lower condition. A possible explanation is that part of that higher condition of the animal is not due to fat storage or yolk only, but to flesh and muscles that increase the mass of the females and are not available directly for reproduction (Madsen and Shine 1996). Notice that the females with higher CIs also have higher CIs condition after the breeding event (Figure 5-7), so the amount of energy used in the reproduction is not dependent on the condition, but it is based in other variables. The fact that the CI after birth is correlated to the CI before birth further suggests that the relative investment of energy for reproduction is not dependent on the condition of the animal; this is similar to the findings of Ford and Seigel (1989b).

Condition index

Contrary to the trend found in the general population of anacondas (Chapter 4), the condition index (CI) of the population of pregnant females decreases with their size (Figure 5-3). This indicates that larger animals are breeding at a lower CI (relatively thinner) than the smaller animals. This is not a trend of larger sizes alone since in the graphs with animals from the all sizes classes this trend is not present (Figure 4-3). It is possible that the larger female are constrained by their mass. A very heavy snake might be constrained in crawling on dry land, which anacondas must often do in the seasonal savanna. Thus, as the female grows in length she is constrained by the maximum mass that her body can carry on land. I believe that not having a rib cage (for the lack of a sternum) there may be a mass limit where the mass of the body might constrain breathing while the animals is on land. This consideration might set an upper mass limit that forces the females to grow thinner as she grows longer. Another possible explanation is that larger animals only need to gather the energy they need to breed and need not reserves for growth and, thus they can breed as soon as they have the minimum surplus of energy to do

so. However, they would benefit of breeding with higher CI, since they could produce larger clutches (Figure 5-8).

The lower condition of larger females may also be related to the lower breeding frequency found. Another explanation in a different level is that larger females might take longer to gather the energy they need and thus, they need a longer time to recover from a breeding event and also to attain a high CI. Larger females might also have a harder time coming across the amount of energy needed for reproduction. Larger *Liasis fuscus* have been found to have problems gathering the amount of energy they need to be able to breed with the result that the larger individuals do not breed as often as the younger ones and the largest might not breed at all (Madsen and Shine 1996). Due to the large amount of potential prey found in the llanos (capybara, caiman, turtles and wading birds) in the very areas where the anacondas occur, it is hard to believe that food availability might be limited.

Larger animals have a different diet than smaller ones. The latter feed primarily on birds and the larger snakes feed mostly on mammals and reptiles (Chapter 3). In anacondas there is the added cost of healing from the wounds that larger snakes are more likely to experience (Chapter 3). It is also possible that larger animals cannot catch birds quite as easily as smaller ones as the female have become much heavier. To feed on larger prey might involve a lower rate of feeding, since it takes longer to catch, to process, and to digest them. Larger prey are less common than smaller ones, and the processing of them must take longer simply by virtue of the larger mass to be digested, and the scaling of the digestive process (Calder 1996); this could explain both the lower CI and the lower breeding frequency. Alternatively the amount of food and energy in a bird might no longer compensate for the metabolic investment of digestion for larger females. Large snakes that practice a sit-and-wait strategy of feeding and regularly go for long time without a meal, have developed certain adaptations to that kind of feeding strategy. Side-winder rattlesnake (*Crotalus cerastes*) can undergo atrophy of the intestine and digestive organs in order to lower the metabolic expenses of an idle digestive system. Upon feeding there is a tremendous metabolic expenditure in regenerating the atrophied organs in order to trigger digestion (Secor et al. 1994). Burmese pythons (*Python molurus*) can experience an increase of 44 times its standard metabolic rate while digesting a large meal (Secor and Diamond 1997). It is a testable hypothesis that the energy contained in small birds does not pay for the expenditures of triggering reproduction. Birds are a very lean prey item that maintains low body fats due to the constraints that flying imposes to the body mass. Thus, although abundant in the llanos, birds might not be very profitable for a large snake, and feeding on a bird might forfeit the chance of feeding on a more profitable prey item. This may explain the reported apparent switch in diet (Chapter 3). Although, larger snakes have a lower relative energy expenditure per unit of body mass, their absolute metabolic expenditures are considerably larger than those for smaller ones (Secor and Diamond 1997). Shine et al (1998b) found a similar switch in the diet of reticulated python (*Python reticulatus*) in approximately the same size classes. They also found a decrease in the feeding and breeding frequency of larger females (Shine et al. 1999b).

If it takes longer for a large female to reach a very heavy condition; it is possible that as soon as she is fat enough to breed, she is found by males who start courting her. The courtship and mating itself can play an important role in inducing ovogenesis (Whittier and Crews 1989; Whittier et al. 1987). This phenomenon might happen more with larger

females than with smaller ones, since larger females are more sought out by the males (Chapter 6), and smaller females may need less food to reach a high condition. Support for this idea is that Olivia (a very large female) mated in 1995 with a lower condition than the threshold for reproduction (8.5 short of 8.75). Olivia was found the following year extremely weak, with many capybara wounds. Shortly afterwards, she was attacked and killed by a caiman no larger than other caiman that I had seen her eating (Chapter 3; Rivas et al. 1999). If the ovulation is induced by the male's courtship or mating, then it is possible that a female being marginal enough to breed can be induced into an inconvenient breeding event. Olivia was a very large female, with her skin all covered by scars. She did not grow at all in the five years that I followed her and was, perhaps, a very old individual. She gave birth in the wild so I could not collect reproductive information such as RCM or clutch size. The question remains as to whether she made an extremely large breeding investment (perhaps suicidal since her RRS was low at her old age) that produced her weak state, or whether she was simply too thin to breed (as suggested by the condition index) and mating in that year was a wrong decision.

The idea of a maladaptive mating induced by courting males contradicts the conventional wisdom that female snakes emit pheromones to attract the males (Ford and O'Bleness 1986; Ford and Low 1984). However, it is a working hypothesis for a species that is probably not fully adapted to the llanos and might have evolved in different environments (see below).

There are some fertility-independent costs of reproduction such as the long fast of the females, the weakness associated with the postpartum activities, and complications giving birth (see below), among others (Madsen and Shine 1993a). If females skip reproduction for a year they would be able to produce an even larger clutch due to their higher CI (Figure 5-8). To produce a larger number of offspring would reduce the fertility-independent cost of reproduction for the female making the breeding investment more profitable. Why are the larger females going into reproduction as soon as they reach breeding condition instead of gathering resources for the following year, when they could have larger clutches? It is possible that the females breed as soon as they meet minimal conditions in order to avoid the risk of dying while foraging on dangerous prey. To die at that point without breeding in that year would involve losing a clutch that was already large, and the increase of clutch size for having a higher condition might not be high enough to compensate for the risk. It is likely that the females are playing it safe when they breed in a relatively thin condition.

The lower condition found in larger females could be related to the apparent decrease of the breeding investment in larger animals (Figure 5-11). The switch in biomass makes the snake switch to a different temporal scale in which they need more time to gather the surplus of energy needed. The reproductive effort (RE) must be measured considering three aspects: 1) the potential cost of fecundity (which RCM measures), 2) the time needed to gather the energy, and 3) the reduction in the survival rate that the female suffers by reproduction (Shine and Schwarzkopf 1992). In light of this interpretation larger females could have a comparable RE than smaller females despite the apparent difference in RCM.

Relative Investment per Offspring and size at maturation.

Neonate anacondas are relatively small in mass compared to the size of the females. This RIO is comparable to the RIO reported in other large-sized species (Madsen and Shine 1996; Shine et al. 1998a). However, they are at birth the size of adult of many

species of snakes. The female would get most benefit from making as many neonates as possible. Why is she not producing many more smaller animals? One explanation is that the neonates need at least a minimum size in order in order to gather the food they need in the llanos. Data that I have collected show that they prey on small birds, such as *Jacana jacana* (approximately 70 g), and small passerines associated with the water bodies (Table 3-4; Chapter 3). The snakes also could catch neonate caimans (from 40 to 50 g). The mass of a neonate anaconda is 200 g at birth so a prey of 40 to 70 g is a relatively convenient size for it to eat (Secor and Diamond 1997). Neonates might not be able to catch fish due to the fast movements of this prey item (Chapter 3). Another possible prey item that I have not found in the diet, but are very abundant in the areas is juvenile turtle (*Podocnemis vogli*). *Podocnemis vogli* are small prey items but with a particular shape that make them very difficult or even dangerous to swallow if the snake is not large enough (Chapter 3). I believe that if the neonate anacondas were smaller they would have a very hard time coming across the food they need and probably they would have a very high mortality. Thus the minimum size of the neonates might be a selection pressure influencing the RIO that the female exhibits, and the minimum size at maturity. The fact that smaller females have a higher RIO than larger ones could be a consequence of the minimum size that the neonates require in order to be viable.

There is another possible explanation that is on a different level of analysis: phylogenetic correlation. The selection for large size on some sector or stages of the population produce changes on other traits that are not the actual target of selection (Halliday and Arnold 1987). Females may be very large for several selected reasons and the large size of the neonates is thus simply a consequence being born to a very large female.

Larger females, with larger oviducts, should be able to produce larger eggs. However, RIO decreases with female's size. The females could produce larger offspring that have a better chance of survival instead, they produced larger clutches. Perhaps the females "choose" to make smaller neonates and produce larger number of them turning into a more r-strategy (Lemen and Voris 1982). The decrease in RIO does not seem to jeopardize the theoretical chances of survival of the offspring; its larger size allows the female to incur in a smaller investment per offspring while still producing larger offspring than the smaller females. This is a similar trend to that found in *Thamnophis butleri* (Ford and Killebrew 1983), several marine snakes (Lemen and Voris 1981) and in Australian elapids (Shine 1978b). Smaller females need to produce comparatively larger offspring which produces an apparent ontogenetic switch in RIO. Thus the minimum size of neonates might be a selection pressure pushing upwards the age (and size) of the females at maturity. This decrease in RIO suggests that the females are optimizing the clutch size producing as many offspring as possible that have a good chance of survival (Lack 1968).

Stillborn animals

Stillborn animals were fully developed and their size did not differ from live ones. The reasons these individuals died before birth are not clear. It is likely that the female had problems expelling embryos and they are asphyxiated in the womb. Animals that give birth late seem to have a higher likelihood of having many stillborn offspring. This suggests that such females have some problems delivering.

Belluomini and Hoge (1957/58) report on a wild caught female anaconda that had problems giving birth in captivity. They performed a cesarean surgery and found many

stillborn in similar conditions to the ones I found; they also found a few individuals that were alive. Ford (1980) found that animals that had clutches larger than average were at a higher risk of having stillborn or developmental anomalies. The relatively larger proportion of large females that had lower reproductive success (Figure 5-17) could be attributed to this problem. In some cases I believe that the handling of the animals might have influenced in the production of many stillborn. Some of the females acted very shy during pregnancy, spending much time under the water, and might not have basked as much as needed to develop the clutch. This problem has been reported to lead to dystocia, the death of the clutch, or even the female (Ross and Marzec 1990).

Egg size and embryo's growth

Egg size is surprisingly similar among the females of varying sizes. There does not seem to be any clear tendency. If anything, we seem to appreciate the same pattern that we see in the RCM where smaller females have a larger variance than larger ones (Figure 5-18). While the neonates from larger females are larger than those from smaller ones, this trend is not present in the eggs. This involves a differential growth rate in embryos of females of the different sizes. Indeed, the growth rate of the embryos from larger females was faster than the embryos from smaller ones (Figure 5-18). The different in size of neonates that developed from eggs of equivalent size suggests that the females transfer nutrient differentially to the neonate in the embryos through some sort of placenta (Stewart et al. 1990).

Why do larger females produce relatively smaller eggs than small females? Allometric growth of the reproductive organs would predict that the embryos from large females would be proportionally larger (Huxley 1932). Furthermore, production of a placenta for the development of eggs and additional provisioning of it means an extra investment of energy and structures. It is possible that the females do not invest in larger eggs in order to not have too many resources committed in some eggs that might not be fertilized, since the females do not seem to be able to reabsorb the energy invested in the unfertilized eggs. The fact that some eggs were fertilized but did not develop could involve two different phenomena. The first could be incompatibility between the sperm and the egg (Zeh and Zeh 1996), and the second, the possibility that the females might exert control on the paternity of her clutch by developing selectively some eggs (see Chapter 6). Both of these are fascinating possibilities that deserve further attention.

.Breeding frequency

It is not easy to answer the question of how often an anaconda breeds. It seems certain that they do not breed every year. The only two cases where that situation was suggested by the condition index were in very young individuals that perhaps skipped reproduction in the first year (Table 5-2). The fact that parturition is at the end of the year (Figure 5-5) and the mating is early in the year (Chapter 6; Figure 6-4) gives very little time for a female to find, process and assimilate enough food to engage in a new breeding event. Furthermore, it has not been possible to breed animals in captivity that eat *ad libitum* in two successive years (Holmstrom personal communication and Strimple personal communication). Although some animals do breed every other year, it is not the rule. Some other individuals seem to take a longer time to recover and engage in a new reproductive event. Some animals skipped reproduction for three years and bred in the fourth (Olivia) and some other animals did not attained breeding condition for four straight years (Chinga)

(Table 3). Chinga was missing the tip of the snout which was an open wound. This probably affected her foraging efficiency and she might not have been able to gather the energy needed to breed. A biennial pattern in reproduction of other snakes have been suggested or found in several species (Madsen and Shine 1992, 1996; Martin 1992; Sazima 1992). Decrease in breeding frequency with the size also has been reported (Madsen and Shine 1996; Shine et al 1998a, 1999a, b) and it even seems to be the rule in large-sized species.

Younger individuals that benefit by growing more might be expected to breed less often than larger ones, but this is not supported by the data. Smaller individuals breed more often than larger ones (Figure 5-20). This might be related to the lower CI found in larger females and perhaps due to a lower feeding frequency (see above). Furthermore, the fact that smaller females end up with larger condition indexes after birth than larger females suggests that they might need less time to recover to optimal breeding condition.

The sample of the population illustrated in Figure 5-2 shows two peaks, where females of snout-vent length of approximately 3 and a half meters are not very abundant. This is consistent with the decrease in the proportion of pregnant females found in the total sample at this size interval (Figure 5-20). Here I summarize four possible non-exclusive explanation for this findings. First, it might be a consequence of their development. Thus, the first peak one sees is when females just reach adulthood and initial breeding size. The next gap in breeding females corresponds to the year (or years) after breeding while the animals are recovering and also growing until they reach the next size class. The difference in the heights of both peaks does not support this idea since the number of females in the older peak is much lower than the former one. This would necessitate high mortality between both peaks which is not supported by the data. The growth rate of the animals in general is not fast enough for the females to move from one peak to the other (in one year or a few; Figure 3-7).

As second reason for the bimodal distribution is that there may be a switch in strategy, whereby smaller females breed as often as they can until they reach a size when the growth they can achieve in one year increases their reproductive output sufficiently to make it worthwhile to skip reproduction (Figure 5-1; Bell 1980; Kozloswki and Weigert 1986). However, at this point, not enough is known about growth rate of every size class to evaluate this hypothesis. However, Figure 5-9 does not suggest a change in strategy or any particular increase in fertility that could be related to it.

The third possibility is that females start breeding when they are large enough to produce viable offspring; it would not pay for her to skip reproduction to grow to a larger size because of the risk of being eaten by caiman or other predator (Chapter 3). Healthy females larger than 3.3 meters are not likely to be eaten by any predator; thus, after the females reaches this size it would pay for them to skip reproduction for a few years to reach a much larger size and have larger clutches.

Finally, the fourth possibility is that, the gap of breeding females between 3.3 to 3.8 m could be a byproduct of energy intake, since larger animals feed on more dangerous prey than smaller animals. The switch in diet from birds to mammals and reptiles seems to coincide with this gap in reproductive females. At the body size at which an anaconda switches her diet for energetic, foraging, of other reasons, a females might go through a period when she cannot find the right amount of food due to her lack of experience in finding, subduing, or ingesting it. Consistent with this gap in reproductive females and the

decrease in breeding condition is the finding that after 3.3 meters the snakes seem to have more wounds than before (Figure 3-15). A detailed study on the diet of anacondas as well as the energetic benefit of different prey items is needed to shed light on whether the gap is due to the switch in the diet. These four hypotheses are not mutually exclusive and some of them could be acting simultaneously. The idea that the first peak is due to some animals that breed early to ensure some offspring due to the risk of predation, and the hypothesis that the gap is due to a dietary transition are fully compatible and deserve future attention.

The proportion of females that breed in a given year seems to be related to the precipitation of the two former years. It is probably related to an increase in the density of prey that benefit by abundant water such as capybara, caimans, wading birds, and the fish that the wading birds feed on. The reason that the relationship is with the amount of precipitation for the two former years is because the female might need to forage for two years to gather the energy needed for reproduction. Furthermore, the increase in biomass that benefits the anacondas might be delayed due to the time needed for the prey population to increase.

5.5 GENERAL DISCUSSION

Selection for large size

Large size is normally constrained by high mass, high cost of mobility, and a conspicuousness that both constrains stalking prey and increases detection by predators. The aquatic environment, where anacondas live, diminishes all those problems. The rich fauna found in the area provides abundant food to sustain a large biomass of predators. In the case of anacondas, larger size offers more advantages besides the above mentioned benefits of increased fertility, such as larger number of prey available and resistance to times of shortage. Furthermore, the fact that anacondas are live bearers confers a stronger selection pressure for a large coelomic capacity (Shine 1994b).

The following scenario might explain the large sizes obtained by anacondas, especially females. Females need to produce relatively large offspring in order to enhance the offspring's survival by escaping predators and subduing sized prey items. This pushes upwards the size of the female at first reproduction, and might be an important reason leading to large size in anacondas. Skipping reproduction to increase future reproductive output has been found in other species such as *Elaphe guttata* (Ford and Seigel 1994). They found that animals that skip reproduction and thereby increased growth have a higher reproductive output after the third year than animals that breed at an earlier age. This strategy of delaying reproduction increases the LRS only if the survival to older age is high. If, on the other hand, the female suffers high mortality during the main growing period she is better-off by breeding as soon as she reaches the minimal size to breed. Several fertility-independent costs of breeding such as risk of death for dystocia, long-term fasting and weakness after birth; along with the minimum size needed for the neonates to survive, may lead the size of first reproduction upwards.

Larger females produce larger clutches of larger individuals at a lower relative total reproductive investment (RCM) and investment per offspring (RIO) than smaller animals. This benefit for large size might contribute to selection for large body size in females. Notice that fertility of the females does not plateau at the larger sizes (Figure 5-9). This suggests that females would benefit by reaching even larger sizes. However, the breeding frequency decreases with the size of the female so a point might be reached where the

benefit of larger clutches might be balanced out by the decrease in frequency. The optimum size for the females would be to have a large size so they can have larger clutches, but remain small enough to find their food supply, locomote easily, and breed regularly. Are the females growing too large for their own good? I calculated the reproductive value of a female at any given time and labeled it Current Reproductive Value (CRV). I calculated it by multiplying the expected breeding frequency for her size by the expected clutch size for her mass. The CRV of females increases with body size despite the decrease in breeding frequency and the relative decrease in clutch size due to the lower CI (Figure 5-24). Not only would females benefit from being larger by increasing the number of offspring they can produce, but larger animals would perhaps also be able to eat larger prey and subdue them easier and with less risk. If an anaconda grows sufficiently large, she would also benefit from increase availability of prey such as pecaries (*Pecari tajacu*, *Tayassu pecari*) tapirs (*Tapirus terrestris*), *Podocnemis expansa*, and *Crocodilus intermedius*. The two latter species were surprisingly abundant historically (Humboldt 1799-1804/1985).

So, why are not the females even larger? The following reasons support for an even larger size in female anacondas: First, the high abundance of prey and their concentration in dry season, which suggests that yearly food supply may not be a problem. Second, due to their aquatic habitats and crypsis, the problems of conspicuousness to their prey, that other predators have, do not seem to apply either. Third, really large size animals might be predator-free, so being detected by predators is not a selection against large size. I have discussed how parasites can affect the animals, and as the time passes, the individuals is expected to reach a larger load of them just as a consequence of the longer time that she is exposed; however, to date there are no field data to document this. One possible limitation to a larger size is the long time that it takes for them to reach it. If animals suffer some size-independent mortality over the years (such as that produced by parasites or accidental injuries by their prey) its life expectancy decreases as it grows older, so the upper range of sizes found is perhaps a direct consequence of their ages. Another possible limit may derive from the seasonal availability of their food supply. Although there is abundant prey item in a given part of the year, the strong seasonality may prevent the females to feed regularly thus imposing on the animals an intermittent feeding schedule that may decrease they growth rate, and also make less profitable the prey consumed (Seccor and Diamond 1997).

How large can an anaconda be?

The maximum size that anacondas can reach has been subject of longstanding debate among herpetologists. There are many accounts of snakes around 9 to 11 meters (Gilmore and Murphy 1993; Beebe 1946; Blomberg 1956, Pope 1961, among others). A lot of the controversy concerns the credibility of the records, the confusion created by the fact that the skins stretch when the snakes are skinned, and animals whose length was merely estimated or measured with unreliable methods. I do not intend to revisit that discussion, but it is striking that the largest snake that I have caught, out of 780 animals, is only a little more than 5 m. What is the reason for such a difference? Why have I not found any animal anywhere near 9-11 m? The slow growth rate of the anaconda (Chapter 3) requires a long time to reach a large size. My study area is a cattle ranch where the protection of wildlife is a recent practice (last 15 years). Presumably the really large animals might have been killed off earlier and the animals that occur now in the areas where I studied

(relatively close to human activities) might have not had enough time to grow to really large sizes. One important gap in the knowledge needed to address this issue is the longevity of the species. There are no good records of longevity and no field data to document it. In captivity it has been reported that they can live more than 20 years but this record was from an animal obtained as an adult (Snider and Bowler 1992). I will address the questions regarding maximum size in the rest of this section using the information that I have gathered.

Measurement errors

I discussed in chapter 3 the problems of measuring a large snake and how the reliability of the measure is low if the animal struggles and when the measure is taken by people without the proper experience. Doing it following a tight methodology I found that the measuring error could be as high as 5% of the length of the animal which translates into 45 cm in a snake 9 meters long. This could be even higher when measuring a very large snake that is not muzzled by people who are struggling with the animal as they try to stretch it. However, I doubt that this can account for the large difference found.

Constraints on large body size

The condition index of larger females decreases with body size. I have argued that mass can be a problem for animals when crawling on dry land, so larger animals might be constrained by their mass. Larger females might alternatively have problem coming across the energy they need to reach the breeding condition (see above). It would not pay for the females to grow to a size where she cannot find enough food to breed. It would not pay for her either to be so large that her mass would be unbearable (and risky). Regardless of the reasons, extrapolating the decrease in condition index I found that a female with 600 cm snout-vent length (approximately 670 cm total length) would have a condition index of 0.875, which is the cutoff below which females do not breed. In other words, if this trend holds for larger animals, a female larger than 6.7 meter would never reach breeding condition. Thus, there would not be any reason for the female to reach this size. However the theoretic limit (670 cm TL) and the actual maximum (521 cm TL) are still far apart.

Assuming that larger females do attain breeding condition, I also calculated the decrease of the expected breeding frequency with size using the regressions presented above. The expected breeding frequency of a female of 8.46 m total length is zero; this size is reasonably closer to the reported values; and the difference could be explained by the measuring error. However, does it really pay for the female to be that large? I calculated the CRV of the animals extrapolating the breeding frequency and the clutch size from the data collected (Figure 5-25) Notice that the CRV peaks slightly before 5 meters of SVL. Natural selection would not favor any increase in size beyond that point. This is about the largest SVL that I found (477 in the total population; Table 3-1). Thus largest range of sizes is optimized to increase the number of offspring than the female bears. Figure 5-25 summarizes the selection pressures affecting female size.

A five meters limit for female size is consistent with the calculation that Pritchard (1994) made about the maximum size of the snakes. In a revision of the size at maturity and maximum size of several North American snakes, he found that the maximum size of the snakes was about 1.5 to 2.5 the size at maturity. The minimum size of a breeding anacondas that I found was 210 cm SVL (Table 6-2) so the expected maximum size of the anacondas following this pattern would be 525 cm.

Explaining the records in the literature

How does one explain all the records of much larger animals documented in the literature? The above analysis suggests two limits on the development of larger size. One is the decrease in the condition index that sets a 6.7 m limit above which the females does not reach breeding condition. The other is the decrease in breeding frequency as the female grows larger that makes it unprofitable for the female to grow beyond 5 meters. Both limits are probably dependent on the environment and are likely to be less important when the females live in more permanent water or in an area without a long dry season. For instance if the anaconda lives in a river or deeper lagoon, or in the aseasonal rainforest, she would not face the constraints of gravity and hence the limitations of mass to crawl on dry land would not exist. Both the decrease in CI and breeding frequency might well be related to a decrease in the feeding frequency or a lower supply of highly profitable prey items due to seasonality. Will a very large female have enough food intake in the new habitat to reach breeding condition and a relatively higher reproductive frequency in more permanent water bodies? I have shown that the amount of water during the previous two years has an influence on the breeding frequency of the anacondas (Figure 5-22), so that the hypothesis that breeding frequency is higher in more wet areas is plausible.

In the savanna, a large female might face some limitation due to the strong seasonality. Early in the arrival of the dry season there is a period of high prey encounter rates as the water level drops over a period of 1 or 2 months. During this time the female might be able to take one or two good meals, but shortly after this, the peak of the dry season arrives and conditions turn overly hot and dry, forcing the female to seek refuge from the heat to places where she is perhaps unlikely to find food. After the wet season starts, the prey encounter rate drops dramatically. With all the savanna flooded the anacondas might not encounter many prey during the wet season either. So the prey availability of the llanos provides some food intake, but this might not be constant. This restricts females to a relatively small number of prey every year, and also imposes the high metabolic expenditures that their switch in metabolism involves (Secor and Diamond 1997, 1998; Secor et al. 1994).

Although predators are not a problem for large females, living in more permanent water body might be safer for large anacondas too. They would not be exposed to overheating, since they do not have to crawl on dry land (Chapter 3). If an anaconda attacks prey in, or near, a river, she can drag it down to the water with less risk of being attacked by the relatives of the prey (e. g. capybara). The risk of being wounded during predation is relatively high for anacondas in the savanna, (chapter 3). Bringing the prey to an unfamiliar environment where the mobility of the prey is constrained may be a further strategy to increase the efficiency of the subduing behavior (Rivas and Andreadis, in prep.). Thus, in a river habitat (or neighboring oxbows and lagoons) a large female might be able to locate their food in a safer manner, allowing her to use her energy and skills in a more efficient way, since there would be less risk of injury.

There are other selection pressures for the female to grow larger in the rivers as well. A 5 m long anaconda can kill and subdue all the native animals occurring in the savanna so there would not be any advantage in the food intake for the female to grow larger. However, in the rivers and associated gallery forest live larger and more diverse prey items such as pecaries, tapirs, crocodiles, and Arrau turtles that can be important food sources. A larger size would benefit the females by enabling them to capture those prey. Also other

less dangerous (non-social) rodents such as agoutis (*Dasyprocta* spp) and pacas (*Agouti paca*) become available on a more regular basis than in the savanna. An animal living in rivers cannot take advantage of a season of high concentration of prey when the female anaconda, an ambush hunter, has a high probability of encountering some prey. Instead, animals living in the more stable but prey-unpredictable environment probably must take advantage of more diverse prey (size and species). So, larger body size would benefit females by increasing their range of potential prey. Also, in more wet places where smaller anacondas might have less predation due to the lower encounter rate with predators, the females would have less mortality at early ages. Thus females in more permanent water bodies would be selected to grow large and also would have the food supply and added safety enabling them to grow to larger sizes and live longer. Due to the search method I used, I could not catch or find animals that used deeper waters, so the decrease in the condition index and breeding frequency could be particular traits of the animals that live in the hyper-seasonal savanna.

If these assumptions are correct I can make the following predictions. First, females in wetter habitats would start breeding at a larger size due to the lower encounter rate with predators in the dry season, which would make it more profitable for an “adolescent” female to keep growing in order to produce larger clutches. Indeed, a female that was born in captivity and fed *ad libitum* throughout her life started breeding at 3.1 meters svl (3 years of age; Holmstrom 1982), much closer to the second peak of Figure 5-20. Second, females would produce larger offspring at maturity, since the neonates would need a larger size to cope with the unpredictable food supply. Again this new selection pressure can increase the size at reproductive maturity. Third, females would have a more uniform condition index across different sizes of reproductive females; alternatively, the decrease in condition with size would be less steep, since they are not constrained by gravity. Fourth, females would maintain a higher breeding frequency as they grow since they can forage throughout the year. The breeding frequency would probably still decrease, since the amount of food needed for larger animals increases with the size, and the time to gather it should increase proportionally. As a consequence of all these predictions, I expect the females to reach larger size. In fact, the model described in Figure 5-25 is very sensitive to changes in the breeding frequency so the optimum size can easily shift upwards if the snakes live in different environments. Indeed, most of the records of very large snakes come from rivers (Gilmore and Murphy 1993). Furthermore, the opportunistic capture of a female anaconda in the Tiputini river (Ecuadorian rainforest, Napo province) supports these ideas. The individual caught (Silvia) was 522.8 cm in TL and 459.8 cm SVL. This is the second largest individual in SVL in the 8 years I have been studying them. The one animal caught in the rain forest was larger than 779 of the individuals caught in the seasonal savanna. Furthermore Silvia presented only 13 scars total which is remarkably lower than the expected for an animal of its size (Figure 3-15) supporting the hypothesis that animals in more aseasonal habitats face less risk of wounds from their preys.

Notice that human activities would preclude the meeting of the conditions that would favor the development of very large size in a snake. Larger prey items (tapirs, capybara, peccaries, crocodiles and Arrau turtles) are decreased by human presence and human activities; indeed, most of these species are themselves endangered. Human presence also would prevent the anacondas from growing to large sizes due to the propensity of people to kill snakes (Chapter 7). Thus, truly large snakes are probably restricted to permanent waters with little or no human intervention.

Two different strategies

To this point I have assumed that size and ages are perfectly related and this is not necessarily the case. Old individuals that have had a low energy income can retain a relatively small size, while young animals that eat well can grow to larger sizes in short time. Thus differences in the behavior of animals associated with their individual metabolic activities can produce confusion in these trends (Ford and Seigel 1989a, 1992, 1994). Additionally, differences in how the animals invest their energy can produce even more confusion. There might be some animals that invest a lot of energy in reproduction or that breed very often and might not have a lot of energy left to grow. Thus, these might well be older individuals that have not grown much due to the fact that they invest all the energy in reproduction and very little in growth. These animals would be playing optimum strategy for smaller sizes. Thus, the animals that breed less often acquire larger size because they devote a larger part of their resources into growth. These explanations would account for the high variance found in smaller sizes since smaller individuals could be young animals with a Small-RCM-Grow-Large (SRGL) strategy or older animals with a Large-RCM-Grow-Small (LRGS) strategy. The difficulty in assessing the ages of reptiles prevents an easy solution to this question. The presence of some relatively small animals that did not grow over several years suggests that individual differences in growth do exist. A more detail analysis of growth rates of the population is needed. The presence of two different tactics co-existing with similar conditions has been documented before. Ford and Seigel (1994) found that some captive reared *Elaphe guttata* fed with high energy diet breed at 20 months of ages while other individuals grown in the same conditions did not breed until 32 months of ages. The earlier breeders had a lower LRS than the later breeders.

Why would two strategies exist?

Two alternative strategies may coexist if both are in equally efficient. I calculated the contribution to the population of individuals below 340 cm of svl (first peak) and of individuals larger than 340 cm (second peak) by adding up the estimated clutch size of all the breeding females of those two size-groups. The second peak, despite the lower number of females, is responsible for 59.5% of the offspring born in a given year. Furthermore, the offspring of larger females have a higher expectancy of survival due to their larger size so their contribution to the recruitment in the population must be even higher. If SRGL females produce more offspring than the LRGS ones, they are expected to replace them in the population. However, the smaller animals are less conspicuous, and use water with more vegetation; so the possibility exist that this group might be under represented in the sample; so the competitiveness of each strategy in the local environment can only be hypothesized.

Animals with a slow growth rate (LRGS) might produce higher contributions to the population than SRGL individuals in times of prolonged shortage (several strong dry seasons or relatively dry wet seasons) when the larger animals have problems finding food and are more constrained by the drought and SRGL individuals might do better in wetter periods. Changes in the efficiency of these strategies between the years might prevent them from replacing each other in the population. Assortative mating may prevent individuals with different strategies from interbreeding (see Chapter 6).

Alternatively, the genetic flow from other areas might prevent adaptation to local conditions. The savanna (bajío) is adjacent to the river banks, and the seasonal

overflowing of them is part of the water supply of the savanna. There may be gene flow between these populations, however, the habitats are quite different and the selection pressures are expected to be different too. Animals with a SRGL strategy are probably adapted to the river and animals with a LRGS strategy may be fitter for the savanna. The shape of the Figure 5-10 may result from two strategies adapted to two different habitats. Is it really likely that in the same population two alternative strategies coexist? Anacondas in the savanna live right next to the gallery forest and the river. The sample (collected in the savanna) is probably composed by some LRGS individuals (first peak) and SRGL individuals as well, which are fittest in the neighboring river. The constant migration of animals from one side to the other prevents the population to fully adapt to the conditions they are on. This lack of adaptation to the local conditions due to genetic flow from neighboring population has been reported to occur in other species (Riechert 1993*a*, 1993*b*). Genetic studies are needed to test these hypotheses.

This is the first naturalistic study of the reproductive biology of any large snake. The ability to follow up animals for several years gives new information about their natural history. The natural history of the anacondas presents unexpected challenges for the conventional wisdom regarding life-history traits. The condition and size of the females seem to play important roles in the reproductive output of the species, where larger animals produce more and larger offspring. However, larger animals seem to breed less often and perform smaller reproductive investment than smaller ones. Ecological constraints regarding the interaction with predators at different ages and prey availability can be responsible of the trends found. Further studies on the population dynamics, mobility, and genetic structure of the populations are needed to understand better the trade-offs and the selection pressures that are affecting the population.

Table 5-1. Body measures of the anacondas caught and some statistics of the reproductive output calculated from the parturition. TL = Total Length, SVL= Snout-vent length, M = Mass, RCM = Relative Clutch Mass (see text for formula), RIO= Relative investment per offspring (%); CL = clutch size, NTL= Neonate total length, NSVL = Neonate Snout-Vent length, NM= Neonate Mass, N= sample number. The sex ratio was calculated over the 21 clutches from which I have full data.

	Mean	SD	Min	Max	N
TL (cm)	365.5	77.5	242.7	521.3	45
SVL (cm)	318.8	71.37	210.7	466	42
M (Kg)	30.82	18.22	9.25	70.00	45
RCM	0.39	0.095	0.17	0.52	27
RIO	1.03	0.43	0.32	2.15	32
CL	29.4	18.4	5	74	39
NTL	78.77	4.21	63.9	91.43	504
NSVL	68.42	4.01	54.7	80.93	492
NM	216.8	35.6	145	330	578
Sex Ratio	1.25	0.59	0.38	2.6	21

Table 5-2. The breeding frequency of the adult female anacondas that were caught in several years. Y and N stand for Yes and No indicating whether they bred in a given year.

Year	1992	1993	1994	1995	1996	1997	1998
E101C	Y	--	Y	--	--		
Lina	N	Y	--	--	--	--	
Kathalina	N	Y	--	--	--	--	
E145C	Y	N	--	--	--	--	
E155C	Y	--	Y				
E161C	N	Y	--	--	--	--	
Hermelinda	Y	N	--	--	--	--	
Sarah	Y	--	--	N			
Laura	--	Y	--	Y	--	--	
Araine	--	Y	N	--	--	--	
E200C	--	N	--	N	--	--	
E223C	--	N	Y	--	--	--	
Renée	--	--	Y	--	N	--	
Diega	--	Y	--	--	--	Y	
E436C	--	N	N	--	--	--	
E486C	--	--	Y	--	Y	--	
Guaratarita	--	--	Y	--	N	--	
E78C	N	Y	--	--	--	--	
Julia	--	--	--	--	Y	Y	
Mary-Jo	--	--	--	--	N	Y	
Alice	--	--	--	N	Y	--	
Courtney	--	--	--	N	Y	--	
E90C	Y	--	N	--	--	--	
Yuang-Ly	--	--	--	N	N	Y	
E204C	--	N	Y	Y	--	--	
Zuca	N	N	--	--	Y	--	
Musiua	N	N	--	--	Y	--	
Mirna	Y	N	N	--	--	--	
Mónica	Y	N	--	--	Y	--	
Antonieta	--	Y	--	Y	N	--	
Andrea	--	Y	--	Y	N	N	
Marion	N	N	Y	--	Y		
E437C	--	Y	--	N	N	Y	
Chinga	N	N	N	N	--	--	
Joan	--	--	N	Y	N	N	
Olivia	N	N	N	Y	N	--	
Madonna	Y	N	--	N	N	Y	N
Sue	Y	N	N	Y	N	Y	



Figure 5-1. Anacondas basking on termite mounds (Judy-Lee) during the wet season. Photo Bob Caputo.

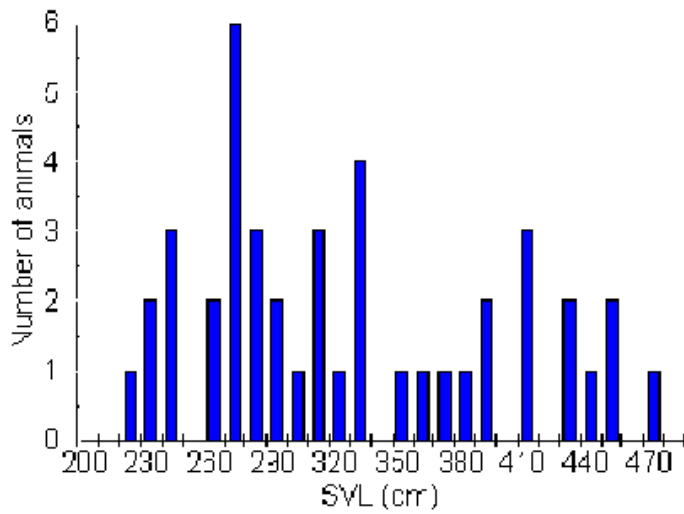


Figure 5-2. Size distribution of the breeding 42 female anacondas.

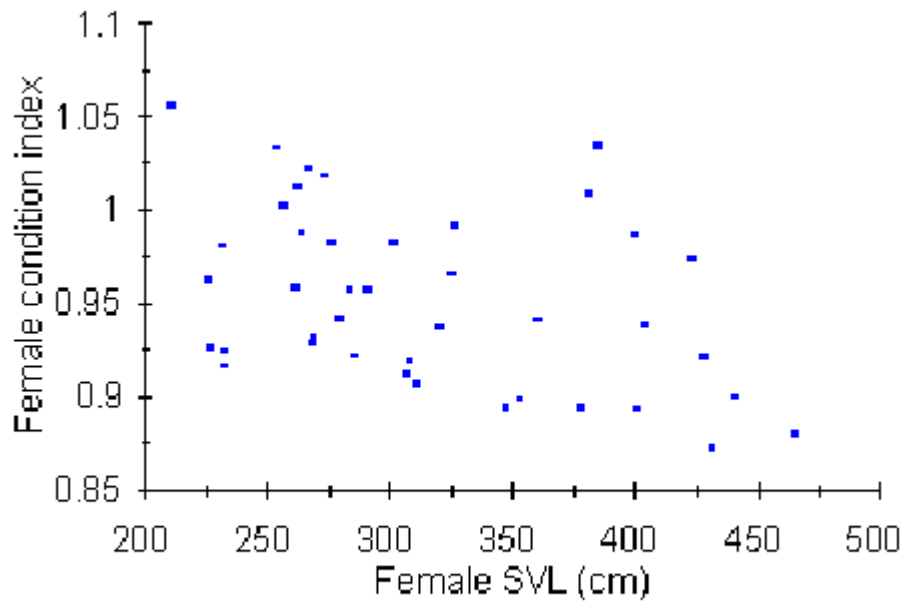


Figure 5-3. Size related changes on the condition index before birth of pregnant female anacondas in relation to female snout-vent length ($r = -0.42$; $p = 0.007$; $n = 41$).

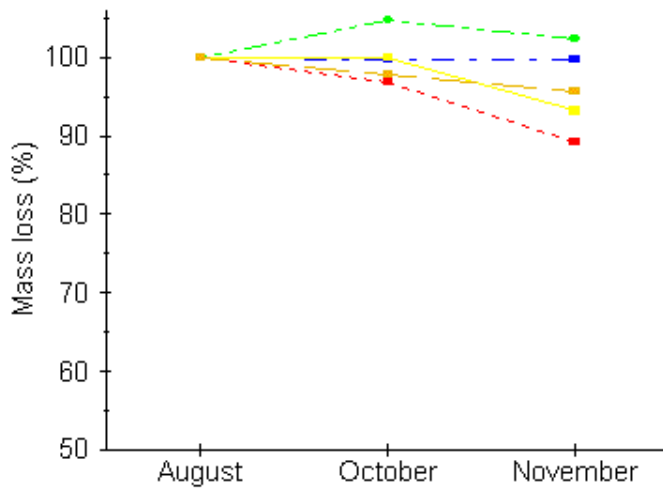


Figure 5-4. Mass loss during pregnancy of a sample of five female anacondas.

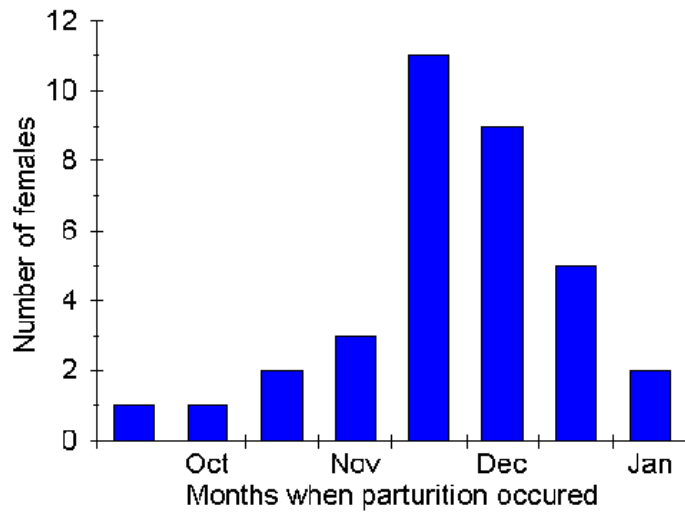


Figure 5-5. Timing of parturition of anacondas during the year.

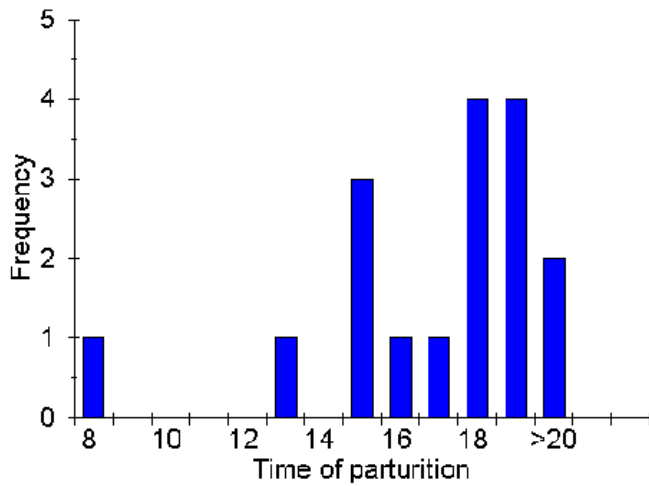


Figure 5-6. Time of the day when the anaconda parturition occurred. All the births that occurred after 19:30 were scored as > 2000.

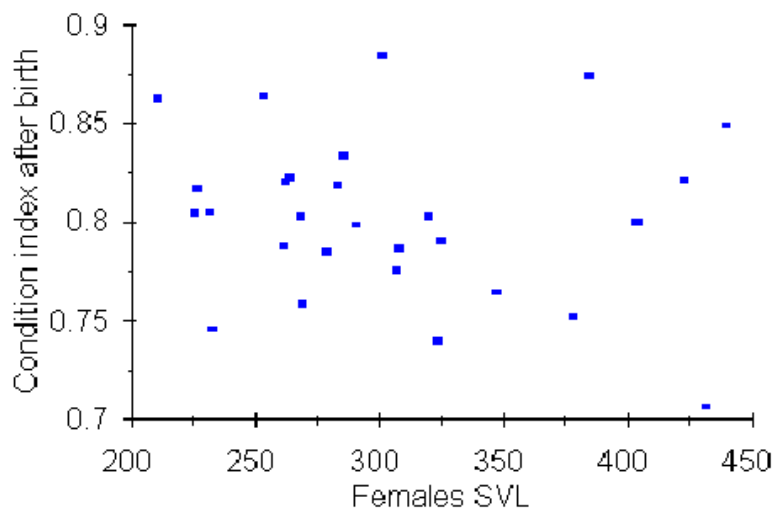


Figure 5-7. Condition Index of female anacondas after birth vs female snout-vent length ($r = -0.12$; $p = 0.49$; $n = 28$).

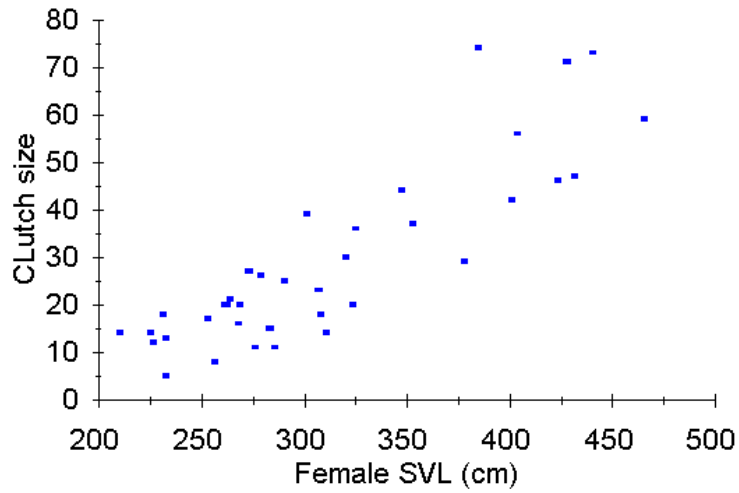


Figure 5-9. Relation between the clutch size of female are anacondas and their snout-vent length ($r=0.83$; $p<0.000$; $n=36$).

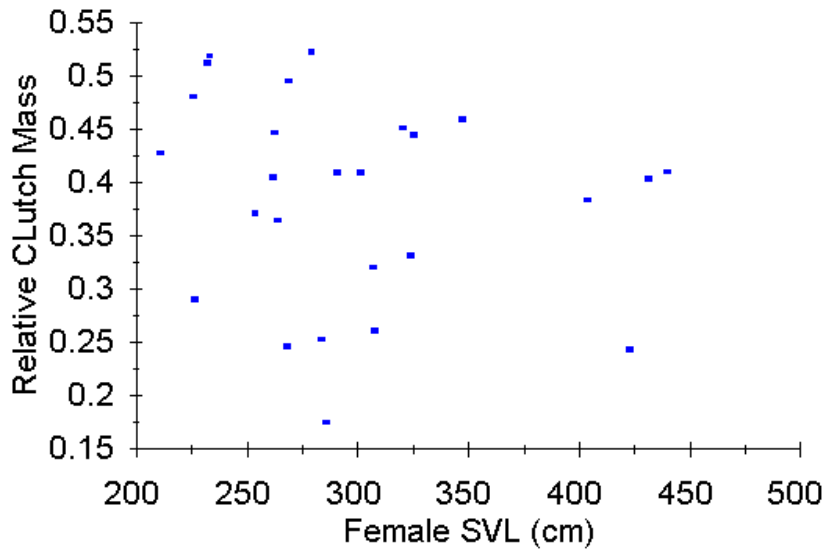


Figure 5-10. Relation between Relative Clutch Mass and female Snout-Vent-length ($r=0.23$; $p<0.28$; $n=25$). The Relative Clutch Mass is calculated by dividing the mass of the clutch by the mass of the females after birth.

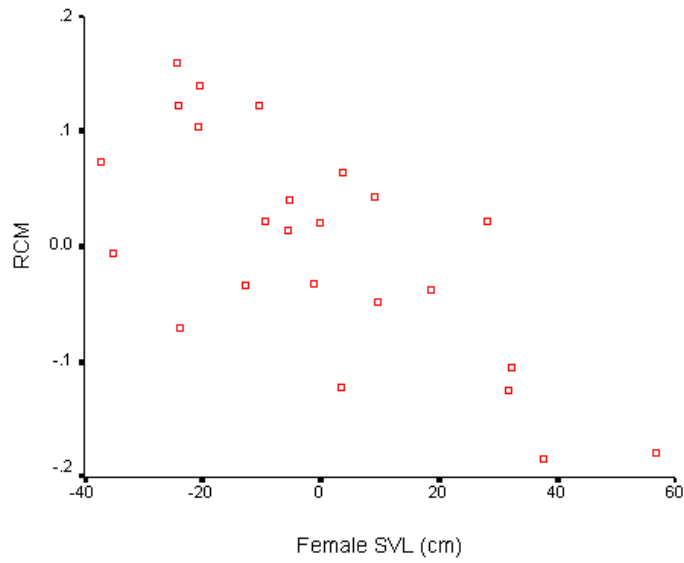


Figure 5-11. Partial correlation plot of anaconda relative clutch mass and SVL holding clutch size and condition index constant ($r= 0.71$; $p = 0.000$; $n = 20$)

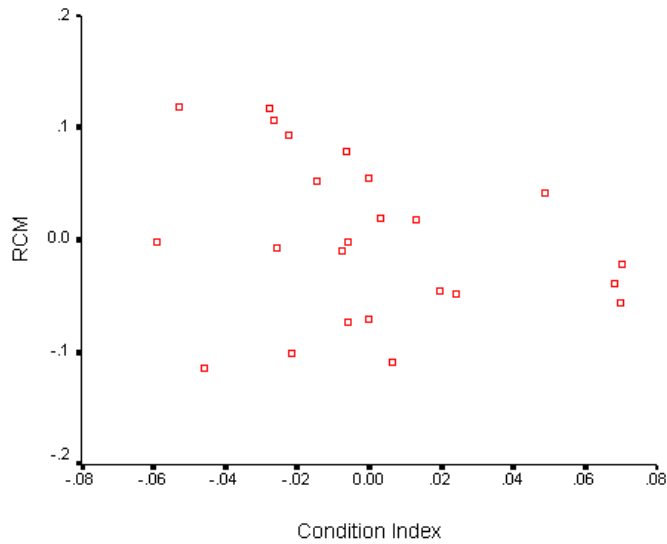


Figure 5-12. Partial correlation plot of anaconda relative clutch mass and condition holding SVL and clutch size constant ($r= -0.27$; $p = 0.23$; $n = 20$)

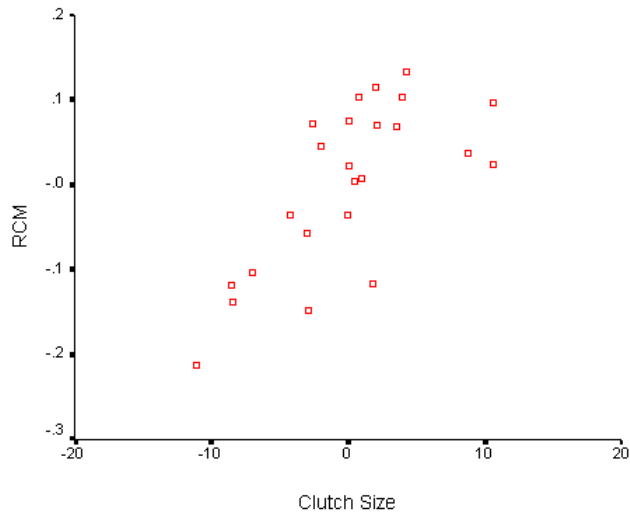


Figure 5-13. Partial correlation plot of anaconda relative clutch mass and clutch size holding SVL and condition constant ($r = 0.7$; $p = 0.000$; $n = 20$).

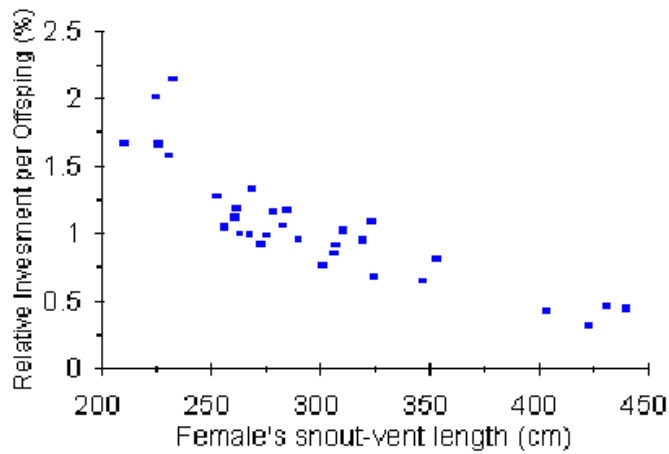


Figure 5-14. Relation between the relative investment per offspring and the SVL of female anacondas ($r = -0.899$; $p < 0.000$; $n = 30$).

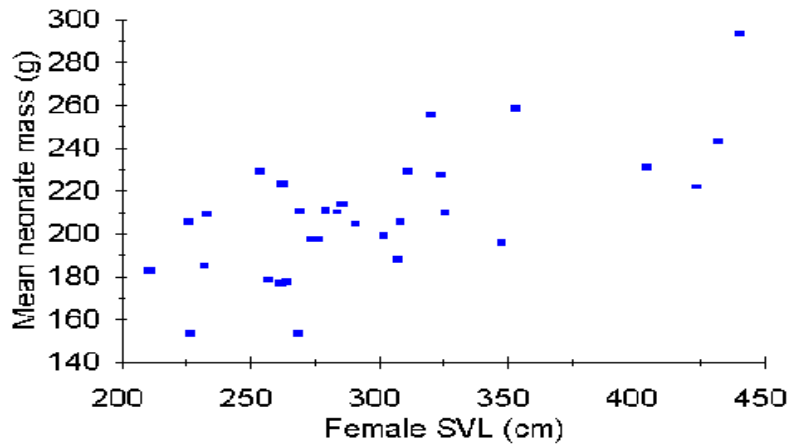


Figure 5-15. Relationship between the mass of neonate anacondas and the SVL of the female ($r= 0.604$; $p< 0.000$; $n= 30$).

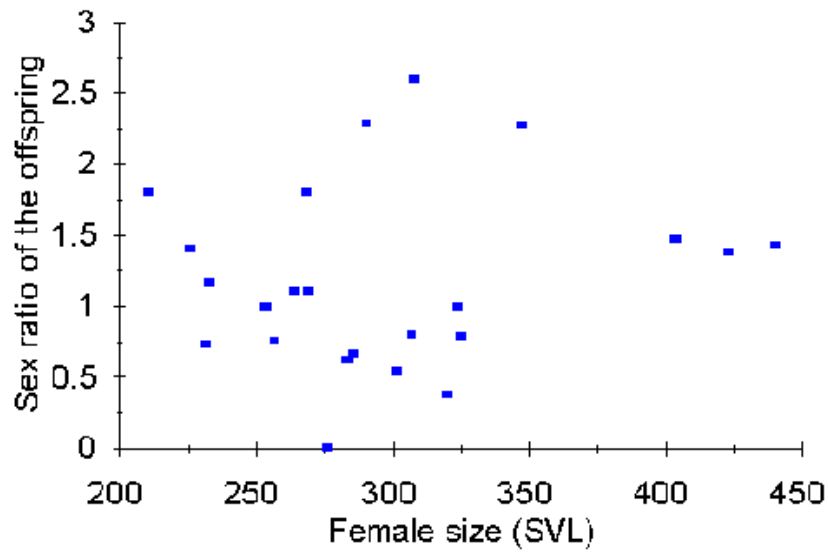


Figure 5-16. Variation in the sex ratio of the clutches of 21 anaconda clutches ($r = -0.013$; $p< 0.964$; $n= 21$); see Table 6-2 for statistics.

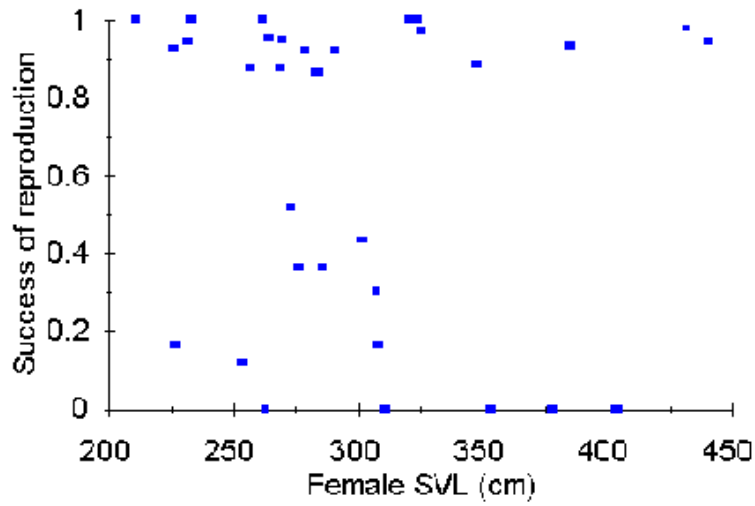


Figure 5-17. Reproductive success of anacondas that gave birth in captivity. The success of reproduction of individual females is calculated by dividing the number of live neonates by the sum of live, stillborn and infertile eggs.

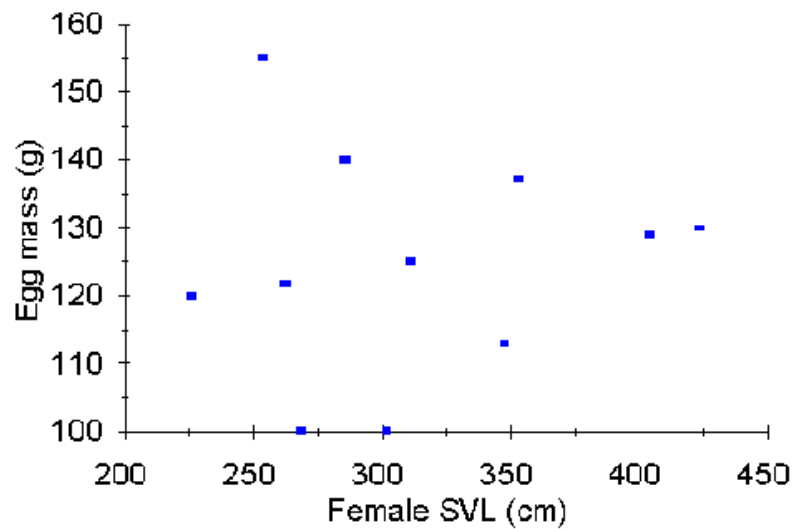


Figure 5-18. Egg mass of female anacondas in relation to female SVL ($r=0.14$; $p=0.69$; $n=11$).

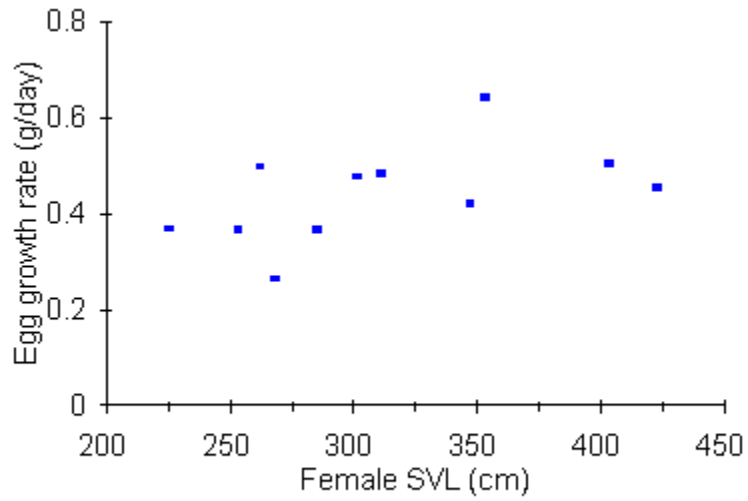


Figure 5-19. Growth rate of embryos of female anacondas in relation to female SVL ($r=0.527$; $p=0.096$; $n=11$)

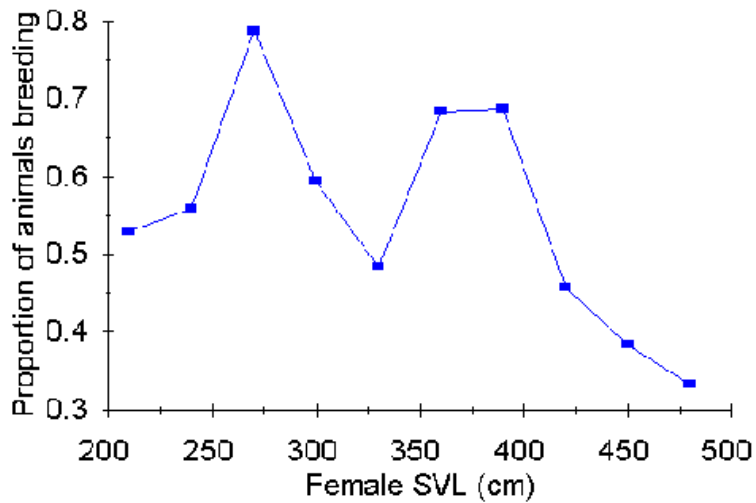


Figure 5-20. Proportion of pregnant females anaconda in different size classes. Calculated from the capture of the total sample ($n=222$) by using the condition index.

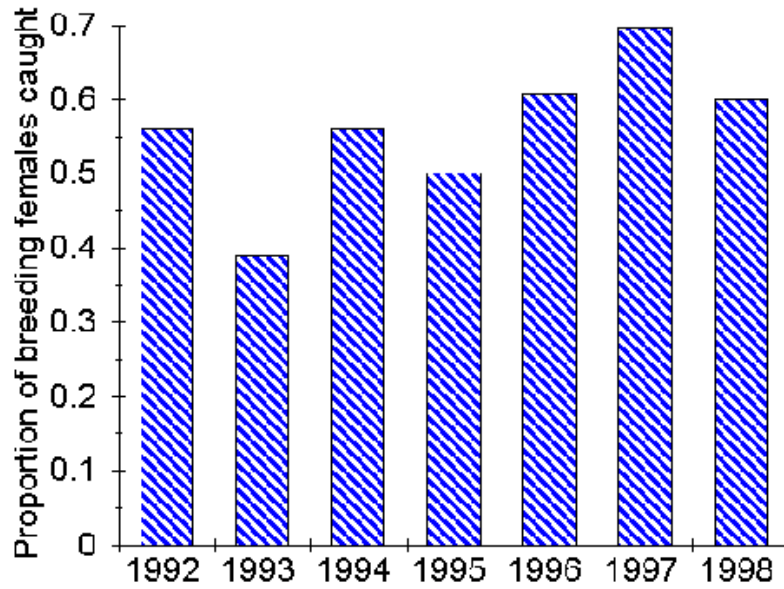


Figure 5-21. Proportion of breeding female anaconda per year of study calculated with the condition index.

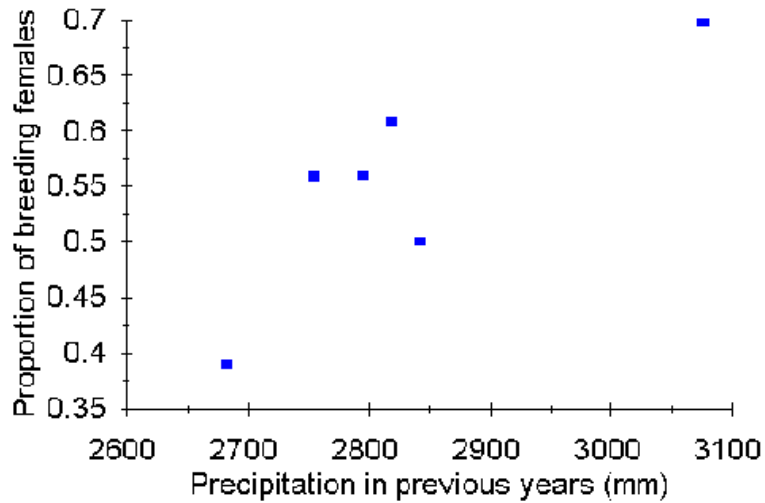


Figure 5-22. Proportion of breeding female anacondas found in relation to the total precipitation of the two former years ($r = 0.66$; $p = 0.16$; $n = 6$).

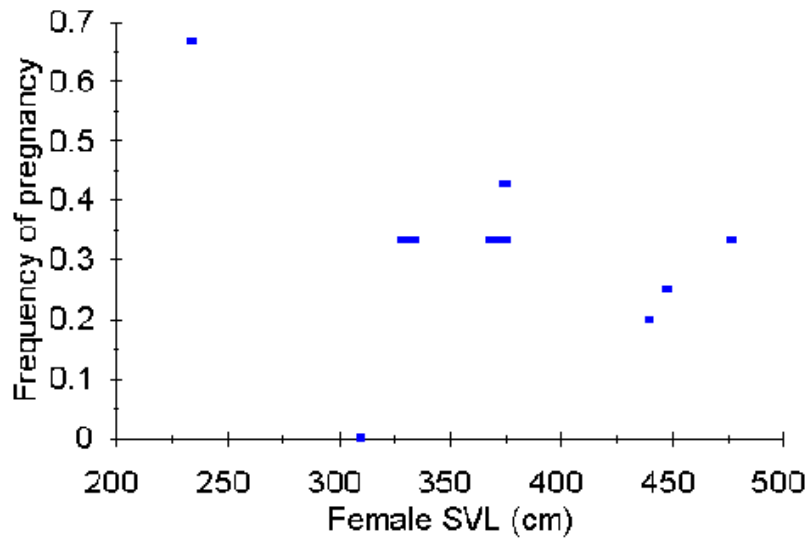


Figure 5-23. Frequency of reproduction of female anacondas that were caught in several consecutive years and were breeding in at least one ($r = -0.805$; $p = 0.005$; $n = 10$).

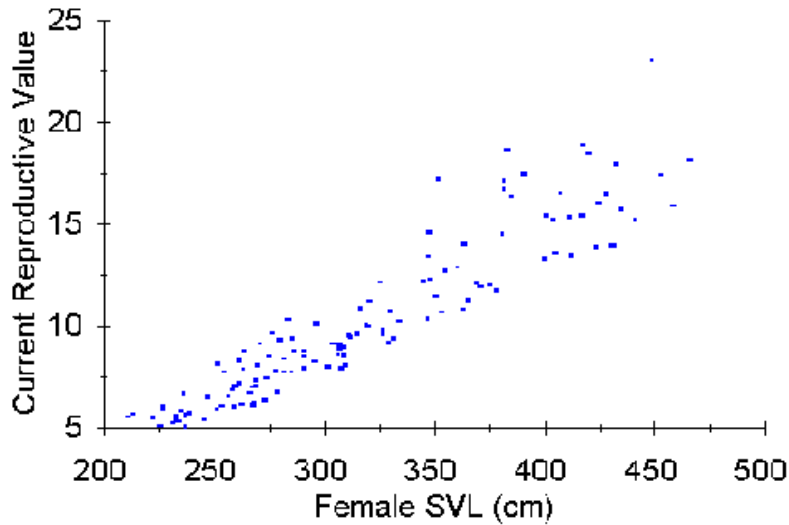


Figure 5-24. Relationship between the reproductive value (breeding frequency multiplied by the clutch size) and SVL of female anacondas ($r = 0.902$; $p < 0.000$; $n = 129$).

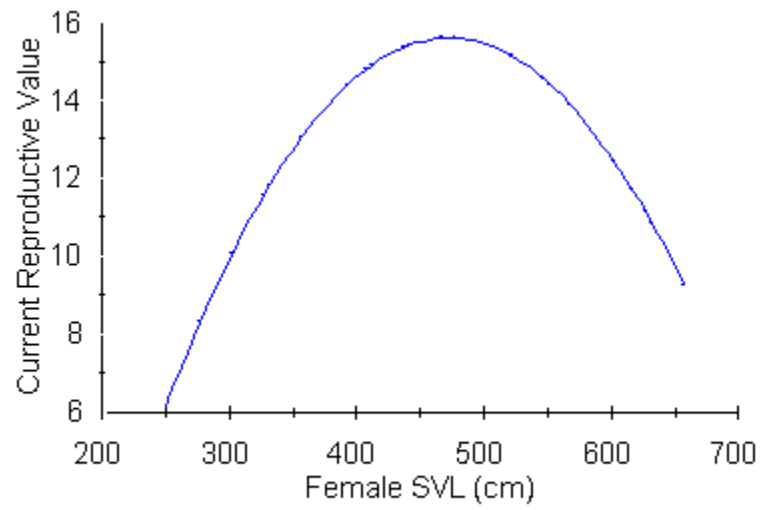


Figure 5-25. Model for anacondas of the expected ontogenetic change in the Current Reproductive Value (CRV). The CRV is calculated by multiplying the expected breeding frequency by the expected clutch size.

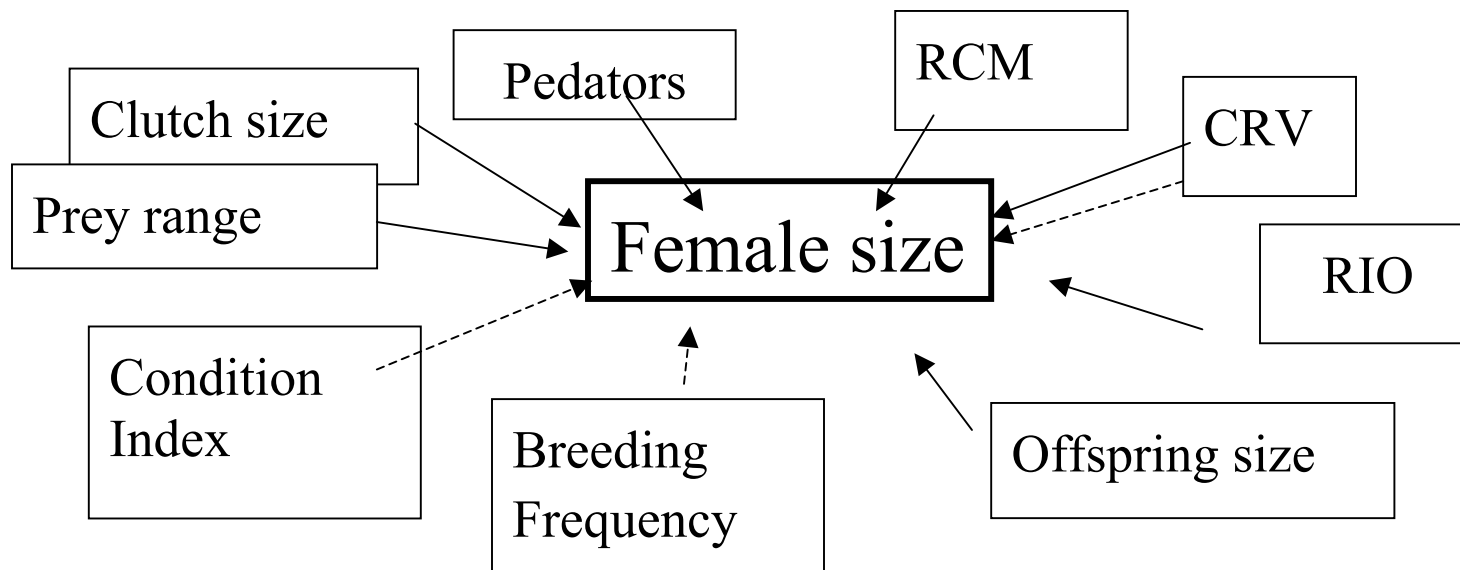


Figure 5-26 Selection pressures affecting female size in green anaconda. Those selection pressures that increase the female size are represented with a solid line. Those that limit or decrease female size are represented with a dashed line. RIO = relative investment per offspring, RCM = relative clutch mass, CRV = current reproductive value. CRV represents the yearly clutch size of the animal in a given size and selects for an optimum female size; so, it is represented with both arrows.