

# Egg laying duration in the olive ridley turtle *Lepidochelys olivacea* and its relevance for the estimation of mass nesting population size

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The olive ridley sea turtle *Lepidochelys olivacea* is known for its unusual behaviour of locally nesting en masse in the eastern Pacific and the east coast of India, leading to difficulties in quantifying population size. In the present study we estimate nesting population sizes in the state of Orissa, taking average egg laying or oviposition duration (OD) into account. We observed 182 and 263 nesting turtles during mass nesting events of 2009 and 2010 to estimate ODs of  $19.8 \pm 5.8$  and  $17.2 \pm 5.9$  minutes, respectively. Clutch size averaged  $123.1 \pm 10.5$  for 2009 and  $124.3 \pm 18.6$  for 2010, which is 10 to 20% more than the global clutch size of 100–110 eggs, and appears to have resulted in a longer OD than previously assumed. Using the average OD from this study,  $172,407 \pm 7509$  and  $134,478 \pm 6204$  turtles were estimated to have nested during 2009 and 2010 respectively, contrasting the 200,000–250,000 turtles reported by the Forest Department. Our results suggest that a minimum of 150 egg laying turtles across each night of the mass nesting period is required to be observed for an accurate quantification of OD.

*Key words:* arribada, clutch size, *Lepidochelys olivacea*, olive ridley turtle, oviposition duration, population estimation

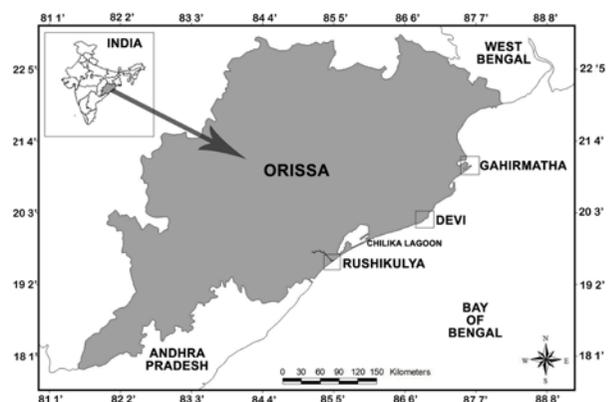
## INTRODUCTION

Population assessments of sea turtles are mainly made at nesting beaches (Meylan, 1982; Schroeder & Murphy, 1999), and are age- or stage-specific with only adult females counted. Female turtles emerge either solitarily or in small groups to nest, and direct counts of nesters or their nests are generally straightforward. However, it can be logistically challenging to identify each individual nesting female. Indirect methods such as counting tracks on beaches along with an assumed mean clutch frequency are generally used to estimate their actual numbers, although the latter aspect is associated with a high level of uncertainty. In very dense aggregations turtles can also obscure each other's tracks, and not all females necessarily lay eggs every time they emerge on a beach (Gates et al., 1996; Limpus et al., 2003; Swaminathan & John, 2010).

Apart from nesting solitarily, ridley turtles are characterized by synchronized mass nesting events known as *arribada* (Carr, 1967; Hughes & Richard, 1974). During an *arribada*, up to 100,000 females can nest on a short stretch of beach within a few days. Not all *arribada* turtles necessarily lay eggs, returning repeatedly to the beach until they do so. Therefore, generating accurate abundance estimates of nesting females during *arribada* events is generally difficult, as individuals are virtually

impossible to count, tracks are immediately obscured and nest numbers are not necessarily indicative of numbers of turtles (Meylan, 1982; Magnuson et al., 1990).

The olive ridley turtle *Lepidochelys olivacea* is found throughout the tropical waters of the world (Pritchard, 1997), and is currently listed as Vulnerable by the IUCN (Abreu-Grobois & Plotkin, 2008). It nests in large numbers at the Pacific coast of Costa Rica and Mexico, as well as along the east coast of India in the state of Orissa (Márquez et al., 1995; Pandav et al., 1997; Pritchard, 1997), where three mass nesting sites are known:



**Fig. 1.** Map showing the location of the three olive ridley turtle *arribada* beaches along the east coast of India.

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Gahirmatha, Devi and Rushikulya (Bustard, 1976; Kar, 1982; Pandav et al., 1994). Gahirmatha has been reported as the world's largest sea turtle rookery, and estimates of nesting turtles ranging between 100,000 and 800,000 (Patnaik et al., 2001). The reliability of these estimates has however been questioned (Tripathy, 2002; Shanker et al., 2004; Pandav & Shanker, 2001), and the census techniques are thought to have grossly overestimated turtle abundances (Shanker et al., 2006).

Different techniques to estimate turtles in an *arribada* include single strip transects (Hughes & Richard, 1974), aerial transects (Meylan, 1982), use of catch effort, tagging and direct census (Márquez et al., 1982), empirically modelling the distribution over time (Márquez & van Dissel, 1982), fixed quadrant sampling (Cornelius & Robinson, 1982), instantaneous count methods (Gates et al., 1996) and strip transects in time (Gates et al., 1996; Valverde & Gates, 1999). The latter has been recommended by the IUCN/SSC Marine Turtle Specialist Group (MTSG) for estimating turtles on mass nesting beaches (Eckert et al., 1999), and has been used at the Gahirmatha and Rushikulya rookeries with modifications (Shanker et al., 2003). Using this method, Shanker et al. (2004) arrived at an estimate of 180,000 turtles for the 1999 *arribada* at Gahirmatha, compared with an estimate of 300,000 turtles by the Orissa Forest Department. Similarly, Tripathy (2008) arrived at an estimate of 23,561 turtles for the 2004 *arribada* at Rushikulya, compared with 200,000 turtles reported by the Forest Department.

Here, we report on the importance of determining the average egg laying or oviposition duration (OD) and its significance in estimating the size of nesting populations. Valverde & Gates (1999) reported a value of 15 minutes as the OD for turtles nesting in Nancite, Costa Rica, which was used by Pandav & Choudhury (2000) for estimating the size of the *arribada* at Gahirmatha in 1999. For the same *arribada*, Shanker et al. (2004) used 13.5 minutes as the duration based on observations of 20 ovipositing turtles, whereas Tripathy (2008) used 14.5 and 13.5 minutes as the average OD for his estimates at Rushikulya. The variation in average OD lead us to believe that this may be a source of substantial error in

estimating the number of individuals during *arribadas* (see also Valverde & Gates 1999).

## MATERIALS AND METHODS

### Study site

The Rushikulya mass-nesting beach (19° 22' N; 85° 02' E; see Fig. 1) is 320 km south of the Gahirmatha turtle rookery in the state of Orissa. *Arribada* nesting at Rushikulya coincides with the dry season from November to May (Dash & Kar, 1990), whereas at eastern Pacific sites it occurs during the rainy season (Cornelius, 1986; Bernardo & Plotkin, 2007). *Arribada* nesting at Rushikulya occurs mainly along a 5 km beach stretch extending from the Rushikulya river mouth to the Kantiagada fishing village. Further nesting occurs on the dynamic sand-spits and sand bars formed at the river mouth. The mainland beach has a width of 80–100 m during the nesting season, and annual temperatures vary between 18°C and 40°C. Moderate to dense plantations of Casuarina (*Casuarina equisetifolia*) border the beach except near the river mouth. Fishermen from two villages (Purnabandha and Gokurkhudha) use parts of the beach for boat landing and other fishery-related activities, and a temporary suspension of these activities is imposed during an *arribada* event.

Six observers stationed at different parts of the beach to record nesting turtles made observations on the OD during the *arribadas* of 2009 and 2010, using a digital watch to record egg-laying duration. Turtles that were near to completion of nest digging were chosen for observation. The observers widened the entrance to the nest immediately behind the carapace and placed one hand into the nest and below the cloaca, so that eggs dropped into their hands. The time between the release of the first egg and the turtle beginning to push the sand into the nest with the hind flippers was recorded as the OD, simultaneously with counting the eggs laid. Non-parametric comparisons of means (Mann-Whitney U-test and Kruskal-Wallis *H*) were carried out to assess the variation in OD and clutch size among years and among *arribada* days. In order to predict the minimum number of ovipositing turtles required for estimating

**Table 1.** Average OD±SD and percent coefficient of variation (% CV) for the 2009 and 2010 mass nesters recorded at Rushikulya, southern Orissa. Range and sample size given in brackets.

Date	Average OD in minutes	% CV	Date	Average OD in minutes	% CV
15.02.09	22.3±7.9 (11–39, 46)	35.4	15.03.10	15.8±4.7 (9–44, 71)	29.7
16.02.09	18.8±5.8 (10–34, 35)	30.9	16.03.10	18.1±6.2 (8–48, 122)	34.3
17.02.09	19.7±4.7 (7–32, 54)	23.9	17.03.10	16.7±6.8 (8–37, 25)	40.7
18.02.09	18.3±3.4 (11–28, 47)	18.6	18.03.10	16.8±4.3 (10–26, 39)	25.6
All days, mass nesters	19.8±5.8 (7–39, 182)	29.5		17.2±5.9 (8–48, 263)	34.2

**Table 2.** Average clutch size $\pm$ SD and percent coefficient of variation (% CV) for the 2009 and 2010 mass nesters recorded at Rushikulya, southern Orissa. Range and sample size given in brackets.

Date	Average clutch size	% CV	Date	Average clutch size	% CV
15.02.09	122.1 $\pm$ 11.1 (98–146, 46)	9.1	15.03.10	120.5 $\pm$ 17.9 (87–152, 71)	14.9
16.02.09	123.2 $\pm$ 9.8 (92–142, 35)	8.0	16.03.10	128.4 $\pm$ 17.7 (89–168, 122)	13.8
17.02.09	122.7 $\pm$ 12.0 (87–153, 54)	9.8	17.03.10	125.8 $\pm$ 20.1 (91–158, 25)	16.0
18.02.09	124.6 $\pm$ 7.7 (109–143, 39)	6.2	18.03.10	117.7 $\pm$ 20.0 (74–158, 39)	17.0
All days, mass nesters	123.1 $\pm$ 10.5 (87–153, 174)	8.5		124.3 $\pm$ 18.6 (74–168, 263)	15.0

the average OD for the two years, a simulated random sampling from the entire data set was conducted by systematically varying sample size from 10 to 200 nests. The average OD for each sample size estimated was based on 5000 randomizations conducted in R software (R version 2.14.0, 2011). All other analyses were carried out with SPSS version 16.0.

## RESULTS

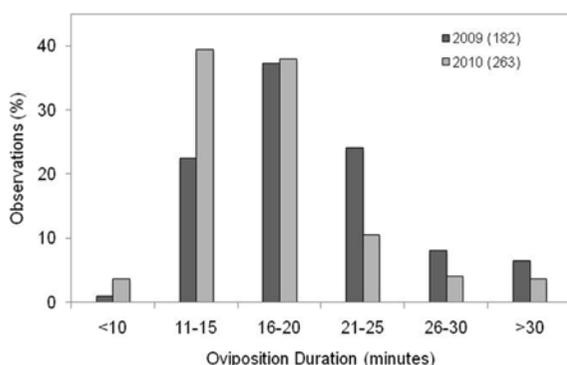
The 2009 *arribada* at Rushikulya started on 15<sup>th</sup> February and continued for five days with turtles nesting from 1900 hours through to 0700 hours. In 2010, the *arribada* started on the 15<sup>th</sup> of March and again continued for a period of five days. A total of 13,716 nesting turtles were counted within the sampling blocks during the 2009 *arribada*, 182 of which were observed for the entire ovipositing duration. In 2010, a total of 7,862 nesting turtles were counted, 263 of which were observed for oviposition. The nesting population was estimated by the Forest Department to be 200,000 to 250,000 turtles.

The average OD of mass-nesters observed in 2009 and 2010 was 19.8 and 17.2 minutes, respectively (Table 1), and were significantly different across years ( $U=-5.564$ ,  $p<0.05$ ). The OD recorded for all the turtles during the

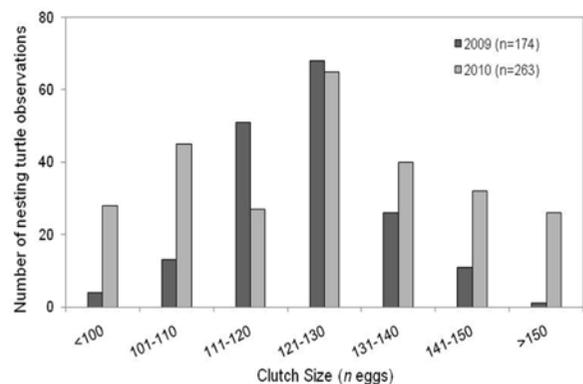
two years ranged between 7 and 48 minutes (Fig. 2). There was no significant difference in OD across the four days of the 2009 and 2010 *arribadas* (2009:  $H=6.923$ ,  $df=3$ ,  $p=0.074$ ; 2010:  $H=6.156$ ,  $df=3$ ,  $p=0.104$ ; Table 1).

The average clutch size was 123.1 $\pm$ 10.5 and 124.3 $\pm$ 18.6 in 2009 and 2010, respectively (Table 2), without significant difference between years ( $U=-1.060$ ,  $p=0.289$ ). Clutch sizes during the two *arribadas* ranged between 74 and 168 eggs, with the majority of observations recording 121 to 130 eggs (2009:  $n=68$ , 2010:  $n=65$ ; Fig. 3). With a coefficient of variation of 15%, clutch size was considerably more variable in 2010 compared to 2009 (8.5%). Clutch size was not significantly different across days in 2009 ( $H=0.871$ ,  $df=3$ ,  $p=0.832$ ), while in 2010 the differences was found to be significant ( $H=12.403$ ,  $df=3$ ,  $p<0.05$ ; Fig. 4).

The 13,716 and 7,862 egg laying turtles were counted at hourly intervals from 1800 to 0600 hours within sampling blocks along the entire nesting beach and during four days of the 2009 and 2010 *arribadas*. Based on this data and using an overall OD of 19.8 min, a total of 174,149 $\pm$ 7,583 turtles were estimated to have nested during 2009; using an overall OD of 17.2 min, a total of 132,914 $\pm$ 6,133 turtles were estimated for 2010. Average OD strongly influenced the estimated *arribada*



**Fig. 2.** Frequency of observations on OD showing a significantly larger number of turtles taking 16 to 20 minutes to oviposit during the 2009 and 2010 *arribada* at Rushikulya.



**Fig. 3.** Frequency of observations of nesting turtles laying a significantly higher number of eggs in the range of 121 to 130 eggs during the 2009 and 2010 *arribada* at Rushikulya.

**Table 3.** Variability in the olive ridley turtle *arribada* population estimates at Rushikulya for 2004, 2005 and the study years estimated assuming different oviposition durations (OD). Figures in *italics* are estimates obtained using the average OD recorded in the present study.

Parameters	2004*	2005*	2009 <sup>#</sup>	2010
Total area of nesting (m <sup>2</sup> )*	150,000	200,000	300,000	186,100
Duration of <i>arribada</i> (minutes) <sup>#</sup>	780	780	780	780
Total number of egg laying turtles counted during the transects	1,144	3,908	13,716	7,862
Width of the transects (m)	20	20	20	20
Number of sampling periods	13	13	13	13
Total length of transects (m)	1,500	2,000	3,580	1,920
20 min OD	17,160	58,620	<i>172,407</i>	114,306
19 min OD	18,063	61,705	181,481	120,322
18 min OD	19,067	65,133	191,564	127,007
17 min OD	20,188	68,964	202,832	<i>134,478</i>
16 min OD	21,450	73,275	215,509	142,883
15 min OD	22,880	78,160	229,877	152,408
13.5 min OD	25,422	86,844	255,419	169,342

\* Except for the 2010 *arribada* the total nesting area (m<sup>2</sup>) reported here for others is a simple measure of the length and average width of the nesting beach. For 2010, the area of the nesting beach was delineated accurately using a GPS.

# The 780 minute durations reported across years here is the fixed time period (13 one hour sessions starting from 1800 to 0600 hours), when the nesting turtle counts are made during each day of the *arribada*.

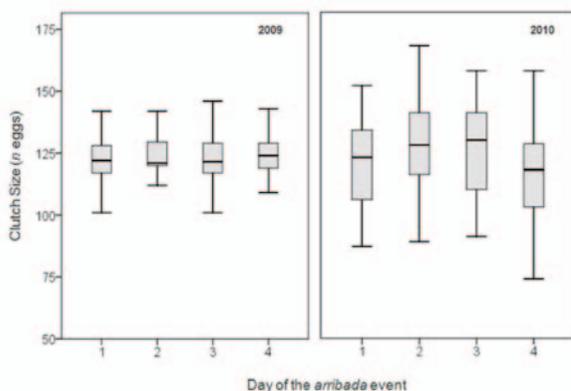
population (Table 3). When the average OD recorded for each day of the 2009 and 2010 *arribadas* was used, the combined population estimates were 172,729±7,541 and 136,949±6,288, respectively (Table 4). Simulated random sampling of the entire datasets over the two years showed an increasing precision as sample size increased, stabilising when sample size reached about 150 turtles for both *arribadas* (Fig. 5).

## DISCUSSION

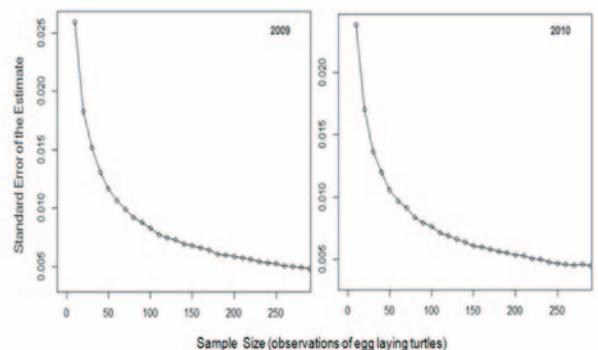
The results of this study clearly demonstrate erroneous *arribada* population size estimates when using an incorrect OD. The average OD for 2009 and 2010 at Rushikulya was at least 4–5 minutes longer than the 13–14 minutes used for population estimates in the past. If an OD of 13.5 minutes is used, an overestimate

in the range of 33% and 21% nesting turtles is revealed. Applying ODs of 19.8 min to the 2004 and 2005 *arribadas* (Tripathy, 2008) leads to an approximately 25% and 30% reduction in the estimated numbers of turtles (Table 3). Past *arribadas* were likely substantially smaller than estimated at Rushikulya and other turtle rookeries.

The average OD of olive ridley turtles at Rushikulya differed from eastern Pacific populations (Nancite beach: 15 min, Valverde & Gates 1999). Clutch size at Nancite and Ostional averaged 99.6±1.58 ( $n=115$ ) and 107.4±2.14 ( $n=66$ ), respectively (Cornelius et al., 1991), lower than in the present study. Previous studies on *arribada* nesters at the Rushikulya rookery also reported larger clutch sizes of 127.9±19.2 (Pandav & Choudhury, 2000) and 130.6±16.0 (Tripathy, 2005), consistent with our findings. Thus, the *arribada* nesters at Rushikulya appear to lay on average 10 to 20% more eggs than the average global



**Fig. 4.** Box plot (median and inter-quartile range) showing the variation in clutch size observed across each of the four days of the two *arribada* years



**Fig. 5.** The minimum number of ovipositing turtles ( $n=150$ ) required to be sampled during an *arribada* based on the standard error of the estimate of mean oviposition duration.

**Table 4.** Daily and combined estimates of the nesting turtle population  $\pm$  C.I. along with standard error and coefficient of variation for the two *arribada* years of the study

Year	Dates	OD (min)	Population Size Estimate (with 95% Confidence Intervals)	Standard Error	Coefficient of Variation
2009	15.02.2009	22.3	49,591 $\pm$ 2080	513.5	4.2 %
	16.02.2009	18.8	71,434 $\pm$ 3208	787.8	4.5 %
	17.02.2009	19.7	37,951 $\pm$ 1583	282.3	4.2 %
	18.02.2009	18.3	13,751 $\pm$ 670	75.3	4.9 %
	Combined		172,729 $\pm$ 7541		
2010	15.03.2010	15.8	65,518 $\pm$ 2662	350.6	4.06 %
	16.03.2010	18.1	40,774 $\pm$ 2007	309.9	4.92 %
	17.03.2010	16.7	21,051 $\pm$ 1136	122.5	5.39 %
	18.03.2010	16.8	9,606 $\pm$ 483	33.0	5.03 %
	Combined		136,949 $\pm$ 6288		

clutch size of 100–110 eggs (Pritchard & Plotkin, 1995; Miller, 1997), resulting in a longer OD.

The average OD of the two *arribadas* of this study differed by three minutes, which again might be a result of differences in clutch size, which in turn may be as a result of clutching behaviour. Olive ridleys lay more than one clutch of eggs in a reproductive season (Miller, 1997). In the 2010 *arribada*, 28% of turtles had a clutch size below 110 eggs, compared to 9% in 2009. Smaller clutch size may be due to an earlier nesting event. In 2010, the Rushikulya *arribada* took place after an earlier *arribada* at Gahirmatha (25 February–5 March).

The variation in OD observed across the nights of the *arribada* and across the two years of this study highlights the need for adequate sampling of ovipositing turtles for estimating nesting population size. We recommend a minimum of 150 egg laying turtles to be observed for OD during *arribada*. Future research could measure hormonal levels in turtles nesting at the rookeries along the Orissa coast in order to understand the reproductive cycle of this population, which is critical in the long-term monitoring and management of this threatened species.

## ACKNOWLEDGEMENTS

This work was carried out with support from the Sea Turtle Tracking Project of the Wildlife Institute of India, funded by the Directorate General of Hydrocarbon, Ministry of Petroleum & Natural Gas, Government of India. We thank Damburu, Madhusudan, Surendra, Shankar, Kedar, Sri Ramalu and many people from Puranbandha and Gokurkhudha for their untiring assistance during field work. We also acknowledge the co-operation and support provided by the Orissa State Forest Department during this work, especially DFO, Berhampur. The first author wishes to thank Barry Noon, Qamar Qureshi, Abishek Harihar and Mousumi Ghosh for helpful discussions and analyses of the data. Further, thank the two anonymous reviewers for their comments and help in improving the manuscript.

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Accepted: 19 August 2012