

DENSITY MEASUREMENTS FOR BIODIVERSITY STUDIES: THE
SPONGE *POLYMASTIA JANEIRENSIS* (BOURY-ESNAULT, 1973)
FROM BRAZIL

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KEY WORDS

Sponge, Porifera, *Polymastia*, density, biodiversity, statistical sampling.

ABSTRACT

Studies on the sponge *Polymastia janeirensis* (Boury-Esnault, 1973) were conducted at an average depth of 3 m on the southeast side of Praia Brava, Buzios, Rio de Janeiro, Brazil during March 4-6, 2001. The sponges measured an average of 17.5 cm x 23.9 cm (oval in shape) and their distribution was moderately aggregated (standardized Morisita Index: 0.49). Densities of *P. janeirensis* were determined using various methods. Density estimates were as follows: actual count 1.7/m²; stratified random sampling 5.8/m²; point center quarter 0.7/m²; 3rd nearest neighbor 1.6/m²; Weinberg 1.2/m²; Strong-Eberhardt 2.0/m²; and Nishiyama medium 1.1/m². The method tentatively recommended for estimating densities is the 3rd nearest neighbor, based on preliminary results from this study and other research projects.

INTRODUCTION

A recent worldwide interest is that of biodiversity. One primary problem has been attempts to estimate densities of sessile or non-mobile organisms (*e.g.*, sponges, trees) in a rapid and accurate manner in order to detect changes in the biota over time, due to pollution, deforestation, etc. The objective here is to simplify the sampling process for many people with environmental interests but no formal training, as well as making biologists and paleontologists aware of opportunities to improve their sampling operations. For these reasons we have used some established as well as new techniques to determine which is the most simple, rapid and accurate method of estimating population densities.



Fig. 1a. Brazil. Box indicates the general region of the study.

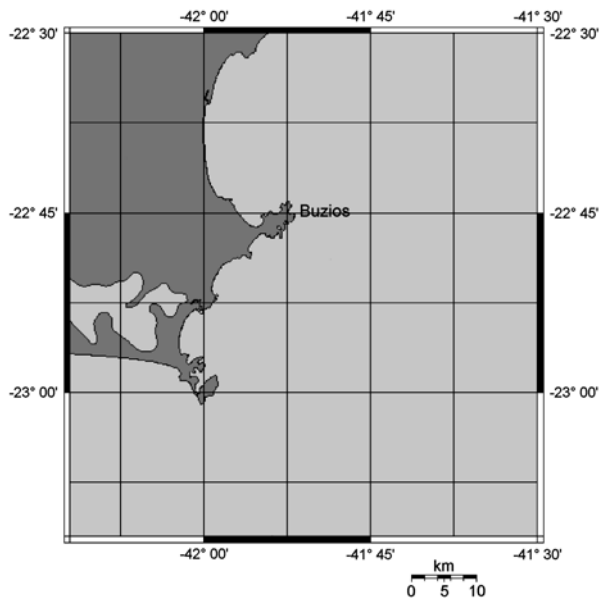


Fig. 1b. Cabo Frio region, Brazil.



Fig. 1c. The arrow indicates the study site, located 20-30 m to the left of the large rock, south side of Praia Brava, Buzios region, Brazil.

MATERIALS AND METHODS

Studies on the sponge *Polymastia janeirensis* (Boury-Esnault, 1973) (see also BOURY-ESNAULT *et al.*, 1994) were conducted at an average depth of 3 m about 30 m offshore on the southeast side of Praia Brava, Buzios, Rio de Janeiro, Brazil during March 4-6, 2001 (Figs 1a-c). Praia Brava is located about 190 km east of the city of Rio de Janeiro. The substratum was relatively flat, water temperature 27° C, the seas often choppy with underwater surge, and the water somewhat turbid. The benthic community was dominated by sponges (*P. janeirensis*), sea urchins [*Arbacia lixula* (Linne', 1758) and *Paracentrotus gaimardi* (Blainville, 1825)] in holes and under ledges, the seastar *Echinaster braziliensis* Müller and Troschel (1840), and calcareous red algae.

Ten transect lines, each 10 m long and 2 m apart, were arranged on a relatively flat seafloor beginning about 30 m offshore and perpendicular to the rocky coast (Fig. 1c). A count of sponges was made across the 10 transect lines within an area of 100 m². Stratified random sampling using 0.25 m² quadrats was conducted every 2 m (randomly selecting one of four quadrats using a table of random numbers) along each of the 10 transect lines. The quadrats were quickly constructed using a 2 m long folding carpenter rule, which is also very useful for short distance measurements. Cases where individuals of *P. janeirensis* were difficult to tell apart were minimal. Five transect lines were selected from the original 10 transects using a table of random numbers. Ten random points were selected along each of the five transect lines for the 3rd nearest neighbor and point center quarter (PCQ - see below) measurements. Line intercept data were collected from the five transect lines for the Weinberg (intercept lengths), Strong-Eberhardt (maximum orthogonal widths), and Nishiyama medium measurements (mean length and width of intercepting sponges). The null hypothesis states that there are no significant differences between the various density estimates used. Computer programs for the various density estimating methods were written in C++ by G.J. Bakus. These programs were tested earlier against known data for accuracy.

Equations for the density estimation techniques are found in WEINBERG (1981) and KREBS (1999). The unpublished equation for Nishiyama medium (designed for organisms 0.1 to 1.0 m in size) is as follows:

$$D = \frac{N (0.45 (W/L) + 0.59)}{\frac{CL}{L}}$$

Where D = density (No. org./m²)
 N = number of organisms intercepting the transect line
 W = mean width of organisms
 CL = contour transect line length on substratum*
 L = mean length of organisms

* the length of the tape pressed on the substratum so that it follows the contour of the reef. This length is longer than the length of the tape if held straight or taut, as there are ridges, dips, and even boulders on many rocky reefs.

RESULTS

Results of the surveys are presented in Tab. I. The sponges (n = 30) measured an average of 17.5 cm x 23.9 cm and were oval in shape. Their distribution or dispersion was moderately aggregated [standardized Morisita Index: 0.49 - see KREBS (1999)].

Tab. I. Density of *Polymastia janeirensis* calculated from different sampling techniques

Sampling Technique	No. Measurements	Density (No./m ²)	Difference in Density from Actual Count	*S.E (+ -)
Actual Count	100 m ² area	1.7	-----	-----
Stratified Random	50 (0.25 m ² quadrats)	5.8	4.1	0.2
Point Center Quarter	50 random points x 4 = 200	0.7	1.0	0.1
3 rd Nearest Neighbor	50	1.6	0.1	0.5
Weinberg	30	1.2	0.5	0.2
Strong-Eberhardt	30	2.0	0.3	0.6
Nishiyama medium	30	1.1	0.6	0.9

* standard error of mean density values

Morisita index of dispersion = 1.29

Standardized Morisita index of dispersion = 0.49

DISCUSSION AND CONCLUSIONS

The null hypothesis was rejected as significant differences were observed between density estimates. Theoretically, stratified random sampling would be the most accurate density estimate for organisms with uniform distributions but this is unlikely with a moderately aggregated distribution. The stratified random sampling

technique with 0.25 m² quadrats was the poorest sampling technique and took considerable time underwater because quadrat frames had to be placed in randomly selected positions. It is probable that a quadrat size of 1 m² would improve accuracy but square meter quadrats are even more difficult to handle underwater. The time to run both PCQ and 3rd Nearest Neighbor (3NN) sampling is about one-half of that for stratified random sampling. The standard error of the 3rd Nearest Neighbor is high, indicating considerable scatter in the data.

From our experience with similar studies in mountains, deserts, coral reefs, the marine intertidal or littoral zone, and simulation experiments, the 3rd Nearest Neighbor technique may provide the easiest and most accurate density estimate. The PCQ method usually works best with a random distribution. The PCQ method is the equivalent of measuring the 1st, 2nd, 3rd, and 4th nearest neighbors. Results are variable with aggregated distributions as there are many types of aggregation (KREBS, 1999). The Strong-Eberhart method occasionally offers good results with trees. The Weinberg method often works well with circular organisms such as hard corals. The Nishiyama medium method did not provide accurate density estimates and had the largest standard error of the mean.

The following scenario is tentatively suggested by the first author as the best overall technique for conducting density estimates in a relatively rapid and accurate manner.

i - Conduct a preliminary study using 3NN. Be certain to sample each part of a heterogeneous study site. Limit sampling to a small number of transects placed either perpendicular or parallel to the shoreline depending on the type of question asked. Alternatively, use random points (or random coordinates), or random plots in a grid system, superimposed on a map of the study region.

ii - Conduct a definitive density study in the same area. Proportion the sampling effort (*i.e.*, number of transects) according to the sizes of the heterogeneous parts. For example, if one-third of the substratum has boulders and two-thirds relatively flat rock, assuming 10 transects, sample three transects in the boulder region and seven transects on smooth rock. Assuming the transects are 30 m long, for example, select five random numbers between one and 30 from a table of random numbers (*e.g.*, 6, 9, 15, 24, 8) for each transect. Go to the first random number (*i.e.*, 6) along the first measuring tape or first transect line. Although 3NN is a plotless method of sampling (one lacking boundaries such as quadrats or plots), set some arbitrary distance as a boundary for convenience. This is critical for regions with high species richness as many species may be uncommon or rare and difficult or impossible to locate. It is also critical to have boundaries for underwater studies as it may take far too much time to locate species. As one moves away from a random point, the boundaries become more difficult to locate. Measure the distance from the random point to the third nearest individual of the species of interest. Continue the measurements for the remaining random points and transects. A minimum of 50 random points is advisable, based on recommendations for the PCQ method (WARD & PETRANKA, 1981).

iii - Third nearest neighbors may be missing, especially in species rich areas and underwater. WARD & PETRANKA (1981) produced a PCQ table with corrections for this. This correction would also work well for 3NN because the correction table is

based on the ratio between the number of zeros (*i.e.*, organisms missing) and the total number of measurements.

iv - Calculate a separate density estimate for each heterogeneous area.

v - Average the densities for the entire area if an overall density is desired. The computer program 3NN, written in C++ by the first author, will do the calculations.

The 3NN method is desirable for studying the densities of one to a few species. If densities of many or all species are desired then belt or strip transects (*e.g.*, 30 m X 1 m) are preferable. Direct counts of species can then be made by swimming along a tape holding a one meter rod perpendicular to the tape and recording the results.

Organisms that cannot be counted (*e.g.*, some algae) are another problem. Quadrats will have to be used and the percentage cover estimated.

Effective visual estimates of percentage cover have been made with 10 cm by 10 cm quadrats (DETHIER *et al.*, 1993). Percentage cover can also be obtained by determining total intercept lengths and dividing by the total transect length (BAKUS & NISHIYAMA, 1999).

Rapid methods of estimating densities include swimming along a transect line using a digital videocamera. Species of interest can be marked then automatically counted in randomly selected digital frames using the computer program PointCount 99 (available from Dr. Phil Dustan, Department of Biology, College of Charleston, Charleston, SC 29424. E-mail: dustanp@cofc.edu). This method assumes that selected species can be identified in all random frames and that no organisms are covered by algae or corals, or live in cracks and crevices.

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A PDF file of this paper and a computer program for 3NN and other density measurement techniques are available from: bakus@worldnet.att.net

REFERENCES

- BAKUS G.J., NISHIYAMA G.K., 1999 - Sponge distribution and coral reef community structure off Mactan Island, Cebu, Philippines. *Mem. Queensl. Mus.*, **44**: 45-50.
- BOURY-ESNAULT N., 1973 - Campagnes de la Calypso au large des cotes atlantiques de l'Amerique du Sud (1961-1962). 29. Spongiares. *Résult. Scient. Camp. Calypso*, **10**: 263-295.
- BOURY-ESNAULT N., HAJDU E., KLAUTAU M., CUSTODIO M., BOROJEVIC R., 1994 - The value of cytological criteria in distinguishing sponges at the species level: the example of the genus *Polymastia*. *Can. J. Zool.*, **72**: 795-804.
- DETHIER M.N., GRAHAM E.S., COHEN S., TEAR L.M., 1993 - Visual versus random-point percent cover estimations: 'objective' is not always better. *Mar. Ecol. Prog. Ser.*, **96**: 93-100.
- KREBS C.J., 1999 - Ecological Methodology. Addison Wesley Longman, Inc., Menlo Park, CA, 620 pp.
- WARDE W., PETRANKA J.W., 1981 - A correction factor table for missing point-center quarter data. *Ecology*, **62**: 491-494.
- WEINBERG S., 1981 - A comparison of coral reef survey methods. *Bijdr. Dierkd.*, **51** (2): 199-218.