



Combining UAVs and multi-sensor dataloggers to estimate fine-scale sea turtle density at foraging areas: a case study in the central Mediterranean

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ABSTRACT: Knowledge of the distribution and density of marine species is key to understanding habitat use and interactions with human activities. Yet such information for sea turtles remains scarce, especially at foraging areas, where low turtle density represents an additional challenge in comparison to turtle aggregations at coastal breeding areas. Aerial surveys with aircraft are an efficient method for collecting these data over broad scales, while more novel unoccupied aerial vehicles (UAVs) are better suited for finer-scale data collection. However, their use is less developed, especially in offshore areas. Here we explored, for the first time in the Mediterranean, the potential of UAV surveys to estimate turtle density (surface and total) at foraging areas and its temporal trend. Between 2017 and 2023, we conducted 427 flights in the Pelagian Islands Archipelago (PIA), Italy, a regionally important foraging area of the loggerhead sea turtle *Caretta caretta*. To convert from surface to total density, we used data from multisensor biologgers deployed on 22 turtles to calculate the proportion of time turtles are visible from aerial surveys (availability time proportion, ATP). Results show that the mean surface turtle density in the PIA (0.336–0.477 turtles km⁻²) is among the highest reported globally for a loggerhead turtle foraging area. Such densities make it possible to assess population trends through periodic UAV surveys, which are less expensive than aircraft surveys and which can minimize the typical biases of aerial surveys (distance sampling, perception, and misidentification). A standardized methodology is needed for meaningful comparisons, including ATP at the visible depth layer vs. surface.

KEY WORDS: Drones · Loggerhead turtle · *Caretta caretta* · Aerial surveys · Availability bias · Datalogger

1. INTRODUCTION

Generating data on the spatial distribution and population trends of marine species can be challenging, yet it is necessary to understand habitat use (Houstin et al. 2022), identify potential areas of overlap with human activities (Awbery et al. 2022), and inform conservation and management efforts (Bovery & Wyneken 2015). Given the logistical challenges associated with observing marine animals offshore,

indirect approaches like satellite tracking (Godley et al. 2008), oceanographic features (i.e. highly productive areas as fronts; Scales et al. 2014), and bycatch rates (Lewison et al. 2014) are often used as proxies for abundance and distribution. Nevertheless, direct observation, typically via boat or aircraft surveys, represents the most robust approach to obtain such data (Williams & Thomas 2007, Gannier & Epinat 2008, Lauriano et al. 2011, Panigada et al. 2017). Aerial surveys by aircraft or helicopters are efficient, especially

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in remote areas, and have been widely used to study marine megafauna such as dolphins (Slooten et al. 2004, Durden et al. 2017), manatees (Langtimm et al. 2011, Alves et al. 2016), dugongs (Holley et al. 2006), sharks (Rowat et al. 2009, Westgate et al. 2014), seals (Furst et al. 2023), seabirds (Certain & Bretagnolle 2008), porpoises (Hammond et al. 2002, Gilles et al. 2009), whales (Andriolo et al. 2006), and sea turtles (Gómez de Segura et al. 2006, Fuentes et al. 2015). However, there is an inherent bias due to the failure to count animals that are present but not visible at the surface (termed availability bias; Marsh & Sinclair 1989a). Marine megafauna sightings are usually largely affected by this error, which can be influenced by environmental conditions (e.g. water turbidity), survey type, and animal diving patterns (Marsh & Sinclair 1989a,b, Pollock et al. 2006). Estimating total density (throughout the entire water column) from observed (surface) density therefore requires a correction factor that can account for the proportion of time that animals spend at visible depths (i.e. detection layer; e.g. Pollock et al. 2006, Fuentes et al. 2015).

Being able to fly at high altitude and speed over largely spaced (e.g. 10–15 km spacing; Pierantonio et al. 2023) and wide transect strips (e.g. up to 400 m; DiMatteo et al. 2022), aerial surveys by aircraft or helicopters are generally used for large areas and are less suited to fine-scale surveys (10s of km wide or less), but they can still sometimes be used for this purpose. However, to obtain an adequate sample size in areas where animals occur at low densities, multiple surveys over the same transects would be needed, which is logistically challenging for aerial surveys by these occupied vehicles. They typically require several operators, specialized crew (pilots), airfield availability, and expensive fuel, causing aerial crewed surveys to be costly (i.e. >1000 USD per survey hour; Colefax et al. 2018), making multiple surveys unlikely for intra- or inter-annual monitoring.

Compared to the limitations associated with aircraft/helicopter surveys, unoccupied aerial vehicles (UAVs) have several advantages, such as being logistically simpler, and having higher operator safety, repeatability, and lower operational costs (Verfuss et al. 2019). UAVs can increase data accuracy and quality through their automated features to follow pre-designed transects, collect videos and images at high (frame) rates and resolution, low altitude and perpendicular perspective, potentially viewable countless times (Rees et al. 2018, Schofield et al. 2019, Yaney-Keller et al. 2021). In addition, with multiple surveys on closely spaced transects conducted on the same small area (a few km wide), UAVs can provide a high

number of sightings, making it possible to provide estimates of surface density at a fine scale (i.e. with high spatial resolution within a small area) as well as spatial or temporal comparisons. In particular, UAV surveys would make it possible to investigate temporal trends (intra- and inter-annual), in contrast to aircraft-based surveys, where repeated surveys (if any) are carried out after several years due to the aforementioned logistical and financial constraints. While coarse-scale density estimates (such as those obtained by aircraft-based surveys) might not accurately reflect the conditions throughout the entire area considered, fine-scale estimates (such as those obtained by UAV-based surveys) might not capture the general patterns and optimal habitats for the studied animals. Therefore, integrating data derived from diverse spatial scales may provide a more comprehensive understanding of the status of animal populations. However, while coarse surveys by aircraft have been widely used so far (see above), fine-scale aerial surveys by UAVs are relatively recent (Rees et al. 2018).

UAVs also have a lower environmental footprint than aircraft and boats, cause minimal or no disturbance to marine megafauna (Bevan et al. 2015, 2018, Christiansen et al. 2016), are allowed to fly at low altitude for better visibility, and have greater freedom of movement during flight (e.g. sudden course changes to approach sighted animals). Moreover, the vertical view removes the bias due to reduced detection as the distance of the subject from the observer increases (distance sampling bias), typical of aircraft-based surveys (with the exception of aircraft equipped with glass bottoms/floors), and reviewing videos/photos multiple times minimizes misclassification (false positives; Aniceto et al. 2018) and perception (false negatives) biases (Hodgson et al. 2013, 2018). Like crewed aircraft, UAVs can also be equipped with specific instruments such as RGB cameras with variable resolutions and near-infrared and infrared cameras, which can detect animals in poor visibility conditions (e.g. during night, among vegetation; Gooday et al. 2018, Whitworth et al. 2022, Román et al. 2023). Drawbacks are represented by the limited area covered by UAVs (due to battery limits and airspace regulations), the need for optimal weather conditions (e.g. less resistance to gusts and strong winds than aircraft), and the need for time-consuming post-processing of videos (Colefax et al. 2018).

Most of the current information on sea turtle distribution is limited to fractions of each population or derived from data opportunistically collected and potentially biased. Typically, sea turtles are easier to observe at breeding or nesting sites (Robinson et al.

2023), either as counts of egg clutches laid by adult females at nesting beaches (the most common index of sea turtle population abundance and distribution) or as breeding adults aggregating at nearby marine areas. Not surprisingly, UAVs have been used for a variety of sea turtle studies at breeding sites (e.g. Schofield et al. 2017, Yaney-Keller et al. 2021, Sellés-Ríos et al. 2022, Staines et al. 2022) or for stranding events at beaches (Escobar-Flores & Sandoval 2021). Unfortunately, adults only constitute a fraction of the population (e.g. Casale & Heppell 2016), while juveniles, representing the bulk of the population, can only be found in foraging areas at significantly lower densities. In these areas, sea turtle distribution or density was estimated by UAVs only in a few cases (Robinson 2020, Odzer et al. 2022), while offshore areas were reached with a maximum of 3 km from the coast (Sykora-Bodie et al. 2017, Gray et al. 2019). Limitations for this approach may include the distance from the coast, the need to launch UAVs from vessels, and the generally lower turtle density expected to be lower at foraging areas than at breeding sites, which are frequented by a higher number of individuals during the mating season (Dickson et al. 2021, 2022).

In contrast to the shallow waters generally found at breeding areas, foraging areas can be much deeper. In such conditions, turtles may only be visually spotted when they are near the surface. Hence, the counts of individuals observed through aerial surveys in offshore deep waters typically overlook those below visible depths. The associated availability bias arising from the missed sighted turtles and affecting the total density estimates is corrected by a correction factor usually provided by other studies using surface or depth sensors. However, data for estimating correction factors are usually limited to some areas, and their use assumes that the proportion of time spent at surface is spatially and temporally uniform, which is not the case (Thomson et al. 2012, Hochscheid 2014). Despite the necessity of applying appropriate correction factors to ensure the accuracy and comparability of densities derived from aerial surveys over time and across spatial areas, a standardized methodology that incorporates multi-layer correction is missing.

In the Mediterranean Sea, most information about sea turtle density and distribution at sea has been derived from satellite tracking (Stokes et al. 2015, Levy et al. 2017), capture-mark-recapture methods (Casale et al. 2007b, Baldi et al. 2023), and bycatch (Casale 2011, Casale et al. 2015) or stranding records (Tomás et al. 2008, Casale et al. 2010, Türkozan et al. 2013). Information from aerial surveys is limited to areas in the western and central Mediterranean Sea (Gómez de

Segura et al. 2003, 2006, Lauriano et al. 2011). Two studies have used aerial surveys to estimate turtle density at coarse scale (at basin and sub-basin levels; DiMatteo et al. 2022, Pierantonio et al. 2023, respectively). However, fine-scale in-water sea turtle densities and trends are lacking, especially for foraging areas where the generally expected lower density and logistical challenges have discouraged the use of UAVs so far.

The Tunisian continental shelf is one of the main neritic foraging areas for Mediterranean loggerhead turtles *Caretta caretta*. Turtles frequenting this area exhibit a wide size range (18.2–87 cm curved carapace length [CCL]; Casale et al. 2007a, 2016), opportunistically feeding on both epipelagic and benthic prey (Casale et al. 2008). Satellite tracking and genetic analyses have revealed strong connectivity with several rookeries in Greece, Cyprus, Italy, Turkey, and Libya (reviewed in Casale et al. 2018, Cerritelli et al. 2022). High levels of turtle bycatch also occur in this area (Casale et al. 2007a, Echwikhi et al. 2010, 2012, Cambiè et al. 2020).

The Pelagian Islands Archipelago (PIA; Italy, Fig. 1), located at the northeast edge of the Tunisian platform, consists of 3 islands: 2 on the continental shelf (Lampedusa and Lampione) and 1 in oceanic waters (Linosa). Offshore areas located west of Lampione and south of Lampedusa have been identified as a conservation hotspot due to high densities of sea turtles (Casale et al. 2013, 2020). In the PIA, there are 4 Natura 2000 sites (a European network of protected areas covering valuable and threatened species and

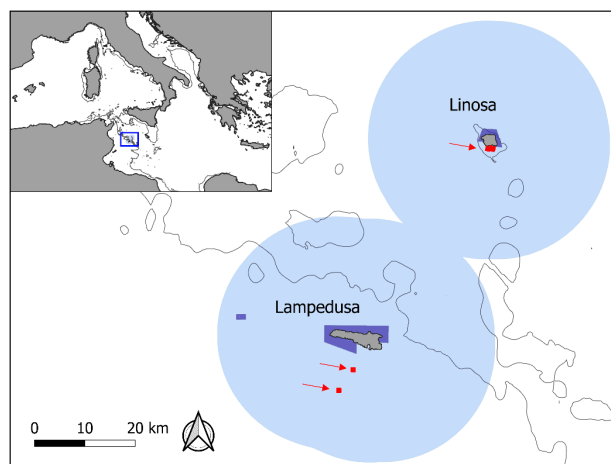


Fig. 1. Offshore (Lampedusa Island) and nearshore (Linosa Island) surveyed areas (red squares; indicated by red arrows) in the Pelagian Islands Archipelago (central Mediterranean Sea; blue square). Thin black line: 200 m bathymetry; light blue areas: ITA040013 Natura 2000 sites; dark blue areas: ITA040014 Natura 2000 sites

habitats) (ITA040001, ITA040002, ITA040013, ITA040014), and 2 of them (ITA040014 and especially the larger ITA040013) extend to offshore areas surrounding the islands, emphasizing the biological importance of this area and the adjacent waters.

The present study used UAVs to quantify the density and distribution of sea turtles in the PIA. Our specific objectives were to (1) estimate the surface density of loggerhead turtles in the PIA, (2) calculate correction factors to convert surface density into total density in the PIA, (3) initiate monitoring inter-annual changes in turtle density, and (4) validate the use of UAVs for assessing fine-scale spatio-temporal density variation of sea turtles at foraging areas.

2. MATERIALS AND METHODS

2.1. Study site

The PIA study area encompasses both neritic and oceanic regions. Although Lampedusa and Linosa islands are relatively close to each other (around 40 km apart), Lampedusa is part of the African continental plate (Tunisian shelf) and is surrounded by gently sloping seafloors that do not exceed 200 m in depth. In contrast, Linosa has a volcanic origin, and its seafloor plunges rapidly to depths of up to 1000 m.

2.2. Aerial surveys

UAV surveys were conducted in the PIA using a DJI Phantom 4 PRO (DJI, Shenzhen) flying at an altitude of 74 m, at a speed of 40 km h⁻¹, and recording videos with a 90° camera angle (nadir position) in 4K and 60 fps resolution. The flight altitude was empirically determined to achieve a desired swath width of 100 m at the sea surface. Surveys were conducted only when the Beaufort sea state (BSS) was <3, with wave height <0.5 m and wind speed <10 km h⁻¹. UAVs flew autonomously along transects designed and transmitted prior to each flight by the LITCHI app (<https://flylitchi.com/>, VC Technology). The flight operations were performed in visual line of sight (VLOS) and in compliance with flight regulations.

Two survey types (offshore and nearshore) were used to maximize the area covered during each flight and to address both the logistic issues related to the marine offshore environment and the coastal morphology of the nearshore areas. One survey type comprised offshore aerial surveys that were conducted in 2 arbitrarily chosen subareas, located at 5 km

(LMP5, take-off/landing central point: 35° 27' 26" N, 12° 34' 8" E) and 10 km (LMP10, take-off/landing central point: 35° 25' 15" N, 12° 32' 17" E) south of Lampedusa Island in 2017 and 2018 (Jul–Oct 2017; Jun–Sep 2018). In both areas, we used a squared (1 × 1 km) concentric flight path 10.6 km long, covering a 1 km² area (Fig. S1 in the Supplement at www.int-res.com/articles/suppl/n054p395_supp.pdf). Such a transect design minimized UAV distance from the pilot and facilitated fast UAV retrieval in the case of any problems or when battery level was low. UAVs took off and landed on a 5 m long motorboat, kept with the engine off during drone flight phase. In the same areas, a turtle-shaped decoy (Fuentes et al. 2015) was submerged 9 times at depths of 1–4 m and BSS 0–2 to assess the maximum depth of detection with the same flight settings as above. The other survey type comprised nearshore aerial surveys that were conducted in four 1 km² coastal areas of Linosa Island (take-off/landing points: 35° 51' 21" N, 12° 52' 6" E; 35° 51' 22" N, 12° 52' 11" E; 35° 51' 23" N, 12° 52' 16" E; 35° 51' 22" N, 12° 51' 35" E) between 2020 and 2023 (Aug–Nov 2020; May–Sep 2021; Jul–Oct 2022; Jul 2023). UAVs took off and landed on the coast. The same transect design (Fig. S2) was used for all areas, consisting of 11 strip lines arranged in a flag shape and covering an area of 1 km² over a total path length of 10.2 km.

For both offshore and nearshore surveys, only 1 area per day was surveyed with multiple consecutive flights (up to 10) over the same transect. Each flight constituted a single survey, providing 1 turtle density. Multiple flights over the same transects allowed us to obtain an adequate number of sightings to calculate an average density (representing the response variable) by avoiding potentially confounding factors such as the specific area. Since the surveyed area was 1 km², the resulting density was equivalent to the count, and the response variable was treated as a count for convenience. Multiple observations of the same individual across different flights did not affect the density estimate, which is an average of densities from each flight.

2.3. Surface turtle density

All turtle sightings were assumed to be loggerhead turtles due to the rarity of other species in the study area (Casale et al. 2018) and the high flight altitude complicating visual discrimination between species. Videos were reviewed on a 4K screen by different independent observers with the same video analysis experience. Methods for videos from Lampedusa and

Linosa differed slightly as follows. Two and 3 observers reviewed the Lampedusa and Linosa videos, respectively. To minimize perception bias (false negatives) in Lampedusa videos, a first observer watched all videos, labelling all turtles as C (certain; Fig. S3a) or U (uncertain; Fig. S3b). This step was repeated by the same observer, without checking data from previous views, until the total number of sightings (C+U) did not increase anymore (a stage reached with 3 views). Then, to minimize misidentification bias (false positives), a second observer watched only the sections of the videos including C and U sightings without knowing the categories assigned by the previous observer. A final classification in 3 categories N/C/U was achieved by assigning N (not a turtle) if the sighting was labelled N by the second observer, U if it was labelled U by at least 1 observer, and C if it was labelled C by both observers. To minimize perception bias (false negatives) in Linosa videos, 2 observers independently watched all videos just 1 time, labelling sightings as C or U. Then, to minimize misidentification bias (false positives), a third observer selectively watched and assigned all the sightings reported by previous observers to a final category (N/C/U). Two count values were considered as the minimum and maximum number of turtles observed: C and C+U.

A drawback of the above transects is that the same turtle can be counted again if it moves within the surveyed area fast enough to reach another transect line. To address this potential problem, a double counting correction (DCC) was performed as follows. For each sighting, the GPS position was derived by time, UAV speed, and flight path coordinates. Then 2 sightings in the same flight were considered as possible sightings of the same turtle if the hypothetical swimming speed (hss) needed to move between the 2 sighting points was lower than the theoretical maximum speed ($s_{\max} = 3.63 \text{ km h}^{-1}$; maximum speed observed in a group of 10 turtles tracked in the same area; Casale et al. 2012a). This comparison was performed for all pairwise combinations of turtles present in the same flight. Possible duplicate sightings (i.e. of the same turtle) were removed, leading to 2 additional corrected counts: corrected C+U (C_c+U_c) and corrected C (C_c). Mean surface density and 95% CI were estimated by the bootstrap method (no. of repetitions = 10 000; Puth et al. 2015) for each count (C, C+U, C_c , C_c+U_c). For Linosa, where data over 4 years were available, the same variables were estimated for each year to investigate interannual differences. The consistency over time (years) of the proportion of possible duplicate sightings in Linosa was investigated through a chi-squared test using the `chisq.test` R function.

To assess the possible effect of BSS and sun glare on the detection capacity of turtles through video analysis, a generalized linear mixed model (GLMM; `glmmTMB` R package; Brooks et al. 2017; binomial distribution) in R (R Core Team 2022; v. 4.1.3) was run on offshore data (Lampedusa; where environmental data were recorded) with the formula $Y \sim \text{BSS} + \text{SUN} + (1|\text{date: flight}) + (1|\text{date})$, where Y is the sighting category (being C as 1 and U as 0), SUN is the proportion of screen affected by sun glare in each sighting as a 3 level factor (0, 1–25%, 25–50%, converted to 0, 1, and 2, respectively), and BSS as a numeric variable with 3 values (0, 1, 2; see Hodgson et al. 2013). Flight and date were included as random effects, with flight nested within date.

2.4. Interannual changes

Annual differences in surface density were investigated in Linosa (where data from multiple years were available) for both C+U and C values, assuming no interannual difference in DCC (as supported by results) and therefore no need for DCC (and C_c+U_c and C_c) for interannual comparisons. A GLMM (R package `glmmTMB`; Brooks et al. 2017) with a Poisson distribution (log linked) was run in the form: $\text{counts} \sim \text{year} + (1|\text{month: date})$, where counts is the number of sightings, year is the year as a factor, and date nested into month (month as factor) are random factors to account for possible day and seasonal effects. A type II Wald test was performed using the `Anova` function ('`car`' package; Fox & Weisberg 2018) to assess the effect of the year factor to counts. Finally, the possible occurrence of temporal (annual) trends was evaluated through a GLMM model like to the one above but with year as a numeric variable.

2.5. Total turtle density

Total turtle density was only estimated at Lampedusa offshore as this was the only site where we were also able to calculate the correction factor, i.e. the proportion of time spent by the turtles in the detection depth layer (availability time proportion, ATP). To calculate ATP, we used data from 4 different sources. Three ATP values were estimated from data obtained through a prototype biologger consisting of a camera, a depth sensor, and a VHF and ARGOS-GPS transmitter. This biologger was deployed on 22 loggerhead sea turtles (CCL: 53.1–75.5 cm, mean = 65.1 cm) during July 2019, October 2021, and June–

September 2022. A wood support was glued with epoxy resin on the second central scute of carapace and the biollogger was attached to it through rubber bands joined by galvanic time releases (GTRs), in a position that placed the turtle's head as visible in the camera frame. After 3–12 h (depending on the GTR model), the biollogger detached and floated at surface. Using the VHF and ARGOS-GPS transmitters, the device was located and recovered using a motorboat. Three ATP values were obtained from the biollogger data: (1) ATP based on video observations (camera outside water; no. of turtles = 21) (Video ATP); (2) ATP based on the pressure sensor, with a depth of 0 m reflecting only the surface (no. of turtles = 19) (Depth0 ATP); and (3) a depth of 0–2 m reflecting the layer where turtles are visible from the UAV (no. of turtles = 19) (Depth2 ATP). A fourth ATP value (here referred to as SWS ATP) was obtained from selected satellite tracking data of a group of 11 turtles (Casale et al. 2012b, 2013) while they were within a selected area of the Tunisian continental shelf (<200 m bathymetry) southwest PIA and in the period June–October (same as the UAV period) (Fig. S4). SWS ATP was calculated from data obtained by the salt water switch (SWS), which detects when the tag is in or out of water. Tags on 9 turtles provided proportion of time spent underwater (TUW). The other 2 tags provided the mean dive duration (MDD) and the dive number (DN) that occurred within a time interval (Ti; 6 h), and TUW was calculated as $(MDD \cdot DN) / T_i$. Only data relative to daylight hour slots (06:00–18:00 h CET) were considered. TUW was converted to ATP using $TUW = 1 - ATP$. A possible spatial effect on ATP was investigated by equally dividing the entire area of tag selection into 4 subareas (bottom-left, BL; bottom-right, BR; upper-left, UL; upper-right, UR) and running a GLMM (with beta distribution; package `glmmTMB`; Brooks et al. 2017) with the formula: $ATP \sim \text{Subarea} + (1|ID)$, where ID is the individual turtle (random factor).

Mean ATP and 95% CI were estimated by a basic bootstrap method ($n = 1000$), using the 'boot' package (Davison & Hinkley 1997) in R (R Core Team 2022). To assess the effect of the 4 ATP sources above on ATP estimates, a Kruskal-Wallis test ($ATP \sim \text{source}$) was performed, where ATP is the ATP value obtained from each turtle.

Mean total density and 95% CI were estimated by using mean ATP as conversion factor: $D_t = D_s / ATP$, where D_t is the total density and D_s is the surface density estimated for Lampedusa offshore areas. D_t was calculated for the 4 counts (C+U; Cc+Uc; C; Cc) and 1 ATP type (Depth2).

2.6. Power analysis

To evaluate the feasibility of detecting spatio-temporal differences by UAV surveys, the sample size (number of surveys) needed to detect a hypothetical spatial or annual difference of 10%, 20%, and 30% from an observed mean surface density was estimated by conducting a power analysis through a simulation approach. First, for each of the 4 counts (C+U; Cc+Uc; C; Cc) of the offshore area and the 2 counts (C+U and C) of the nearshore area, the mean number of counts was calculated as well as a hypothetical mean increased by 10, 20 and 30%. Second, assuming a Poisson distribution, for each of the above 6 count types, 2 groups of n surveys were simulated through the `rpois` function, one with observed lambda and the other with the hypothetical lambda. Third, a GLM with Poisson distribution was run in the form $\text{count} \sim G$, where G is the group (2 groups: with observed and with hypothetical lambda) and tested with the `anova` function. This step was repeated 1000 times and the proportion of cases with $p < 0.05$ gave the power level. The second and third steps were repeated with increasing values of n (starting from 100, with steps of 1) until the resulting power was at least 80%.

3. RESULTS

We conducted a total of 128 flights (Table S1) in offshore Lampedusa (2017–2018) and 399 in nearshore Linosa (2020–2023) areas (Table S1). From the offshore surveys, we recorded 61 turtle sightings (45 C and 16 U), while in the nearshore area there were 197 turtle sightings (65 C and 132 U) (Table S1). The maximum detection depth in the offshore area was 2 m (Table S2). BSS and sun glare did not show any significant effects on the sighting category (C/U) (GLMM; $n = 61$).

3.1. Surface turtle density

Mean surface density in the offshore area in Lampedusa ranged from 0.336 (C) to 0.477 (C+U) turtles km^{-2} (Table S3, Fig. 2). The mean proportion of potential duplicate sightings was -6.5% and -4.4% for C+U and C, respectively. Mean surface density in the nearshore area in Linosa ranged from 0.158 (Cc) to 0.504 (C+U) turtles km^{-2} (Table S3; annual means in Table 1). The mean proportion of potential duplicate sightings was -21.4% and -5.9% of C+U and C, respectively. This proportion was not significantly different among years for both C+U and C counts (Pearson chi-squared test; $n = 268$; Table S4).

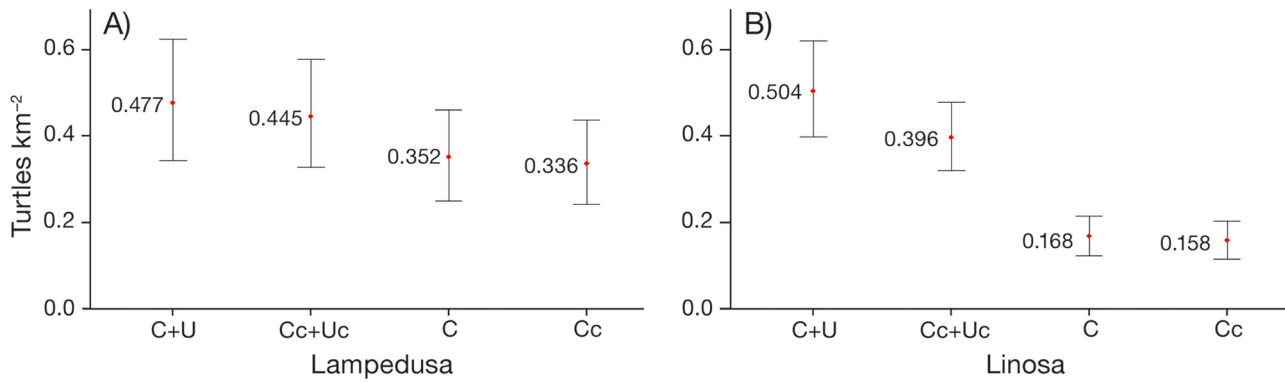


Fig. 2. Surface turtle density at (A) Lampedusa (years 2017–2018) and (B) Linosa (2020–2023). Red point: mean; vertical bars: 95% CI; C+U: all certain + uncertain (C+U) sightings; Cc+Uc: C+U corrected for duplicates; C: C sightings; Cc: C corrected for duplicates

3.2. Interannual changes in the nearshore area (Linosa)

Both C+U and C counts did not significantly differ among years (GLMM and Wald chi-squared test; $\chi^2 = 7.081$, $df = 3$, $p = 0.069$, $n = 399$ for C+U; and $\chi^2 = 6.7521$, $df = 3$, $p = 0.080$, $n = 399$ for C). No significant temporal trend of C+U counts was detected, while C counts showed a negative relationship with years that was close to significance (GLMM; $est = -0.468$; $p = 0.052$; $n = 399$).

3.3. Total turtle density in the offshore area (Lampedusa)

The 3 ATP estimations obtained from biologgers (Video; Depth0; Depth2) were derived from data from 22 turtles. The ATP type obtained from satellite tags (SWS) was estimated from 11 turtles, and no significant effects of subarea on this ATP were detected

Table 1. Annual surface density (bootstrapped mean and 95% CI, no. of repetitions = 10000) from 2020 to 2023 measured as turtles km^{-2} relative to Linosa flights. Dataset C+U: all certain + uncertain (C+U) sightings; dataset C: only C sightings

Dataset	Year	Mean (95% CI)
C+U	2020	0.851 (0.436–1.213)
	2021	0.648 (0.451–0.835)
	2022	0.287 (0.202–0.367)
	2023	0.308 (0.038–0.500)
C	2020	0.277 (0.149–0.394)
	2021	0.253 (0.121–0.363)
	2022	0.080 (0.032–0.117)
	2023	0.115 (0–0.231)

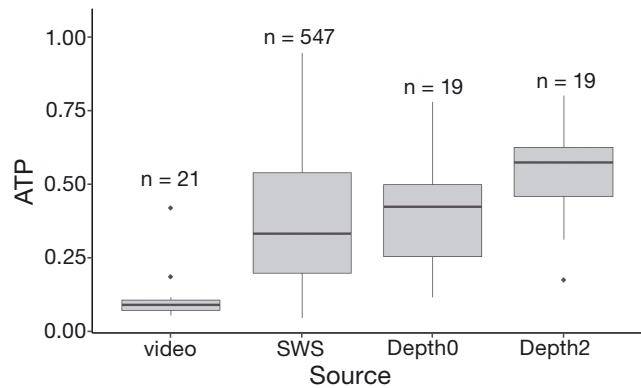


Fig. 3. Distribution of the availability time proportion (ATP) values (proportion of time spent in the detection depth layer; range: 0–1) for each ATP source. Video: camera; SWS: satellite tag; Depth0: depth sensor considering surface = 0 m; Depth2: depth sensor with surface ≥ -2 m. Box: lower (Q1) and upper (Q3) quartiles; whiskers: values within Q1 or Q3 + (1.5 \times interquartile range); black dots: outliers; thick black line: median. Sample size shown above the boxes

(GLMM; BL: reference category; BR: $p = 0.993$; UL: $p = 0.193$; UR: $p = 0.784$). Mean and 95% CI for the 4 ATP types (Video; Depth0; Depth2; SWS) are provided in Table S5. ATP estimated from different sources were significantly different (Kruskal-Wallis $\chi^2 = 57.741$, $df = 3$, $p < 0.001$; $n = 606$, Fig. 3). Specifically, video ATP significantly differed from all the other ATP types (Dunn test; $p < 0.01$; $n = 606$). SWS ATP was significantly different from all the other ATP types (Dunn test; $p < 0.01$; $n = 606$), except Depth0 ATP. The most appropriate type of conversion factor in this study was assumed to be Depth2 ATP, because in the surveyed area, UAVs can detect a turtle 2 m deep (see above). This type of ATP represented the highest ATP mean value among the 4 ATP estimates and led to the lowest total density estimate compared to the other ATPs. Mean total density estimates

ranged from 0.621 to 0.882 turtles km⁻² depending on the count type (C+U, Cc+Uc, C, and Cc) (Table S3).

3.4. Power analysis

The sample size (no. of surveys) needed to detect a significant difference of 10, 20 and 30% from the observed mean surface density with a power of 80% varied from 402 to 4864 depending on the area (off-shore, nearshore), the difference level, and the count type (Table 2).

4. DISCUSSION

We explored, for the first time, the use of UAVs for estimating sea turtle density in Mediterranean foraging areas, proposing a detailed correction methodology to address biases affecting density estimates. This UAV application constitutes an alternative method that can provide finer-scale spatial and potentially temporal resolution than aircraft-based surveys.

4.1. Turtle density in the PIA

The present study represents the first endeavour to estimate surface and total density of loggerhead turtles at a fine scale on the Tunisian shelf and the nearby oceanic area. Results support the importance of the Tunisian shelf as a neritic foraging area for loggerhead turtles in the Mediterranean Sea, as previously indicated by indirect approaches such as bycatch records (Casale et al. 2010, Casale 2011), genetic markers (Garofalo et al. 2013, Karaa et al. 2016), and satellite tracking (Casale et al. 2012a, 2013, Mingozzi et al. 2016). Moreover, the area is also

frequented by small juveniles (min. CCL reported by bycatch data in the PIA area: 18.2 cm; Casale et al. 2016) that may not be detected by aerial surveys (Barco et al. 2018); therefore, the real turtle density may be even higher.

High turtle density in the shallow waters around Linosa Island may be due to a 'sea mount effect' (Fiori et al. 2016, Vassallo et al. 2018), whereby marine megafauna aggregate close to these geological structures due to their high productivity. However, citizen science reports (Casale et al. 2020) revealed a heterogeneous surface turtle density in the PIA, with higher values between Lampedusa and Linosa islands than around each island, in agreement with aerial surveys that reported higher surface turtle density in oceanic waters (Pierantonio et al. 2023). Expanding UAV surveys in the oceanic zone of the PIA would enhance our understanding of turtle distribution over a broader area and allow us to investigate the influence of island proximity and bathymetry on sea turtle aggregation.

The present results (0.158–0.504 turtles km⁻²) are also similar to the latest estimates of loggerhead density provided for the wide area (109 709 km²) of the Sicily Channel (0.665 turtles km⁻²; Pierantonio et al. 2023), which is the highest surface turtle density in the whole central and northwestern Mediterranean Sea. The turtle density values observed in this study in the PIA area are similar to or higher than the values reported from the oceanic (both surface and total density; Gómez de Segura et al. 2003, 2006) or neritic (surface density; Carreras et al. 2004, Cardona et al. 2005) foraging areas in the western Mediterranean, respectively.

Considering that the total density from the above-mentioned area of the western Mediterranean was probably overestimated (by a correction factor obtained from SWS ATP that underestimates the time spent in visible depth layers; see Section 4.3), the total

Table 2. Number of flights needed to detect a spatial or annual difference of 10, 20 and 30% in mean surface density with $p < 0.05$ and power = 80%. Estimates provided by iterative power analysis. C+U: all certain + uncertain (C+U) sightings; Cc+Uc: C+U corrected for duplicates; C: C sightings; Cc: C corrected for duplicates

	Dataset	Mean surface density (95% CI)	Sample size needed to identify difference between means of:		
			10%	20%	30%
Offshore area	C+U	0.477 (0.336–0.609)	3372	872	402
	Cc+Uc	0.445 (0.320–0.563)	3577	930	427
	C	0.352 (0.242–0.453)	4681	1172	537
	Cc	0.336 (0.234–0.430)	4864	1238	573
Nearshore area	C+U	0.504 (0.383–0.609)	3115	860	381
	C	0.168 (0.113–0.201)	9563	2552	1138

turtle density in the neritic zone of the PIA is probably much higher than in those areas of the western Mediterranean. Moreover, the latter is also frequented by loggerhead turtles of Atlantic origin (Clusa et al. 2014), while the PIA is an exclusive foraging area for the Mediterranean loggerhead population. For these reasons, monitoring sea turtle density in this area would be valuable for investigating trends in the Mediterranean population.

In general, the loggerhead turtle surface density within the Tunisian shelf appears to be one of the highest worldwide estimated at foraging areas by aerial surveys, as well as total density, especially when comparing estimates derived by ATP based on depth layers, like in the Middle Atlantic (Barco et al. 2018) and the East Pacific Ocean (Seminoff et al. 2014) (Table S6).

4.2. Potential value of UAVs for monitoring sea turtle density at foraging areas

Even though turtle density at foraging areas is much lower than at breeding areas, results show that the turtle density in the study area is high enough to allow statistical comparisons among areas and years using data collected through UAV surveys. In this respect, the multiple-year survey campaign carried out in the present study (i.e. Linosa surveys) represents the first attempt to use UAV-based surveys for detecting inter-annual differences at turtle Mediterranean foraging areas. Only very strong trends could have been detected in such a short study period, and a higher number of surveys is needed, as shown by the power analysis. Nevertheless, the present results showed the feasibility of long-term fine-scale monitoring at sea through UAV surveys. Given the geographical position of PIA and the ecological role of the wider area for turtles, PIA is a good candidate for establishing long-term monitoring programmes both in the neritic and oceanic zones for monitoring sea turtle population trends.

Surveying offshore areas with UAVs is challenging because it requires taking off and landing on stations at sea. For this reason, very few studies worldwide have conducted such surveys, other than the present one (Yaney-Keller et al. 2021, Odzer et al. 2022). Notwithstanding, smart planning of the drone flight path (e.g. squared concentric) and careful coordination of logistical operations (e.g. multiple batteries and precision in motorboat positioning) can make UAV surveys in offshore areas feasible.

4.3. Availability bias and other methodological aspects

Selecting an appropriate ATP is crucial for determining total density and abundance. The present results complement a previous study (Barco et al. 2018) in showing the impact of maximum detection depth on ATP and consequently on total density values. ATP referred to the surface layer (SWS and Depth0), commonly used as a conversion factor, may be greatly underestimated because turtles can be detected by aerial surveys also at deeper layers, resulting in an overestimated total turtle density. For instance, Depth0 and SWS ATP values were significantly smaller (25–29%, respectively) than Depth 2 ATP. These 2 surface ATP (SWS and Depth0) are affected by slightly different biases: SWS ATP would report only periods when the transmitter is well out of water — but the turtle may be at surface even if the transmitter is totally or partially submerged — while Depth0 ATP would report periods spent at depths greater than 0 m, due to sensor accuracy (as shown by a comparison between videos and depth data in this study).

Since ATP depends on a variety of biological and environmental factors, it probably varies geographically, undermining meaningful comparisons of surface densities from distant areas. For the same reasons, caution is needed when applying a single ATP value to surface densities over wide areas (e.g. DiMatteo et al. 2022, Pierantonio et al. 2023). Finer-scale estimates would be more appropriate.

The ATP values here observed lay within the range of the Mediterranean ATP (0.06–0.59; Hochscheid et al. 2007, Revelles et al. 2007, see Table S7). Such a great variability may be, at least in part, due to turtle size and life stage, diel cycle and behavioural mode (Hochscheid 2014, Patel et al. 2015), seasons, and the frequented habitat (oceanic vs. neritic; Thomson et al. 2012), as well as the different depth ranges considered as 'surface'. ATP in neritic zones where turtles can reach and feed at the sea bottom and in oceanic zones where turtles cannot reach the sea bottom are probably different; therefore, turtle surface densities in different habitats cannot be directly compared. Thus, spatial resolution is an important factor to consider, and total densities derived from proper corrections are needed to make meaningful comparisons over time and space.

The need for estimating total turtle abundance can be relaxed for trend analyses. If annual ATPs are not available — which is likely the case — the same correction factor (ATP value) is used for the study period,

making the value of adding the uncertainty associated with the abundance estimate (e.g. Benson et al. 2020) questionable. In such cases, if the availability bias is assumed to be constant and area-specific, surface turtle density would be sufficient to monitor temporal trends in the same area, as attempted in the present study and others based on aerial (Barco et al. 2018) or boat surveys (Archibald & James 2016). Aerial surveys in very shallow coastal waters where even turtles at the sea bottom are visible (Dickson et al. 2021) do not need to account for availability bias, at least theoretically. However, even in such cases, habitat features such as algae, mangroves, rocks, etc., may interfere with turtle observations and lead to miscounts (Odzer et al. 2022) and underestimated turtle density.

The present results indicate that environmental conditions do not affect the certainty of the detected sightings and, as also reported by other studies (Marsh & Sinclair 1989b, Gómez de Segura et al. 2006, Lauriano et al. 2011, Hodgson et al. 2013), suggest that $BSS \leq 2$ represents a reasonable threshold to conduct safe and reliable UAV surveys at sea. Transects designed as in the present study bear the problem of possible double counting, a problem that increases with increasing total densities, making a correction necessary. Strip line transects do not have this problem, but they may not be feasible with UAVs because of coastline conformation or UAV flight regulations. For instance, the requirement to fly while maintaining constant visual contact with the drone severely limits the potential application of UAVs, which are constantly evolving and achieving ever-better performance (e.g. capable of several km of autonomous flight), especially at remote locations and open sea.

The above considerations, also based on the present results, advocate for incorporating multiple corrections to counts obtained from aerial surveys (in addition to distance sampling bias needed in aircraft-based surveys): availability bias (underwater animals not available to observation), perception bias (missed animals, i.e. false negatives), misidentification bias (objects mistaken for animals, i.e. false positives), and double counting (of the same animal). However, most studies considered only distance sampling bias (if aircraft-based) and availability bias. Few studies based on aerial surveys considered perception bias (Fuentes et al. 2015, Sykora-Bodie et al. 2017), misidentification bias (e.g. through categories of detection confidence like certain vs. uncertain sightings; Sykora-Bodie et al. 2017, Stokes et al. 2023), and double counting (Barco et al. 2018), with only 1 study, other

than the present one, correcting density for all the biases (Barco et al. 2018).

In addition to the enhanced accuracy of UAV surveys attributable to automated flight, camera resolution and angle, and image reviewing, the possibility of conducting multiple flights in the same area and in short time intervals allows for the estimation of a range of potential density values, thereby enhancing the reliability of the final estimate. In particular, for fast-breathers such as turtles, the number of individuals in the visible range may vary considerably among flights. Then, the corrections described in this study can improve the precision of the density estimate.

4.4. Conclusions and recommendations

UAV surveys can greatly improve our knowledge of the fine-scale distribution of turtles and on seasonal or interannual trends. Although limited to the northern part of the Tunisian continental shelf, the present results confirm the importance of this neritic foraging area for Mediterranean loggerhead turtles. In particular, PIA may represent a suitable station for long-term monitoring of the Mediterranean population, and other offshore stations in the Tunisian shelf and the adjacent oceanic zones are desirable to provide valuable indications of the overall turtle abundance and distribution in this important neritic foraging area. Such stations could also provide insights into possible seasonal variability.

Aerial surveys are subject to several biases, and failing to correct them through adequate methods may provide erroneous density estimates and/or make comparisons among different areas impossible, limiting their value for conservation. A standardized methodology is needed to ensure accurate and comparable densities of turtles. Specifically, we recommend carefully planning transect design to minimize double counting and performing a power analysis to evaluate the sample size feasibility. Corrections for perception and misidentification biases are possible with UAVs and should be always considered. When correction for availability bias is sought to estimate total turtle density, ATP should be calculated as the proportion of time spent within the detection layer and obtained from the same or very close areas.

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