Incubation temperatures, sex ratio and hatching success of leatherback turtles (*Dermochelys coriacea*) in two protected hatcheries on the central Mexican coast of the Eastern Tropical Pacific Ocean

J. García–Grajales, J. F. Meraz Hernando, J. L. Arcos García, E. Ramírez Fuentes

Abstract

*Incubation temperatures, sex ratio and hatching success of leatherback turtles (*Dermochelys coriacea*) in two protected hatcheries on the central Mexican coast of the Eastern Tropical Pacific Ocean.* Incubation temperatures, sex ratio and hatching success of leatherback turtles have received little attention in conservation programs in Mexico. This study was carried out from October 2014 to May 2017 in two enclosed hatchery sites. To determine temperature parameters in the nest chamber environment and their variation during the incubation period, we placed data loggers in the centre of the egg mass in relocated nests. We then buried other data loggers in the sand near the relocated nests, inside and outside the hatchery. A total of 46 nests were examined over three nesting seasons. Mean nest temperature showed no statistical difference between nests in either the San Juan Chacahua hatchery or in the Palmarito hatchery nests. The mean sex ratio based on average temperature during the middle third of incubation duration was 96.3 % skewed to female production. Hatching success in both San Juan Chacahua and Palmarito was high. Our findings support the common pattern of a female–dominated leatherback turtle sex ratio. Furthermore, hatching success rates in the shade–cloth hatchery were higher than those in the natural nests observed in other populations.

Key words: Hatcheries, Secondary beaches, Clutch size, Nests temperatures, Shading net

Resumen

*Temperaturas de incubación, proporción de sexos y éxito de eclosión de la tortuga laúd (*Dermochelys coriacea*) en dos criaderos protegidos en la costa central mexicana del océano Pacífico tropical oriental.* Las temperaturas de incubación, la proporción de sexos y el éxito de eclosión de la tortuga laúd han recibido poca atención en los programas de conservación en México. Este estudio se realizó entre octubre de 2014 y mayo de 2017 en dos criaderos cercados. Para determinar los parámetros de temperatura en el ambiente de la cámara de anidación y su variación durante el período de incubación, colocamos registradores de datos en el centro de cada nido trasladado. Posteriormente, se enterraron otros registradores en la arena cerca de los nidos trasladados, dentro y fuera del criadero. Se analizó un total de 46 nidos durante tres temporadas de anidación. No se observaron diferencias estadísticas en la temperatura media entre los nidos del criadero de San Juan Chacahua ni entre los de Palmarito. La proporción de sexos basada en la temperatura media durante el segundo tercio del período de incubación fue del 96,3 % en favor de la producción de hembras. El éxito de eclosión en los criaderos de San Juan Chacahua y de Palmarito fue alto. Nuestros resultados concuerdan con el patrón habitual de proporción de sexos observado para la tortuga laúd, que está dominado por la presencia de hembras. Además, los índices de éxito de eclosión en el criadero protegido con sombra artificial fueron más altos que los observados en los nidos naturales en otras poblaciones.

Palabras clave: Criaderos, Playas secundarias, Tamaño de nidada, Temperaturas de nidos, Malla de sombreado

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Introduction

The leatherback turtle (*Dermochelys coriacea*) inhabits a wide range of coastal and pelagic waters in tropical and temperate ecosystems. They are found in both hemispheres, from the equator to sub–polar regions, although nesting activity is confined to tropical and subtropical latitudes (Benson et al., 2015). This species is globally listed as vulnerable under the International Union for the Conservation of Nature (IUCN) criteria (Wallace et al., 2013), and trends and status in the Pacific Ocean basin have declined precipitously in recent decades, with declines of more than 90 % in Mexico (Sarti et al., 2007).

For more than two decades, considerable efforts and broader conservation strategies have been devoted to the protection of sea turtles in Mexico (García et al., 2003). These efforts include the protection of nesting beaches with regular patrols against human poaching, widely implemented nest translocation to protected hatcheries, and other general strategies such as a complete ban on the exploitation of turtles and their eggs (García et al., 2003). However, specific conservation efforts for leatherback turtles in Mexico have been focused on four index beaches (Mexiquillo, Tierra Colorada, Cahuitán and Barra de la Cruz) selected due to their intense nesting activity (Sarti et al., 2007; Santidrían et al., 2017). Nevertheless, secondary beaches where turtles nest regularly can also be considered important nesting sites (Santidrían et al., 2017). In all of these beaches, nests are protected by relocating freshly laid clutches to protected hatcheries —a common practice used at sea turtle rookeries around the world— to increase hatchling recruitment (Baskale and Kaska, 2005; Maulany et al., 2012; Santidrían et al., 2017). In this context and knowing that temperature plays an important role in the life–history of sea turtles (Binckley and Spotila, 2015), it is important to understand temperature regimes in enclosed, protected hatcheries and corresponding hatchling sex ratios and hatching success because few detailed studies have been conducted on these topics and on the effectiveness of hatchery management on leatherback turtle nests laid on secondary beaches in the Mexican Pacific (Vannini and Rosales, 2009; Vannini et al., 2011)

The goals of this study were: 1) to compare the temperatures of leatherback turtle nests in two protected hatcheries, 2) to estimate the sex ratio of hatchlings, 3) to compare the differences in the incubation period, the number of dead embryos, and hatching success between hatcheries, and 4) to obtain information about the relationship between nest temperature and the incubation period in hatcheries on the Central Pacific Coast of Oaxaca, Mexico.

Material and methods

Field work

The study took place in San Juan Chacahua and Palmarito beaches in the Central Pacific Coast of Oaxaca. San Juan Chacahua beach is 12 km in length, and is part of the Lagunas de Chacahua National Park, while Palmarito is about 16 km in length, extending from San José Manialtepec River in the northwest to Punta Colorada in the southeast. The climate is tropical, hot, and humid, and characterized by well–defined dry and rainy seasons. The mean annual temperature is 27.5 ºC and mean annual rainfall is 800 mm, concentrated between July and October; the dry season can last 8 months, from November to June.

This study was carried out from October 2014 to May 2017, comprising three annual leatherback–breeding seasons (October–May). Community groups at each beach protect and relocate the nests to increase hatching success (García et al., 2003; Vannini and Rosales, 2009; Vannini et al., 2011). The enclosed hatchery sites were constructed to cover a total area of 80 m^2 (10 x 8 m), which was sufficient to accommodate 100 nests and high enough not to be inundated by high tides. The distance between nests was set at 1 m in order to reduce interaction and to allow hatchery personnel to walk without stepping on the nests. Likewise, in order to protect them from the intensity of the sun, the hatcheries were covered by a sheet of shading net at a height of 1.50 m during all breeding seasons. Hatcheries were moved each year around the area to avoid accumulation of bacteria and other kinds of contamination.

Community groups patrolled both beaches at night from 21:00 to 06:00 h, using an all–terrain vehicle ATV, to record any sea turtle activity. All nests recorded were collected and numbered and cloth size was recorded. Nests were transported in clean plastic bags to the enclosed hatcheries. These sites were closely monitored daily for threats from natural predators. All relocated nests were buried in the hatchery at a depth of 80 cm, the mean depth of leatherback nesting activity reported for Pacific populations.

To determine temperature parameters in the nest environment and their variation during incubation period, we placed a data logger in the center of the egg mass in some relocated nests. Two types of data loggers were used between 2014 and 2017: HOBO® Pro v2 Temp/HR (Onset Comp. Corp., Bourne, MA, USA) in 6 nests on both beaches, and HOBO® UA–002–08 (Onset Comp. Corp., Bourne, MA, USA) in 40 nests on both beaches. Additionally, to study the effect of metabolic heating, one data logger was buried outside the hatchery (approximately 10 m from the hatchery) at the same depth (called 'R1', reference 1). A second group of two temperature data loggers was distributed inside the hatchery; one was buried alone in the ground at the same depth (called 'R2', reference 2), and another was placed at the environmental level under the shading net (called 'R3', reference 3). Due to logistical situations, we put the reference data loggers only in the last two seasons of the study, and we only recorded the incubation temperature data in nests of San Juan Chacahua in the first season of the study (2014–2015). All data loggers were programmed to record temperature every 30 min.

Nest contents were excavated twenty–four hours after the emergence of the first hatchling, as suggested by Patiño–Martínez et al. (2010), and data loggers were retrieved. The total number of eggs (the number of eggs laid in the nest) and hatching success were calculated

by counting unhatched eggs, dead hatchlings in eggs, and dead hatchlings in nests, and by excluding the shelled albumen globes (SAGs). The hatching success for each nest was calculated as the percentage of hatchlings in the clutch, and the incubation period per nest was determined as the numbers of days from the date of egg deposition to the date of the first hatchling emergence (Yalçın–Özdilek et al., 2007).

Data analysis

Daily thermal fluctuation was calculated from the difference between maximum and minimum daily temperatures for each nest. After testing for normality and homogeneity of variances, we used a parametric one–way ANOVA test to examine differences in mean daily temperature and daily thermal fluctuation in nests and between nests, years and hatcheries.

Metabolic heat is measured from the difference between clutch temperature and soil temperature (without eggs), and has a daily cyclic variation depending on clutch and the differences in the soil temperature. For this work, the metabolic heat was defined as the difference between the nest temperature and the data-logger reference temperature (without eggs, outside of the hatchery) during the incubation period (Broderick et al., 2001). Levene's t–tests were used to compare variance between nest temperatures and reference–site temperatures during the middle third of incubation, which corresponds to the temperature sensitive period (TSP), and the entire incubation duration. Welch t–tests were used to compare temperatures between nest and reference sites.

The mean middle–third temperature for each monitored clutch was calculated individually. For this work, we used the mean temperature during the middle third of the incubation period to estimate the sex ratio, and used and adapted the equations of sex ratio as a function of temperature calculated by Mrosovsky et al. (2002), as follows:

$$Y = 100.06 / (1 + Exp (+ 188.78 - 6.37*X))$$

where Y is the sex ratio and X is the temperature.

The hatching success was determined using the following formula:

$$[(total\ eggs - unhatched\ eggs)/total\ eggs] \times 100$$

Mean hatching success was calculated by hatchery and by year. As hatching success is not a continuous variable, an arcsine transformation of data was implemented. Then we tested the normality and homogeneity of variances of data, and a parametric one–way ANOVA test was used to examine differences between years and hatcheries.

Results

Clutch size and nest temperatures

A total of 46 nests were examined for nest temperatures during three nesting seasons in the two hatcheries from the Central Pacific Coast of Oaxaca. For each season,

the nests were placed at different times throughout the breeding season. The mean clutch size in leatherback turtle nests was 84.1 ± 10.6 eggs (range 62–104 eggs), excluding the SAGs. The incubation period was recorded between 52 and 62 days, with a mean of 57.04 days ± 2.7 SD (standard deviation). Mean nest temperature in San Juan Chacahua hatchery showed no statistical difference among nests (ANOVA $F_{25, 584}$ = 147.4, $p > 0.05$; fig. 1), nor was there any statistical difference among Palmarito hatchery nests (ANOVA $F_{19, 367}$ = 118.7, $p > 0.05$; fig. 2). The overall average temperature by seasons for the San Juan Chacahua hatchery was 31.01 ± 0.46 °C in 2014–2015, 30.74 ± 0.96 °C in 2015–2016, and 30.43 ± 0.78 °C in 2016–2017, but this difference was not significant (ANOVA $F_{2, 54}$ = 1.33, $p > 0.05$). The overall average temperature by seasons for the Palmarito hatchery was 30.61 ± 0.23 °C in 2015–2016, and 30.53 ± 0.41 °C in 2016–2017, again without significant differences (ANOVA $F_{1, 18}$ = 1.83, $p > 0.05$).

The daily thermal fluctuation varied among nests (ANOVA $F_{25, 38}$ = 3.72, $p < 0.05$) in the San Juan Chacahua hatchery, and also varied (ANOVA $F_{19, 36}$ = 3.16, $p < 0.05$) in the Palmarito hatchery. However, or we found no differences in daily thermal fluctuations between seasons in the San Juan Chacahua hatchery (ANOVA $F_{2, 19}$ = 4.41, $p > 0.05$) or in the Palmarito hatchery (ANOVA $F_{1, 16}$ = 3.38, $p > 0.05$).

Length of incubation period

The length of the incubation period was obtained for 46 nests with known mean incubation temperature. We found a statistical relationship between the length of the incubation period and the mean nest temperature (r = –0.97; $F_{1,47}$ = 22.260; $p < 0.05$; fig. 3A), as well as between th e length of the incubation period and the mean temperature during TSP (r = –0.86; $F_{1, 53}$ = 24.75; $p < 0.05$; fig 3B).

Metabolic heat

Metabolic heat was evident in all monitored clutches and most evident during the TSP in all clutches. However, heating for all clutches was greater during the final third of incubation ($F_{2, 45}$ = 7.41, $P < 0.05$; fig. 4), followed by a gradual decline in nest temperature toward the end of incubation, compared with the first or the middle third. Mean temperature during the entire incubation period differed between nests and reference sites (t = 0.847, $p < 0.05$). Intensity of metabolic heat ranged from 0.7 °C to 3.2 °C, with a mean of 1.88 °C ± 0.52 °C for the entire incubation period.

Sex ratio and hatching success

Table 1 shows the sex ratios of hatchlings for all nests estimated from curve equation. The mean sex ratio based on average temperature during the middle third of incubation duration (T°) was 96.3 %, and ranged between 44 and 100 %, but the sex ratio between nests did not vary significantly (Kruskal–Wallis test, H = 17.469, $P = 0.13$). Only one of the 46 nests (nest
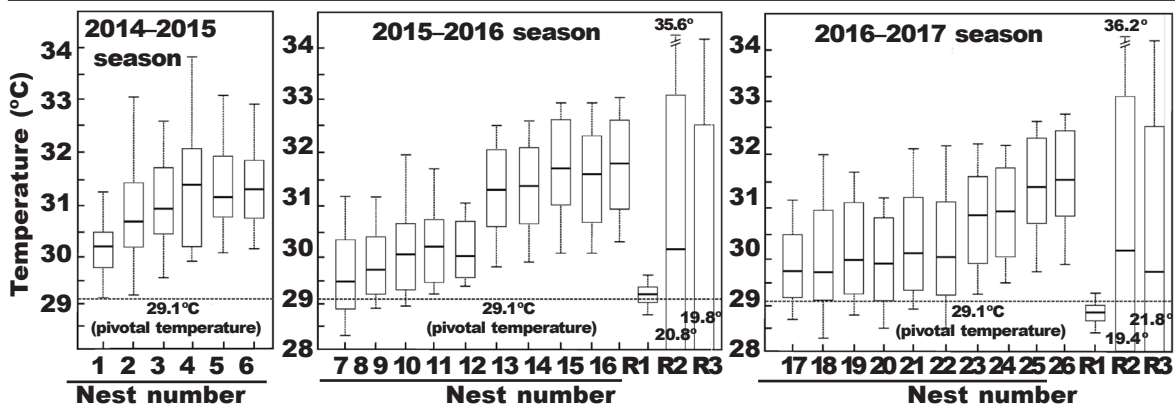
Fig. 1. Mean nest temperature in San Juan Chacahua hatchery during three seasons. Dark lines inside each box represent median temperature. Whiskers represent the maximum and minimum values recorded. The horizontal dashed lines correspond to the proposed nest pivotal temperature for leatherback turtle: R1, reference sensor 1; R2, reference sensor 2; R3, reference sensor 3.

Fig. 1. Temperatura media de los nidos en el criadero de San Juan durante tres temporadas. Las líneas oscuras dentro de los recuadros representan la temperatura mediana. Los bigotes representan los valores máximos y mínimos registrados. Las líneas horizontales discontinuas corresponden a la temperatura umbral de nido propuesta para la tortuga laúd; R1, sensor de referencia 1; R2, sensor de referencia 2; R3, sensor de referencia 3.
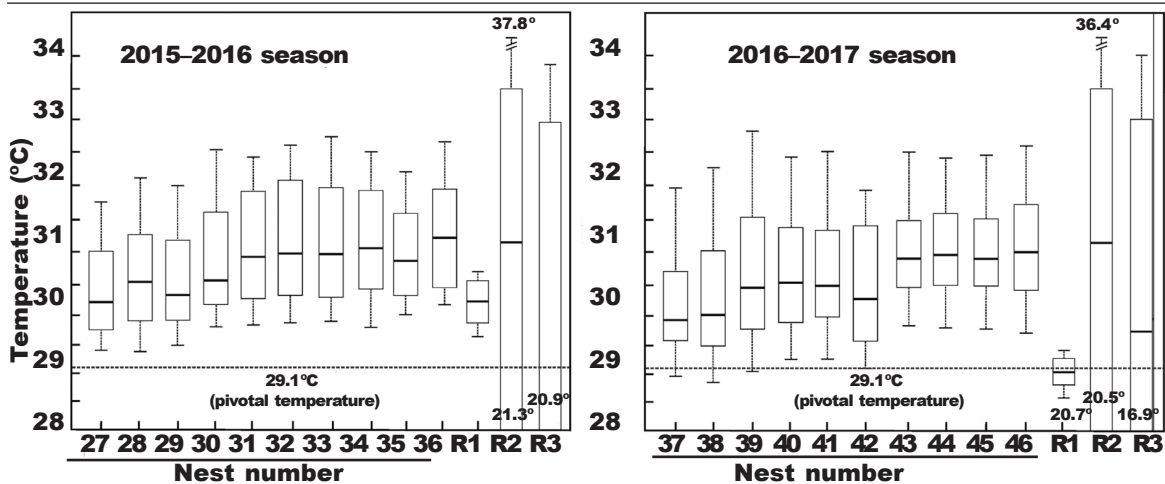

Fig. 2. Mean nest temperature in Palmarito hatchery during two seasons. Dark lines inside each box represent median temperature. Whiskers represent the maximum and minimum values recorded. The horizontal dashed lines correspond to the proposed nest pivotal temperature for Leatherback turtle: R1, reference sensor 1; R2, reference sensor 2; R3, reference sensor 3.

Fig. 2. Temperatura media de los nidos en el criadero de Palmarito durante dos temporadas. Las líneas oscuras dentro de los recuadros representan la temperatura mediana. Los bigotes representan los valores máximos y mínimos registrados. Las líneas horizontales discontinuas corresponden a la temperatura umbral de nido propuesta para la tortuga laúd: R1, sensor de referencia 1; R2, sensor de referencia 2; R3, sensor de referencia 3.
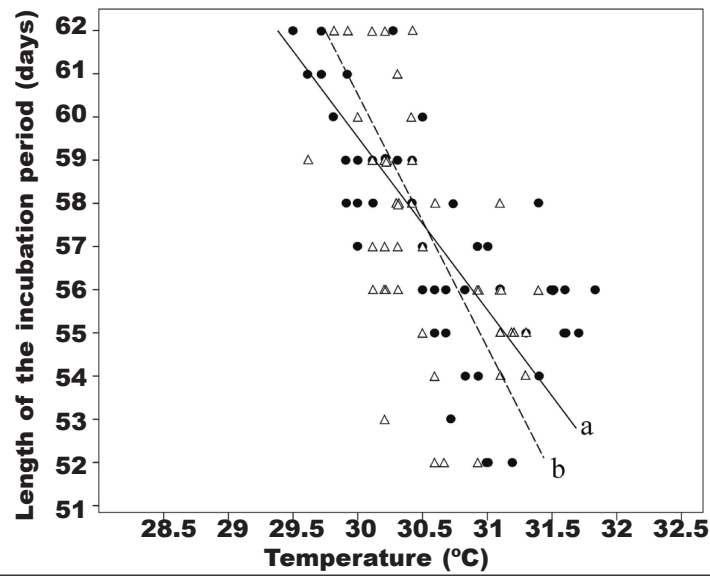
Fig. 3. Relationship between the length of the incubation period and a) mean nest temperature, and b) mean TSP; obtained for 46 nests in both hatcheries. Filled circles represent San Juan Chacahua hatchery and open triangles represent Palmarito Hatchery.

Fig. 3. Relación entre la duración del período de incubación y a) la temperatura media de los nidos y b) el período sensible a la temperatura; obtenida para 46 nidos en ambos criaderos. Los círculos negros representan el criadero de San Juan y los triángulos blancos, el de Palmarito.
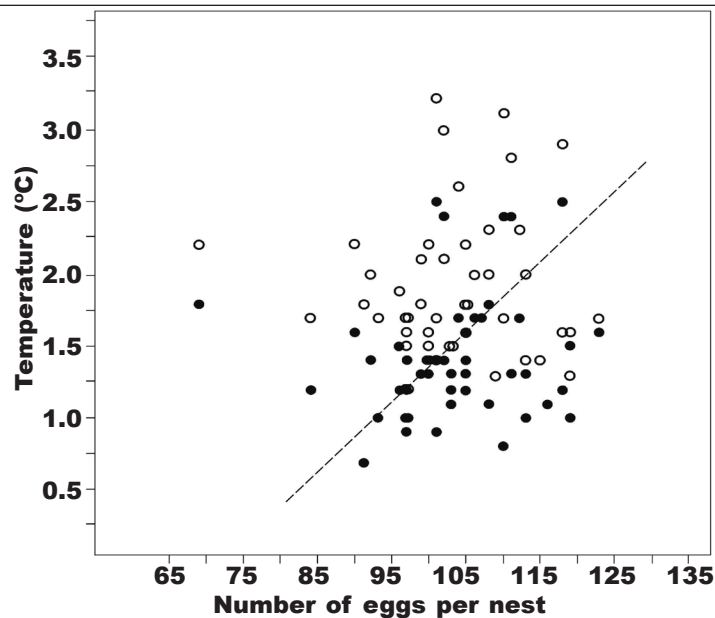
Fig. 4. Metabolic heat during the final third of the incubation period, and the relationship with the number of eggs per nest. Filled circles represent San Juan Chacahua hatchery and open circles represent Palmarito Hatchery.

Fig. 4. Calor metabólico durante el último tercio del período de incubación y la relación con el número de huevos por nido. Los círculos negros representan al criadero de San Juan y los blancos, al de Palmarito.

Table 1. Sex ratios of hatchlings incubated in two protected hatcheries on the central coast of Oaxaca, Mexico: Nn, nest number; Nf, number of fertile eggs; N, number of SAG's; T, total number of eggs; Id, incubation duration (days); WIP, whole IP; Ft, first third; St, second third (PTS); Lt, last third; Mh, metabolic heating (in ºC); Hs, hatching success number (%).

Tabla 1. Proporción de sexos en las crías incubadas en dos criaderos protegidos en la costa central de Oaxaca, en México: Nn, número de nidos; Nf, número de huevos fértiles; N, número SAG; T, número total de huevos; Id, Duración de la incubación (en días); WIP, IP completo; Ft, primer tercio; St, segundo tercio (PTS); Lt, último tercio; Mh, calor metabólico (en ºC); Hs, número de eclosiones con éxito (%).

| | | | | | | Incubation temperature (ºC) | | | | | | | | |
| | | | | | | Nest | | | | Sand | | | | |
Nn	Lay date	Nf	N	T	Id	WIP	Ft	St	Lt	Ft	St	Lt	Mh	Hs
San Juan Chacahua														
2014–2015														
1	13/10/2014	86	24	110	62	30.3	29.7	30.4	30.9	29.1	29.7	30.2	0.7	35 (40.7)
2	28/10/2014	78	18	96	58	30.7	29.8	31.1	32.1	29.5	29.9	30.3	1.8	28 (35.9)
3	05/11/2014	92	16	108	57	30.9	29.8	30.3	31.7	29.6	30.1	30.4	1.3	34 (37)
4	07/11/2014	81	12	93	56	31.5	30.1	31.1	32.3	29.6	30.1	30.6	1.7	36 (44.4)
5	18/11/2014	87	15	102	55	31.3	30.1	31.3	32.2	29.5	29.9	30.1	2.1	46 (52.9)
6	05/12/2014	69	28	97	54	31.4	30.3	31.1	31.7	29.8	30.2	30.5	1.2	32 (46.4)
2015–2016														
7	08/11/2015	68	31	99	62	29.5	28.9	29.8	30.9	28.2	28.5	28.8	2.1	57 (83.8)
8	12/11/2015	94	14	108	61	29.7	29	30.3	31.3	28.3	28.5	29	2.3	75 (79.8)
9	23/11/2015	79	26	105	59	30	29	29.6	30.9	28.2	28.4	28.7	2.2	63 (79.7)
10	09/12/2015	85	21	106	58	30.1	29.6	30.4	31.1	28.4	28.7	29.1	2	68 (80)
11	13/12/2015	79	25	104	58	29.9	29.5	30.3	31.6	28.3	28.6	29	2.6	57 (72.2)
12	24/12/2015	93	18	111	56	31.5	30.1	30.9	32.1	28.5	28.5	29.3	2.8	71 (76.3)
13	27/12/2015	103	15	118	56	31.6	30.1	31.1	32	28.7	28.6	29.1	2.9	76 (73.8)
14	03/01/2016	65	37	102	55	31.7	30.5	31.2	32.4	28.5	28.8	29.4	3	52 (80)
15	05/01/2016	89	21	110	55	31.6	30.7	31.2	32.4	28.6	28.8	29.3	3.1	64 (71.9)
16	13/02/2016	78	23	101	56	31.8	30.8	31.4	32.6	28.7	28.9	29.4	3.2	59 (75.6)
2016–2017														
17	28/10/2016	97	15	112	62	29.7	28.7	30.2	31.3	28.3	28.5	29	2.3	71 (73.2)
18	02/11/2016	84	12	96	61	29.6	28.6	29.9	30.8	28.1	28.4	28.9	1.9	69 (82.1)
19	05/11/2016	72	18	90	61	29.9	28.8	30.1	31.1	28.3	28.5	28.9	2.2	58 (80.6)
20	16/11/2016	96	17	113	60	29.8	28.7	30	31.1	28.5	28.7	29.1	2	68 (70.8)
21	27/11/2016	101	18	119	59	30.1	28.9	30.2	31	28.4	28.7	29.2	1.8	82 (81.2)
22	07/12/2016	69	31	100	58	30	29.1	30.1	31.3	28.6	28.8	29.1	2.2	53 (76.8)
23	09/12/2016	73	19	92	57	31	29.2	30.3	31.2	28.5	28.9	29.2	2	58 (79.5)
24	05/01/2017	87	18	105	56	31.1	29.1	30.2	31.1	28.6	28.8	29.3	1.8	62 (71.3)
25	10/01/2017	95	13	108	56	31.5	29.4	30.3	31.2	28.7	28.6	29.2	2	74 (77.9)
26	08/02/2017	62	7	69	55	31.6	29.7	30.5	31.6	28.4	28.7	29.4	2.2	49 (79)
Palmarito														
2015–2016														
27	03/11/2015	82	23	105	59	30.2	29.9	30.4	31.1	28.8	29.1	29.5	1.6	68 (82.9)
28	09/11/2015	65	32	97	56	30.5	29.7	30.2	31.1	28.9	29	29.4	1.7	51 (78.5)
29	24/11/2015	86	13	99	58	30.4	29.9	30.6	31.2	28.9	29.2	29.4	1.8	67 (77.9)
30	07/12/2015	76	21	97	57	30.5	30.1	30.4	31.2	29.1	29.4	29.7	1.5	62 (81.6)

Tabla 1. (Cont.)

| | | | | | Incubation temperature (ºC) | | | | | | | | |
| | | | | | Nest | | | | Sand | | | | |
Nn	Lay date	Nf	N	T	Id	WIP	Ft	St	Lt	Ft	St	Lt	Mh	Hs
31	15/12/2015	84	19	103	52	31	30.2	30.7	31.5	29.3	29.6	30	1.5	68 (81)
32	08/01/2016	79	21	100	56	30.6	30.5	31.1	31.8	29.5	29.7	30.1	1.7	58 (73.4)
33	13/01/2016	94	17	111	56	30.7	30.2	30.9	31.7	29.4	29.6	30.2	1.5	73 (77.7)
34	26/01/2016	75	22	97	55	30.7	30.6	31.2	31.7	29.5	29.8	30	1.7	63 (84)
35	06/02/2016	86	17	103	55	30.6	30.4	31.1	31.6	29.7	29.9	30.1	1.5	73 (84.9)
36	12/02/2016	92	31	123	54	30.9	30.5	31.3	31.8	29.4	29.7	30.1	1.7	76 (82.6)
2016–2017														
37	13/10/2016	84	13	97	59	29.9	29.7	30.1	30.9	28.8	29.1	29.7	1.2	72 (85.7)
38	17/10/2016	97	22	119	57	30	29.6	30.1	30.8	28.9	29.1	29.5	1.3	81 (83.5)
39	10/11/2016	73	11	84	59	30.4	29.8	30.2	31.1	28.7	29	29.4	1.7	59 (80.8)
40	18/11/2016	88	15	103	60	30.5	29.8	30.4	31.1	28.7	29.1	29.5	1.6	71 (80.7)
41	13/12/2016	96	19	115	58	30.4	29.7	30.3	31	28.9	29.2	29.6	1.4	83 (86.5)
42	28/12/2016	78	23	101	59	30.3	29.7	30.2	31.1	28.9	29.3	29.7	1.4	65 (83.3)
43	05/01/2017	104	9	113	53	30.7	29.8	30.2	31.2	28.9	29.2	29.8	1.4	74 (71.2)
44	13/02/2017	83	18	101	52	31	30.1	30.6	31.4	29	29.2	29.7	1.7	69 (83.1)
45	28/01/2017	96	22	118	54	30.8	30.2	30.6	31.5	29.1	29.4	29.9	1.6	77 (80.2)
46	08/02/2017	89	16	105	52	31.2	30.2	30.9	31.6	29.1	29.3	29.8	1.8	73 (82)

number seven, see table 1) was predicted to produce more males; t the majority of the nests were thus predicted to produce more females.

Hatching success varied between years in both hatcheries. In the San Juan Chacahua hatchery it was 42.8 % in 2014–2015, 77.1 % in 2015–2016, and 77 % in 2016–2017 (ANOVA $F = 105.84$, $p < 0.0001$), while in the Palmarito hatchery it was 80.5 % in 2015–2016, and 81.5 % in 2016–2017 (ANOVA $F = 0.568$, $p < 0.05$). Hatching success also varied between hatcheries (ANOVA $F = 12.771$, $p < 0.0$).

Discussion

In this study, the mean clutch size in leatherback turtle nests was 84.1 ± 10.6 eggs in both hatcheries. This is higher than the clutch size (62 ± 17.9) reported in the index of beaches of the Mexican Pacific (Sarti et al., 2007). Most other leatherback nesting populations have a smaller clutch size (Eckert et al., 2015; Sotherland et al., 2015). Some researchers have shown that clutch size increases with body size in other marine turtles (Wallace et al., 2007). However, we did not collect female size data.Santidrían and Swiggs (2015) mention inconsistencies in the reports of clutch size and in the terms used to describe hatching of eggs and emergence of hatchlings, mainly because some consider the total number of eggs including SAGs, which are not real eggs because they lack yolk (Sotherland et al., 2015).

Temperature may be the single most important variable affecting egg development and hatchling output in leatherback turtles, influencing the developmental rate, hatching success, emergence rate, proportion of female hatchlings, and fitness of hatchlings (Santidrián and Swiggs, 2015). Our results provide evidence of daily thermal fluctuation within the egg chamber of *Dermochelys coriacea* nests in protected hatcheries. Likewise, it should be taken into account that the mean temperature of nests remained relatively homogeneous, probably as a result of the hatchery shading. This shading strategy has been proposed as a focused technique to mitigate the effects of temperature (Van de Merwe et al., 2006; Hill et al., 2015) and can be performed to facilitate survival from the nest and to increase reproductive output, principally because hatcheries with shade cloth decreased sand temperatures to the upper limit of the optimal incubation temperature range (Hamann et al., 2010).

We found that the mean incubation period of all hatchery nests (57.04 days) was shorter than that of natural nests (59.9 days) in the East Pacific (Santidrian and Swiggs, 2015). Similarly, for temperature–recorded nests, the mean nest temperature of hatchery nests (30.6 ºC) was similar to that of natural nests (30.6 ºC) in the East Pacific (Santidrian and Swiggs, 2015).

To the best of our knowledge, this is the first study to investigate and compare metabolic heating in hatcheries in Mexico, including the comparison of nest centre and adjoining sand temperatures (reference site). In the past, quantifying metabolic heating within nests was difficult due to the expense and unreliability of temperature loggers (Limpus et al., 1983; Broderick et al., 2001). Currently, however, the miniaturization and the capacity of the loggers make accurate measurements of nest temperatures feasible (Broderick et al., 2001). Our results show that the general pattern of metabolic heating was similar to that found in previous studies for other species (Morreale et al., 1982; Broderick et al., 2001; Tapilatu and Ballamu 2015; Candan and Kolankaya, 2016; Özdilek et al., 2016), with metabolic heating recorded mainly during the second half of incubation with a peak, followed by a gradual decline in nest temperature toward the end of incubation. Godfrey et al. (1997) recorded the temperature in clutches of the leatherback turtle (Matapica beach, Suriname) in addition to recording the sand temperature to the side of the clutch. Nest temperatures were found to vary, on average, from control temperatures by 0.82 °C during the TSP, suggesting that metabolic heating may play some role in influencing hatchling sex ratios. In addition, it has been suggested that metabolic heating can only be important if it elevates the nest temperature by > 1 ºC during the middle third of incubation (Yntema and Mrosovsky, 1980).

Özdilek et al. (2016) mention that metabolic heating during incubation periods should not be ignored as a cause for the increasing nest temperatures found during incubation. This study showed that mean temperature in the nest chamber increases 3.2 ºC with respect to the reference sites, and heating was observed in all monitored nests. Yntema and Mrosovsky (1980) stated that a change of 1–2 °C can make a considerable difference in the sex ratios of hatchlings, and some studies have documented sufficient metabolic heating in nests during TSP to significantly alter the hatchling sex ratios (Broderick et al., 2001; Kaska et al., 2006; Jribi et al., 2013; Tapilatu and Ballamu, 2015; Özdilek et al., 2016). This is because sexual differentiation in sea turtles is strongly influenced by ambient incubation temperature (Standora and Spotila, 1985; Mrosovsky, 1994); more specifically, the embryo is exposed to a continuous temperature during the middle trimester of incubation, which determines the eventual gonadal differentiation and sex of the hatchling (Wibbels, 2003). Nevertheless, incubation studies in leatherback turtles indicate that the most temperatures produce either all males or females, given the narrow transitional range of temperature (Mrosovsky and Pieau, 1991; Godfrey et al., 1997; Binckley and Spotila, 2015), and there is only a narrow time range when nest temperature determines hatchling sex (Mrosovsky and Pieau, 1991). Although in this work we provide evidence of metabolic heat, this was most pronounced during the last third of development with variations, and possibly sex was already determined (Mickelson and Downie, 2010).

The reported sex ratio for leatherback turtles is generally female–dominated (Binckley and Spotila, 2015), as we report in this study. Possibly, increases in global temperature will affect leatherback hatchling sex ratios (Binckley et al., 1998; Patiño–Martínez et al., 2012), and survival of hatchlings (Saba et al., 2012; Spotila et al., 2015). Several studies have used controlled incubation temperature to measure and explain temperature effects on sex determination in leatherback turtle eggs (Rimblot et al., 1985; Chan and Liew, 1996; Binckley et al., 1998; Chevalier et al., 1999). However, the pivotal temperature may vary with species and among populations in natural nests (Binckley and Spotila, 2015). Nevertheless, the hatchery management strategy implemented in Mexico (García et al., 2003) focuses only on increasing hatching success (Sönmez et al., 2013; Sari and Kaska, 2017) and does not contemplate the effects of temperature on the nests.

With regards to the hatching success of temperature–recorded nests, we found the mean hatching success (74.4 %) to be higher than that of natural nests (47 %, Playa Grande Costa Rica; Santidrian and Swiggs, 2015), which is consistent with our overall finding on hatching success showing that hatchery management increases hatching success. However, the use of a hatchery site may not always guarantee the hatching success (Pazira et al., 2016; Vannini et al., 2011). For example, in a study on leatherback turtle eggs in the Southwestern Caribbean Sea, Patino–Martínez et al. (2012) found that the hatching rate was higher in natural nests (79.9 %) than in those transferred to the beach hatchery (67.7 %). Hence, although relocation is suggested to be a common strategy for conservation of declining sea turtle populations (Baskale and Kaska, 2005; Pfaller et al., 2008), there is no consensus among researchers about whether relocation is an effective conservation tool for sea turtles (Sari and Kaska, 2017). More details about the advantages and disadvantages of hatchery management can be found in Sari and Kaska (2017). This study attempts to elucidate the impact of nest relocation and the effectiveness of community conservation in Mexico. It also provides a basis for further studies related to reproductive ecology of the leatherback sea turtle, an endangered species.

Conclusions

San Juan Chacahua and Palmarito beaches are considered secondary nesting beaches for leatherback turtles on the Mexican Pacific Coast (Sarti et al., 2007; Santidrian et al., 2017). The nest temperatures recorded inside the chamber nests were near the upper tolerance limits for incubation of leatherback turtle eggs and hence the results of the present study were in agreement with the common pattern of leatherback turtle hatchling sex ratios from beaches in the Eastern Pacific, which is female–dominated. Hatching success rates in the shade–cloth hatchery were higher than the natural nest rates observed in other populations. Finally, the conservation aspects related to sex ratio and hatching success, as well as research on metabolic heating need to be continuously monitored over several years.

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