South American Journal of Herpetology, 3(2), 2008, 168-174

A FIRST ESTIMATE OF THE POPULATION SIZE OF THE CRITICALLY ENDANGERED LANCEHEAD, *BOTHROPS INSULARIS*

MARCIO MARTINS[1], RICARDO J. SAWAYA[2] AND OTAVIO A.V. MARQUES[2]

[1] *Departamento de Ecologia, Instituto de Biociências, Universidade de São Paulo, 05508-090 São Paulo SP, Brazil. E-mail: jararaca@ib.usp.br*
[2] *Laboratório Especial de Ecologia e Evolução, Instituto Butantan, Av. Vital Brazil 1500, 05503-900 São Paulo, SP, Brazil.*

ABSTRACT. Although *Bothrops insularis* is critically endangered, there is no estimate of its population size based on field quantifications. Here we provide the first estimate of its population size based on plot surveys conducted at the Queimada Grande Island. We distributed 26 quadrat plots regularly in a trail that crosses the island. Plots were sampled twice in 2002. Of the 26 plots we sampled, 21 were located in forests and five in grassy areas. For the first survey, mean estimates of population size for plots located in forests and grassy areas were 2134.3 and 224.0 snakes, and the Percentage Relative Precision for these estimates were 38.6% and 277.6%, respectively. For the second survey, no snakes were found in grassy areas, whereas the mean estimate of population size for plots located in forests was 1304.3 snakes and the Percentage Relative Precision for these estimates was 93.7%. Although suffering from relatively low precision, our best estimate of population size in *B. insularis* is around the lower end of the guesses found in the literature (2000-4000 individuals for the entire island). Furthermore, based on encounter rates obtained in a non-systematic way in the last 12 years, we have the impression that the density of *B. insularis* decreased in this period and we have evidence for the illegal removal of snakes from the island in the last few years. Our results suggest the urgent need of enforcement to restrain the illegal removal of snakes from the island and of a monitoring program to track future changes in the population size of *B. insularis.*

KEYWORDS. conservation, Queimada Grande Island, southeastern Brazil, Atlantic Forest.

INTRODUCTION

One of the most useful kinds of information needed to assess the conservation status of a given species is its population size (e.g., Rabinowitz *et al.,* 1986). With this information, it is possible, for instance, to assess its vulnerability to extinction and to track changes in population size (with further estimates using the same census method; see, e.g., Traill *et al.,* 2007).

The golden lancehead, *Bothrops insularis* (Fig. 1A), is endemic to Queimada Grande Island, a very small island (about 43 ha) located 33 km off the coast of São Paulo State, southeastern Brazil (Amaral, 1921; Duarte *et al.,* 1995; Fig. 1B). The island was inhabited by humans (the lighthouse keeper and his family) in the beginning of the last century, but it is now uninhabited (Marques *et al.,* 2002). The lighthouse is now automatic and kept sporadically by the Brazilian Navy. The low forests which cover about 24 ha of the island (Lower Montane Rain Forest, cf. Oliveira-Filho and Fontes, 2000; Fig. 2A) are the main habitat of *B. insularis,* (Fig. 3A, B). The remaining areas are bare rock and open areas covered mostly by grasses (Fig. 2B, 3A, B); many of these open areas resulted from the removal of the forest by humans (Marques *et al.,* 2002). Although most commonly found inside the forest, *B. insularis* may also be found in grassy, open areas.

Bothrops insularis was described in 1921 by Afrânio do Amaral (Amaral, 1921) and studied by Alphonse R. Hoge and collaborators in the 50's (Hoge, 1950; Hoge *et al.,* 1959). Duarte *et al.* (1995) provided a review of the knowledge on *B. insularis* and some new field data and recently Marques *et al.* (2002) provided further information on its biology resulting from additional field data. Presently, *B. insularis* is included as critically endangered in the IUCN Red List of Threatened Species (IUCN, 2008) and in the Brazilian List of Endangered Animals (Machado *et al.,* 2005). Although there is no estimate on the population size of *B. insularis* based on field quantifications in the literature, guesses based on field experience are found in Hoge in Grzimek *et al.* (1975) and in Marques *et al.* (2002): 3000-4000 and 2000-4000 snakes, respectively.

Here we provide the first estimate of the total population size of *B. insularis* based on two plot surveys carried out in 2002 at the Queimada Grande Island.

METHODS

We used 10 x 10 m quadrat plots to sample *B. insularis* at Queimada Grande Island. Plots were delimited by ropes, installed after removing the shrubby

FIGURE 1. A – An adult male of *Bothrops insularis* at the Queimada Grande Island, southeastern Brazil. B – The Queimada Grande Island.

FIGURE 2. A. The interior of the low forest which is the main habitat of *B. insularis* at the Queimada Grande Island, southeastern Brazil. B. Patches of grassy areas (within low forests), where *B. insularis* may also be found.

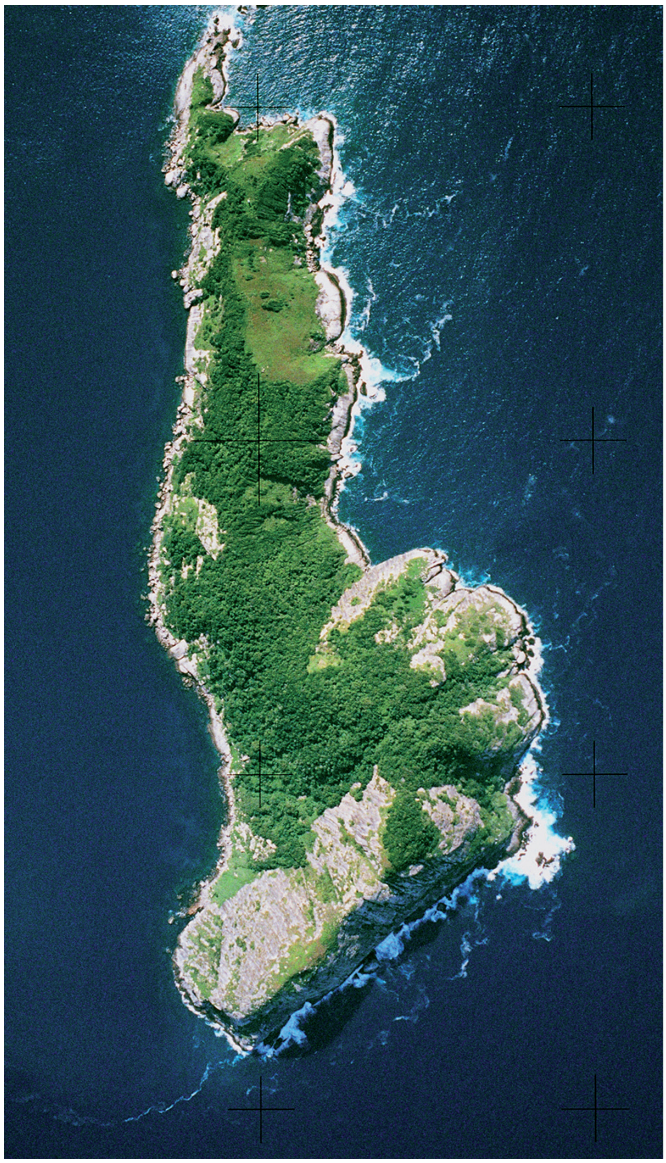
vegetation. Usually, eight people sampled each plot. One person was located at each side of the plot during the cutting of the shrubby vegetation, installation of the ropes, and sampling, in order to capture escaping snakes. At least four people searched for snakes visually, two of them on the ground and two on the vegetation, including the canopy (inspected from the ground and/or from tree branches located as close to the top of the canopy as possible). Besides air temperature and relative air humidity, we measured in each plot environmental variables that will be treated elsewhere. All snakes found were captured, measured (body and tail length), marked (with pit-tags), and released after the sampling.

We distributed 26 plots along a trail that crosses the island in the North-South direction (Fig. 3B), regularly at every 50 m (elevation ranged from ca. 50 m to about 170 m). We are aware of the problems associated with sampling at regular intervals (e.g., Greenwood, 1996), but the relief of the island would make a random sampling design almost impossible. Indeed, our working trail follows the generally narrow ridge of the island, which separates the eastern and western slopes (Fig. 1B). We sampled all 26 plots twice in 2002: in 10 to 18 February (first survey) and in 30 April to 2 May (second survey). Since in over ten years of fieldwork in the island we found snakes in bare rock only occasionally (much less than 1% of the snakes found), we did not include areas of bare rock in our plot samples.

A map of the island ground cover was made with the use of an aerial photograph (1:4000) obtained in 17 May 2002 (Fig. 3A). The photograph was digitized and georeferenced in ArcGis 9.0 (ESRI, 2004). The classification of ground cover (into bare rock, grassy areas, open forest, and forest; Fig. 3B), which was made visually, and the calculation of polygon areas, were made in ArcGis 9.0 (ESRI, 2004). We called open forests those forests with interrupted canopy. To avoid problems related to small sample sizes, we consider forests and open forests together in all analyses.

We divided the trail into five sectors (A to E) in order to compare the aggregation of snakes between the two surveys. Each sector included five consecutive plots, except for sector E which included six plots. Sector A corresponded to plots located at 50-250 m of the trail, with predominantly open areas but also low forest, relatively level ground, and elevation about 50-60 m. Sector B corresponded to plots located at 300-500 m of the trail, with predominantly low forest, relatively level ground, and elevation about 60-70 m. Sector C corresponded to plots located at 550-750 m of the trail, with predominantly low forest, relatively sloping ground, and elevation about 70-90 m. Sector D corresponded to plots located at 800-1000 m of the trail, with low to high forest, sloping ground, and elevation about 90-120 m. Sector E corresponded to plots located at 1000-1300 m of the trail, predominantly high forest, sloping and relatively level ground, and elevation about 120-170 m.

The mean and 95% confidence limits for the estimates of total population size in each vegetation type were calculated from the estimates of total population size for each plot. Thus, we generated 21 estimates of total population size for forests in both surveys and five for grassy areas in the first survey (we found no snakes in plots in grassy areas in the second survey). For comparisons of precision between estimates for different vegetation types and different times of the



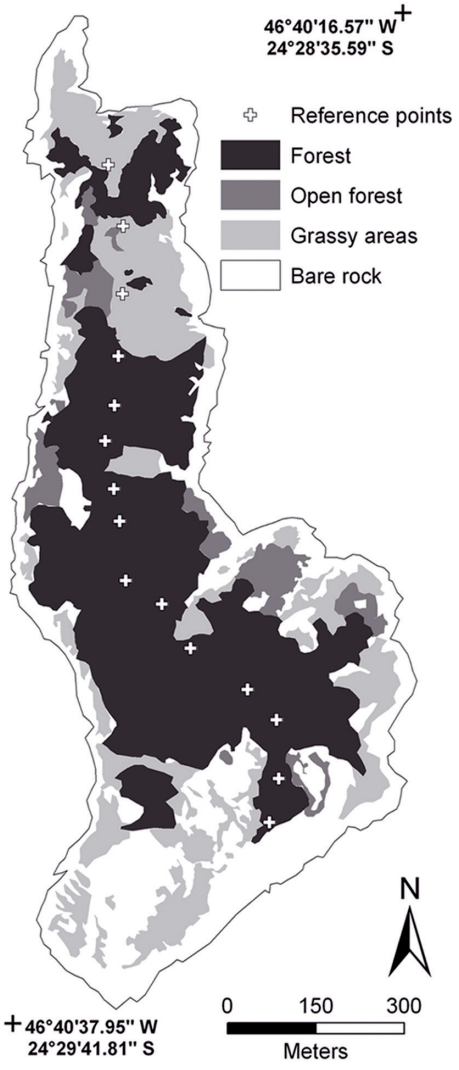


Figure 3. A – An aerial photograph of the Queimada Grande Island obtained in 17 May 2002; B – Classification of soil cover (based on the aerial photograph in A) showing reference points of the trail where quadrat plots were surveyed.

year, we also provide Percentage Relative Precision, which is the difference between the estimated population size and its 95% confidence limits, expressed as a percentage of the mean (Sutherland, 1996). Thus, higher values of Percentage Relative Precision indicate lower precision.

The differences in the number of snakes captured in plots and sectors were compared by Mann-Whitney and Kruskall-Wallis tests, and differences were considered significant when $p < 0.05$ (Zar, 1999). All statistical analyses were performed in Statistica (StatSoft, 2003).

Results

Forests cover 24.9 ha of the island, whereas grassy areas cover 11.2 ha (Fig. 3B). Of the 26 plots we sampled, 21 were located in forests and five in grassy areas. Climatic conditions were similar in both surveys: air temperatures 26.9 ± 3.4 (mean ± 1SD) and 28.3 ± 1.3°C, relative air humidity 35.2 ± 8.6 and 36.3 ± 6.1%, respectively (n = 26 in both surveys). The number of snakes found per plot was significantly higher in the first survey (January) than in the second (April/May; medians 0 and 2 snakes, respectively; $z = 2.35$, $p = 0.041$; Table 1).

In the first survey, plots located in forests contained 0-2 snakes (median 1), whereas those in grassy areas contained 0-1 snake (median 0; Table 1). Mean estimates of population size for plots located in forests and grassy areas were 2134.3 and 224.0 snakes, and the Percentage Relative Precision for these estimates were 38.6% and 277.6%, respectively (Table 1). These estimates of population size correspond to means of 0.86 and 0.20 snakes per 100 m^2 plot or 86 and 20 snakes per ha, respectively, multiplied by 24.9 ha of forest.

In the second survey, plots located in forests contained 0-3 snakes (median 0) and no snakes were found in plots located in grassy areas (Table 1). The mean estimate of population size for plots located in forests was 1304.3 snakes, and the Percentage Relative Precision for this estimates was 93.7% (Table 1). This estimate of population size corresponds to a mean of 0.52 snakes per 100 m^2 plot, or 52 snakes per ha, multiplied by 24.9 ha of forest.

In the first survey, the number of snakes found in plots located in forests (median 1) was larger than the number of snakes found in plots in grassy areas (median 0), although marginally nonsignificant ($z = 1.72$, $p = 0.085$). In the second survey, the number of snakes found in plots located in forests (median 0) was not different from the number of snakes

Table 1. Summary of 10 x 10 m quadrat plot sample results for *Bothrops insularis* at the Queimada Grande Island. Higher values of percentage relative precision indicate lower precision in the estimates.

	First survey 10-18 February 2002	Second survey 30 April to 2 May 2002
Forests (24.9 ha)		
number of plots sampled	21	21
total number of snakes found	18	11
median of snakes/plot	1	0
number of plots with 0 snakes	7	16
number of plots with 1 snake	10	2
number of plots with 2 snakes	4	0
number of plots with 3 snakes	0	3
mean estimate of population size	2134.3	1304.3
confidence limits (95%)	1310.3, 2958.3	82.5, 2526.0
percentage relative precision	38.6%	93.7%
Grassy areas (11.2 ha)		
number of plots sampled	5	5
total number of snakes found	1	0
median of snakes/plot	0	0
number of plots with 0 snakes	4	5
number of plots with 1 snake	1	0
mean estimate of population size	224.0	-
confidence limits (95%)	-397.9, 845.9	-
percentage relative precision	277.6%	-

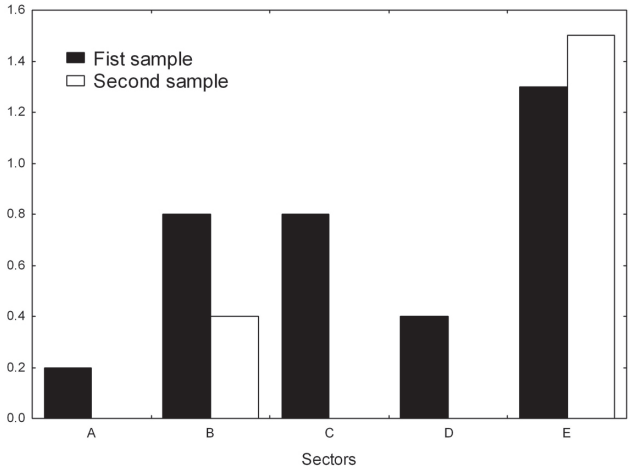


FIGURE 4. Number of individuals of *B. insularis* found per plot in each trail sector in the first (black bars) and second (white bars) surveys conducted at the Queimada Grande Island in January and April/May 2002, respectively. Sectors are: A (plots located at 50-250 m of the trail), B (300-500 m), C (550-750 m), D (800-1000 m), and E (1000-1300 m). See text for descriptions of sectors.

found in plots in grassy areas (median 0; $z = 0.81$, $p = 0.416$).

In both surveys, the number of snakes found per plot varied along our working trail (Fig. 4). In the first survey, the number of snakes found per plot in each sector of the trail was different (Fig. 4), although marginally nonsignificant ($H_{[4,26]} = 8.24$, $p = 0.083$). A post-hoc test indicated that there was a significantly higher number of snakes in sector E in relation to sector A. However, these results are not evident when sector A, which includes four plots in grassy areas, is removed from the analysis ($H_{[3,21]} = 4.75$, $p = 0.191$). There was a tendency to find more snakes in sector E also in the second survey (Fig. 4), although the difference is marginally nonsignificant ($H_{[4,26]} = 8.60$, $p = 0.072$). However, this result is not evident when sector A is removed from the analysis ($H_{[3,21]} = 6.17$, $p = 0.103$).

DISCUSSION

The estimate of population size for *B. insularis* we obtained for grassy areas are not reliable because of its very low precision. Therefore, we here discuss only those estimates obtained for forests. Indeed, based on our field experience in the island in the last 12 years, we infer that the open areas may harbor less than 10% of the individuals of *B. insularis* at any time considered. Thus, our conclusions based only on the estimates for forests may be very close to those we would reach if we had good estimates for grassy areas.

The density of *B. insularis* is high, but far from being the highest density of an insular snake. If we consider the total island area, our higher estimates (first survey, including the gross estimate for grassy areas) indicate a density of about 55 snakes per hectare (2358 snakes in 43 ha). A similar estimate for the Shedao Island pitviper, *Gloydius shedaoensis,* in China (Li, 1995), is about 200 snakes per hectare (14,600 snakes in 73 ha; Huang, 1990). Indeed, it is suggested that in suitable habitats the density of *G. shedaoensis* may be as high as one snake per square meter (see Shine *et al.,* 2002).

Our results indicate that snakes were more aggregated in the more elevated parts of the island (where the forest is higher) during both surveys. Birds could be more abundant in these areas, which could lead to an aggregation of snakes in them. However, recent bird census data do not indicate that migrant birds (potential prey of *B. insularis* adults; Marques *et al.,* 2002) tend to be more aggregated in the more elevated parts of the island at any time (A. Macarrão, pers. com.). Thus, the hypothesis of an aggregation of snakes due to an aggregation of potential prey is not corroborated by bird census data. Additionally, the higher forest of the more elevated areas may provide more suitable resources to the snakes, like better thermal environments (lower and/or less variable air temperatures).

Our best estimate of population size in *B. insularis* (2134 snakes in forests) is around the lower end of the guesses found in the literature: 2000-4000 individuals for the entire island (Hoge in Grzimek *et al.,* 1975; Marques *et al.,* 2002). These latter estimates originated from field experience in the island in the 50's (Hoge in Grzimek *et al.,* 1975) and 90's (Marques *et al.,* 2002). Indeed, between 1995 and 1998, we found 31 to 66 snakes in nine two-day trips to the island, in which we conducted snake searches in one afternoon and one morning (what resulted in 46 ± 12 snakes/day; pers. obs.). In five recent trips to the island (during 2007-2008), we found similar numbers of snakes, but in three days of fieldwork (with a similar sampling effort; 22 ± 7 snakes/day). Based on these encounter rates, we have the impression that the density of *B. insularis* decreased in the last 12 years, and this impression is shared by all people who worked in the island during this period. Thus, perhaps the upper estimates of Hoge in Grzimek *et al.* (1975) and Marques *et al.* (2002) were close to the actual population size at the times these estimates were done.

But what factor or factors could be responsible for a supposed decrease in population size in *B. insularis?* We have evidence for the illegal removal of snakes from the island in the last few years (Marques *et al.,* 2002). Although there was never an official program by the Brazilian government to breed or to export individuals of *B. insularis,* there are websites offering to sell them and discussion groups on the Internet where people state that it is easy to keep them in captivity and even to collect them at the Queimada Grande Island. Indeed, in our trips to the island, we occasionally find in our working trail waste and garbage from other people than us and the marines who sporadically keep the automatic lighthouse (and restrict their movements in the island to the 280 m-trail that connects the sea to the lighthouse and apparently do not use our working trail at all). Additionally, the sailors who used to take us to the island stated that once they took to the island people who identified themselves as staff from Instituto Butantan, and we found out latter that nobody from Instituto Butantan has been in the island at that time. And recently (March, 2008), when we arrived from a trip to the island, two unknown men said to two members of our field team that they knew people in the Santos harbor (the biggest port in Brazil, located 11 km from the point where we disembarked) who would buy a *B. insularis* for about US$ 30,000.00 (K. N. Kasperoviczus and A.B. Barros, pers. comm.).

Bothrops insularis is included as critically endangered (CR B1ab[iii]+2ab[iii]) in the IUCN Red List of Threatened Species (IUCN, 2008) and in the Brazilian List of Endangered Animals (Machado *et al.,* 2005) because: (1) a very small extent of occurrence (less than 100 km^2; criterion B1); (2) its restriction to a single location (criteria B1a and B2a); (3) an observed decline in the quality of its habitat (forest destruction by marines who keep the lighthouse; criteria B1b[iii] and B2b[iii]); and (4) a very small area of occupancy (less than 10 km^2; criterion B2). Our plot sample results and the evidence described above strongly indicate that the population size of *B. insularis* is decreasing. Therefore, two additional criteria, both of them related to a decline in the number of mature individuals (B1b[v] and B2b[v]), should be added to the criteria already used in the IUCN and Brazilian lists. The evidence above also indicate the urgent need of enforcement by the Brazilian government to restrain the illegal removal of snakes from the island and of a monitoring program to track future changes in the population size of *B. insularis*.

Resumo

Embora *Bothrops insularis* esteja criticamente em perigo, não existem estimativas do tamanho de sua população a partir de quantificações em campo. Neste estudo fornecemos a primeira estimativa do tamanho de sua população com base em amostragens de parcelas conduzidas na Ilha da Queimada Grande. Distribuímos 26 parcelas regularmente ao longo da trilha que corta a ilha. As parcelas foram amostradas duas vezes em 2002. Das 26 parcelas amostradas, 21 estavam em áreas de floresta e cinco em áreas de gramíneas. Na primeira amostragem, a média das estimativas do tamanho da população para parcelas localizadas em florestas e em áreas de gramíneas foi de 2134,3 e 224,0 serpentes, e a precisão relativa percentual para essas estimativas foi de 38,6% e 277,6%, respectivamente. Na segunda amostragem, nenhuma serpente foi encontrada nas parcelas em áreas cobertas por gramíneas, ao passo que a média das estimativas do tamanho da população para parcelas localizadas em florestas foi de 1304,3 serpentes e a precisão relativa percentual para essas estimativas foi de 93,7%. Embora apresente problemas de precisão, nossa melhor estimativa para o tamanho populacional de *B. insularis* está nos limites inferiores de suposições disponíveis na literatura (2000-4000 indivíduos para toda a ilha). Além disso, com base em taxas de encontro obtidas de maneira não sistemática ao longo dos últimos 12 anos, temos a impressão que a densidade de *B. insularis* diminuiu durante esse período, e existem evidências de remoção ilegal de serpentes da ilha nos últimos anos. Nossos resultados indicam a necessidade urgente de fiscalização de forma a coibir a remoção ilegal de serpentes da ilha e de um programa de monitoramento para acompanhar as futuras mudanças no tamanho populacional de *B. insularis.*

Acknowledgments

We thank F. S. Avino, C. A. Brasileiro, C. Monteiro, G. Nilson, M. E. Oliveira, A. C. T. P. Santos, A. M. Tozzetti, and K. Zamudio for invaluable help in fieldwork; G. Nilson and H. W. Greene for inspiring discussions on the biology of *B. insularis;* two anonymous referees for providing helpful suggestions on an earlier draft of the manuscript; D. F. Bertani, L. Tambosi, A. Igari, and LEPaC (Laboratório de Ecologia da Paisagem e Conservação, Instituto de Biociências, Universidade de São Paulo) for GIS-related laboratory work; M. Rodrigues for the compilation of capture data for encounter rates; CNPq, FAPESP, Fundação O Boticário de Proteção à Natureza and National Geographic for financial support; and FAPESP for providing a post-doctoral fellowship to RJS.

Literature Cited

Amaral, A. 1921. Contribuição para o conhecimento dos ophidios do Brasil. A Parte II. Biologia de uma nova espécie. Anexos das Memórias do Instituto Butantan, 1:39-44.

Duarte, M.R., G. Puorto, and F.L. Franco. 1995. A biological survey of the pitviper *Bothrops insularis* Amaral (Serpentes,Viperidae): an endemic and threatened offshore island snake of south-eastern Brazil. Studies on Neotropical Fauna and Environment, 30:1-13.

ESRI, 2004. ArcGIS, Release 9.0. Environmental Systems Research Institute, Redlands.

Greenwood, J.J.D. 1996. Basic techniques; pp. 11-110. In: W.J. Sutherland (Ed.), Ecological Census Techniques: A Handbook. Cambridge University Press, Cambridge.

Grzimek, B. (ed.) 1975. Grzimek's Animal Life Encyclopedia. Volume 6: Reptiles. Van Nostrand Reinhold, New York.

Hoge, A.R. 1950. Notas erpetológicas. 7. Fauna erpetológica da Ilha da Queimada Grande. Memórias do Instituto Butantan, 22:151-172.

Hoge, A.R., H.E. Belluomini, G. Schreiber, and A.M. Penha. 1959. Sexual abnormalities in *Bothrops insularis* (Amaral, A. do) 1921 (Serpentes). Memórias do Instituto Butantan, 29:17-88.

Huang, M. 1990. The present state of *Agkistrodon shedaoensis* population; pp. 271-272. In: E. Zhao (Ed.), From Water Onto Land. China Forestry Press, Beijing.

IUCN 2007. 2007 IUCN Red List of Threatened Species. <http://www.iucnredlist.org>. Downloaded on 10 March 2008.

Li, J-L. 1995. China Snake Island. Liaoning Science and Technology Press, Dalian.

Machado, A.B.M., C.S. Martins, and G.M. Drummond. 2005. Lista da Fauna Brasileira Ameaçada de Extinção. Fundação Biodiversitas, Belo Horizonte.

Marques, O.A.V., M. Martins, and I. Sazima. 2002. A jararaca da Ilha da Queimada Grande. Ciência Hoje, 31:56-59.

Oliveira-Filho, A.T. and M.A.L. Fontes. 2000. Patterns of floristic differentiation among Atlantic forests in Southeastern Brazil and the influence of climate. Biotropica, 32:793-810.

Rabinowitz, D., S. Cairns, and T. Dillon. 1986. Seven forms of rarity and their frequency in the flora of the British Isles; pp. 182-204. In: Soulé, M. E. (Ed.), Conservation Biology, the Science of Scarcity and Diversity. Sinauer, Sunderland.

Shine, R., L. Sun, M. Kearney, and M. Fitzgerald. 2002. Thermal correlates of foraging-site selection by Chinese pit-vipers (*Gloydius shedaoensis,* Viperidae). Journal of Thermal Biology, 27:405-412.

StatSoft, Inc. 2003. Statistica (data analysis software system), version 6. <http://www.statsoft.com>.

Sutherland, W.J. 1996. Introduction; pp. 11-110. In: W.J. Sutherland (Ed.), Ecological Census Techniques: A Handbook. Cambridge University Press, Cambridge.

Traill, L.W., C.J.A. Bradshaw, and B.W. Brook. 2007. Minimum viable population size: A meta-analysis of 30 years of published estimates. Biological Conservation, 139:159-166.

Zar, J.H. 1999. Biostatistical analysis. 4th ed. Prentice-Hall, New Jersey.

Received 20 March 2008
Accepted 27 June 2008