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# ASSESSING BIAS IN AERIAL SURVEYS FOR THREATENED CETACEANS: RESULTS FROM EXPERIMENTS CONDUCTED WITH THE FRANCISCANA (*PONTOPORIA BLAINVILLEI*)

Federico Sucunza<sup>1,2,3</sup>, Daniel Danilewicz<sup>2,3</sup>, Eduardo R. Secchi<sup>4</sup>, Artur Andriolo<sup>1,3</sup>, Marta Cremer<sup>5</sup>, Paulo A. C. Flores<sup>6</sup>, Emanuel Ferreira<sup>7</sup>, Luiz Claudio P. de S. Alves<sup>3</sup>, Franciele R. de Castro<sup>3</sup>, Dan Pretto<sup>6</sup>, Camila M. Sartori<sup>5</sup>, Beatriz Schulze<sup>5</sup>, Pablo Denuncio<sup>8</sup>, Martin Sucunza Perez<sup>2</sup>, Paulo H. Ott<sup>2</sup> and Alexandre N. Zerbini<sup>3,9,10</sup>

1- Laboratório de Ecologia Comportamental e Bioacústica, Programa de Pós-graduação em Ecologia, Universidade Federal de Juiz de Fora, Juiz de Fora, MG, Brazil.

2- Grupo de Estudos de Mamíferos Aquáticos do Rio Grande do Sul, Torres, RS, Brazil.

3- Instituto Aqualie, Juiz de Fora, MG, Brazil.

4- Laboratório de Ecologia e Conservação da Megafauna Marinha, Instituto de Oceanografia, Universidade Federal do Rio Grande, Rio Grande, RS, Brazil.

5- Laboratório de Ecologia e Conservação de Tetrápodes Marinhos e Costeiros, Unidade São Francisco do Sul, Universidade da Região de Joinville, São Francisco do Sul, SC, Brazil.

6- Centro Mamíferos Aquáticos, Instituto Chico Mendes para a Conservação da Biodiversidade, Florianópolis, SC, Brazil.

7- Associação R3 Animal, Florianópolis, SC, Brazil.

8- Instituto de Investigaciones Marinas y Costeras, Departamento de Ciencias Marinas, Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Mar del Plata, Mar del Plata, Argentina.

9- Joint Institute for the Study of the Atmosphere and Ocean, University of Washington and Marine Mammal Laboratory, Alaska Fisheries Science Center, Seattle, WA, USA.

10- Marine Ecology and Telemetry Research, Seabeck, WA, USA.

## Summary

1. Line transect aerial surveys are widely used for estimating abundance of biological populations, including threatened species. However, estimates obtained with data collected from aircrafts are often underestimated because of visibility bias or bias in estimating group sizes from a fast-moving platform.

2. An assessment of multiple sources of bias in aerial surveys were carried out in southern Brazil by experiments multiple survey platforms (e.g., boats and aircrafts). These studies focused on evaluating visibility bias (perception and availability bias) and potential differences in the estimation of group sizes from different types of platforms used in franciscana (*Pontoporia blainvillei*) abundance surveys. The ultimate goal was to develop of correction factors to improve accuracy of estimates of density and population size for this threatened dolphin.

3. Estimates of density and group sizes computed from boats were assumed to be unbiased and were compared to estimates of these quantities obtained from an airplane. A correction factor ( $CF=4.42$ ,  $CV=0.04$ ) was computed as the ratio of the density estimated by boats  $2.99 \text{ ind/km}^2$  ( $CV=0.23$ ) and by the aircraft  $0.68 \text{ ind/km}^2$  ( $CV=0.28$ ). Availability of franciscana groups was estimated at  $0.39$  ( $SE = 0.009$ ).
4. Visibility bias was substantial and accounted for  $\sim 70\%$  of the total bias. Group sizes estimates from the boats were significantly different ( $\sim 30\%$  larger) than those from the aircraft and accounts for a relatively large proportion of the bias in the aerial survey estimates of density.
5. The correction factor reported above can be used to refine range wide abundance estimates of franciscanas given certain assumptions are met. The lack of observed effects of environmental variables (e.g. depth and water transparency) on franciscana groups availability indicates the potential use of the independent estimated availability bias along all the species range.

## **1. Introduction**

Aircrafts are widely used to conduct surveys for wildlife populations, mainly because they provide the opportunity to search large and/or inaccessible areas in a relatively short period of time (e.g. Hiby and Hammond 1989, Andriolo et al. 2010, Sucunza et al. 2015, McLellan et al. 2019). However, aerial surveys are commonly plagued by imperfect counts of individuals or groups that are within the sampling area (Caughley 1972, Barlow et al. 1988, Heide-Jørgensen et al. 2007, Sucunza et al. 2020). Bias results from a variety of factors and, if not accounted for, can lead to equivocal conservation actions.

Aerial surveys for cetaceans are often carried out using line transect methods (Buckland et al. 2001, 2004). These methods assume that all individuals or group of individuals are seen on the survey trackline ( $g(0) = 1$ ) and that group sizes are accurately estimated (Buckland et al. 2001). Because cetacean species spend periods of time unavailable to be seen neither of these assumptions often hold during aircraft surveys (e.g. Laake et al. 1997, Pollock et al. 2006, Sucunza et al. 2018, Boyd et al. 2019). Marsh and Sinclair (1989) defined two categories for

visibility bias (animals missed on the survey line): availability bias occurs when animals are unavailable to be detected during a passing observer (e.g. on a plane) because they are submerged and perception bias occurs when animals are available but not detected (e.g. due to observer fatigue). In addition, a variable proportion of the individuals within a group may be available at the same time to be counted which makes the estimation of group size of marine mammals species imprecise (Gilpatrick 1993, Gerrodette et al. 2018, Boyd et al. 2019). The relatively high speed of the aircrafts reduces the time an observer has to search through a given area, resulting in a higher proportion of undetected animals as well as in underestimation of the total number of individuals in a group. In this sense, experiments to investigate the magnitude of bias in aerial surveys are essential to produce robust results and, consequently, promote conservation.

The franciscana (*Pontoporia blainvillei*) is a small dolphin endemic to coastal waters off the eastern coast of South America. The species occurs in waters typically shallower than 30 m (Danilewicz et al. 2009) between Itaúnas, Brazil (18°25'S) and Golfo San Matías, Argentina (41°10'S) (Crespo et al. 1998, Siciliano et al. 2002). The species is regarded as one of the most threatened small cetaceans in the western South Atlantic Ocean due to high, possibly unsustainable, bycatch levels as well as increasing habitat degradation throughout its range (Ott et al. 2002, Secchi et al. 2003, Secchi 2010) and is listed as Vulnerable by the IUCN Red List of Threatened Species (Zerbini et al. 2017).

Aerial surveys have been considered the most appropriate survey method to estimate abundance of franciscanas (e.g. Secchi et al. 2001, Crespo et al. 2002). However, developing abundance estimates from aerial surveys for this species can be challenging because franciscanas are difficult to detect from fast-moving platforms. In addition, surface-based observations have suggested that franciscana groups seen from airplanes are often two to four times smaller than those seen from stationary or slow moving platforms (Bordino et al. 1999, Cremer and Simoes-Lopes 2008, Crespo et al. 2010, Danilewicz et al. 2010), indicating that biases in estimates of the size of groups from an fast-moving, aerial platform can be substantial.

In this study, experiments to investigate potential sources of visibility bias and group size bias in aerial survey of franciscanas are described and correction factors to improve/correct estimates of abundance of the species are proposed.

## 2. Methods

Two experiments were conducted to estimate visibility bias and group size bias in abundance numbers from data recorded during aircraft surveys: Experiment 1 used simultaneous aerial and boat surveys to assess differences in density and group sizes of franciscanas between the two platforms and experiment 2 used helicopters to evaluate behavior of franciscanas observed from aerial platforms.

### 2.1. Experiment 1

#### 2.1.1. Study Area and Survey Design

Concomitant aerial and boat-based surveys were conducted in Babitonga Bay (26°16'S, 048°42'W), southern Brazil from 13 to 24 February 2011. Babitonga Bay is a shallow (average depth 6 m) small estuarine area in northern Santa Catarina State (SC), southern Brazil (Cremer and Simões-Lopes 2008) (Fig. 1). This area presents a number of advantages for the type of study intended here: (1) it is a region where franciscanas predictably occur in relatively large densities throughout the year and show limited or no avoidance to small boats (Cremer and Simões-Lopes 2008), (2) group sizes seen in the bay are believed to be representative of those seen through most of the franciscana range and (3) the bay is relatively protected and therefore provides good weather conditions (e.g. relatively calm waters) for sighting surveys.

A planned area of 160km<sup>2</sup> (Area A, Fig. 1) was defined based on locations where franciscanas were known to occur (e.g. Cremer and Simões-Lopes 2008). Aerial and boat surveys followed design-based line transect methods (Buckland et al. 2001). A sampling grid of 16-17 equally spaced (at 600 m from each other) tracklines was proposed. To ensure sampling was random and independent for each platform, the starting point of the grid was randomly selected for each realization of the design for both survey platform types. The total trackline length (74 km) of the design was specified in a way that the planned area could be fully surveyed by two boats in a period of four hours. This period was chosen to maximize sampling during calm weather, typically observed in this region between dawn and noon. In this four-hour period, the airplane could complete 3-4 realizations of the trackline design.

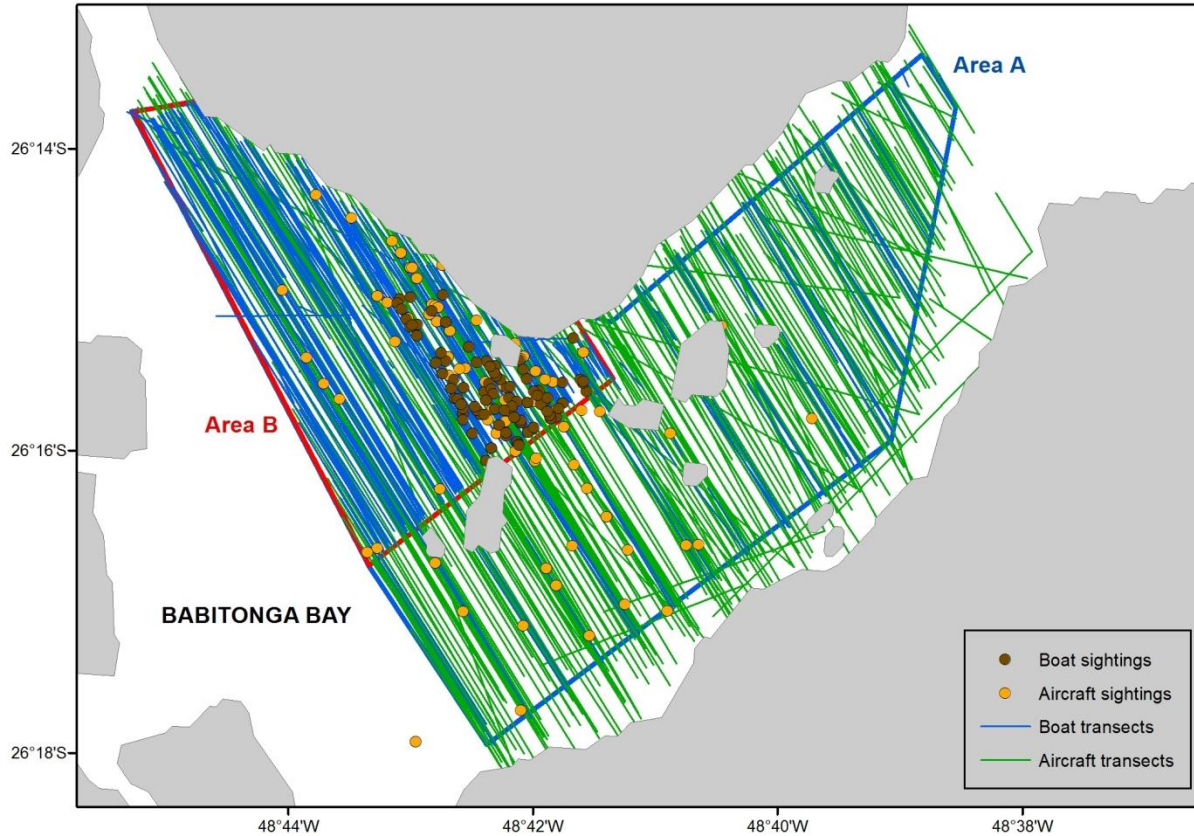


Figure 1. Map of Babitonga Bay, southern Brazil, showing survey areas, realized trackline effort and franciscana sightings for both aircraft and boats.

After the first two survey days, it became clear that franciscanas were concentrated in a smaller region within the planned region. Based on this information and because of identified restrictions for the navigability of some planned tracklines, the sampling area for the boat surveys was reduced (Area B - 16.48 km<sup>2</sup>, Fig. 1) to maximize records of franciscana groups. The trackline design, however, maintained the same line spacing as the original design. The sampling strategy was not modified for the airplane because it could cover the entire survey area (Area A) much faster and because sample sizes collected on the first two days indicated that sufficient sightings (60-80 records, Buckland et al. 2001) would be recorded for estimation of detection probability for this platform. For the purpose of the analysis presented here, only data collected in Area B for both platforms is considered for density estimation.

### 2.1.2. Field Methods

Sampling occurred under good weather conditions and calm seas (Beaufort Sea State < 3). Water transparency was measured with a Secchi disc at the beginning, middle and end of every boat transect and cloud cover was registered once changes were observed. Surveys were conducted in “passing mode” for both the aircraft and the boats.

#### 2.1.2.1 Aerial surveys

Visual surveys were made from a high-wing, twin-engine *Aerocommander 500B* aircraft at an approximately constant altitude of 150 m (500 ft) and a speed of 170-200 km/h (~90-110 knots). The aircraft had four observation positions (two on each side of the plane), with bubble and flat windows available for front and rear observers, respectively. Different window configuration resulted in a partial overlap in the front and rear observer’s field of view (beyond 80 m from the trackline). Observers worked independently during on effort periods, with neither visual nor acoustic communication. The beginning and the end of each transects were informed to the observers by the pilot. Data were recorded by each observer on audio digital recorders and every record was time-referenced based on digital watches synchronized to a GPS. Environmental data (e.g. Beaufort sea state, water transparency, intensity of glare) was recorded at the beginning and end of each transect or whenever conditions changed. When a group of dolphins was detected, the species and the size of the group were recorded. The declination angle between the horizontal and the sighting was obtained using an inclinometer when the group passed a beam of the observer. Additional information such as presence of calves, Beaufort sea state, and water transparency were also recorded along with each sighting.

#### 2.1.2.2. Boat surveys

Visual surveys were conducted with two small (5 and 6 m) open boats equipped with 40 and 60 hp outboard engines and a crew of four people: two observers, a data recorder and a pilot. The observers were located at the bow of the boat and searched for dolphins groups with the naked eyes. Observers on the left and right of the bow searched for a 0-50° to the port and starboard,

respectively. Once a group was detected, information on the estimated radial distance to the group, the radial angle (measured with an angle board), the species and the group size were relayed to the recorder and registered in a standard data sheet. The recorder was not involved in searching or distance estimation, but assisted the observers in identifying species, tracking detected groups, and estimating group size and group composition. Sightings recorded during transit between transects or from or to the harbor were considered off effort sightings.

There is evidence that group size estimation during passing mode can be biased low because observers do not spend sufficient time to obtain an accurate count of the individuals in a group (e.g. Gerrodette et al. 2018). To assess whether this occurred in this study, the boats returned to areas of high density after the end of certain transect lines and randomly approached franciscana groups. A count of individuals in the group during these ‘off-effort’/‘closing’ approaches was then compared to group size estimation on the transect lines.

#### 2.1.2.3 Distance calibration experiments

Because observers on the boats estimated the radial distance for the groups with the naked eyes, experiments were conducted to assess measurement error in distance estimation and to correct for such error for each individual observer. The experiment was repeated three times during the study, the first before the surveys started, the second halfway through the survey period and the last one at the end of the study. During these experiments, five observers (two for each boat and a standby observer) stood in a fixed platform and independently estimated their distance from a moored object painted with colors resembling the franciscana color pattern. This object was placed at various known distances (measured with a GPS) from the platform. The experiment was conducted in a location with similar visibility conditions to those found in the survey area and the distances at which the moored object was placed from the observers were within the range franciscanas were seen in boat surveys previously conducted in Babitonga Bay (Cremer and Simões-Lopes 2008). For each of the three experiments, 12 distance estimates were obtained for each observer. True (measured) and estimated distances were used to correct for bias in radial distance estimation in a regression framework (Williams et al. 2007).

### 2.1.3. Analytical Methods

#### 2.1.3.1. Magnitude of bias in group size estimates from the airplane

A generalized linear model (GLM) with a Poisson error structure was used to assess differences in group sizes estimated from the boats and the airplane. This potential difference is interpreted here as the bias in groups size estimates from the airplane, assuming that estimates from the boats were unbiased. The GLM takes the following form:

$$\text{Log}(\mu) = \beta_0 + \beta_1 x_1 + \beta_k x_k + \varepsilon$$

Where:  $\mu$  is the response variable (group size-1),  $\beta_0$  is the intercept,  $\beta_1 \dots \beta_k$  are the coefficients for the  $x_1 \dots x_k$  predictor variables (distance - numerical covariate, and platform - factor covariate with two levels "boat" and "airplane") and  $\varepsilon$  is an error term.

Four models were proposed and Akaike weights  $w_i$  were calculated for each model as a representation of the probability of the model be the actual “best model” within the full set of models (Burnham and Anderson 2002). Inference about the relative importance (RI) of each predictor variable in determining the group size was based on the sum of Akaike weights of each variable across all candidate models containing the variable, and ranged from 1 (most important) to 0 (least important). Model averaging was conducted across a set of models including all possible permutation of the two predictor variables, and model-averaged parameters were estimated for each predictor variable, with unconditional standard errors incorporating model uncertainty (Burnham and Anderson 2002). Model assumptions were verified by plotting residual versus fitted values and versus each covariates in the model (Zuur and Ieno 2016). Model averaging was performed using the package MuMIn (Barton 2017).

Because the perspective from what constitute a group may differ for observers searching from boats or airplanes, in this study observers from both platforms were trained to use the same group definition: an aggregation of dolphins in close proximity of each other (within ~10 body lengths), moving in the same direction and in apparent association (Shane 1990).

#### 2.1.3.2. Estimation of Detection Probability

Detection probability was estimated using Conventional (CDS) and Multiple Covariate Distance Sampling (MCDS) methods (Buckland et al. 2001, Marques and Buckland 2003). MCDS differs from CDS as it allows for the inclusion of multiple covariates in the estimation of detection probability (Marques and Buckland 2003). Only the half-normal and the hazard-rate detection functions were proposed to fit distance data for both platforms. Exploratory analyses indicated that adequate fits were obtained by modeling grouped distance data for both platforms (plane grouping intervals: 0-30m, 30-60, 60-130, 130-200m, 200-270m; boats grouping intervals: 0-25m, 25-55m, 55-90m, 90-130m, 130-180m). Beaufort sea state (factor covariate with two levels: "calm", Beaufort sea state between 0 and 1, and "high" between 2 and 3), glare (factor covariate with two levels "presence" and "absence") and group size (numerical covariate) were considered as covariates to model distance data from the airplane. For the boat data analyses, only Beaufort sea state (factor covariate with two levels: "calm", Beaufort sea state  $\leq 0$ , and "high" between 1 and 2) was considered as a covariate. Models were ranked according the Akaike Information Criterion (AIC), and model averaging were performed to incorporate unconditional model selection variance in the estimates and confidence intervals (Burnham and Anderson 2002). Analyses were performed using a set of customized functions (mrds v.2.2.0, Laake et al. 2018) in R (R Development Core Team 2018). Only data recorded by the front observers in the airplane (bubble windows) are considered in the analysis presented in this study because of the field of view between front and rear observers only partially overlapped (Sucunza et al. 2019). Perpendicular distance estimated from the boats were corrected for each observer considering the calibration experiments described above prior to estimation of detection probability for that platform.

#### 2.1.3.3. Group Size, Density, and Abundance Estimation

Abundance of groups ( $N_g$ ) and individuals ( $N_i$ ) was estimated using the Horvitz-Thompson (H-T) estimator as follows (Marques and Buckland 2003):

$$\hat{N}_g = \sum_{i=1}^n \frac{1}{\hat{p}(z_i)}$$

$$\hat{N}_i = \sum_{i=1}^n \frac{s_i}{\hat{p}(z_i)}$$

Where:

$n$  – number of groups recorded;  $s_i$  – group size of each recorded group  $i$ ;  $\hat{p}(z_i)$  – detection probability for vector of sighting-specific covariates  $z$  for each recorded group  $i$ .

Expected group size was estimated by dividing  $N_i/N_g$  (Innes et al. 2002). Variance was estimating using the analytical estimator of Innes et al. (2002) and Log-normal 95% confidence intervals (Buckland et al. 2001) were computed after unconditional variance was derived (Zerbini et al. 2006).

#### 2.1.3.4. Computing a Correction Factor for Aerial Surveys

A factor to correct for visibility and group size biases in aerial survey-based estimates of density was computed from the following ratio:

$$CF = \frac{\hat{D}_{boat}}{\hat{D}_{plane}}$$

and variance for this CF was approximated by the delta method (Seber 1982).

This CF assumes that no visibility bias occurred in the density estimated by the boat survey (i.e.  $g(0)_{boat} = 1$ ) and that the size of the group detected from this platform was accurately estimated (i.e. underestimation of group size by the boat observers would result in an underestimation of the CF and vice versa).

## 2.2. Experiment 2.

### 2.2.1. Assessment of availability of franciscana groups

Data on availability of franciscana groups was obtained from helicopter surveys conducted in Babitonga Bay from 23 to 31 January 2014 (Sucunza et al. 2018), and in Ubatuba (23°28'S, 045°03'W), State of São Paulo (SP), southeastern Brazil from 28 November to 15 December 2016. Studies conducted in Babitonga Bay have proved useful to assess availability of franciscanas (Sucunza et al. 2018). However, the visibility conditions (typically murky and shallow waters) in this region is similar to that in only part of the range of the species. Therefore, sampling in more heterogeneous habitats were required for correction factors to be more representative of all franciscana habitats. In these sense, new helicopter surveys were conducted in Ubatuba, a relative high-density area for this species and a region with contrasting environmental conditions from those of Babitonga Bay (i.e., clearer and deeper waters).

A four-seat helicopter Robinson R44 was used during visual surveys in both regions. Flights were conducted at 150 m (500 ft), an altitude consistent with that flow during aerial surveys to estimate abundance of franciscanas (e.g. Secchi et al. 2001, Danilewicz et al. 2010, Crespo et al.

2010, Sucunza et al. 2019). Surveys were carried out during the morning in calm conditions (Beaufort sea state < 3), and had an average duration of 4 h. To maximize visibility for the observers, the doors of the helicopter were detached. Two observers with substantial experience in aerial surveys and familiar with the identification of franciscanas searched for groups of dolphins on the left side of the helicopter. Once a group was detected, the pilot hovered over it and each observer recorded surfacing and dive times independently. A group was defined as an aggregation of dolphins in close proximity of each other, moving in the same direction and in apparent association (Shane 1990). Each observer was responsible for recording biological (e.g., group size, presence of calves) and environmental (e.g. Beaufort sea state, water color) variables. Depth and water transparency (measured with a Secchi disc) at the location of each sighting were recorded from boats operating in the same area and in radio communication with the helicopter. A detailed description of data collection is presented in Sucunza et al. (2018).

A surfacing interval was defined as the period of time in which at least one individual in a group of franciscanas was visually available, at or near the surface, to the observer in a helicopter while a diving interval was defined as the period of time in which all individuals of the group were not visible. A surface-dive cycle was defined as the period from the beginning of one surfacing to the next. The proportion of time at surface was calculated as the ratio between a surfacing period and a surface-dive cycle.

Generalized linear mixed-effects model (GLMM) were used to evaluate the effects of biological and environmental predictors on the proportion at surface (the response variable) using the package nlme (Pinheiro and Bates 2019). Model-averaged parameters were estimated for each predictor variable following the modeling processes described in Sucunza et al. (2018).

To estimate the probability of one franciscana group be visually available within the visual range of a passing observer in a fixed-wing aircraft, or availability of franciscana groups, the model proposed by Laake et al. (1997) was used:

$$\widehat{Pr} = \frac{E(s)}{E(s)E(d)} + \frac{E(m)[1 - e^{-\frac{w(x)}{E(m)}}]}{E(s) + E(d)}$$

where  $E(s)$ ,  $w(x)$ , and  $E(d)$ , correspond, respectively, to the mean time of each individual surfacing interval, the window of time during which a franciscana group is in the observer's view at a distance  $x$  ( $w(0) = 6$  seconds, Sucunza et al. 2018) and the mean time of each individual diving interval. Standard errors and confidence intervals of  $\widehat{Pr}$  were estimated with 1,000 replicates of a nonparametric bootstrap procedure (Manly 2006).

Additional data on franciscana availability was obtained using an artificial franciscana model. The model was constructed using a fresh carcass from a franciscana by-caught in southern Brazil, which makes it identical to an adult franciscana. The experiment followed the methods proposed by Pollock et al. (2006). The model was positioned at different depths in the water column, and each observer in the helicopter recorded if the model was or not recognizable for detection of a passing observer in a fixed-wing aircraft.

### 3. Results

#### 3.1. Experiment 1

The realized effort in areas A and B by boat and aircraft are summarized in Table 1. In nearly 1,900 km of trackline sampled by both platforms, a total of 356 franciscana groups were recorded.

Table 1. Survey effort conducted by boats and airplane to estimate density of franciscanas in Babitonga Bay, southern Brazil, in February 2011. Survey effort in Area B represents the effort used for density estimation.

	Boats	Airplane
Total survey effort (km) in Areas A and B	551	1,396
Survey effort (km) in Area B	447	476

### 3.1.1. Group Size

Group size statistics for the franciscana aerial and boat surveys in Babitonga Bay are summarized in Table 2. Group sizes varied between 1 and 7 individuals for both platforms.

Table 2 – Summary of average (SE in parenthesis) group sizes of franciscanas in Babitonga Bay, southern Brazil in February 2011.

	Boat		Plane					
	All		Front		Rear		All	
	Mean (SE)	n	Mean (SE)	n	Mean (SE)	n	Mean (SE)	n
On effort groups	2.96 (1.20)	114	2.18 (0.96)	91	1.95 (1.20)	60	2.09 (1.06)	151
Off effort groups	2.87 (1.08)	50	2.35 (1.47)	31	2.4 (1.26)	10	2.36 (1.40)	41
Total		164		122		70		192

The most parsimonious approximating GLM to assess the influence of distance and platform to the group sizes estimates included only platform as the predictor variables (Table 3). This model suggested that group sizes estimates from the aircraft were significantly smaller than those from the boat. Predicted group sizes for each platform computed from the model-averaged predictor coefficients indicated that groups seen from the boat is, on average, 33% greater than those seen from the airplane (Table 4).

Table 3 – Models proposed to assess differences in group size estimation between boat and aircraft.  $w_i$  = Akaike weights.

Model	Explanatory variables	AIC <sub>c</sub>	$\Delta$ AIC <sub>c</sub>	$w_i$
#1	Platform	565.80	0.00	0.52
2	Distance and platform	565.94	0.14	0.48
3	Null	579.16	13.36	0.00
4	Distance	581.21	15.40	0.00

Table 4. Model-averaged parameter estimates. SE = standard error. RI = relative importance.

Parameter	Mean	SE	RI	$P$
Intercept	0.594	0.107	-	<0.001
Platform (plane)	-0.491	0.127	1.00	<0.001
Distance	1.602	1.149	0.48	0.166

There was no significant difference in group sizes estimated by observer on the boat while surveying the transect lines (passing mode, mean = 2.96, SE = 1.20) and when groups were approached off effort (mean = 2.87, SE = 1.08) for a more accurate estimation of the number of individuals in the group ( $p$ -value = 0.71).

### 3.1.2. Distance Calibration

Radial distance data was log-transformed to address the observed heteroscedasticity problem in the least-square regression. One out of five observers tended to underestimate distance by 9% on average. All other observers overestimated distance by on average 8-40%. Results of the calibration experiment are summarized in Table 5

Table 5. Observer bias in estimating radial distance from calibration experiments.

Observer	Bias	p-value
1	+34%	<0.001
2	+8%	0.002
3	-9%	<0.001
4	+19%	0.083
5	+40%	0.421

### 3.1.3 Density and Abundance Estimates and Correction Factor Computation

The hazard rate model with size covariate or with Beaufort sea state covariate provided the best fit for perpendicular distance data for airplane and boats, respectively (Fig. 2, Table 6). Boat ( $2.99 \text{ ind/km}^2$ , 95% CI = 1.92-4.66) and plane ( $0.68 \text{ ind/km}^2$ , 95% CI = 0.39-1.16) densities were significantly different and the ratio of the two resulted in a correction factor of 4.42 (CV=0.04). Quantities related to density and abundance estimation are summarized in Table 6.

Table 6. Quantities used for estimation of density of franciscanas in Babitonga Bay, southern Brazil in February 2011. Coefficients of variation are shown in parenthesis when applicable.

	Boats	Airplane
Survey effort	447	476
On effort sightings in Area B	114	56*
Encounter rate	0.69 (0.21)	0.12 (0.24)
Number of sightings used in fitting the detection function	108	88
Average detection probability ( $p$ )	0.65 (0.08)	0.67 (0.09)
Expected group size <sup>1</sup>	2.91 (0.04)	2.04 (0.09)
Density	2.99 (0.23)	0.68 (0.28)
Abundance	49 (0.23)	11 (0.28)

\*Sightings recorded only from front observers; <sup>1</sup>Expected group size was computed after truncation and fitting a detection probability function.

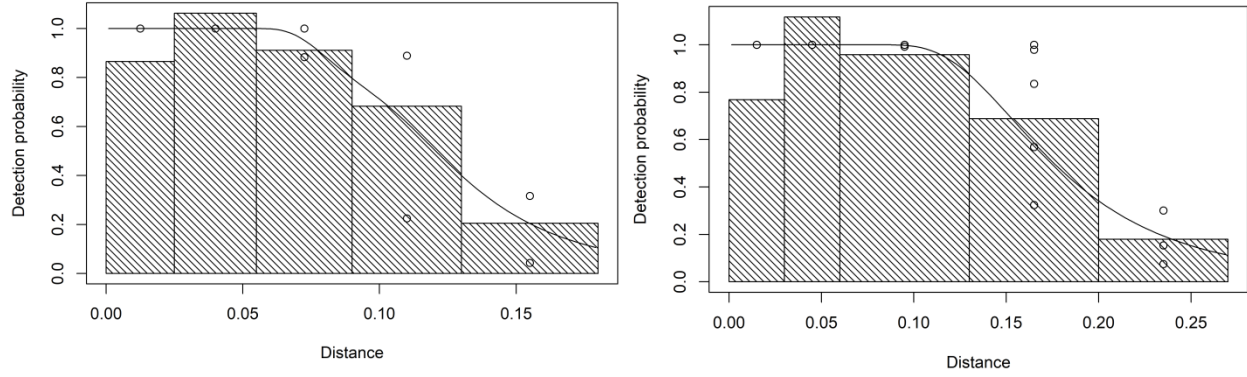


Figure 2. - Detection probability functions fit to perpendicular distance data collected in Babitonga Bay by the boats (left) and the aircraft (right).

### 3.2. Experiment 2

A total of 45 hours was flown during the helicopter experiments in Babitonga Bay (15hs) and Ubatuba (30hs). A total of 373 complete surface-dive cycles were recorded for 167 franciscana groups. Biological and environmental variables recorded in both areas are summarized in Table 7. The depth at which the franciscana model became recognizable to an aerial platform at 150 m of altitude vary between the areas from 1.40 m in Babitonga Bay and 2.25 m in Ubatuba.

The most parsimonious GLMM only included group size as the predictor variable, suggesting a significant positive effect on the proportion of time at surface (Table 8). Group was the most important predictor variable ( $RI = 1.0$ ) and was significant in some but not all models. All the other predictor variables were non-significant in all models. Model was validated by plotting residuals versus fitted values and versus each covariate in the model.

Surfacing and dive intervals were significantly smaller in Babitonga Bay than in Ubatuba ( $p < 0.001$ ), but the proportion of time at surface did not vary significantly between the study areas (Babitonga Bay = 0.36, Ubatuba = 0.34,  $p = 0.32$ ) (Table 9). The estimated window of time  $w(0) = 6$  seconds, resulted in an estimation of availability of 0.39 ( $SE = 0.009$ ) for both areas combined.

Table 7. Summary of biological and environmental variables recorded in Babitonga Bay and Ubatuba region and tested in the generalized mixed-effects models. SE= standard error.

Variable	Factor/Numeric	Levels	Mean	SE
Group size	Factor	<i>small</i> (1-3) and <i>large</i> (4-7)	3.03	1.13
Presence of calves	Factor	yes and <i>no</i>	0.33*	-
Water transparency (m)	Numeric	0.77-7.16	2.49	1.84
Depth (m)	Numeric	4.4-17.3	9.90	3.62

\*Proportion of groups (n = 167) with calves (n = 56)

Table 8. Model-averaged predictor coefficients and relative importance (RI).  $\beta$  = coefficients values for the averaged model, SE= standard error.

Parameter	<i>B</i>	SE	RI	<i>P</i>
Group size - <i>large</i>	0.20	0.07	1.00	0.007
Transparency	0.01	0.01	0.24	0.31
Depth	0.008	0.009	0.23	0.33
Presence of calves - yes	0.003	0.07	0.14	0.97

Table 9. Summary of franciscana groups surface-dive cycles data recorded during helicopter experiments in Babitonga Bay (BB) and Ubatuba region. n-groups = total number of groups, n-cycles = total number of surface-dive cycles. Standard errors shown in parenthesis when applicable.

Area	n-groups	n-cycles	Mean surface (sec.)	Mean dive (sec.)	Proportion at surface
BB	101	248	16.10 (9.75)	39.77 (29.06)	0.36 (0.23)
Ubatuba	66	125	38.78 (13.07)	77.26 (19.98)	0.34 (0.09)
Total	167	373	23.70 (15.33)	52.33 (31.75)	0.35 (0.19)

#### 4. Discussion

This study clearly demonstrates that estimates of franciscana density/abundance from aircraft are biased low to a relatively large extent if no correction is applied for visibility and group size bias. Bias in these two quantities correspond to the main factors affecting estimates of other cetacean species' occurrence and abundance (e.g. Cockcroft et al. 1992, Gu and Swihart 2004, Fuentes et al. 2015, Williams et al. 2016, Williams et al. 2017), and, although a variety of techniques have been developed to correct for these biases (e.g. Marsh and Sinclair 1989, Laake et al. 1997, Borchers et al. 2006, Pollock et al. 2006, Thomson et al. 2012, Gerrodette et al. 2018), address it is a challenge frequently not achieved. The present results indicate that abundance estimates computed from aerial surveys data underestimate the true abundance of franciscanas by about 4 times.

Because cetaceans remain at the surface for relatively short periods of time, observers tend to underestimate the number of individuals in a group (Gilpatrick et al. 1993, Gerrodette et al. 2018, Boyd et al. 2019). The fast speed of the aircraft reduces the period of time that a dolphins group is within the observers view, reducing the time available to precisely count and thus increasing the magnitude that group sizes are underestimated by an observer in an aircraft.

Results of this study shown that there is a significant negative bias (~30%) in the estimated size of franciscana groups in aerial surveys. This relies on the assumption that observers from the surface (i.e. boats) and from the aerial (i.e. aircraft) platforms used the same group definition and that estimates of group size from the boats were unbiased. Both assumptions were considered to be achieved in this study because there were no doubt between observers about group definition, and because groups seen off effort during boats surveys (i.e. those for which group sizes were estimated after observers spent significant more time with the animals) were not statistically different from those seen during passing mode while sampling survey lines. In addition, the range and mean group size estimated from the boats in this study (mean = 2.91, range = 1-7) were identical to those obtained during an independent experiment conducted from helicopter in the same area at a different time period (mean = 2.90, range = 1-7, Sucunza et al. 2018), suggesting that group definition was consistent between surface- and aerial-based observers and that estimates of group size from the boats were unbiased. However, if group sizes estimated from the boats are biased low, the ~30% group size bias computed here for the airplane is also negatively biased.

Another way of assessing bias in group size estimates from the aircraft would be to compare the expected group sizes computed with the Horvitz-Thompson abundance estimates. In the estimates presented above, groups estimated from the plane (mean = 2.04, CV = 0.09) are 43% smaller ( $p < 0.001$ ) than those seen from the boats (mean = 2.91, CV = 0.04). This figure is different, but comparable to that computed with the GLM analysis (a 33% difference between boat and airplane estimates of group size). The difference between the average group sizes computed from the GML analysis and the H-T estimator likely occurs because different factors are considered in their computations. The GLM analysis is preferred here because it takes into account perpendicular distance at which groups were estimated from the trackline.

During aircraft surveys, the window of time that an observer has to search on an specific area of the ocean is primarily conditioned by the aircraft speed (Caughley 1974). Increasing speeds, negatively affect the probability of detection of available groups (perception bias) as well as the probability that a group becomes available during the passage of the aircraft (availability bias). Although perception bias can be computed from data recorded during line-transect surveys (e.g. Laake and Borchers 2004, Pollock et al. 2006, Southwell and Low 2009, Hammond et al. 2013),

estimation of availability typically requires additional effort, such as the independent estimates of the availability processes produced in this study.

Environmental variables (e.g., water transparency, depth) have been demonstrated to affect availability of marine species (Slooten et al. 2004, Pollock et al. 2006, Thomson et al. 2011). However, in the present experiments, only the size of the group had a significant effect on the availability of franciscana groups. This apparent lack of effects of environmental variables on the availability process of franciscanas was previously reported by Sucunza et al. (2018), who credited it to the relative narrow range of the values recorded of the environmental covariates in Babitonga Bay. In this study, data from Sucunza et al. (2018) were combined with surface-dive data recorded in Ubatuba waters, which are deeper and clear than those in Babitonga Bay. Although the mean surface and dive intervals varied significantly between both areas (e.g. Table 7), the proportion of time at surface was very similar, which explains, at least partly why environmental covariates may have little effect on the availability of franciscana groups seen from the air.

A potential shortcoming of the present analysis is that no information is available on the surface-dive cycles of franciscana in shallow and clear waters. Although such features are not typical of the franciscana habitat the availability of individuals in areas where the bottom can be seen should equal 1 (Pollock et al., 2006). Based on the observations of the franciscana model, it can be assumed that franciscanas are available to be seen when they are within 1m from the surface irrespective of the transparency of the water.

If one assumes that 33% of the bias in estimates of franciscana abundance from aerial surveys comes from underestimation of group sizes the fraction of the correction factor computed above that correspond to visibility bias is 2.96 ( $=4.42 \cdot (1-0.33)$ ), which is equivalent to an estimate of  $g(0) = 0.338$ . Once the availability of franciscana groups estimated in this study is equal to 0.39, the proportion of groups available that were missed by the observers can be estimated at 13% ( $=1-(0.338/0.39)$ ). Similar values of perception bias were reported using mark-recapture distance sampling methods (MRDS, Borchers et al. 1998, 2006) during aerial surveys for franciscana in south and southeast Brazil (perception bias = 13% - 23%, Sucunza et al. 2020). It is interesting to note that some of the observers changed between the present study and the surveys reported in Sucunza et al. (2020), but all had relatively similar experience. Thus, these results suggest a

similar rate of miss-detection of franciscana groups between observers with similar experience. Laake et al. (1997) reported that experienced observers missed 14% of available groups while inexperienced observers missed up to 77% of the available groups during aerial surveys for harbor porpoise, as species with similar characteristic to the franciscana, in coastal waters of Washington State. In the present study perception bias was not assessed because inconsistencies in determining groups that were seen by only front, rear or both observers during the experiments in Babitonga Bay.

#### 4.1. Application of the Correction Factor to Existing and Future Franciscana Abundance Estimates

The use of the correction factor computed here to adjust existing and future estimates of franciscana abundance requires considerations about the field of view and the speed of the aircraft, flight altitude and experience of the observers. If differences between aircrafts result in different field of view such as in previous abundance estimates of franciscanas in southern Brazil and Argentina (e.g. Secchi et al. 2001, Crespo et al. 2010), the correction factor is not applicable. For surveys using the same aircraft and observers with similar experience the use of the correction factor is valid and should be performed.

The new estimates of availability of franciscana groups reported in this study as well as the independent estimate of groups size bias can be used independently of the assumptions described to the correction factor. Experiments to address availability of franciscana groups to aerial platforms are recommended in other regions to compute improved and/or area-specific correction factors. However, the availability of franciscana groups reported here appears to be a robust estimate considering that surveys were carried out in two locations with different environmental characteristics but consistent with those found throughout most of the species range. Therefore, correction factors for availability and group size bias provided here should be applied in range-wide aerial surveys to improve abundance estimates even if surveys are conducted in relatively different survey conditions (e.g., different aircrafts and different observers).

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