

ARTICLES



RESCUE AND POST-MORTEM IDENTIFICATION OF SEA TURTLES ON THE SOUTHWEST COAST OF INDIA

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INTRODUCTION

Sea turtles, as contemporary representatives of an ancient lineage of marine reptiles, offer unique insights into evolutionary history and adaptation. Their ecological roles as grazers, predators, and nutrient transporters make them keystone species vital to the structure and resilience of marine ecosystems (Hamann *et al.*, 2010). Globally, seven species of sea turtles are recognised, of which five occur in Indian waters: olive ridley (*Lepidochelys olivacea*), green (*Chelonia mydas*), hawksbill (*Eretmochelys imbricata*), loggerhead (*Caretta caretta*), and leatherback (*Dermochelys coriacea*) turtles (Shanker & Pilcher, 2003). Only three species nest regularly in India. Olive ridley turtles demonstrate synchronised mass nesting events (“arribadas”) in Odisha (Shanker *et al.*, 2004) and the Andaman and Nicobar Islands (Namboothri *et al.*, 2015) and display solitary and sporadic nesting along the coastline of Andhra Pradesh and other states (Tripathy *et al.*, 2003). Green turtles predominantly nest in the state of Gujarat and on some offshore islands (Venkatesan *et al.*, 2004; Sunderraj *et al.*, 2006). Leatherback turtle nesting occurs in the Andaman and Nicobar Islands (Andrews & Shanker, 2002; Swaminathan *et al.*, 2011).

All sea turtles face severe anthropogenic pressures, including bycatch, entanglement in abandoned fishing gear, coastal development, light and other pollution, vessel strikes, and ingestion of marine debris (Fuentes *et al.*, 2023). Turtles that have been injured or killed by any of these threats are often found floating at sea or washed ashore, alive or dead. Accurate species identification of dead turtles is important for effective monitoring and management. Morphological traits such as carapace shape, number of carapace scutes, and number of prefrontal scales are typically used for identification (Wyneken, 2001). However, advanced decomposition and/or external damage can obscure these features. In such cases, molecular tools like mitochondrial

cytochrome c oxidase subunit I (COI) barcoding can provide robust species identification and strengthen stranding datasets (Naro-Maciel *et al.*, 2010).

In this study, we report on dead sea turtles from the southwest coast of India and the rescue of a live turtle found entangled in abandoned, lost, or discarded fishing gear (ALDFG) associated with a fish aggregating device (FAD). We employed an integrated approach combining detailed morphometric analyses with DNA barcoding for species identification and confirmation. Furthermore, we document the epibionts on a green turtle and interpret these observations within both biological and ecological contexts.

METHODS

Field surveys were conducted along the southwest coast of India from 2024 to 2025 as part of a marine mammal and sea turtle monitoring programme. During this period, three sea turtle encounters were recorded. On 15th October 2024, two olive ridley turtles were encountered: one dead individual (9° 13.9' N, 76° 17.8' E) and one live individual entangled in ALDFG (9° 13.9' N, 76° 17.9' E). Additionally, one dead green turtle was encountered off Kochi (09° 58.1' N, 75° 59.2' E) on 6th May 2025 (Figure 1). Morphological identification for all specimens was based on species-specific diagnostic traits following Pritchard & Mortimer (1999). Standard morphometric measurements were collected from all individuals following the protocols of Bolten (1999). Tissue samples were collected from the front flippers of the dead turtles using sterile scalpels and preserved in 95% ethanol for subsequent DNA extraction and molecular species confirmation.

Genomic DNA was extracted using the DNeasy Blood and Tissue Kit (Qiagen, Germany; Cat. No. 69504) according to the manufacturer's instructions. The mitochondrial COI gene, a standard marker for species

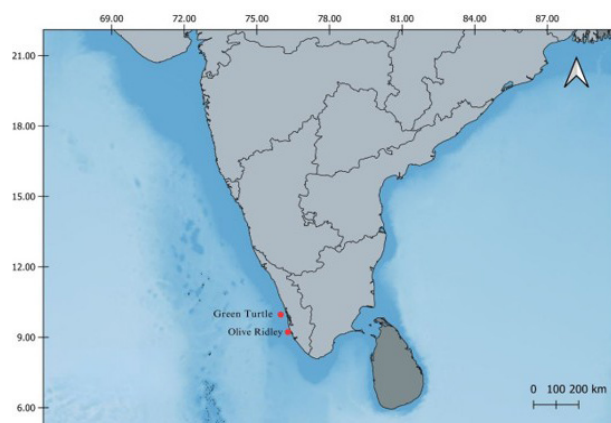


Figure 1. The Location of a Dead Green Turtle and Live and Dead Olive Ridley Turtles Examined in this Study.

identification, was amplified using primers: COI-F (5'-TCAACCAACCACAAAGACATTGGCAC-3') and COI-R (5'-TAGACTTCTGGGTGGCCAAAGAATCA-3') (Ward *et al.*, 2005). Each polymerase chain reaction (PCR) was performed in a 25 μ L Maxiamp PCR tube containing EmeraldAmp[®] GT PCR Master Mix (Takara Bio, Japan), molecular-grade water, the same primer pair, and template DNA. PCR amplification was performed on a SimpliAmp[™] thermal cycler (Applied Biosystems, USA) with standard barcoding cycling conditions. Amplified products were separated on a 1.5% agarose gel stained with ethidium bromide (EtBr) and visualised under ultraviolet illumination to verify amplicon size and integrity. Amplified products were purified and directly sequenced using the same primer pairs. The resulting sequences were compared against reference data in GenBank and the Barcode of Life Data Systems (BOLD) using BLASTn to confirm species identity. A maximum likelihood phylogenetic tree was constructed using the Tamura–Nei model with gamma-distributed rate variation (TN93+G). The freshwater turtle *Podocnemis unifilis* was included as an outgroup to validate tree topology.

RESULTS AND DISCUSSION

Morphological examination was used to identify the dead olive ridley and green turtles and the live olive ridley turtle found entangled in a FAD associated with ALDFG. The species of the dead turtle was also confirmed by molecular analysis. The dead green turtle exhibited external epibionts, notably *Chelonibia testudinaria*.

Morphological characteristics

The morphometric and morphological profiles of the three encountered sea turtles (Table 1) confirmed the species and indicated mature or immature life stages. The curved carapace length (CCL) and curved carapace width (CCW) for the olive ridley turtles fall within the documented sizes for adult populations along the Indian coast, where nesting and migratory adults typically exhibit a mean CCL of 60–70cm (Behera *et al.*, 2016). The size of the dead green turtle indicated a large sub-adult to emerging adult in the Indian Ocean where physiological maturity occurs between 85–90cm (Frazier, 1980; Wyneken, 2001).

Molecular species confirmation

DNA barcoding of the mitochondrial COI gene provided molecular confirmation of morphological species identification, with >99% similarity to reference sequences in GenBank and BOLD. The olive ridley and green turtle sequences were submitted to NCBI under accession numbers PV437215 and PX218923, respectively, and associated barnacle DNA sequences were also deposited (PX219660, PX226007). This congruence between morphology and genetics highlights the potential role for integrative approaches, particularly when decomposition or physical damage may obscure diagnostic traits (Naro-Maciel *et al.*, 2010).

Table 1. Morphometric data of live and dead sea turtles.

Curved Carapace Length (CCL), Curved Carapace Width (CCW), Total Body Length (TL), Head Length (HL), Curved Plastron Length (CPL), Curved Plastron Width (CPW), Right/Left (R/L). No data recorded (-)

Date	Turtle	Live/ Dead	CCL (cm)	CCW (cm)	CPL (cm)	CPW (cm)	TL (cm)	HL (cm)	Front	Back	# Vertebral Scutes	# Costal Scutes (R/L)	# Marginal Scutes
									Flipper Length (cm)	Flipper Length (cm)			
15 th Oct 2024	Olive Ridley	Dead	60.0	65.5	50.0	65.0	91.0	25.5	29.0	22.0	6	7/7	-
15 th Oct 2024	Olive Ridley	Live	64.0	62.0	-	-	-	28.0	34.0	-	8	-/7	-
6 th May 2025	Green	Dead	88.5	82.0	-	-	-	-	-	-	5	4/4	25–27

The maximum likelihood phylogenetic tree (Figure 2) constructed under Tamura–Nei model with gamma-distributed rate variation (TN93+G) resolved distinct, well-supported clades for each sea turtle species based on mitochondrial COI sequences. The sequences generated in this study (highlighted in blue) clustered within their respective species groups, confirming accurate molecular identification. *Chelonia mydas* samples grouped tightly with reference sequences (bootstrap=100), while *Lepidochelys olivacea* samples also aligned within their corresponding clades with strong support. The freshwater turtle *Podocnemis unifilis*, included as an outgroup, was clearly separated from marine turtle taxa, validating the tree topology.

Epibiont association and ecological insights

The dead green turtle exhibited a distinct macro-epibiotic settlement, characterised by the attachment of specialised turtle barnacles (*Chelonibia testudinaria*) across the carapace (Figure 4). *C. testudinaria* has been documented globally across all seven extant species of sea turtles, though its abundance patterns vary significantly by host species (Zardus & Hadfield, 2004). It is most heavily concentrated on loggerhead turtles (*Caretta caretta*), whose slower swimming speeds and rough carapaces provide an ideal settlement surface (Frick & Ross, 2001). Carapace colonisation by obligate commensals is widely documented in sea turtles, often serving as a biological indicator of prolonged pelagic residency or a decline in host health before mortality (Frick & Pfaller, 2013). While

C. testudinaria settlement is common among healthy foraging populations, excessive macro-epibiont loads can reflect a compromised physiological state. For instance, in loggerhead sea turtles, heavy barnacle infestations are highly prevalent in individuals afflicted by Debilitative Sea Turtle Syndrome (DSTS), where chronic lethargy reduces the host's natural swimming speed and grooming efficiency, thereby facilitating larval settlement (Baruffaldi *et al.*, 2023). In this study, the presence of these macroscopic bio-indicators on the green turtle carcass provides a critical ecological context. It suggests a period of reduced vitality or a prolonged, slow-moving trajectory, either actively migrating or passively drifting, within coastal or pelagic zones. This finding underscores how epibiotic profiling can enhance our understanding of sea turtle health baselines and mortality factors along the southwest coast of India.

Entanglement in ALDFG

The active entanglement event and subsequent rescue of the live olive ridley turtle documented in this study is an example of the threat posed by FADs and ALDFG to sea turtles in Indian coastal waters. Modern synthetic netting materials resist degradation and can persist as marine pollution for years to decades, resulting in prolonged "ghost fishing" dynamics that continually entrap non-target marine wildlife (Thomas *et al.*, 2023; Zainul *et al.*, 2025). Along the southwest coast of India, including states like Kerala, traditional artisanal fishers frequently deploy specialised, floating FADs to

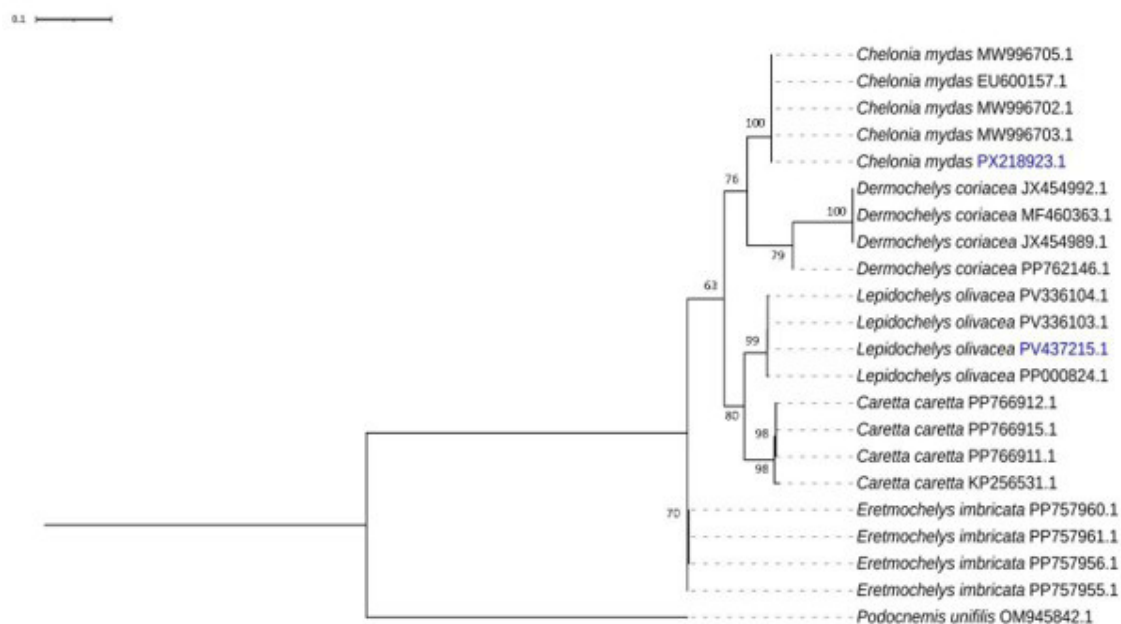


Figure 2. Maximum Likelihood Phylogenetic Tree of Study Sea Turtles Based on Mitochondrial COI Gene Sequences.

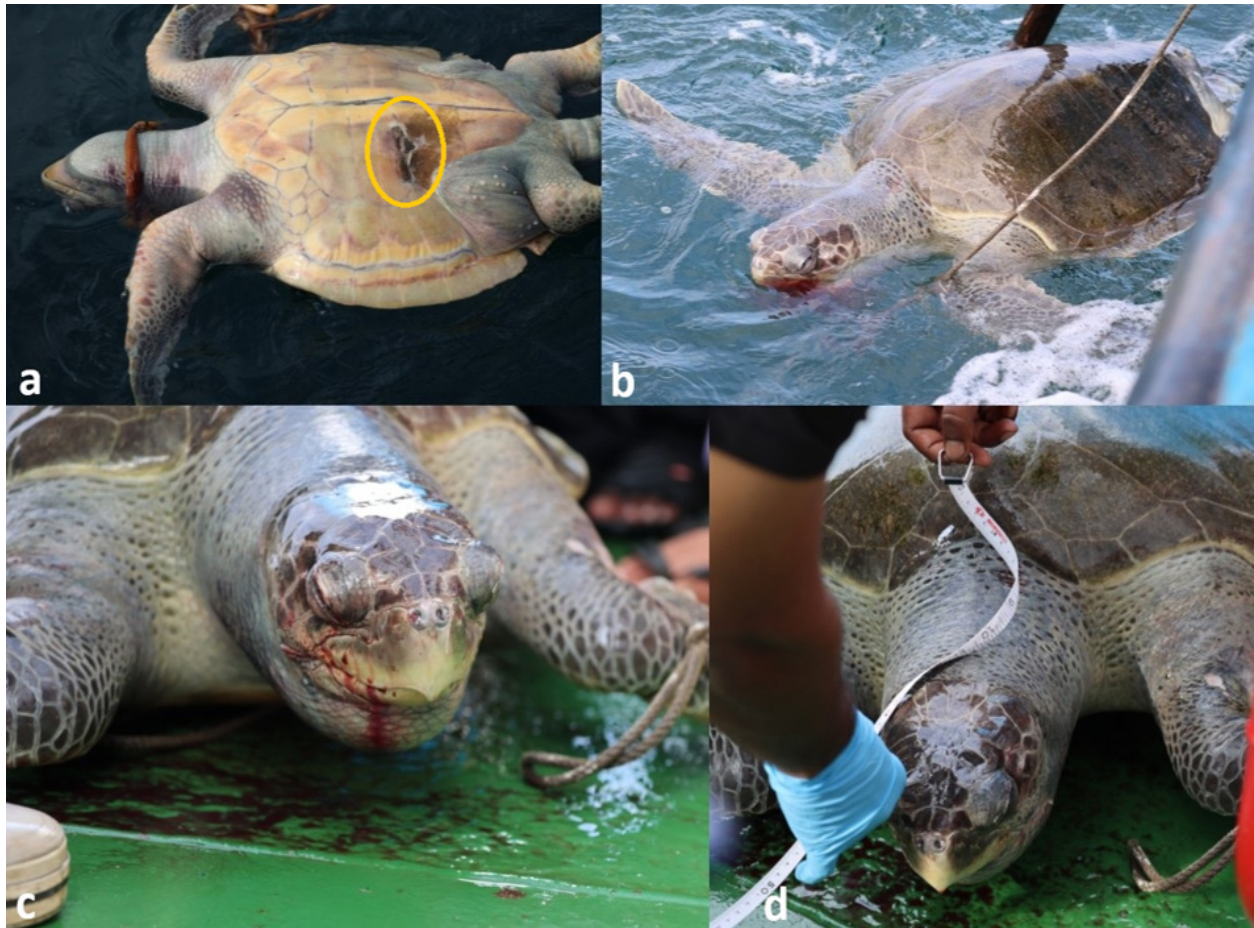


Figure 3. Dead Olive Ridley Turtle from the Southwestern Indian coast. a) Ventral View; Wound Outlined in Yellow. b) Floating Anterolateral View. c) Head, Dorsal Close-up. d) Measuring Turtle on Vessel Deck. (Photo credit: Vishnu C.B.)

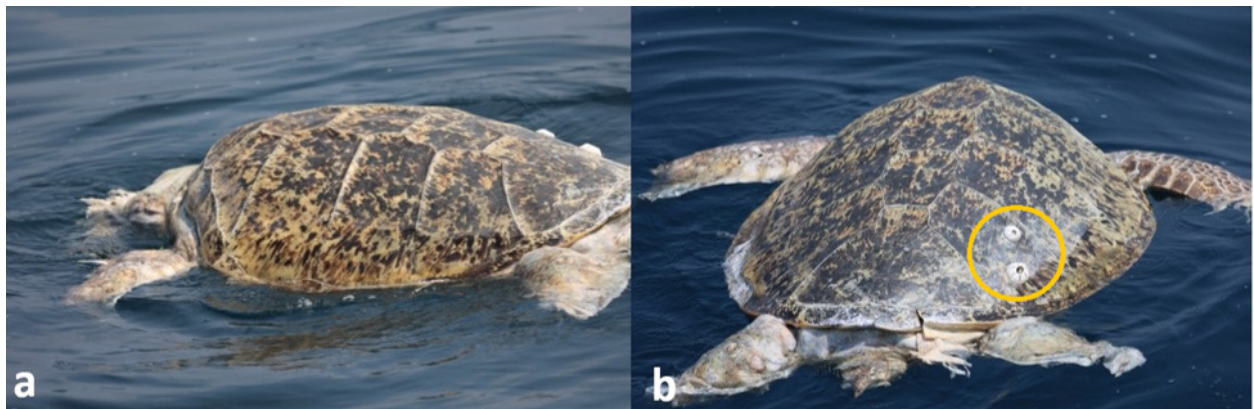


Figure 4. Dead Green Turtle from the Southwestern Indian coast. a) Lateral View of the Carapace. b) Posterolateral View with Two Macro-epibiotic Turtle Barnacles (*Chelonibia testudinaria*) Circled in Yellow. (Photo credit: Krishnapriya P.P.)

target cuttlefish. These configurations typically utilise inverted, biodegradable coconut spadices (fronds) to provide attractive, natural egg-laying substrates for cephalopods (Figure 5). However, to maximise efficiency, fishers often fix synthetic monofilament or gillnets directly around these organic structures. While

the core FAD components are biodegradable, these units can aggregate lost netting and synthetic marine debris over time. The combination of floating organic material and highly entangling synthetic mesh creates a cryptic, lethal hazard for marine megafauna; it is highly plausible that the live olive ridley rescued during this monitoring



Figure 5. Rescue of an Olive Ridley Turtle. a) Entangled in ALDFG Comprising Synthetic Netting, Structural Ropes, and Floating Plastic Bottles. b) Post-release Swimming Behaviour. c) FAD, the Coconut Spadices (Outlined in Yellow). (Photo credit: Zainul Abid P.M.)

period became entrapped within such a setup, a critical interaction that would have inevitably resulted in mortality without immediate human intervention. Managing ALDFG requires targeted, gear-specific conservation interventions and community-led ghost net mitigation frameworks.

CONCLUSION

This study documents dead olive ridley and green sea turtles along India's southwestern coast, using a reliable combination of morphometrics and morphological characteristics combined with DNA barcoding to confirm species identity. Incorporating these molecular methods makes regional wildlife monitoring more accurate when decomposition or physical injury makes it hard to identify a turtle species by its external features alone. Additionally, documenting entanglement in ALDFG highlights the human pressures these animals face in Indian waters. Our study demonstrates the information that could be collected by a long-term monitoring and stranding network on the Kerala coast.

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