Identifying critical marine habitats of the largest nesting population of loggerhead sea turtles in the Mediterranean: insights from stable isotope markers and satellite telemetry

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Introduction

Southern Kyparissia Bay (37.3399°N, 21.6952°E) on the west coast of the Peloponnese, Greece (Fig. 1), supports one of the largest nesting populations for loggerhead turtles in the Mediterranean (Casale et al. 2018). Monitoring of the core 9.5km of this rookery has been conducted since 1984 by ARCHELON (Margaritoulis & Rees 2001).

Traditionally, conservation of marine turtles in the Mediterranean Sea is targeted towards nests and nesting females on the easily accessible nesting grounds. This protects only a small proportion of the life cycle and does not protect critical marine habitats. Loggerhead turtles are known to migrate long distances across the Mediterranean Sea to several geographically distinct foraging grounds where they face anthropogenic threats. A key threat in the Mediterranean Sea is bycatch with approximately 44,000 deaths per year (Casale 2011). Therefore, protection of critical marine habitats is required and considered a research priority in Mediterranean marine turtle ecology.

To date, post-breeding migrations and foraging habitat use of loggerhead turtles from Zakynthos (Greece) have been extensively studied with more than 70 satellite tracks published in the scientific literature (Zbinden et al. 2008; Schofield et al. 2020). In contrast, there are no published tracks for turtles breeding at Kyparissia Bay and only four post-nesting migrations have been recorded. The profound lack of knowledge of the critical marine habitats of the Kyparissia Bay population is precluding the formation of evidence-based marine protection and management measures. The level of bycatch varies across the Mediterranean, so individuals using different regions are likely to differ in survivability.

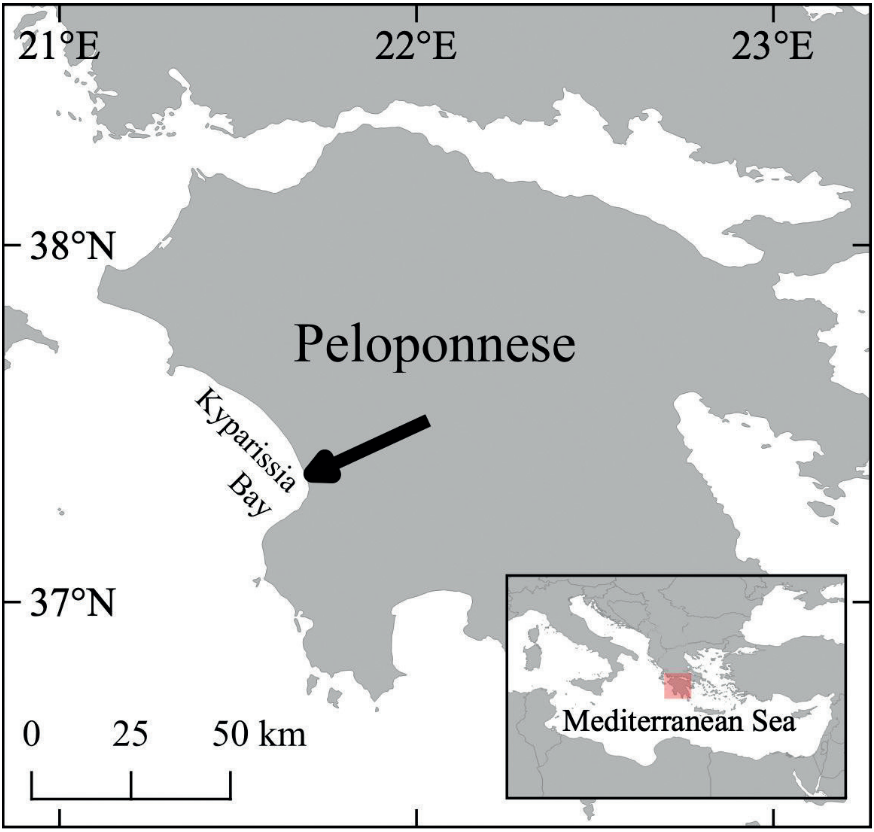

Fig. 1. Location of southern Kyparissia Bay in Greece.

To identify post-breeding movements and habitat use of marine turtles, satellite tracking is commonly used, though this is usually limited to a few individuals due to the expense of satellite tag purchase and remote monitoring (Godley et al. 2008). Stable isotope analysis (SIA) is a powerful tool helping to explain the foraging and spatial ecology of marine turtles globally (Haywood et al. 2019). Baseline isotope gradients occur naturally throughout the oceans due to variations in nutrient cycling (DeNiro & Epstein 1978). The geographical location where food was ingested by a marine turtle relates to these baseline isotopes within the environment and are in turn reflected as isotope signatures in the individual (DeNiro & Epstein 1978). The isotope signatures within a tissue sample from a marine turtle can be measured using SIA and in turn used as geographic markers. Complementing satellite tracking with isotope signatures has enabled the number of females assigned to foraging grounds using satellite tracking to be scaled up for other loggerhead turtle populations within the Mediterranean (e.g. Zbinden et al. 2011; Haywood et al. 2020) and globally (e.g. Ceriani et al. 2017 in the Atlantic), better representing the critical marine habitats at a population scale.

The aim of this study was to determine if we can use isotope signatures to characterise foraging regions and upscale results from tracked turtles to achieve population-level inferences for the largest loggerhead turtle rookery in Greece, Kyparissia Bay.

Methods

To locate turtles, we conducted patrols along sections of the core nesting area between 2300 and 0300 hr in June of 2018 and 2019. We selected turtles for study on completion of a nesting emergence, confirmed by observation of egg laying.

We applied uniquely coded metal tags (National Band and Tag Co., Newport, Kentucky, USA) to both front flippers of un-tagged turtles and recorded any existing tags to ensure that individual identities were assigned. We recorded straight carapace length from nuchal notch to tip of the longest supracaudal scute (SCLn-t; Bolten 1999) of each turtle to the nearest 0.5cm. We collected small (<0.25cm^2) skin tissue samples using a hole punch from the trailing edge of either hind flipper (Fig. 2) from 100 turtles (49 in 2018 and 51 in 2019). Skin tissue samples were stored in 90% ethanol at room temperature until they underwent carbon, nitrogen, and sulphur stable isotope analysis (for details of laboratory methods see Haywood et al. 2020).

We attached Platform Transmitter Terminals (PTTs; Model SPOT-375; Wildlife Computers, Redmond, Washington, USA) to a subset of flipper tagged and sampled loggerhead turtles, to track them using the Argos system (www.argos-system.org). We deployed nine location-only PTTs in 2018 and 12 similar PTTs in 2019. We manoeuvred turtles into a large plastic box to retain them on the beach during PTT deployment (for approximately two hours). We attached the PTT to the carapace of the turtle, centred over the second vertebral scute, at the start of the nesting season, using the attachment kit of Wildlife Computers and their recommended methods (www.wildlifecomputers.com) (Fig. 3).

To determine if we can use isotope signatures to characterise foraging regions, Kyparissia Bay satellite tracked females were assigned to one of the foraging regions identified as isotopically distinct by Haywood et al. (2020) including the Adriatic Sea, offshore Tunisian Plateau, and the eastern Mediterranean; or as pelagic foragers when a distinct foraging ground was not occupied and foraging occurred in waters over 200m in depth. To determine if isotope signatures significantly differed among the foraging grounds an Analysis of Covariance was run taking turtle size into account.

Fig. 2. Nesting loggerhead turtle having a skin tissue sample taken from a fore flipper, photographed during a similar research project. Photo by Olkan Erguler.

Fig. 3. Loggerhead turtle in Kyparissia Bay with a PTT attached after nesting.
Photo by ALan Rees / ARCHELON.

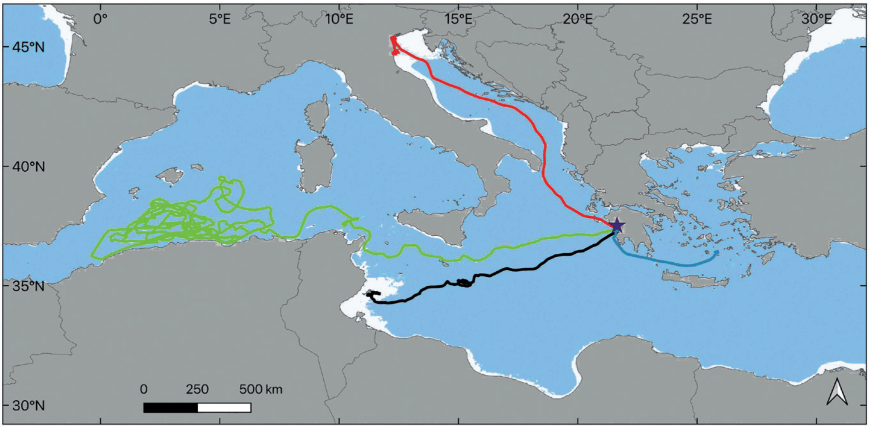

Fig. 4. Examples of post nesting movements of loggerhead turtles from Kyparissia Bay (star) identified using satellite tracking. All tracks are truncated to a one-year duration. Straight portions of the tracks are assumed to be migrations and more convoluted portions are assumed foraging locations/periods. Water less than 50m deep, where turtles are likely to forage on benthic prey, is indicated in white. Black track = Offshore but assumed benthic foraging on the Tunisian plateau with a period of epipelagic (surface water) foraging en route; Red = Direct migration to shallow coastal foraging habitats of the northern Adriatic Sea; Blue = Direct migration to shallow coastal foraging habitat bordering Anafi Island within the Aegean Sea; Green = Directed migration westwards with some epipelagic foraging assumed followed by persistent epipelagic foraging in the western Mediterranean in water depths >200m, indicated by the turtle never settling in one localised foraging spot.

Results

The satellite tracking showed that females from Kyparissia Bay occupied foraging grounds in the Adriatic Sea, Aegean Sea, Tunisian Plateau, and the coast of Tunisia, whilst eight females were identified as pelagic foragers (example routes depicted in Fig. 4). One satellite tracking device stopped transmitting during the nesting season and therefore was not included in the analysis.

Isotope signatures of tracked and untracked females range between -18.94 and -9.82‰ for carbon (mean = -15.71, SD =1.60), 5.83 and 15.91‰ (mean = 9.58, SD = 2.12) for nitrogen, and 9.91 and 20.62‰ for sulphur (mean = 16.87, SD = 2.40; Fig. 5).

Isotope signatures of tracked females did not differ among foraging grounds for carbon ($F(_{3,15})$ = 3.20, p = 0.05) or sulphur ($F(_{3,15})$ = 1.03, p = 0.41), but did differ for nitrogen ($F(_{3,15})$ = 5.75, p = 0.008) due to those foraging in the Adriatic Sea having significantly higher nitrogen values than the Tunisian Plateau foragers (p = 0.01).

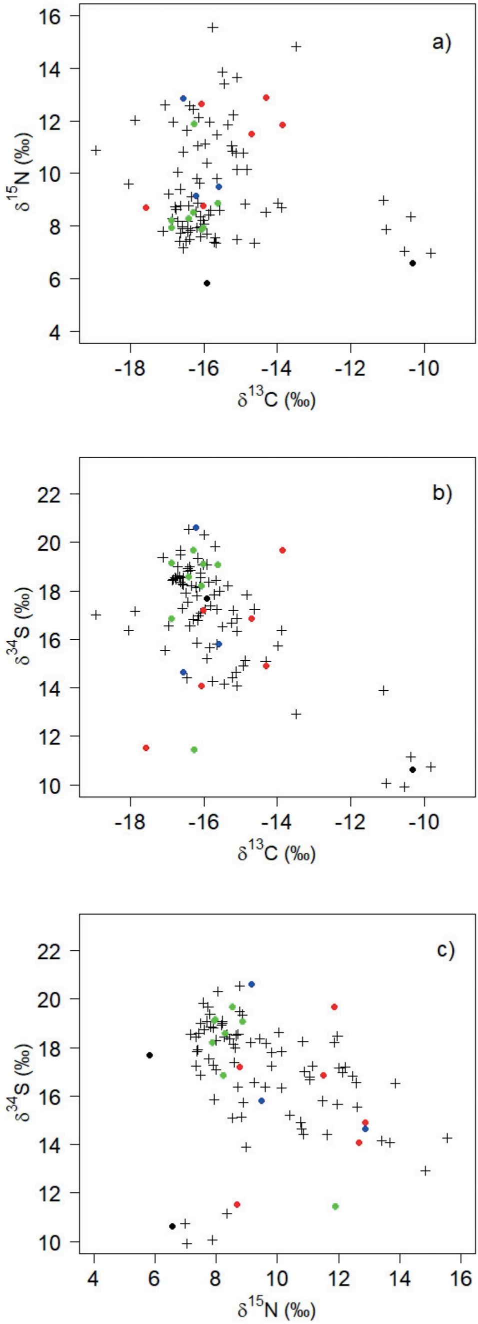

Fig. 5. Bivariate plot of
a) $\delta^{13}C$ and $\delta^{15}N$,
b) $\delta^{13}C$ and $\delta^{34}S$, and
c) $\delta^{15}N$ and $\delta^{34}S$
isotope signatures of untracked (black crosses) and tracked females sampled in Kyparrisia.

Adriatic Sea = red
Tunisian Plateau = black
Eastern Mediterranean = blue
Pelagic foragers = green

Discussion

Tracked females went to similar regions previously identified as important loggerhead turtle foraging grounds, including the Adriatic Sea and the Tunisian Plateau; however, a large proportion of the satellite-tracked females performed pelagic foraging, the extent of which has not been previously recorded in the Mediterranean. Loggerhead turtles are opportunists, likely to forage on invertebrates (for example, crustaceans and molluscs) as well as fish and sponges in neritic habitats, whilst in pelagic habitats foraging on gelatinous prey, such as, jellyfish and tunicates (Bjorndal 1997).

Isotope ratios were in a similar range to those identified by Zbinden et al. (2011) and Haywood et al. (2020) for those foraging in the Adriatic Sea and Tunisian Plateau, suggesting similar trophic levels and foraging grounds are being utilized in these regions. However, Kyparissia Bay females had few individuals with isotope ratios matching the eastern Mediterranean (low nitrogen and high carbon values), suggesting limited numbers migrate to foraging grounds in the eastern Mediterranean. This is supported by previous tracking (e.g. Hays et al. 2014) and isotope studies (e.g. Zbinden et al. 2011) from Greece.

The isotope ratios did not significantly differ between previously identified foraging regions. This may be due to a more dynamic foraging strategy with a more generalist diet used by Kyparissia Bay females, which is supported by the high number of pelagic foragers not previously seen before for Mediterranean nesting females. The lack of isotopically distinct foraging regions in this nesting population prevented untracked females being assigned to putative foraging grounds. Additional analytical techniques such as amino acid compound-specific stable isotope analysis (e.g. Seminoff et al. 2012; Vander Zanden et al. 2013) or analysis of other intrinsic markers, for example trace elements (e.g. Ramirez et al. 2019), may help distinguish foraging grounds and enable untracked females to be assigned to foraging grounds.

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