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**Have we achieved a sustainable balance? Evaluating the effects of  
regulated guano extraction on an important penguin breeding colony  
(2008–2019)**

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Tesis para optar el Título Profesional de Licenciado en Biología

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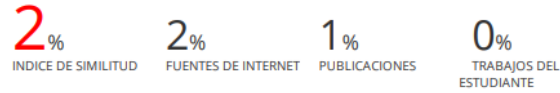
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## RESUMEN

La extracción de guano en el Perú ha causado impactos significativos en las colonias del pingüino de Humboldt (HUPE, *Spheniscus humboldti*). Estos impactos incluyen mayor estrés en individuos, menor número de intentos reproductivos, y mayor abandono de nidos. Estrategias de mitigación para minimizar la interacción entre pingüinos y las campañas se han desarrollado y aplicado desde el 2001 en Punta San Juan (PSJ), una de las áreas reproductivas más grandes de HUPEs. Este estudio busca determinar los efectos de las dos campañas de extracción de guano más recientes en PSJ (2012 y 2019) en la reproducción de HUPEs. Realizamos tres análisis para evaluar si las campañas de extracción de guano alteraron el número de intentos reproductivos (i.e. nidos), o la temporalidad reproductiva (i.e. fenología) de la colonia de estudio. Primero, usando las cuentas semanales de nidos por 11 años (2008-2019), evaluamos si los años de extracción presentaron una diferencia significativa en la fenología y número de nidos en comparación a años sin extracción. Segundo, durante las campañas del 2012 y 2019 evaluamos diferencias en la fenología y el número de nidos entre sectores de la colonia, dada la proximidad de estos a las zonas de extracción. Finalmente, usamos un GLM para estimar la relación entre la distancia de cada sector a las zonas de extracción con el número de nidos durante las campañas de extracción de guano. No evidenciamos impactos de las campañas de extracción de guano en la especie de estudio. Sin embargo, declives poblacionales y variaciones irregulares en la temporalidad reproductiva de los HUPEs en años recientes pueden estar ocultando impactos de la campaña del 2019. Este caso ejemplifica como alcanzar el equilibrio entre intereses económicos y de conservación. Sin embargo, también ilustra la necesidad de identificar los factores causantes del reciente declive poblacional de HUPE en PSJ.

Palabras claves: Perú, Punta San Juan, aplicación de medidas de mitigación, *Spheniscus humboldti*, declive poblacional.

## ABSTRACT

Guano extraction in Peru has caused significant impacts on the endangered Humboldt penguin (HUPE, *Spheniscus humboldti*) colonies. These impacts include HUPE distress, breeding avoidance, and abandonment of nests. Mitigation strategies to minimize the interaction between breeding penguins and guano extraction have been developed and applied since 2001 at Punta San Juan, one of the largest HUPE breeding areas. This study aims to determine the effects of the two most recent guano harvests at Punta San Juan (2012 and 2019) on HUPE breeding. We conducted three analyses to evaluate if such events altered the number of breeding attempts (i.e. nests) or the breeding temporality (i.e. phenology) of the study colony. First, using yearlong nest counts for 11 years (2008–2019), we assessed if extraction years present significant variation in nesting phenology and number of nests compared to non-extraction years. Second, during the 2012 and 2019 guano harvests, we evaluated differences in nesting phenology and the number of nests between sectors of the study colony, given their proximity to extraction activities. Finally, we used a GLM to estimate the relationship between sector distance to extraction areas and the number of active nests on a week-to-week resolution (as nest desertion proxy) during the extraction periods. Results showed no evidence of guano harvest impacts on our study species. However, downward trends in the overall population and irregular variation in HUPE breeding temporality in recent years might be masking possible 2019 harvest effects. While this case exemplifies how a balance between economic and conservation interests can be achieved, it also illustrates the necessity of identifying underlying factors of HUPE population decline and the need for long-term monitoring.

Keywords: Peru Punta San Juan Applied mitigation strategies *Spheniscus humboldti* Population decline



# **Have we achieved a sustainable balance? Evaluating the effects of regulated guano extraction on an important penguin breeding colony (2008-2019)**

## Introduction

Guano extraction was an important practice in Peru during the XIX and XX centuries that caused strong impacts on the local fauna. Off the coast of Peru, thousands of guano birds: Guanay cormorants (*Phalacrocorax bougainvillii*), Peruvian boobies (*Sula variegata*), and Peruvian pelicans (*Pelecanus thagus*), gather every year to breed in islands and peninsulas, producing massive amounts of guano (Hutchinson, 1950; Coker, 1920). In the XIX century, the constant unregulated extraction of this fertilizer in Peru due to its high demand in the agroindustrial international markets caused declines in coastal fauna populations, including the guano birds themselves (Coker, 1920; Murphy, 1936). Consequently, in the early 1900s, the Peruvian government implemented various mitigation measures to prompt the recovery of the guano birds' populations. Guano extraction was to be conducted solely by governmental agencies, permanent guard posts were established in breeding grounds, and exploited areas were rotated after every harvest to minimize ongoing disturbances (Cushman, 2005; Tovar et al., 1987). Furthermore, in the 1940s, concrete walls were built around some peninsulas of the Peruvian coast that had until then been considered secondary breeding grounds for guano birds (Cushman, 2005; Tovar et al., 1987). These isolated headlands acted as artificial islands and helped minimize disturbances caused by humans and other animals (e.g. Andean fox *Lycalopex culpaeus*). Despite these mitigation measures, other non-guano bird fauna was still disturbed during guano harvests (Cushman, 2005; Tovar et al., 1987). Although there was little monitoring of the guano extraction industry's interaction with other species, impacts have been registered for otariids, gulls, condors, red-footed shags, Peruvian diving petrels, and Humboldt penguins (HUPE, *Spheniscus humboldti*; Cushman, 2005, 2013; Zavalaga, 2015).

Historically, guano harvests caused dire impacts on HUPE populations, becoming the main driver of their population decline (Hays, 1984; Murphy, 1936), and are currently still considered to be a threat towards this endangered species (McGill et al., 2021). For the greater part of the XIX and XX centuries, direct impacts during guano harvests, such as the collection of eggs and poaching of adults, were a constant threat to the HUPE population (Hays, 1984; Zavalaga & Paredes, 1997). Since HUPEs burrow in guano during nesting, removing guano deposits in reproductive colonies further affected the species' reproductive success (Hays, 1984; Murphy, 1936; Paredes & Zavalaga, 2001). Additionally, exposure to human disturbances can reduce reproductive outcomes, as reported for various penguin species (Hokey & Hallinan, 1981; Simeone & Luna-Jorquera, 2012; Kitaysky et al., 2003; Ellenberg et al., 2006). Stress in adults can cause individuals to avoid reproduction (Hokey & Hallinan, 1981) or to desert their nests (Ellenberg et al., 2007). For HUPEs, this has been reported to occur during El Niño events, where local prey availability is reduced, and breeding individuals are forced to forage farther away and desert their colonies (Paredes & Zavalaga, 1998, Culik et al., 2000). Furthermore, when adults are disturbed, they can momentarily abandon their nests, exposing their eggs or chicks to predation (Simeone & Luna-Jorquera, 2012). In chicks, exposure to stressful factors can compromise their ability to learn techniques associated with foraging efficiency (e.g. ability to resolve associative learning and spatial tasks), which can consequently affect their survival rate (Kitaysky et al., 2003). HUPEs have been denoted as the most skittish of penguins; these are disturbed by movements > 100 meters away and avoid breeding in areas frequently transited by humans (Ellenberg et al., 2006). Therefore, during guano harvests, workers, trucks, and other stimuli at a close range are considered to have caused impacts on breeding HUPEs (Villacorta, 2003; Zavalaga, 2015).

Punta San Juan holds one of Peru's largest and most researched HUPE breeding areas. At this site, specific mitigation strategies have been implemented since 2001 to reduce the impacts of guano harvests on this species' population (Cárdenas-Alayza & Cardeña-Mormontoy, 2012). In 1987, negative effects of an unregulated guano harvest at PSJ included the poaching of ~100 penguins and the abandonment of ~700 eggs by breeding

adults (Riveros Salcedo, 1999). Given the disastrous consequences of this event, it was recommended that extractions be suspended in this peninsula until mitigation measures could be established, paralyzing extraction at PSJ for 14 years. In 1998, the Peruvian guano management agency (PROABONOS) signed an agreement that established new measures to mitigate guano harvest disturbances in PSJ based on the evidence collected by the long-standing in-situ research Punta San Juan Program (now within the Center of Environmental Sustainability, UPCH). Since the signing of the agreement, four guano harvests have taken place in PSJ in 2001, 2007, 2012, and 2019. The 2001 and 2007 harvests occurred during the HUPE annual molting period (January-March; Cárdenas-Alayza, 2012); therefore, the extraction events did not directly impact this species during its reproduction. However, the 2001 and 2007 guano harvests took place while the main guano bird species was breeding (Guanay cormorant *Phalacrocorax bougainvillii*, October-March) and was consequently disturbed by extraction workers (*personal communication A. Bussalleu*). Therefore, authorities in charge of guano management requested changing the future timing of extraction periods to avoid overlap with the cormorant breeding season (Cárdenas-Alayza, 2012). As a result, the 2012 and 2019 harvests were conducted within the annual HUPE reproductive period. During all four harvests, diverse mitigation measures were implemented, such as temporal regulation of the extraction periods and spatial planning of extraction areas (i.e., distances to potential HUPE breeding areas).

Since the implementation of mitigation measures at PSJ, impacts similar to those of the 1987 guano harvests have not been recorded again (Cárdenas-Alayza et al., 2019). However, despite the applied mitigation measures during the 2012 and 2019 harvests, breeding HUPEs were still disturbed when exposed to extraction stimuli (i.e., encounters with humans entering/leaving extraction areas or transiting guano-hauling trucks; Cárdenas-Alayza, 2012, Cárdenas-Alayza et al., 2019). Therefore, exploring the possible impacts of guano harvests in these years is crucial. This study aims to determine the effect of the 2012 and 2019 guano harvests on breeding HUPEs at the largest reproductive colony of PSJ. Given that HUPE sensitivity to human disturbances is relatively unknown, we evaluated possible impacts at two spatial scales: (i) colony scale,

by assessing the overall impacts of guano harvests on the breeding colony, and (ii) sector scale, by assessing the impacts associated with the extraction area proximity to different colony sub-sections throughout the harvests. Given that the guano extraction advanced parallel to the study colony, each colony sub-section had varying distances to the extraction area. For example, the colony sub-section closest to the extraction area at the beginning of the harvests was the farthest away when the harvests ended. The harvests' parallel progression allows for an ideal natural experiment that compares breeding patterns between colony sub-sections to determine the effects of extraction area proximity on HUPE. We hypothesize that during guano extraction periods, impacts can decrease overall nest abundance and delay breeding onset (as reported during other stressful conditions like El Niño events; Paredes & Zavalaga, 1998, Simeone et al., 2002).

## Methods

### 2.1 Study species

The HUPE is an endemic species of the Great Marine Humboldt Current, distributed from Isla Foca (5°12'S) in northern Peru to Isla Guafo (43°12'S) in southern Chile (BirdLife International, 2018). In Peru, the annual reproductive period for this species extends from March to December and can be separated into two breeding seasons: March-July and August-December (Zavalaga & Paredes, 1997). Reproductive events last approximately 4 months, in which adults occupy nests (1-2 months), incubate (2 eggs per clutch; 6 weeks), and brood their chicks (10-12 weeks) (Zavalaga & Paredes, 1997). Overall, the HUPE population is declining (McGill et al., 2021). The species is classified as "vulnerable" in IUCN's red list of endangered species (BirdLife International, 2018) and is also included in Appendix I of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) (UNEP-WCMC, 2014). In Peru, this species is considered "endangered" by law (Supreme Decree N° 004-2014-MINAGRI).

### 2.2 Study site

This study was conducted at Punta San Juan (hereafter PSJ), San Juan de Marcona, Ica (15.36°S, 75.16°W; Figure 1), a 54-hectare peninsula that forms part of the Guano Islands, Isles, and Capes National Reserve System (RNSIIPG) (Supreme Decree N° 024-2009-MINAM). PSJ harbors one of the largest monitored HUPE breeding colonies in Peru and is also considered one of the most important breeding hot spots of guano birds in the country (Zavalaga, 2015). PSJ is divided into 20 beaches, 10 on the Northside (N0-N9) and 10 on the Southside (S0-S9), all of which lead to steep 8-30 meter cliffs (Figure 1B). At this location, the majority of HUPE nest along the cliff edges and slopes in the guano layer. A 1.2 km concrete wall built in the 1950s surrounds the reserve (Zavalaga & Paredes, 1997). Since the late 1970s, the Punta San Juan Program (hereafter PSJP) of the Center for Environmental Sustainability of the U. Peruana Cayetano Heredia runs a long-term monitoring and research program at this site (Cárdenas-Alayza & Cardeña-Mormontoy, 2012).

The study colony (situated at S7-S8) supports the largest HUPE nest aggregation in PSJ, hereafter referred to as *colony*. To facilitate monitoring, the colony is divided into 12 nesting zones, hereafter referred to as *sectors*. This study only considered sectors situated at cliff tops ("*top*", N = 6; Figure 1C), given that they present the highest number of nests and that they are visually aligned with the guano extraction areas (potential direct contact with extraction workers). Additionally, selecting "*top*" sectors allows for standardization of nesting substrate between sectors, as cliff tops' primary substrate is guano, in contrast to the mixed substrate of stone, gravel, guano, and sand found at cliff slopes and beaches (Paredes & Zavalaga, 2001). To facilitate interpretation, sectors were labeled with letters ranging from A to F. Labels were assigned given their spatial position, being sector A closest to the peninsula headland and sector F the farthest away (Figure 1C; see Table A.1 for original sector name list).

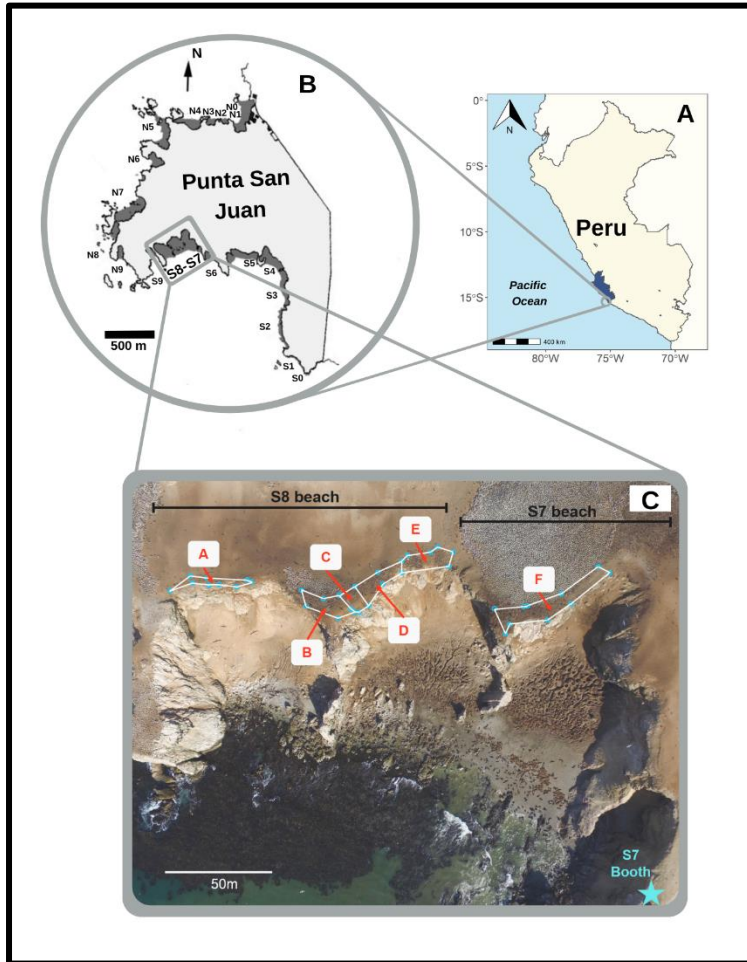


Figure 1. Map of (A) Peru, with the Ica department shaded blue; (B) the Punta San Juan natural reserve and its 20 beaches (darker shades), extracted from Paredes & Zavalaga, 2001; and (C), a photograph of the study site (S7-S8) taken with a drone (DJI Phantom 4) in November 2020 by D. Torres of the PSJP, showing the S7 and S8 beach extensions (black lines), reference points of penguin nesting area (blue circles), sectors of the study colony A-F (labeled in red), and the S7 monitoring booth as a colored star.

### 2.3 Monitoring of active nests

The PSJP personnel has continuously monitored the colony since 2001. During our study period (2008-2019), weekly active nest counts of each colony sector were conducted from a monitoring point at a straight-line distance range of 113-179m (S7 booth; Figure 1C). During censuses, one investigator meticulously scanned each sector with the help

of tally-counters, binoculars, and/or telescopes. A scanning direction is determined during these censuses to avoid double counting. The investigator classified active nests according to their contents as roosting nests (RoostNest, as a proxy for incubation) or nests with chicks (ChickNest, as a proxy for brooding). If a nest presented both a roosting adult and a chick, it was considered to be brooding and therefore classified as a ChickNest. Given this, the nest categories RoostNest and ChickNest are mutually exclusive.

#### 2.4 2012 and 2019 guano harvests at PSJ

Extraction in both harvests advanced gradually from the Southwest to the Northeast of the peninsula, extracting a total of 8,522 tons of guano in 2012 and an estimate of 12,989 tons in 2019 (AgroRural, 2019; Figure 2). The number of guano extraction personnel varied temporally, with a daily maximum of 140 and 200 workers in 2012 and 2019, respectively. By 2012, PSJ had become part of the RNSIIPG protected area, and the application of mitigation measures at this site was coordinated between PSJP, AgroRural (current public entity in charge of guano management), and the Peruvian Agency for Natural Protected Areas (SERNANP). Additionally, groups of PSJP volunteers were present to help enforce and monitor the established measures. To minimize disturbance, activities were initiated at 5:30 am, having workers move to the extraction areas before dawn. Activities ended at 12-3 pm to avoid all interaction with fauna for 12-17 hours before the next working day began (Cárdenas-Alayza et al., 2019). During working hours, PSJP volunteers regulated the crossing of workers and guano-hauling trucks to and from extraction areas (Cárdenas-Alayza, 2012; Cárdenas-Alayza et al., 2019). Furthermore, AgroRural and SERNANP gave talks to sensitize workers on the possible impacts on local fauna. Minimum extraction distances to the HUPE breeding grounds were also established (15m in 2012, 50m in 2019); therefore, the guano used by HUPE as nesting substrate was not collected. A newly implemented mitigation strategy was the use of movable cloth barriers between extraction areas and the penguin colony to diminish direct visual contact (as suggested in Ellenberg et al., 2006).

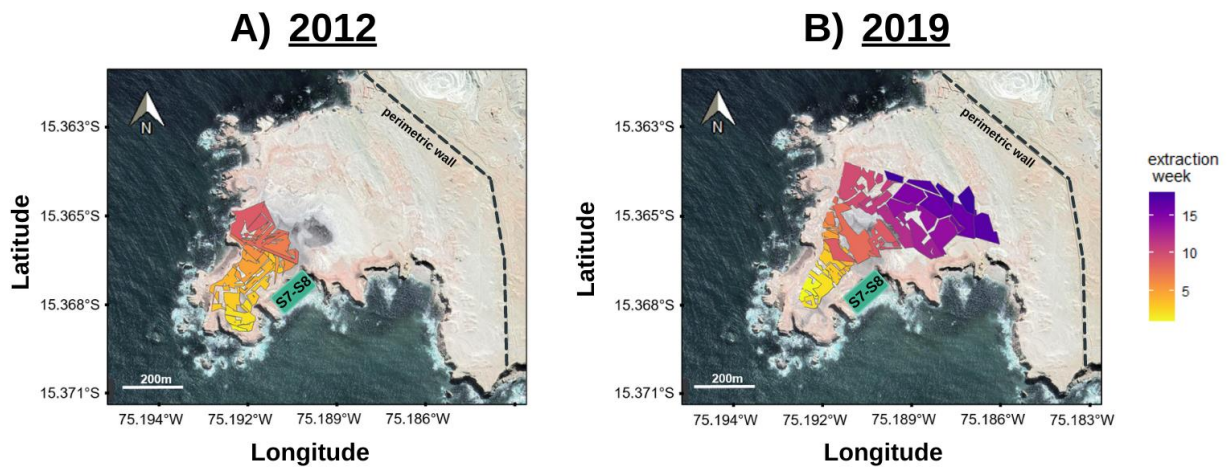


Figure 2. Image of PSJ and the A) 2012 and B) 2019 guano extraction areas. Weekly extraction areas are represented by colored polygons, ranging from 0-9 in 2012 (09 of August – 01 of October) and from 0 – 17 in 2019 (31 May – 30 September). A teal rectangle represents the study colony. Satellite image source: Google Earth, image CNES / Airbus.

## 2.5 Georeferencing and distance estimations

PSJP personnel took photographs of the colony sectors both using a DJI Phantom 4 drone (GPS altitude of 21-22m, precision  $\pm 0.5\text{m}$ ) and a digital camera (60X) from the S7 booth (distance specified in 2.3). From these photographs, we identified predetermined landmarks outlining colony sectors: stones, nests, posts, or PVC tubes. The PSJP personnel have used these landmarks to monitor the colony since early 2000 (see Paredes & Zavalaga, 2001). Then, to georeference sectors, the drone was flown between February-April 2021 and positioned over each landmark as close to the ground as possible (GPS altitude of 0.16-14.6m). Once in position, photographs were collected using the device's camera, in which GPS coordinates of the drone's position were automatically recorded (precision  $\pm 1.5\text{m}$ ). From these, we then estimated polygons to represent colony sectors (Figure 1C).



During the 2012 and 2019 guano harvests, PSJP volunteers monitored the spatio-temporal dynamics of the extraction areas. Using a GARMIN eTrex Summit HC manual GPS logger (95% chance of precision < 10m), volunteers collected ground-based spatial coordinates delimiting the guano-extracted areas every afternoon after daily extraction concluded. Using these GPS points, daily extraction polygons were determined and then aggregated into weekly polygons (Figure 2) to scale to weekly nest census information.

Using the estimated polygons, we calculated minimum distances from the extraction area perimeter to the center of each colony sector for each week during the 2012 and 2019 harvests. Distances were estimated from the extraction area perimeters as to consider the closest these were to colony sectors (i.e. potential highest interaction). On the other hand, distances were estimated to the center of the colony sectors to better represent harvest effects on all active nests in a sector. All spatial data management and distance estimations were done using the *sf* library in R (Pebesma, 2018).

## 2.6 Analysis

Unless specified otherwise, all analyses were done using base functions in R (R Core Team, 2021). We used counts from the yearlong nest census to derive annual phenological parameters for the colony from 2008-2019. We considered phenology as the annual trend of active nest counts. Trends were evaluated independently for RoostNest (incubation) and ChickNest (brooding). HUPE annual phenology was separated into two distinct breeding seasons (BS-1 and BS-2; Paredes et al., 2003, Simeone et al., 2002), based on “cut dates”, i.e. average date between pre-defined first and second annual reproductive peaks with the lowest nest count (reproductive peaks were defined from Paredes et al. 2002 for incubation and from preliminary data exploration for brooding). Based on these estimated breeding seasons and the previously defined active nest classes (RoostNest and ChickNest), the annual breeding period was divided into 4 breeding phases: incubation-1, brooding-1 (for BS-1), and incubation-2, brooding-2 (for BS-2). For each independent breeding phase in our study

period, we then estimated three “phenological parameters” from nest count values to represent their overall temporality: start date (5th percentile date), peak date (when nest count was the highest), and end date (95th percentile date; Figure 3A). Using various phenological parameters allowed for a better representation of possible variations in the temporality of the breeding phases (see Miles et al., 2017). Furthermore, we estimated the maximum nest counts per breeding phase as a proxy for the magnitude of the reproductive output (i.e. size).

A first analysis focused on evaluating colony size and phenology variation between 2008-2019 at the colony scale. From this, we determined differences in the pattern followed by guano extraction years compared to non-extraction years. We first explored overall changes in colony maximum nest counts per breeding phase throughout our study period. Then, we evaluated the inter-annual variation of the colony’s phenology. This was done independently per breeding phase (N=4) and phenological parameter (N=3; overall 12 groups). For each group, we calculated the mean value for all non-extraction years in our study period and their standard deviations. From these, we estimated cumulative probability distributions (Figure 3B). The cumulative probability distributions reflect the inter-annual variation of each breeding phase's start, peak, and end dates. We then identified phenological parameters during the 2019 and 2012 guano extraction periods that fell out of the non-extraction year’s 90% cumulative probability distributions (i.e. “outliers”; two 5% tails, Z-test).

A second analysis focused on comparing size and phenology between sectors of the colony. We then compared guano extraction years at sector scale to non-extraction years. First, to strictly evaluate differences between sectors, we standardized all parameters (sector size and phenological parameters) to each independent breeding phase’s colony value. This allowed us to discard inter-annual variation from this analysis. Sector size values were standardized as a percentage of the colony maximum count for each breeding phase, whilst sector phenological parameters were standardized as the difference in days with their overall colony value. Sector size variation was evaluated per breeding phase (N=4) and sector (N=6; a total of 24 groups), and phenological parameter

variation was evaluated independently per breeding phase (N=4) and parameter (N=4; a total of 16 groups). We estimated their inter-annual mean values and standard deviations for each of these groups. From these, we estimate cumulative probability distributions for each group (Figure 3C). Cumulative probability distributions reflect the inter-annual deviation of sector values from their overall colony values. We then identified sector size or phenological parameter values during the 2019 and 2012 guano extraction periods that were considered outliers of their estimated cumulative probability distributions (i.e. differences between sectors in 2019 and 2012 were similar to those in non-extraction years). Finally, we evaluate if sector size or phenology outliers were associated with the extraction areas' proximity to said sector (i.e. distance).

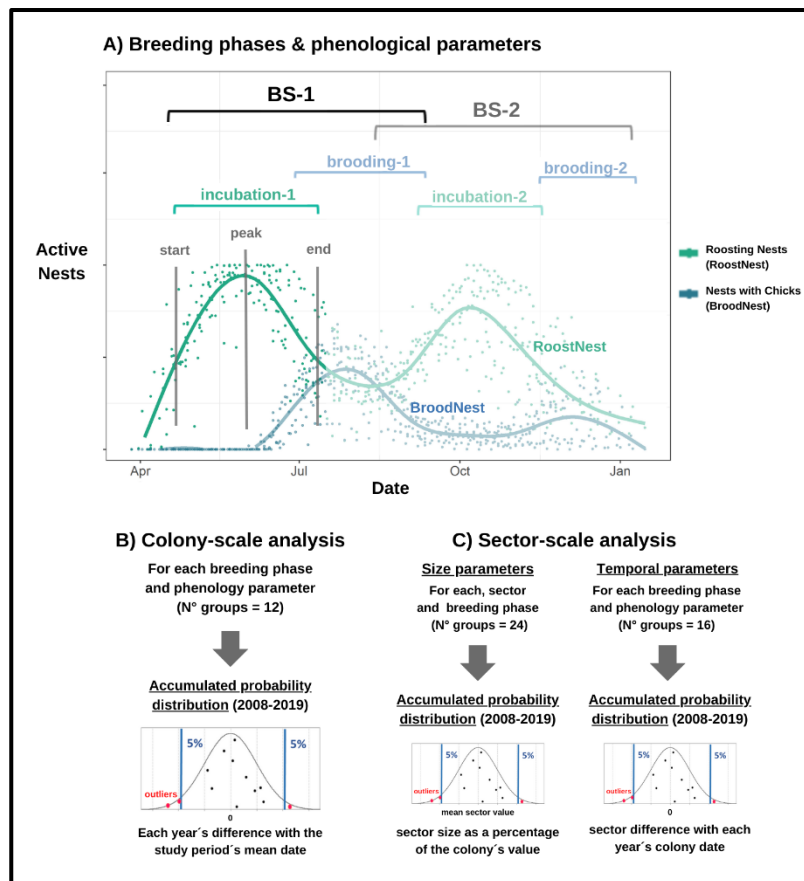


Figure 3. Visual representation of part of the three analyses conducted in this study. (A) Separation of annual phenology in two breeding phases (BS-1 and BS-2) and three phenological parameters (start, peak, and end dates), (B) Colony-scale analysis, and (C) sector-scale analysis.

In a third analysis, we evaluated if extraction area distance to sectors was associated with weekly nest counts (as a nest desertion proxy) using a Poisson distributed Generalized Linear Model (lmer4 library in R; Bates et al., 2015). Models were constructed independently for each harvest year (2019 and 2012) and each breeding stage, with ActiveNest (total nests), RoostNest (proxy of incubation), and ChickNest (proxy of brooding) as response variables. We incorporated an offset for sector maximum nest count to include sector size differences. Random intercepts per sector were also used to account for other sector-specific factors. Additionally, we included “day of the year” as a polynomial independent variable to represent annual trends in the number of nests (i.e. phenology).

## Results

### 3.1 HUPE phenology breeding phases

Annual breeding phases presented low variation throughout our study period (Table 1). Mean start and end dates  $\pm$ SD in days in our study period indicate that the first breeding season (BS-1) ranged approximately from April 4<sup>th</sup>  $\pm$ 7 – August 21<sup>st</sup>  $\pm$ 7, and the second breeding season (BS-2) from July 23<sup>rd</sup>  $\pm$ 6 – Dec 1<sup>st</sup>  $\pm$ 21. Incubation in BS-1 (*incubation-1*) lasted on average 86  $\pm$ 11 days, while incubation in BS-2 (*incubation-2*) lasted an average of 114  $\pm$ 18 days. Brooding in both BS (*brooding-1* and *brooding-2*) lasted on average 76  $\pm$ 6 days and 76  $\pm$ 15 days, respectively.

breeding season	phenology phase	start date	peak date	end date
BS-1	incubation-1	04 Apr (SD=7)	12 May (SD=11)	28 Jun (SD=6)
BS-1	brooding-1	06 Jun (SD=7)	07 Jul (SD=14)	21 Aug (SD=7)
BS-2	incubation-2	23 Jul (SD=6)	13 Sep (SD=22)	14 Nov (SD=16)
BS-2	brooding-2	16 Sep (SD=10)	31 Oct (SD=27)	01 Dec (SD=12)

Table 1: Table of mean start, peak and end dates for each breeding phase for our HUPE study colony from 2008-2019.

### 3.2 Inter-annual variation of colony size and nesting phenology

Although phenological parameters presented low inter-annual variation, a clear decrease in HUPE colony size is evidenced from 2008-2019 (Figure 4). Overall trends of the maximum number of nests (Figure 4A) can be separated into three periods: 2008-2012 (average maximum counts  $1000 \pm 81$  RoostNest), from 2012 to 2015 BS-1 (average maximum counts  $1302 \pm 115$  RoostNest), and from 2015 BS-2 to 2019 (average maximum counts  $590 \pm 243$  RoostNest). In this last period (2015-2019), pronounced decreases are evidenced after the 2015-16 and 2017 El Niño events (Figure 4A). Maximum number of adults, as a proxy to breeding population size in the colony, followed similar trends to that of maximum nest counts. Productivity, as the ratio of the maximum ChickNest count and the maximum RoostNest count in a breeding season, is similar between years, being lowest from 2015-2017 for BS-1 and in 2010, 2015, and 2017 for BS-2 (Figure 4B, row 5). Colony phenological parameters present low inter-annual variability (Figure 4B, rows 1-4). Considering non-extraction years, phenological parameters of the BS-1 presented less variation than those of BS-2 (overall SD being 10 and 12, respectively). For all breeding phases, start dates presented less variation than end dates (SD=5 and SD=7, respectively) and peak dates (SD=14). A higher variation of phenology parameters seems to occur in BS-2 after 2015/17; for example, *incubation-2* end dates are overall delayed after 2017.

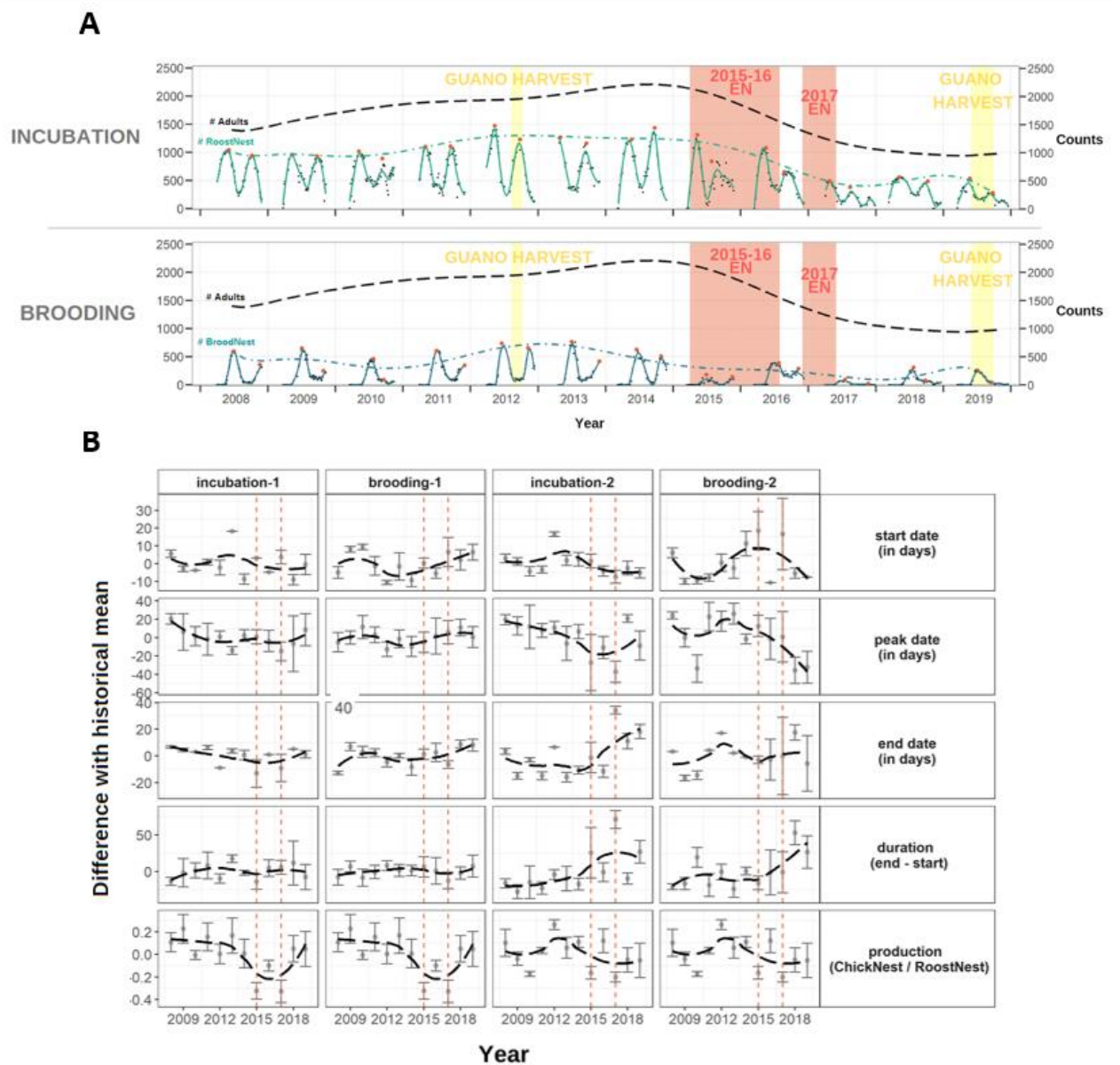


Figure 4. Reproductive trend of the Humboldt penguin (HUPE, *Spheniscus humboldti*) study colony from 2008 – 2019. (A) Historical trend of adults and active nests. Nest counts are separated in panels: RoostNest (incubation; top panel) and ChickNest (brooding; bottom panel). Black dots illustrate counts for each census date. Colored continuous lines delineate annual trends (phenology), with maximum values per breeding season as red circles; and colored dot-dashed lines represent historical trends for each panel. In both panels, trends of the total number of adults in the colony are displayed as a black dashed line. The 2015-16/17 El Niño events are illustrated as red shades. Yellow shades represent the guano harvests at PSJ. (B) Changes in HUPE

phenology temporal parameters. Panels separate breeding phases and phenological parameters (start, peak, and end dates). In each panel, annual phenology values are shown from 2008-2019 as gray points. These values are centered and are shown as the difference between that year's phenological parameter value and the study period average (negative values indicating earlier-than-average values, and positive values later-than-average values). Error bars illustrate the standard deviation of colony sector values for each year. Black dashed lines outline trends in temporal parameter values from 2008-2019. Vertical red dashed lines indicate 2015 and 2017 as referential points for the ENSO events' onsets

The maximum number of nests during the 2012 and 2019 extraction periods at PSJ behave similarly to that of their neighboring years. In 2012, nest counts during the extraction period (BS-2) were one of the highest in our study period (Figure 4A), as in the previous (2011) and consecutive years (2013, this takes into account possible carry-over effects). On the other hand, during the 2019 extraction period (overlap of BS-1 and BS-2), nest counts were low compared to the first years of our study period, but similar values have been evidenced since 2017. During the guano extraction periods, only 2 colony phenological parameters were outliers (fell outside their inter-annual 90% cumulative probability distribution), both occurring in 2019 (Figure A.1). The average number of outliers per year throughout our study period is 2; therefore, 2019 behaves accordingly.

### 3.4 Variation of sector nest counts and phenology during guano extraction periods

During our study period (2008-2019), sector F presented the most nests in the colony, making up, on average, 35% (SD=7) of the maximum colony nest counts. All other sectors made up between 12-18% (max sector SD=7), with sectors B and E usually having the highest values (in 81% of the cases) and sector A usually having the lowest values (44% of the cases). There were no differences in sector size between breeding seasons or reproductive stages (i.e. incubation/brooding), with a maximum SD of 6 between these

groups. The number of sector size values per year that were found on the lower 5% probability distribution tail (smaller than their expected values) varied throughout our study period for non-extraction years, with an average of  $1 \pm 1$  from 2008-2011, 2013-2014 and an average of  $3 \pm 1$  from 2015-2018. Similarly, during guano extraction periods, we found 0 sector size values on their lower 5% distribution tail in 2012 and 4 values in 2019 (Figure A.2).

Similar to phenological parameters at the colony scale, sector start dates were of less variation ( $SD=4$ ) than end dates and peak dates ( $SD=6$  and  $SD=13$  respectively). There were no marked differences in variation between breeding phases, with the lowest being for *brooding-1* ( $SD=6$ ) and the highest for *brooding-2* ( $SD=11$ ). During non-extraction years, the number of phenological parameters that were outliers was, on average,  $4 \pm 3$  from 2008-2014 and, on average,  $9 \pm 3$  from 2015-2018. We found similar patterns during guano extraction periods (Figure A.3). During the 2012 harvest, 0 phenological parameters were outliers, while in the 2019 harvest, there were 10 parameters as outliers.

We did not find any association between the sector parameter outliers (i.e. outside the 90% distribution) during guano extraction periods and the extraction area proximity to said sectors (Figure 5; see Figure A.4). In 2019, we found overall 5 size values and 10 phenological parameters that were outliers. Of these, all size outliers occurred in BS-2 (after September) when the extraction area was over 160 meters away from all colony sectors. Of the 10 phenological parameters that were outliers, 7 occurred in 2019 BS-2 (>160m to all sectors). From the remaining phenological parameter outliers, 1 occurred when the extraction area was over 140 meters away (sector F *brooding-1* peak date). We did find proximity association to outliers for sector C *brooding-1* start date (66m), and for sector A *brooding-1* start date (44m, closest sector to extraction area in that week); however, both phenological parameters only presented a one-week mismatch with the overall colony date. Both these values occurred during the breeding phase with the least variation (*brooding-1*) and for the less deviant phenological parameter (start date); therefore, a one-week difference was sufficient for these values to fall out of their



90% probability distributions. We do not consider this to be evidence of extraction area proximity association with phenological irregularities.

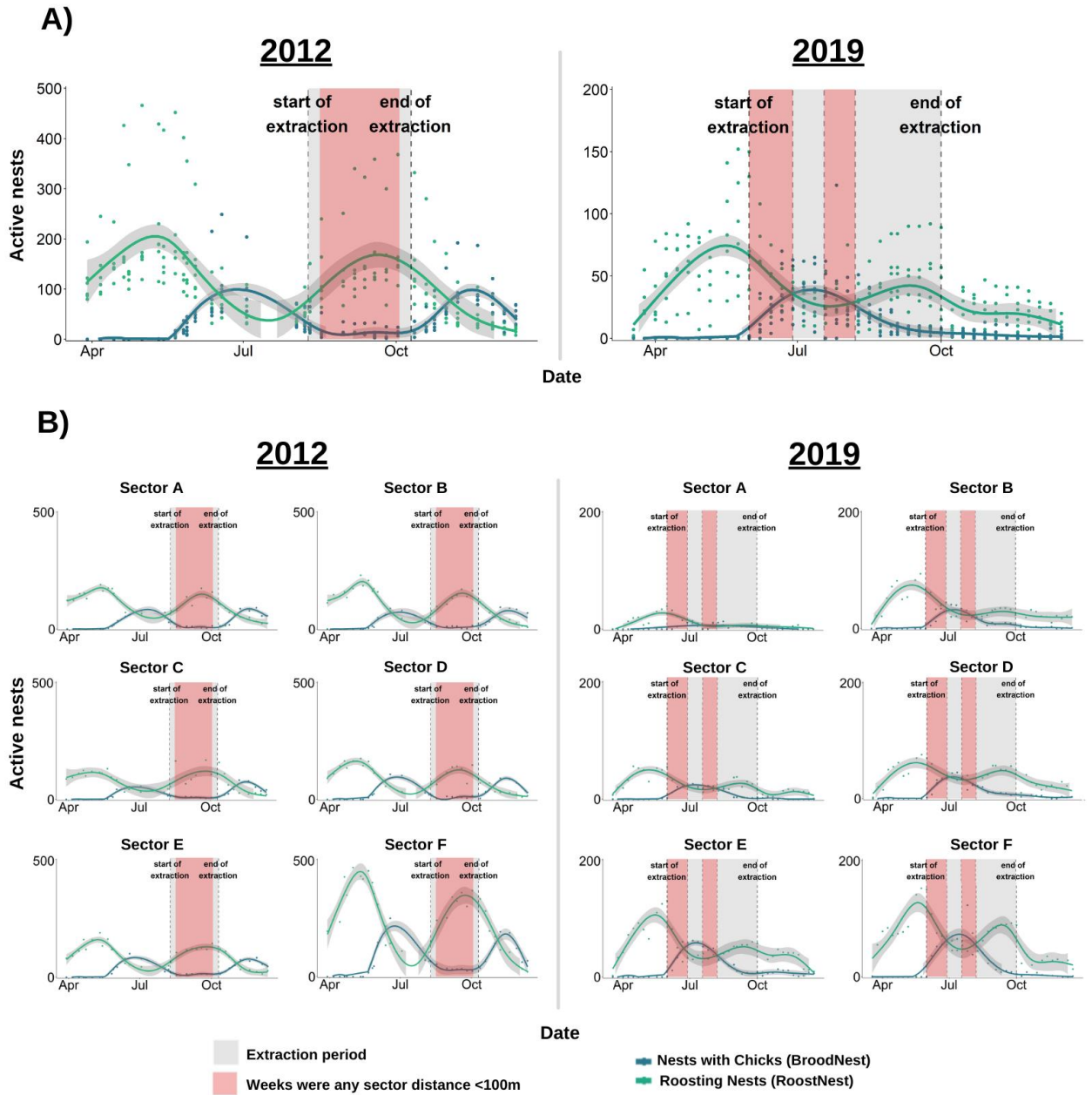


Figure 5. Phenology of (A) the colony and (B) each individual sector during 2012 and 2019, coupled with the guano extraction periods for each year. Each panel presents yearlong active nest counts as dots and continuous lines representing the annual breeding phenology trend. Colors distinguish nest-breeding stages: incubation (RoostNest) and brooding (ChickNest). Gray quadrants outline the extraction periods:

June – November 2019 and August – October 2012. Red quadrants highlight weeks when the distance from the extraction area to any sector was less than 100 meters.

### 3.5 GLM model

All estimated models (per harvest year [2012, 2019], and breeding stage [ActiveNest, RoostNest, ChickNest]) presented low residual deviance, ranging from 76-264 on 45 degrees of freedom for 2012 (N=54 observations) and from 203-261 on 99 degrees of freedom for 2019 (N=108 observations). There was no significant association between distance to extraction areas and active nests for any harvest year or breeding stage (p-values 0.42-0.76).

### Discussion

Overall, no impacts of the 2012 and 2019 guano harvest were found on the PSJ HUPE breeding population (i.e. active nests) or their breeding patterns (i.e. phenology). During the 2012 extraction, nest counts were some of the highest registered values, and phenology was similar to the study period average. On the other hand, during the 2019 guano harvest, we found fewer nests and higher phenology variation overall than in the first years of our study period (2008-2014). Although 2019 breeding size and nesting phenology presented irregularities, similar variations have been evidenced since 2015. Therefore, our results indicate that irregularities seem to be a consequence of the overall trends in the study population rather than to direct guano harvest impacts.

#### 4.1 Recent trends of the PSJ HUPE breeding colony

Colony trends in recent years indicate a strong effect of the 2015-16 and 2017 El Niño events as well as potential effects of ongoing marine heatwaves on the PSJ HUPE

breeding population (Pietri et al., 2021). During El Niño events, HUPEs avoid food scarcity by increasing their foraging efforts and changing prey species (Chiu Werner et al., 2019; Culik et al., 2000; Luna Donoso, 2016). Although this adaptive behavior helps avoid adult mortality, nest desertion increases during these periods (Hays, 1986; Paredes & Zavalaga, 1998; Simeone et al., 2002). Similarly, our results show a steep reduction in the PSJ HUPE breeding population during the 2015-16/17 El Niño events. Similarly, the 1998 El Niño was associated with drastic changes in the HUPE population and abundance distribution (Paredes et al., 2003; Vianna et al., 2014). After said event, various colonies in Chile saw an increase in their population (Vianna et al., 2014), while in Peru, overall numbers were reduced to 5000 individuals, mostly aggregated in only five colonies (Paredes et al., 2003; the authors note that the 1997-98 El Niño event caused a marked decline in the HUPE population, but the precise magnitude of said reduction is unknown). Moreover, our results show that breeding performance remained low in the years following the 2015-16/17 El Niño events, suggesting a recent increase in additional temperature anomalies (i.e. local, short-duration marine heat waves; Pietri et al., 2021) also affected long-term changes in the PSJ population. In this study, the reduction of the breeding population at the study colony after the El Niño events is notable, with overall reductions after the 2015-16 event of 26.17% for adults, 48.28% for RoostNests, and 64.44% for ChickNests. The magnitude and timing of this reduction should be interpreted with caution since population estimates are recommended to be complemented using counts during the molting period, which include breeding and non-breeding adults, as well as juveniles, to analyze the trends of the population as a whole (Paredes et al. 2003). Our results differ from those of Tieber-Runnels (2016), in which HUPE counts at PSJ during the 2016 molting period were similar to those of previous years. These discrepancies can be an important insight into the difference between the number of penguins that molt (i.e. juveniles and adults) and those that reproduce (i.e. breeding adults) in a colony after an El Niño event. Our results do agree with the more recent census of molting individuals that indicates a reduction of the PSJ population after 2017 (Reyes Robles et al., 2019).

Additional to a progressive reduction of breeding adults and active nest counts following the El Niño events, we have evidence that in latter years the annual BS-2 presents both

fewer breeding attempts (i.e. number of nests) as well as decreased breeding performance (i.e. number of brooding nests/number of incubating nests) compared to that of BS-1 (Figure A.5). The capability of HUPEs to present two breeding attempts in a year is thought to be conditioned to high prey availability (Paredes et al. 2002; Simeone et al., 2002). This behavior allows individuals to maximize breeding in a highly productive ecosystem, which is also periodically disturbed by El Niño events. In this study, BS-1 and BS-2 are considered independent events; however, the outcome of a first reproductive attempt can affect the following breeding attempt's temporality (Paredes & Zavalaga, 2002). Similarly, Rebstock & Boersma (2018) found that the onset of breeding phenology in Magellanic penguins, a closely related species in the *Spheniscus* genus (see Vianna et al., 2020), was affected by carry-over effects from non-breeding periods (Rebstock & Boersma, 2018). Therefore, low nest counts during BS-2 in recent years can reflect yearlong pressures on the colony, possibly the same pressures that caused the overall population decline.

#### 4.2 Colony differences between guano harvest years

Given recent declines in the HUPE breeding population at PSJ, it is vital to evaluate the 2012 and 2019 harvests independently and to explore what the population differences between both periods might imply for penguin-guano harvest interactions. In 2019, the minimum distance from guano extraction areas to the breeding colony was over three times that of 2012. Therefore, direct impacts of extraction proximity to the colony (if any) should be more evident in 2012. However, this argument assumes that HUPE sensitivity to guano extraction stimuli (or other disturbances) is the same between years. Our results indicate that during the 2012 harvest, the colony was in its peak condition (2011-2014); contrary to that, the 2019 harvest occurred during the study colony's worst state (2017-2019). These differences can be attributed to various possible underlying pressures. Prey availability and prey quality are key variables influencing penguin breeding (Boersma, 2008). Its recent reduction near PSJ has been hypothesized to affect other top predator populations at this site (Cárdenas-Alayza et al., 2021; Cárdenas-

Alayza et al., 2022). Furthermore, an additional pressure on the study population since the late 2000s is the increasing presence of rats that predate on eggs and chicks (*personal communication M. Cardeña-Mormontoy*). Various efforts are continuously being made to control the rat population at PSJ (see Cárdenas-Alayza et al. 2019); nevertheless, the extent to which this invasive species impacts the study colony is unknown. These underlying pressures can not only have caused the population reduction evidenced in recent years but can also have affected HUPE tolerance to additional stressful factors. Palacios and collaborators (2018) found altered physiological responses to tourism in a declining penguin colony compared to a neighboring growing colony. These authors hypothesize that underlying pressures causing colony trends could be affecting penguin sensitivity to tourism (Palacios et al., 2018). Similarly, we hypothesize recent breeding performance decline at the study colony occurred due to the overall impact of various selective pressures, making additional small-scale disturbances all the more perilous to this population. This emphasizes the need to tailor conservation measures to specific species and populations, given their sensitivity to disturbances. Furthermore, efforts should be directed towards constantly monitoring the state of protected populations and adjusting applied conservation measures accordingly. Additionally, these results highlight the importance of the strict mitigation strategies for guano harvests at PSJ that have been implemented since 2001. Without these measures, the study population would likely have been reduced to a much higher degree.

#### 4.3 Limitations

This study presented certain limitations. For some years, late-onset of nest monitoring affected estimated *incubation-1* start dates (e.g., 2013 onset was 18 days later than the mean BS-1 start date), and early-conclusion of monitoring affected estimated *brooding-2* end dates (e.g., 2009 monitoring concluded 21 days earlier than average BS-2 end date). Furthermore, human error can influence nest counts; this error increases under certain climatic conditions (e.g., foggy days). An additional limitation was the visual perspective from the monitoring booth. Using drone images, nests have been identified

in areas outside the monitoring booth's visual range (*personal communication D. Torres*). These nests were not included in this study. Nevertheless, the limitations of the nest dataset are minimum. The effort of conducting year-long monitoring since the early 2000's allowed us to have historical high-resolution data of reproductive patterns in one of the most important HUPE breeding sites.

The use of colony sectors as the spatial unit presented certain disadvantages. Sectors delimit regions of the colony where HUPES nest, but within certain sectors (specifically in S7), we evidenced areas of higher/lower nest aggregation. As a result, the estimated distances from extraction areas to colony sectors fail to capture high-resolution nest-guano harvest interactions. Furthermore, there is a higher error when recollecting coordinates that outline sectors when using our methodology compared to using manual GPS loggers, given that researchers must position the drone precisely over the reference points. Even so, this methodology avoided the need to directly enter the colony to manually collect spatial coordinates, minimizing the disturbance of local fauna and avoiding the risks researchers present when accessing cliff tops of slippery substrates (i.e. guano).

#### 4.4 Unexplored effects of guano harvests on HUPES

Even though no direct impact was evidenced in our study, possible guano harvest effects on HUPE breeding should not be discarded. In this study, we assumed impacts would be evidenced when extraction areas were closest to the colony (i.e. acute stressor causing a short-term response); however, this might not be the case. Prolonged (i.e., chronic) stress throughout the breeding attempt (Groscolas et al., 2008), or lagged responses to disturbances (Crisuolo et al., 2005), could have caused nest desertion at different moments. At times, the presence of workers and guano-hauling trucks disturbed adults in their transit routes from the colony to the sea or vice-versa. Although the frequency in which these routes were used did not change (Cárdenas-Alayza et al., 2019), disturbance of parents in their foraging trips can affect chick fledging weight and

consequently their first-year survival rate, as reported for Yellow-eyed penguins (Ellenberg et al., 2007; McClung et al., 2004). Guano harvest activities might have caused changes in the study population's behavior patterns with further unknown consequences; for example, various mammal species have been reported to increase their nocturnal activity in response to human disturbances (Gaynor et al., 2018). The use of movable cloth barriers helped mitigate visual interactions, but other stimuli, like harvest noise, might have affected the breeding population. This is seconded by the fact that harvest noise was the main cause of registered HUPE stress responses in the 2019 harvest (Cárdenas-Alayza et al., 2019). Lastly, other indirect interactions are considered important threats to the PSJ colonies, like the increased number of rats due to their attraction to the organic waste produced during harvests. This highlights the importance of further exploring interactions between guano harvests and HUPEs and implementing new measures to achieve sustainable guano extraction and reduce long-term threats that can arise from guano harvests in HUPE breeding colonies.

#### 4.5 Sustainable guano extraction in Peru

The Peruvian guano industry has had an important role in shaping the country's history; achieving a sustainable extraction would not only exemplify the development of a sustainable practice but could also increase its economic value. The use of guano as a fertilizer extends back to pre-Hispanic cultures (Cushman, 2013). In the XIX century, its global commercialisation was crucial to Peru's development (Cushman, 2013). After the guano bird population crashed in the late-1800s due to unregulated guano extraction, the desire to revitalize the guano industry motivated the Peruvian government to apply leading measures to protect marine wildlife (Cushman, 2005; Tovar et al., 1987). A similar case was observed in the Benguela ecosystem in Africa, where uncontrolled guano extraction in the XIX century caused a decrease in the African penguin (*Spheniscus demersus*) breeding success (Frost et al., 1976). Furthermore, guano extraction was halted in this region due to its impacts on seabirds (Makhado et al., 2020). In Peru, the measures to revitalize the guano industry were coupled with a strong incentive for

developing scientific knowledge about guano birds to further prompt guano production (application of evidence-based solutions; Cárdenas-Alayza, 2022). Currently, guano is extracted with the main focus of promoting the growth of small and medium-sized agriculture in Peru by subsidizing it (Agro Rural, 2014). Additionally, an increase in guano production also presents a biological value, being a source of nitrogen and phosphorous input on land and sea ecosystems (Plazas-Jiménez & Cianciaruso, 2020). Altogether, guano extraction has been a crucial piece in the country's development and currently still possesses an important historical, social and biological value. These aggregated values would make achieving a sustainable guano extraction an ideal example of how economic (productivity) and conservation interests can collaborate and further enhance mutual growth. There is recent hope for a Third Guano Age, in which guano values would rekindle (Cushman, 2013) and new strategies would increase the revenue that the current administration generates (Valqui & Cárdenas, 2016). One of the proposed strategies by Valqui & Cárdenas (2016) is to draw on the historical and landscape values that the extraction areas present to increase tourism. This value is linked to the presence of charismatic species in the RNSIIPG islands and headlands, like the HUPE. In that sense, an important economic value could also be attributed to this species' conservation if ecotourism was promoted at these sites. In other *Spheniscus* colonies, penguin tourism has been reported to generate high revenue (Lewis et al., 2012; Skewgar et al., 2009). Lewis and colleagues, (2012) describe how a fast-growing penguin tourism industry became an important local source of income in only 20 years (Lewis et al., 2012). This example is a strong incentive for local authorities to prioritize penguin conservation, as is being done during guano harvests at PSJ.

#### 4.6 Future investigation and recommendations

Guano extraction effects on breeding HUPEs are still unclear, and future investigations should consider other possible disturbances like auditory stimuli and rat population increase. Using information on HUPE abundance at various guano extraction sites, a detailed following of population changes after harvests could be outlined to assess the



overall effects of the practice on this species' breeding colonies. More information is needed on the spatial distribution and survival rates of individuals of different life cycle stages (i.e. adult and juvenile) to fully comprehend HUPE population trends (McGill et al., 2021). Additionally, more studies are required to further understand HUPE responses to human disturbances. This could be explored using other higher-resolution monitoring methods at the individual or nest level (e.g., individual nest checks). From this, various parameters could be estimated (e.g. specific nest-loss dates, chick growth rates, fledging ratios) and associated with anthropogenic activity proximity. Other factors might have influenced penguin interaction with extraction stimuli and should be considered, like individual's personality or age (Ellenberg et al., 2009), previous exposure to human disturbances (Ellenberg et al., 2009, 2012), short-term habituation (Ellenberg et al., 2009; Walker et al., 2006), differences between breeding stages (Ellenberg et al., 2009; Groscolas et al., 2008), and effects of guano harvest stimuli on HUPE predators. Furthermore, colony topography and nest orientation should be included in future studies of similar nature, given that these can influence the interaction between extraction stimuli and the breeding HUPES. The use of physiological measurements could also help shed light on the degree of disturbance occurring during these activities (see, for example, Ellenberg et al., 2006, 2009, 2012; Palacios et al., 2018). We did not explore the effect of other confounding factors on the species' phenology, like environmental variables or fishing efforts near the colony, which could have helped isolate guano harvest effects from other selective pressures. Overall, our results indicate that guano harvest impacts are currently not the drivers of the HUPE population decline at PSJ. In that sense, the mitigation strategies applied at this site have proven to be efficient and similar alternatives should be implemented in other colonies where extraction occurs. Although these findings are a great story of human-induced impact mitigation and conservation, future studies are necessary to identify underlying factors of recent population reduction (e.g. suspected limitations of available prey) and to mitigate future declines expected due to climate change scenarios (Figueroa Nuñez, 2020).

## 5. Conclusions

We evaluated the effects of guano harvests with mitigation strategies on breeding HUPEs at PSJ and no evidence of negative impacts were identified. This study contributes much-needed scientific evidence to the question of the effectiveness of mitigation strategies for guano harvests at PSJ which were developed thanks to collaborative efforts between academic and governmental authorities. Nevertheless, the results of HUPE population trends are concerning for this species' conservation. Similar investigations focused on other threats that consider additional confounding factors are crucial to identifying the underlying causes of recent population declines. This case exemplifies the possibility of achieving a sustainable balance between economic practices and conservation, and most importantly, how future management and research could potentially boost mutual growth.

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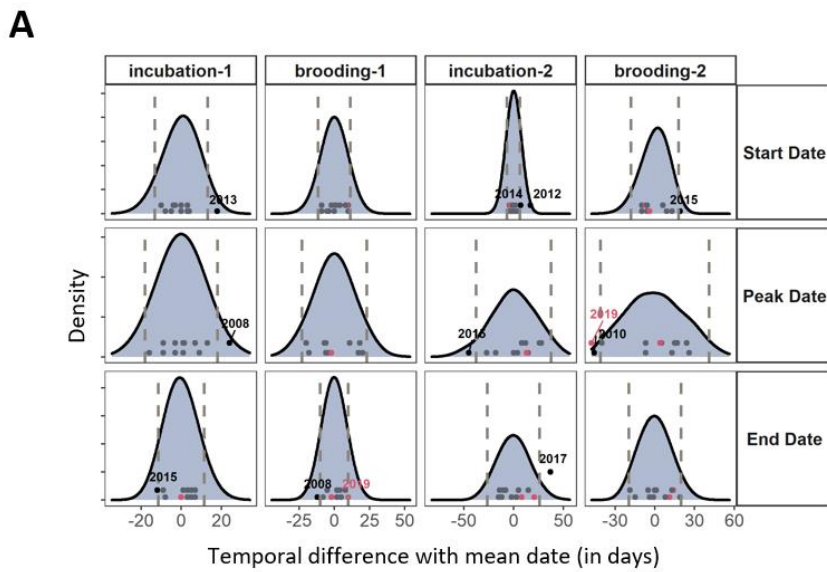
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## Appendix

Beach	PSJP sector label	Study sector label
S8	S8C	A
S8	S8B-izq	B
S8	S8B-area	C
S8	S8B-der	D
S8	S8A	E
S7	S7	F

Table A.1. Humboldt penguin colony sectors. Beaches in which colony sectors are found are specified, as well as original labels designated by the PSJP, and the labels used in this study (A-F).

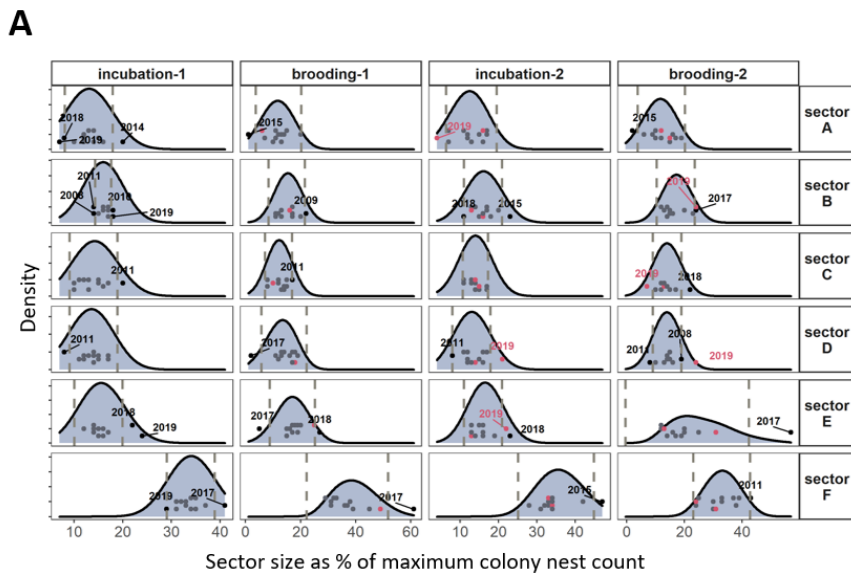


**B**

Campaign Year	Breeding Phase				Total N° phenological parameter outliers per guano extraction period	Average number of phenological parameter outliers per year (2008-2011, 2013-2014 or 2015-2018)
	incubation-1	brooding-1	incubation-2	brooding-2		
2012	-	-	-	-	0	1 ±1
2019	-	end date (p-value=0.048)	-	peak date (p-value=0.027)	2	2 ±1

}

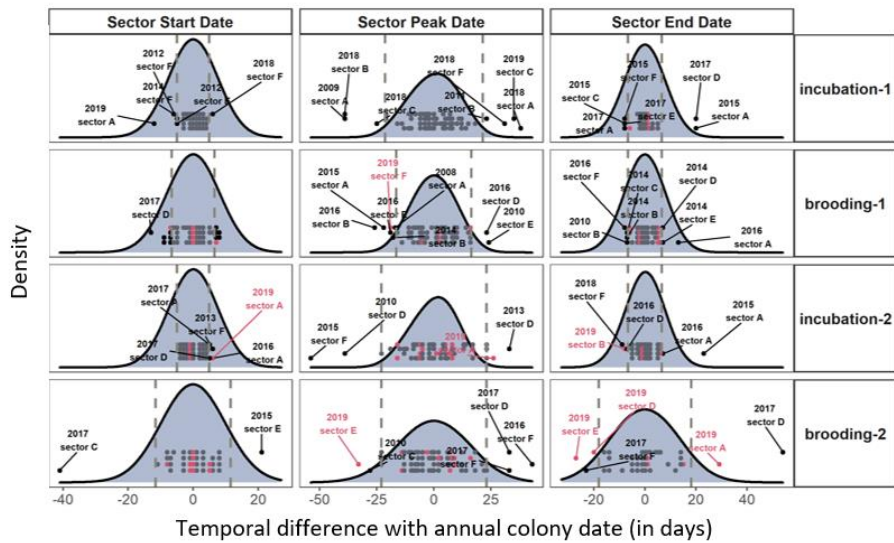
Figure A.1. Colony temporal parameters during the 2012/19 guano harvests that fall out of their 90% estimated probability distributions. A) Density distributions of phenological parameters per breeding phase. Vertical lines delimit 5% and 95% cumulative probability distribution lines. Parameters during extraction periods (BS-2 2012 and 2019) are colored in red and labeled if they are outside the vertical lines. B) Table of phenological parameters during guano extraction periods that fall out of their estimated 90% probability distributions.



**B**

Campaign Year	Breeding Phase				Total N° size outliers (lower 5% tail) per guano extraction period	Average number of size outliers per year (2008-2011, 2013-2014 or 2015-2018)
	incubation-1	brooding-1	incubation-2	brooding-2		
2012	-	-	-	-	0	1 ± 1
2019	-	-	Sector D (p-value=0.004) Sector E (p-value=0.023)	Sector B (p-value=0.04) Sector D (p-value=0.0004)	4	3 ± 1

Figure A.2. Sector size values during the 2012/19 guano harvests that fall in their lower 5% estimated probability distribution. A) Density distributions of sector size as % of maximum colony nest counts per breeding phase. Vertical lines delimit the 5% cumulative probability tails. Parameters during extraction periods are colored in red and labeled if they are outside their 90% cumulative probability distribution. B) Table of sector size values during guano extraction periods that fall on the lower 5% of their estimated probability distributions (i.e. lower than expected sizes).



## B

Campaign Year	Breeding Phase				Total N° phenological parameter outliers per guano extraction period	Average number of phenological parameter outliers per year (2008-2011, 2013-2014 or 2015-2018)
	incubation-1	brooding-1	incubation-2	brooding-2		
2012	-	-	-	-	0	4 ±2
2019	-	Sector A start date (p-value=0.04) Sector C start date (p-value=0.04) Sector F peak date (p-value= 0.029)	Sector A start date (p-value=0.023) Sector A peak date (p-value=0.032) Sector B end date (p-value=0.023)	Sector A end date (p-value=0.004) Sector D end date (p-value=0.035) Sector E end date (p-value=0.007) Sector E peak date (p-value=0.009)	10	9 ±3

Figure A.3. Sector temporal parameters during the 2012/19 guano harvests that fall out of their 90% estimated probability distribution. A) Estimated density distributions of sector phenological parameters per breeding phase. Vertical lines delimit the 5% cumulative probability tails. Parameters during extraction periods (BS-2 2012 and 2019) are colored in red and labeled if they fall outside their 90% cumulative probability distribution. B) Table of sector phenological parameters during guano extraction periods that fall out of their 90% estimated probability distributions.

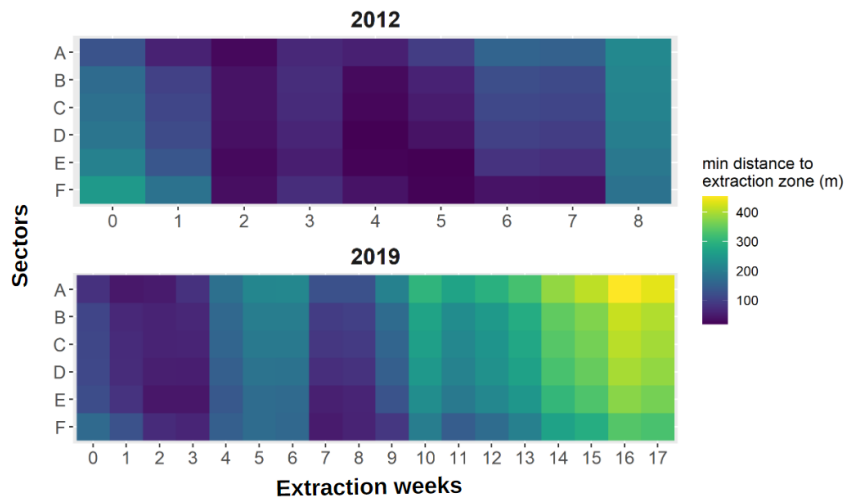


Figure A.4. Distances from the colony breeding sectors to weekly extraction areas during the 2012 and 2019 harvests at Punta San Juan. In 2012, distances ranged between 17-252 meters along 9 extraction weeks; on the other hand, in 2019 distances ranged from 43 to 453 meters in 17 extraction weeks.

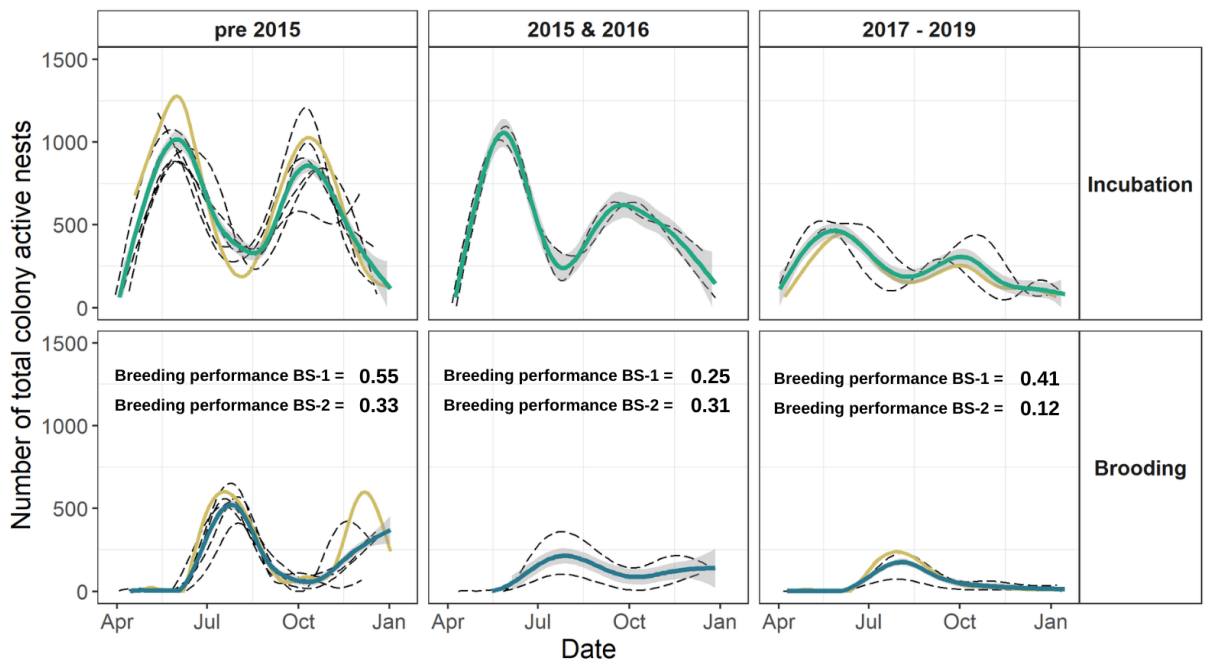


Figure A.5. Humboldt penguin phenology differences before and after the 2015 and 2017 ENSO events. Panels separate colony phenology by breeding stages (incubation and brooding) as well as by year groups: pre-2015, 2015 & 2016, 2017-2019. Black dashed lines represent yearly trends of the number of nests during non-extraction years; while gold continuous lines represent guano extraction years (2012 and 2019). Colored

continuous lines represent mean trends for each panel, with shaded outlines representing their standard deviation. Average breeding performance per breeding season (maximum ChickNest / maximum RoostNest) is shown per year group.