Czech University of Life Sciences Prague

Faculty of Environmental Sciences

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Habitat preference and subgroup association of the common bottlenose dolphin in the Aegean Sea

Master's Thesis

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DIPLOMA THESIS ASSIGNMENT

Bc. Nikola Semeráková

Engineering Ecology
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Thesis title

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Objectives of thesis

The aim of this work is to increase the knowledge on the association of social groups of Mediterranean dolphin subpopulations with the description of long-term habitat preference. This information will subsequently lead to the development of conservation management of these cetaceans in the Mediterranean Sea.

Methodology

By analysing a photo-identification database from 2015 – 2021, the occurrence and interactions of specific bottlenose dolphin individuals in the Aegean Sea will be mapped. The unique shape of the dolphins' dorsal fin is mainly used for self-identification. The occurrence and mutual distribution of specific individuals in time and space (with respect to habitat) will be visualised using GIS software tools.

The proposed extent of the thesis

40 pages

Keywords

habitat preference, group association, bottlenose dolphin, photo-ID

Recommended information sources

Bearzi, G., Fortuna, C., & Reeves, R. (2009). Ecology and conservation of common bottlenose dolphins Tursiops truncatus in the Mediterranean Sea. Mammal Review, 39(2), 92.

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Prague on 27. 03. 2022

Declaration

I declare that I have prepared the master's thesis titled: *Habitat preference and subgroup association of the common bottlenose dolphin in the Aegean Sea* by myself, and I have cited all the information sources that I used in the thesis and I listed at the end of the thesis in the references.

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In Prague on 30th of March ______________________________

Bc. Nikola Semeráková

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Habitat preference and subgroup association of the common bottlenose dolphin in the Aegean Sea

Abstract

The Mediterranean subpopulation of bottlenose dolphins (*Tursiops truncatus)* is vulnerable, according to the International Union for Conservation of Nature Red List (IUCN). Due to its coastal habitats, this species is endangered by cumulative stressors, human activities, and other escalating pressures.

Photo-identification (photo-ID) method was used to identify and create a catalogue of dorsal fins of bottlenose dolphins and provide evidence about the distribution of habitat preference and groups association between the summer (April – September) and winter (October – March) seasons. The photo-ID dataset was collected during boat surveys year-round between January 2015-August 2021 in the eastern Aegean Sea. A geographic information system (QGIS) was used to illustrate sightings of identified dolphins between seasons and group size within bathymetry distribution.

Over seven years, 500 dedicated boat surveys with 22,300 km of active search effort (Beaufort sea states ≤3) were conducted, and 131 bottlenose dolphins were documented and subsequently resighted in the photo-ID catalogue based on their dorsal fins. The encounter rate of sighted bottlenose dolphins was 2.6 dolphins per 100 km and 1.4 photo-identified bottlenose dolphins per 100 km. The main objectives were to investigate the influence of environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, chlorophyll) on bottlenose dolphins' distribution and group size during seasons (summer and winter).

The Mann-Whitney U test showed that environmental factors (bathymetry, distance from shore, sea surface temperature, salinity) significantly influenced the distribution of bottlenose dolphins between seasons, except for chlorophyll concentration. The mean group size was 6.23 individuals, ranging between 1 and 18 dolphins. A Mann-Whitney U test revealed that the group size of identified dolphins was significantly larger during the winter than the summer season. Analysis of a general linear model (GLM) demonstrated no correlation in group size of identified bottlenose dolphins with depth and distance from the coast. The obtained results about habitats and social groups are essential for adequate protection and identifying special management measures, planning, and conservation activities in this study area.

Keywords: *Tursiops truncatus*, photo-identification, seasonal distribution, group size, encounter rate

Preference stanovišť a dynamika sociálních skupin delfína skákavého v Egejském moři

Abstrakt

Středomořská subpopulace delfínů skákavých (*Tursiops truncatus*) je podle červeného seznamu IUCN zařazena v kategorii zranitelných. Vzhledem k jejich výskytu v pobřežních oblastech je tento druh ohrožen kumulativními stresory, lidskými činnostmi a dalšími negativními vlivy.

Pro identifikaci a vytvoření katalogu hřbetních ploutví delfínů skákavých byla využita metoda photo-ID (identifikace hřbetních ploutví pomocí fotografií). Tato metoda poskytla zásadní informace o preferenci stanovišť a o sdružení skupin delfínů skákavých mezi obdobím léta (duben - září) a zimy (říjen - březen). Fotografie hřbetních ploutví delfínů byly shromážděny při průzkumech lodí od ledna 2015 do srpna 2021 ve východní části Egejského moře. Pro zmapování identifikovaných delfínů skákavých a zobrazení jejich velikostí skupin byl použit geografický informační systém (QGIS).

Během sedmi let bylo prostřednictvím lodí uskutečněno 500 průzkumů s celkovou naměřenou délkou trasy 22 300 km. Na základě těchto průzkumů bylo v katalogu identifikováno 131 delfínů skákavých. Četnost setkání všech spatřených delfínů skákavých byla 2,6 delfínů/100 km a v případě identifikovaných jednotlivců 1,4 jednotlivců/100 km. Jedním z hlavních cílů práce bylo prozkoumat vliv proměnných prostředí (batymetrie, vzdálenost od břehu, teplota mořské hladiny, slanost, chlorofyl) na rozšíření delfínů skákavých a rozpoznat změny ve velikosti skupin tohoto druhu během léta a zimy ve zkoumané oblasti.

Pomocí testu Mann-Whitney U bylo zjištěno, že všechny faktory prostředí, s výjimkou koncentrací chlorofylu, významně ovlivnily rozšíření delfínů skákavých mezi letním a zimním obdobím. Průměrná velikost skupiny byla 6,23 delfínů a pohybovala se v rozmezí 1 až 18 jednotlivců ve skupině. Mann-Whitney U test odhalil, že velikost skupiny identifikovaných delfínů byla v zimě výrazně větší než v létě. Zobecněný lineární model (GLM) neprokázal žádnou korelaci velikosti skupiny identifikovaných delfínů skákavých s hloubkou a vzdáleností od pobřeží. Zjištěné výsledky o preferovaných stanovištích a sociálních skupinách delfínů skákavých jsou zásadní pro budoucí plány ochrany, managment a další ochranářské aktivity v této oblasti.

Klíčová slova: *Tursiops truncatus*, foto-identifikace, sezonalita, velikost skupiny, četnost setkání

Table of content

List of abbreviations

ACCOBAMS – Agreement on the Conservation of Cetaceans of the Black Sea, the Mediterranean Sea and Contiguous Atlantic Areas

ASI – ACCOBAMS Survey Initiative

CITES – Convention on International Trade in Endangered Species of Wild Fauna and Flora

CHL – Surface Chlorophyll Concentration

CMS – Convention on Migratory Species

DEP – Depth

ER – Encounter Rate

GLM – General linear model

IMMA – Important Marine Mammal Area

IUCN – International Union for the Conservation of Nature

MMPATF – Marine Mammal Protected Areas Task Force

MPAs – Marine Protected Areas

Photo-ID – Photo-identification

PQ – Picture quality

psu – Practical Salinity Unit

QGIS – Geographical Information System

SCI – Special Conservation Interest

SPA – Special Protection Area

SPAMI - Specially Protected Areas of the Mediterranean

SSS – Sea Surface Salinity

SST – Sea Surface Temperature

1 Introduction

The Mediterranean Sea covers less than one per cent of the world's oceans (Bethoux et al., 1999). Nevertheless, it is exceptional for its different biological, climatic and physical processes. It is a marine biodiversity hotspot, including large cetacean species (Coll et al., 2010). Cetaceans, as umbrella species, are effective indicators of the health and productivity of a marine ecosystem, highly responsive to human-caused effects, and their conservation can improve the protection of other critical elements of the marine ecosystem (Carlucci et al., 2018; Karamitros et al., 2020). Research into cetaceans in the Mediterranean began in the late 1980s (Notarbartolo di Sciara & Bearzi, 2005). However, they have been studied in small regions, and large areas remain unexplored in the Mediterranean basins (Bearzi et al., 2009), especially in the eastern Mediterranean (Giannoulaki et al., 2017).

Common bottlenose dolphins, *Tursiops truncatus* (hereafter, bottlenose dolphins), are among the most frequently observed cetacean species throughout the Mediterranean Sea (Bearzi et al., 2009; Bonizzoni et al., 2019). Bottlenose dolphins are a social cetacean species that live in complex social groupings with varying social strategies (Connor et al., 2000). The associations of populations into communities are influenced by various abiotic and biotic factors, including oceanographic features, geographical area (Bearzi et al., 2009), home range size, habitat modification (Pace et al., 2012), seasonal and yearly changes (Bearzi et al., 1997), food availability (Pace et al., 2012), predator risk and competitor pressure (Heithaus & Dill, 2002), fisheries (Díaz López & Shirai, 2008), boat and noise disturbance and other anthropogenic pressure (Bejder et al., 2006; Pirotta et al., 2015). The group size of bottlenose dolphins is usually small, with less than ten dolphins in a coastal area (Bearzi et al., 2009). The average size of pelagic groups can be about 35 individuals (Bearzi & Fortuna, 2006).

The behaviour of this species can be affected by several environmental and ecological variables, depending on different habitats, different seasons, prey preferences (Shane et al., 1986; Bearzi et al., 2009; Silva et al., 2010; R. Wells et al., 2019). Dolphins are incredibly intelligent mammals with high levels of behavioural flexibility, adaptability to local environmental conditions (R. Wells et al., 2019) and the ability to learn new strategies and techniques (Díaz López, 2009). For instance, dolphins have opportunistically learned to feed in areas with a high concentration of prey around fish farms or chase fishing boats as an easy alternative food source for caught and discarded fish from nets (Gonzalvo et al., 2008; Bearzi et al., 2009; Díaz López, 2017).

In the Mediterranean Sea, bottlenose dolphins inhabit a wide range of habitats due to their high behavioural and ecological plasticity (Bearzi et al., 2008; Tonay et al., 2015). They are usually found over coastal areas, in semi-enclosed waters, and near surrounding islands (Giannoulaki et al., 2017). Dolphins show a full range of movements from the year-round occurrence of individuals with a high degree of site fidelity, regular/occasional stays, seasonal movements, and long-distance movements (Bearzi et al., 2004; Wells & Scott, 2009; Silva et al., 2010; Bearzi et al., 2011). For instance, long-distance movements (up to 265 km) have been observed in Greek waters (Bearzi et al., 2011). The distribution of habitats throughout their range is considerable (R. Wells et al., 2019).

The Mediterranean subpopulation of bottlenose dolphins is vulnerable based on the IUCN (International Union for Conservation of Nature) Red List. A subpopulation decline of at least 30% has been observed since 1946 (Bearzi & Fortuna, 2006) due to the last culling campaign and increasing anthropogenic threats (e.g., prey depletion, habitat degradation and loss, fishing activities, bycatches, underwater noise, chemical pollution and climate change) (Bearzi & Fortuna, 2006; Schipper et al., 2008; Bearzi et al., 2009; R. Wells et al., 2019). Moreover, dolphins are increasingly affected by habitat fragmentation, which causes changes in group size, habitat use, and decline (Bearzi & Fortuna, 2006; Boisseau et al., 2010; Hammond et al., 2012; Tonay et al., 2015).

Observing the conservation status of vulnerable bottlenose dolphins is essential for international deals (Bearzi & Fortuna, 2006; Tonay & Notarbartolo di Sciara G, 2021). Efforts to protect these species are necessary to ensure that the current population persists and thrives in the Mediterranean (Bearzi et al., 2009). It is essential to have valuable baseline information on species distribution, abundance, habitat preferences, subgroup associations and also to understand the environmental and human-caused variables that affect bottlenose dolphins, as scientific evidence is needed for conservation strategies and management planning to prevent further declines (Bearzi & Fortuna, 2006; Bearzi et al., 2009). There is limited information regarding the association structure of bottlenose dolphins and their habitat preferences, abundance, and movements in the eastern Mediterranean Sea.

The study area in the eastern Aegean Sea is a marine biodiversity hotspot for dolphins that are endangered (e.g. short-beaked common dolphin *Delphinus delphis*) and vulnerable (e.g. Striped dolphin *Stenella coeruleoalba)* in the Mediterranean Sea (Coll et al., 2010; Tonay et al., 2015). However, coastal waters of the Aegean Sea are highly affected by cumulative human impacts, including reduced availability of prey caused by overfishing, shipping traffic and habitat degradation (Micheli et al., 2013).

Photo-identification (photo-ID) method from January 2015 to August 2021 was used to catalogue photos from sightings of bottlenose dolphins within the eastern Mediterranean Sea. Each dorsal fin has a unique shape, natural marking, nicks, and notches along the trailing edge of the fin (Wüersig & Jefferson, 1990; Smith et al., 2013). The ability to recognise individuals through photo-ID allows for studying distribution, movements, population dynamics, population size, social structure, and habitat preferences of an individual or group over time. The environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, chlorophyll) may directly or indirectly affect bottlenose dolphins' habitat preferences and association with social groups. These variables were used for statistical tests and map outputs. The study compared habitat preferences and groups of identified dolphins between seasons: summer (April - September) and winter (October - March) (Xoplaki et al., 2016).

1.1 Objectives and hypothesis

This study aims to increase the knowledge of the habitat preference and group association for the Mediterranean subpopulation of bottlenose dolphins by describing and mapping their habitat preferences and group association in the eastern Aegean Sea. To achieve this, the photo-ID method and the environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, chlorophyll) with the comparison with seasons (summer, winter) were used for analysis during the study period (2015-2021). Understanding this information is essential for developing management and conservation plans and ensuring a favourable conservation status for the cetacean populations in the Mediterranean Sea.

Questions of this study:

1. Does the distribution of identified bottlenose dolphins (from the photo-ID catalogue) change due to environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, chlorophyll) between the summer and winter seasons?
2. What is the group size of identified bottlenose dolphins in the study area? Does the group size change during seasons (winter and summer)?
3. Does the group size of identified bottlenose dolphins vary in distance from the coast and depth?

Based on these questions, the following hypotheses were developed:

1. There is a different distribution of identified bottlenose dolphins based on environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, chlorophyll concentration) and seasons (winter and summer).
2. There is a difference in the group size of identified bottlenose dolphins related to the seasons (winter and summer).
3. There is a positive correlation between the group size of identified bottlenose dolphins and the distance from the coast and depth.

2 Literature Review

2.1 Cetacean species in the Mediterranean Sea

Cetacean species (whales, dolphins, and porpoises) play an irreplaceable role and essential part in marine ecosystems (Cañadas et al., 2002). Cetaceans are separated into two major groups: the mysticetes and the odontocetes. The mysticetes are characteristic of baleen plates that filtrate their prey, with larger body size and blowhole divided into two nostrils (Estes et al., 2016). Only one type of mysticete, the fin whale (*Balaenoptera physalus),* regularly appears in the Mediterranean Sea (Tonay & Notarbartolo di Sciara G, 2021). The odontocetes are also known as toothed whales, which include dolphins. A total of 25 cetacean species appear in the Mediterranean: 12 species occur regularly; three species visit the area occasionally; and ten species are very uncommon in the region (Tonay & Notarbartolo di Sciara G, 2021). Figure 1 shows the distribution of resident marine mammals in the Mediterranean Sea, with a high concentration in the western Mediterranean and Aegean seas (Coll et al., 2010). Nevertheless, there is a lack of information about their abundance and habitat preference in many regions in the Mediterranean Sea (Bearzi et al., 2009).

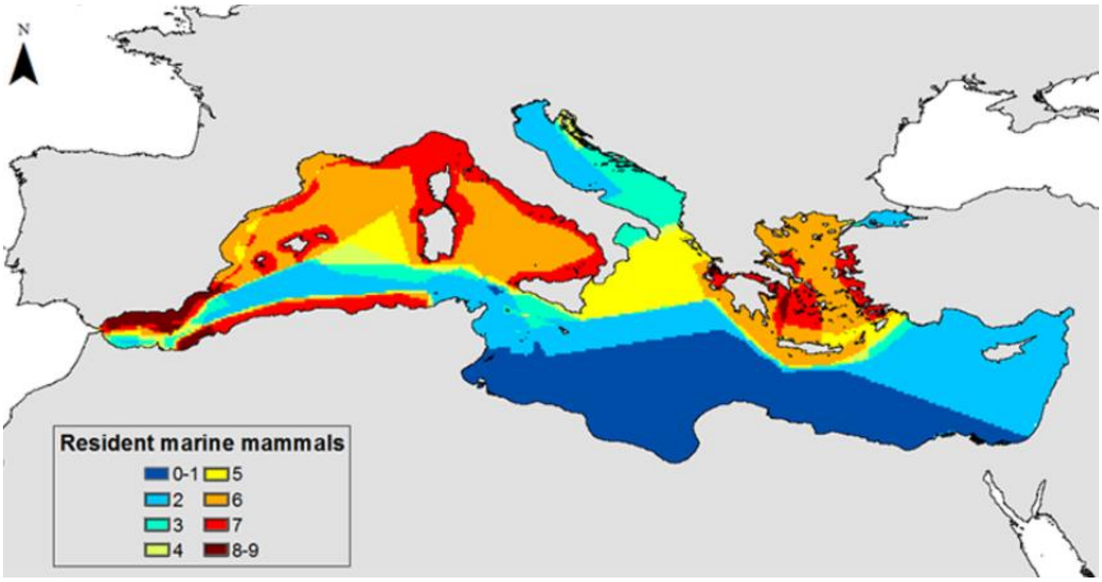


Figure 1. Map of distribution of marine mammals regarding the number of species found in the Mediterranean Sea (Coll et al., 2010).

Historical and present pressures have highly impacted the Mediterranean Sea and its inhabitants (Lejeusne et al., 2010; Micheli et al., 2013). Cetacean species are affected by many human threats, such as overfishing, accidental captures in fishing gear, whale/dolphin watching, direct human disturbance, underwater noise, chemical contaminants, marine litter, and climate change (Notarbartolo-di-Sciara et al., 2008). Up to 62% of fish stock is overfished in the Mediterranean, making the Mediterranean the most overfished sea globally and with a danger of being depleted completely (FAO, 2020). Climate change directly causes increased sea temperature, ocean acidification and earlier phytoplankton blooms in spring (Lejeusne et al., 2010). The latter has possible cascade effects in the food chain, resulting in reduced primary and secondary plankton production (Lejeusne et al., 2010; Birchenough et al., 2013).

Globally the conservation status of marine mammals is of particular concern. An approximated 36% of marine mammals are threatened (Silva et al., 2008). Coastal waters changes are most impactful for small marine mammals such as dolphins (Schipper et al., 2008). Figure 2 shows the distribution of multiple threats across regions in the Mediterranean Sea. The highly affected regions are the Alboran Sea, Tunisian Plateau, the Aegean Sea (especially along the Turkish coast) and the Adriatic Sea (Micheli et al., 2013). According to Micheli et al. (2013), only less than one per cent of these areas remain relatively untouched by multiple cumulative threats.

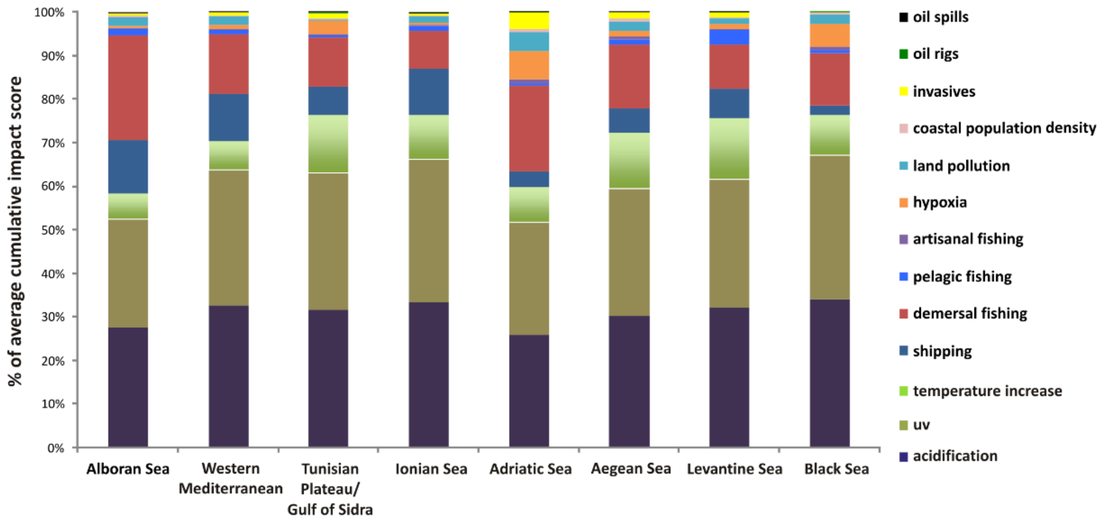


Figure 2. Percentage distribution of human and environmental impacts to the average cumulative impact score in the Mediterranean regions (Micheli et al., 2013).

2.2 The Aegean Sea

The Aegean Sea is a marine biodiversity hotspot for dolphins (Coll et al., 2010) and home to a high concentration of endangered, threatened, and vulnerable species, including sea turtles, birds and an endangered Mediterranean monk seal (*Monachus monachus)* (IUCN-MMPATF, 2017). Twelve cetacean species have been identified in the Aegean Sea (Tonay et al., 2015) and eleven species in Greek waters (Frantzis, 2009). The northern and southern sub-basins of the Aegean Sea (divided by a latitudinal boundary of 38 N) (Figure 3) exhibit significant changes in sea surface temperature (SST), salinity (SSS) and different dynamic features (Skliris et al., 2011). The southern waters have high salinity with high temperature and a higher warming linear trend, whereas the northern basin has a colder temperature with low salinity (Skliris et al., 2011). The Aegean Sea began to heat up rapidly following a cooling effect from the late 1960s to the 1990s (Skliris et al., 2011). The satellite-derived analysis showed seasonal fluctuations in sea surface temperature in this area, with much higher warming rates during summer and autumn than during spring (from April to June) and winter (from January to March), with the lowest temperature in March and highest in August (Skliris et al., 2011). Since 1989, the sea surface temperature has changed to 3.19 ± 1.26 ° C (Kuleli & Bayazit, 2020). The average monthly temperatures in the Aegean Sea range from 8°C (north) to 26°C (south) and salinity from 31.0 psu (north) to more than 39.0 psu (southeast) (Skliris et al., 2011). Simultaneously, sea surface satellites indicated an increase in the intensity, duration and frequency of marine heatwaves that impact sea surface temperature warming, noticed for the first time in 2012 (Darmaraki et al., 2019).

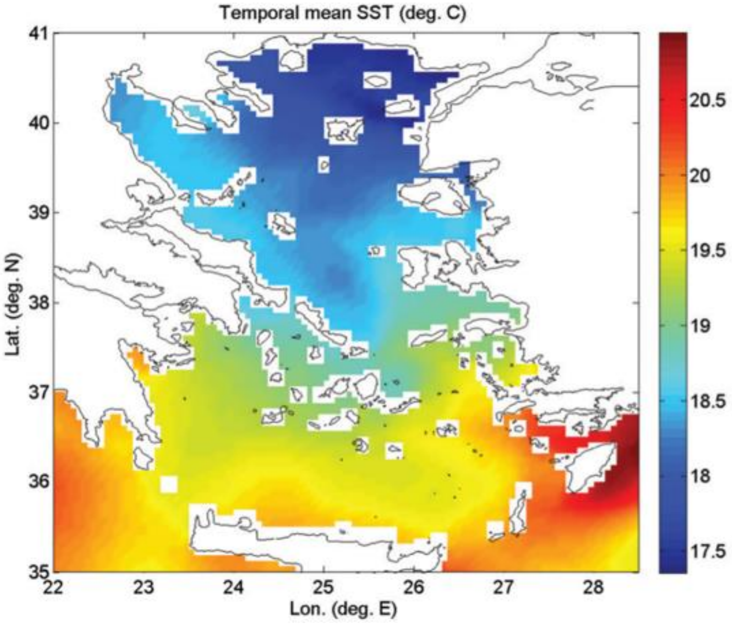


Figure 3. The temporal mean of sea surface temperature in the Aegean Sea (from a statistical database 1985-2008) (Skliris et al., 2011).

2.3 Common bottlenose dolphins

Bottlenose dolphins are among the most well-studied, best-known and widespread cetacean species, occurring in all oceans within a wide latitudinal range (Figure 4) (Reynolds et al., 2000; Bearzi et al., 2009; Tonay & Notarbartolo di Sciara G, 2021). The species display geographically and seasonally different ecological, behavioural, and social characteristics, including different habitat and prey preferences (Silva et al., 2010; R. Wells et al., 2019).

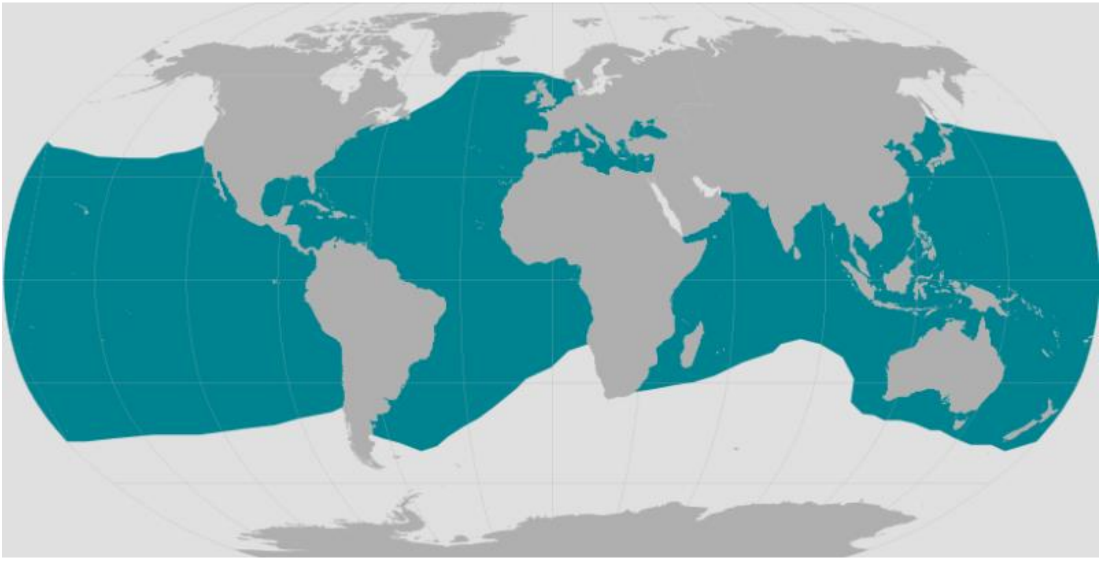


Figure 4. The geographic range of bottlenose dolphins worldwide (NOAA Fisheries, 2021).

Bottlenose dolphins belong to the class of Mammalian, order Cetartiodactyla, suborder Odontoceti, and family Delphinidae (Jefferson et al., 2015). At present, two species of bottlenose dolphins are recognised within the genus *Tursiops,* the common bottlenose dolphin *T. truncatus* and the smaller Indo-Pacific bottlenose dolphin *T. aduncus* (Bearzi & Fortuna, 2006). Bottlenose dolphins are characteristic of a robust body, grey colouration with white/pinkish belly colouration and cream stripes on the side, short snout, and high dorsal fin (Figure 5) (Bearzi et al., 2009). The length of adult bottlenose dolphins is roughly 2.5-3.8 m for males, with bodyweight from 150-650 kg, and females are approximately ten per cent lighter than males (Wells & Scott, 2009; Jefferson et al., 2015). The body size and colouration of dolphins vary depending on geographical location and different ecotypes they inhabit (coastal vs pelagic waters) (Wells & Scott, 2009).

Figure 5. Common bottlenose dolphin from photo-ID catalogue, photographed in the eastern Aegean Sea.

The global population is estimated at 750,000 individuals, and the numbers may vary as a large portion has not been surveyed; therefore, the overall number may be significantly higher (R. Wells et al., 2019). The global status of the population of bottlenose dolphins is assessed as Least Concern by the IUCN Red List (R. Wells et al., 2019). However, several populations worldwide are critically endangered, such as the Fiordland subpopulation in New Zealand (Currey et al., 2009). The subpopulation in the Black Sea is classified as Endangered, with fewer than 1,000 individuals (Bearzi & Fortuna, 2006). The Mediterranean subpopulation of bottlenose dolphins is listed as vulnerable according to the IUCN Red List (Hammond et al., 2012). The total number of individuals of Mediterranean bottlenose dolphins is estimated at a low 10,000 individuals (Bearzi & Fortuna, 2006). In the Mediterranean Sea, a subpopulation of bottlenose dolphins has declined by at least 30% since 1946 (Bearzi & Fortuna, 2006). Some subpopulations in the Mediterranean, such as in the Adriatic Sea, have decreased by 50% since the 1960s (Bearzi et al., 2004). Many other small subpopulations face a high risk of extinction in the Mediterranean basins (Bearzi & Fortuna, 2006).

2.3.1 Mediterranean subpopulation

Based on mitochondrial and nuclear DNA analyses, the populations occupying the Black Sea, eastern North Atlantic Ocean, and Scottish waters genetically differentiate from the Mediterranean population (Natoli et al., 2005). Significant genetic differentiation exists between the eastern versus western Mediterranean Sea populations. Therefore, five populations of bottlenose dolphins were distinguished (Figure 6): eastern North Atlantic, Scottish, eastern Mediterranean, western Mediterranean and Black Sea (Hammond et al., 2012; Natoli et al., 2005). The study also found that physical boundaries (such as the Strait of Gibraltar, the Almeria-Oran front, the Italian Peninsula, Sicily Channel, Turkish Straits) and differences in habitat characteristics restrict individuals' movements and genes flow (Natoli et al., 2005). At the same time, these barriers affect the movement of prey species (Natoli et al., 2005).

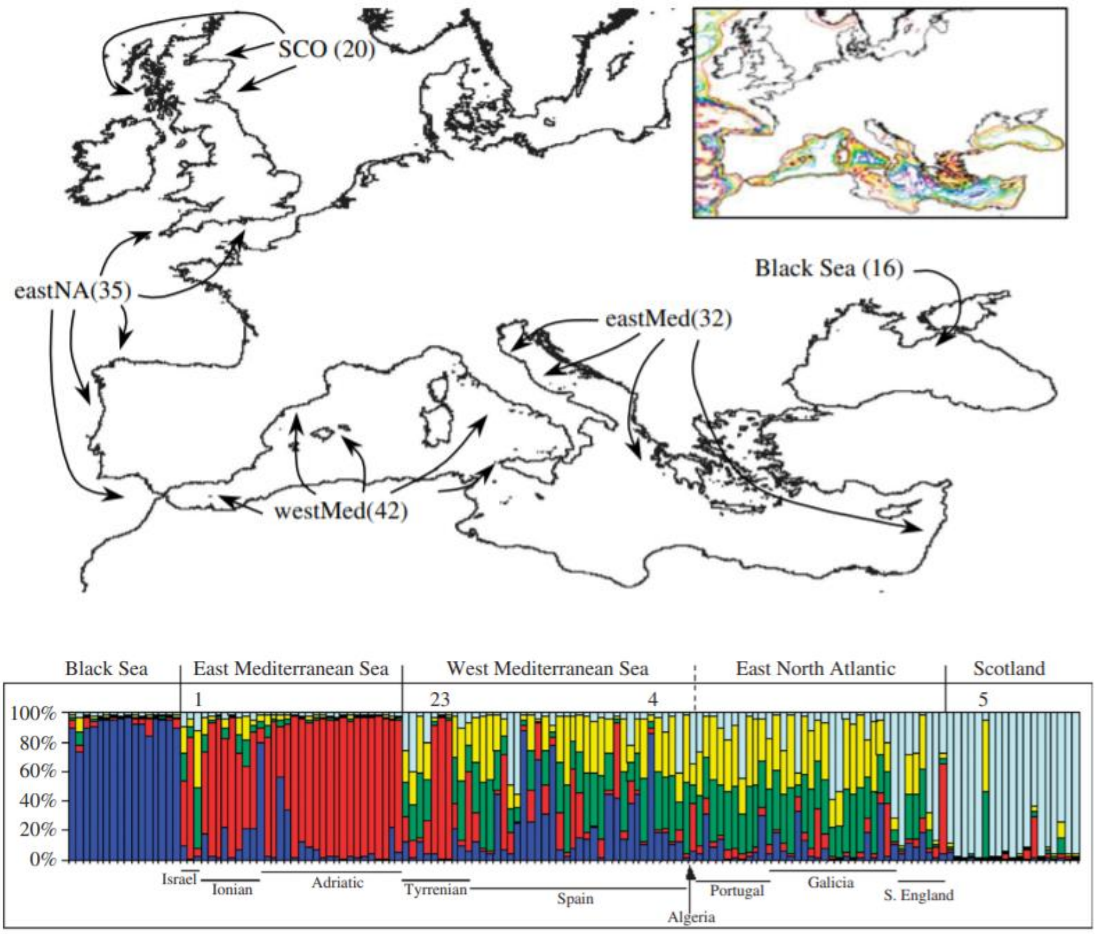


Figure 6. Estimated geographical origin and genome of five distinguished populations of bottlenose dolphins. Colours for each population are in parentheses: Black Sea (blue colour), East Mediterranean Sea (red colour), West Mediterranean Sea (yellow colour), East North Atlantic (green colour), Scotland (turquoise colour) (Natoli et al., 2005).

2.3.2 Behaviour and ecology

Bottlenose dolphins are long-lived cetacean species. Female dolphins live more than 57 years and males more than 48 years (Wells & Scott, 2009). Their generation time is estimated as 23 years (Taylor et al., 2007). Females are capable of sexual maturity from 5 to 13 years and males later from 9 to 13 years (Wells & Scott, 2009). Births have been observed during all seasons, although the most common during the spring-summer months (with a peak from July to August) (Bearzi et al., 1997; Wells & Scott, 2009). The pregnancy time is 12 months, with the offspring remaining with the mother for several years (from 2 to 11 years) (Connor et al., 2000).

Bottlenose dolphins have a high level of behavioural flexibility and adaptability with a significant capability to learn new feeding techniques (Bearzi et al., 2009; Díaz López, 2009). Bottlenose dolphins adjust their feeding strategies to habitat type, geographic area, local and food conditions (Shane et al., 1986; Milani et al., 2018). For instance, in the Ambracian Gulf in Greece (400 km^2), there is an isolated population of bottlenose dolphins where the density of dolphins is higher than in overfished waters nearby (Bearzi et al., 2008). This is primarily related to prey accessibility, specific feeding habitat (dolphins have to learn to hunt in shallow waters of around 30 m), and behaviour adaptations (Bearzi et al., 2008). Dolphins' behaviour also depends on boat traffic and other anthropogenic pressures. For instance, in Italy, it has been observed that bottlenose dolphins prefer to leave and avoid locations with heavy disturbance by motorboats (La Manna et al., 2013). The feeding habits of this species are described as opportunistic, which means that it can develop a multitude of adaptive strategies (Gonzalvo et al., 2008; Bearzi et al., 2009). The species consume a wide range of prey, tending to feed on the most abundant species at any time and location, and are known to use readily available food resources from the surrounding environment (Gonzalvo et al., 2008). Depending on the depth, the dive for prey usually lasts 3-5 minutes, up to 8 minutes (Bearzi et al., 2009). Bottlenose dolphins feed on epipelagic and mostly on demersal prey, such as European hake (*Merluccius merluccius)*, European conger (*Conger conger)*, red mullet (*Mullus surmuletus*), depending on their abundance and availability, including cephalopods, such as common octopus (*Octopus vulgaris)*, common cuttlefish (*Sepia officinalis)*, and occasionally crustaceans (Cañadas et al., 2002; Tonay & Notarbartolo di Sciara G, 2021).

Human-caused food patches, such as trawl fishery and aquaculture, can affect the behaviour of cetacean species by altering predation pressure, the transformation of habitat, changes in food distribution and availability, which may modify social interaction, structure and organisation (Díaz López & Shirai, 2008; Pace et al., 2012). In many world areas, dolphins have opportunistically learned to closely follow fishing vessels to take advantage of fish caught and discarded from nets or by fishermen, although this behaviour can result in bycatch (Gonzalvo et al., 2008; Bearzi et al., 2009).

Furthermore, the habitat of bottlenose dolphins overlaps with aquaculture as a food source in several coastal areas worldwide (Díaz López et al., 2005; Bearzi et al., 2009; Díaz López, 2009; Giannoulaki et al., 2017). Marine aquaculture directly impacts the surrounding environment and ecosystem and represents an easy alternative food source for dolphins, with concentrated fish density (wild and farmed fish) within aquaculture facilities (Díaz López, 2012). Greece and Turkey (Figure 7) are the largest producers of marine aquaculture in the EU (Hofherr et al., 2015; Papageorgiou et al., 2021).

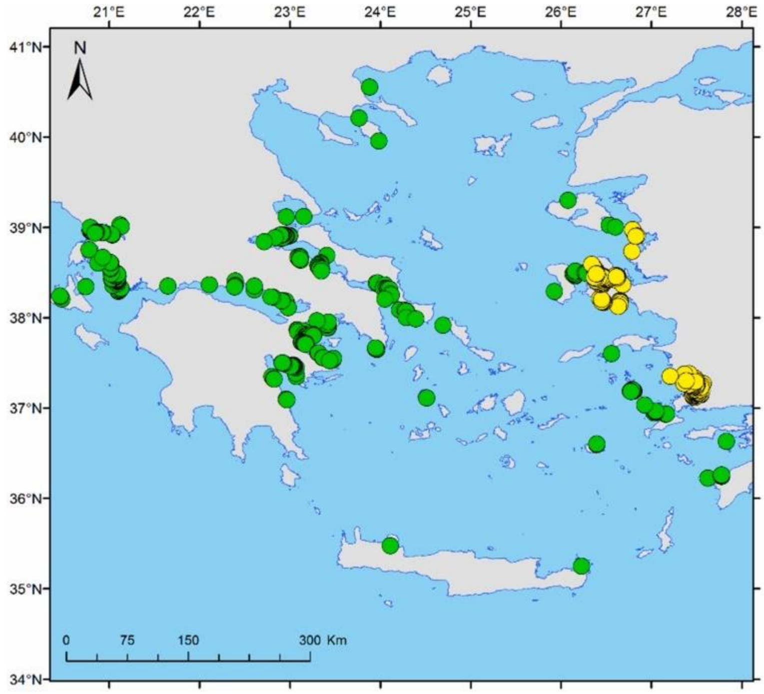


Figure 7. Distribution of aquaculture in Greek (green points) and Turkey (yellow points) waters (Papageorgiou et al., 2021).

2.3.3 Subgroup association

In the Mediterranean Sea, bottlenose dolphins' group size is usually small, consisting of less than ten individuals, although they have also been observed in larger pods (Bearzi et al., 2009). Significant differences in group size between areas have been monitored (Shane et al., 1986; Wells & Scott, 2009). The characteristics of individuals in a group often vary in gender, age, family relationships, history of the association, reproduction condition, and activity level (Wells et al., 1987; Connor et al., 2000). The social groups with various subgroups include nursery school groups, mixed groups of adolescents, individuals of adult males and strongly connected pairs (Wells & Scott, 2009). The groups formed with calves are often larger (mean group size 6-12 individuals) than groups only composed of adults (mean group size 4 individuals) (Bearzi et al., 1997).

Dolphins live in fission-fusion societies where the dynamic of group composition and structure changes daily or hourly depending on the shared or conflicting interests of the individuals in the group (Connor et al., 2000). Group composition plays a more crucial role for bottlenose dolphins than the number of dolphins involved in feeding actions (Díaz López & Shirai, 2008). Open populations (with higher genetic diversity due to migratory behaviours) are more resilient to environmental and anthropogenic threats than closed and fragmented communities (without immigration and emigration) (Papale et al., 2017).

Within feeding, the dolphin association can divide into three groups: acquaintances (dolphins associate at a minimum of one feeding group but never create preferred associations), affiliates (association of dolphins through both categories of feeding), and feeding associates (association of dolphin within only one feeding category) (Díaz López & Shirai, 2008). Based to the authors Díaz López & Shirai

(2008), it has been demonstrated that during opportunistic feeding, cooperation between dolphins influenced by anthropogenic food patches (e.g. aquaculture) decreased due to more effortless prey capture.

Bottlenose dolphins occasionally associate with other cetacean species; they have been observed in mixed groups with short-beaked common dolphins (Cañadas et al., 2002; Bearzi et al., 2009) and long-finned pilot whales (Cañadas et al., 2002; Stephanis et al., 2008) in some areas in the Mediterranean basins.

2.3.4 Habitat preference and distribution

Bottlenose dolphins are a cosmopolitan species that live in temperate and tropical water worldwide, with SST from about 10 to 32°C (Wells & Scott, 2009). They are widespread in the Mediterranean, where they live in various habitats (Bearzi et al., 2009). Bottlenose dolphins usually occur over enclosed seas in coastal areas (including lagoons, semi-enclosed bays, and estuaries), continental shelves, shallow waters, and surrounding islands (Cañadas et al., 2002; Giannoulaki et al., 2017).

Bottlenose dolphins seem to be year-round residents with a high site fidelity within a given area in coastal water of the Mediterranean Sea (Bearzi et al., 2011). However, they can temporarily leave the area to move long-distance from the coast, migrate seasonally, or stay regularly and occasionally (Bearzi et al., 2004, 2009; Wells & Scott, 2009). In Greek waters, bottlenose dolphins have been documented to move between core habitats up to 265 km apart (Bearzi et al., 2011). Furthermore, bottlenose dolphins living in the Azores (Portugal) moved over long distances of almost 300 km with large home areas, showing lower density and uneven distribution of their prey (Silva et al., 2008). Additionally, wide-ranging movements and a lack of territorial behaviour provide an opportunity to maintain genetic differentiation and reduce the inbreeding between dolphins (Silva et al., 2008).

Bottlenose dolphins are frequently found in productive and shallow waters within 300 m (Cañadas et al., 2002; Bonizzoni et al., 2019). Nevertheless, groups of dolphins are sometimes observed at depths of more than 2000 m (Bearzi et al., 2009). According to a study in the Greece Sea (Giannoulaki et al., 2017), dolphins prefer waters with a high probability of sardine presence and concentrations of chlorophyll (more than 0,135 mg m^{-3}), coastal distance (< 7 km). Bottlenose dolphins are occasionally known to enter estuaries and rivers (Sackl et al., 2007).

The distribution of bottlenose dolphins in the Mediterranean Sea appears to be dispersed, fragmented into smaller groups and with low densities due to natural or anthropogenic reasons (Bearzi & Fortuna, 2006; Boisseau et al., 2010; ACCOBAMS et al., 2021). According to Figure 8, in the Mediterranean Sea, species are mainly found in the Strait of Gibraltar, the Alborán Sea, the Balearic Sea, the northern area of the Adriatic Sea, the Strait of Sicily, north of the Tyrrhenian Sea, the Aegean Sea and along the coast, with some sighting further offshore (ACCOBAMS et al., 2021).

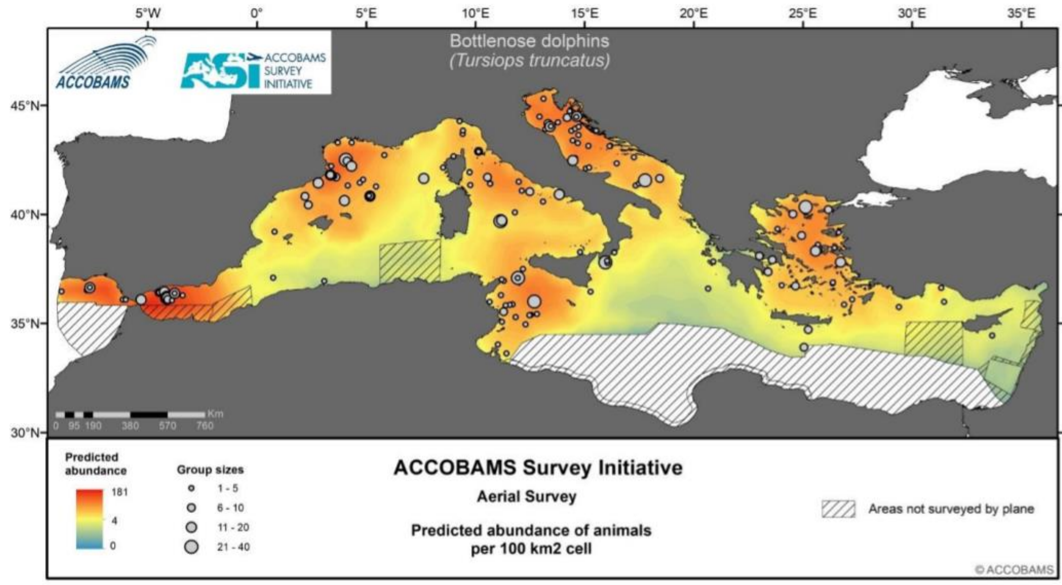
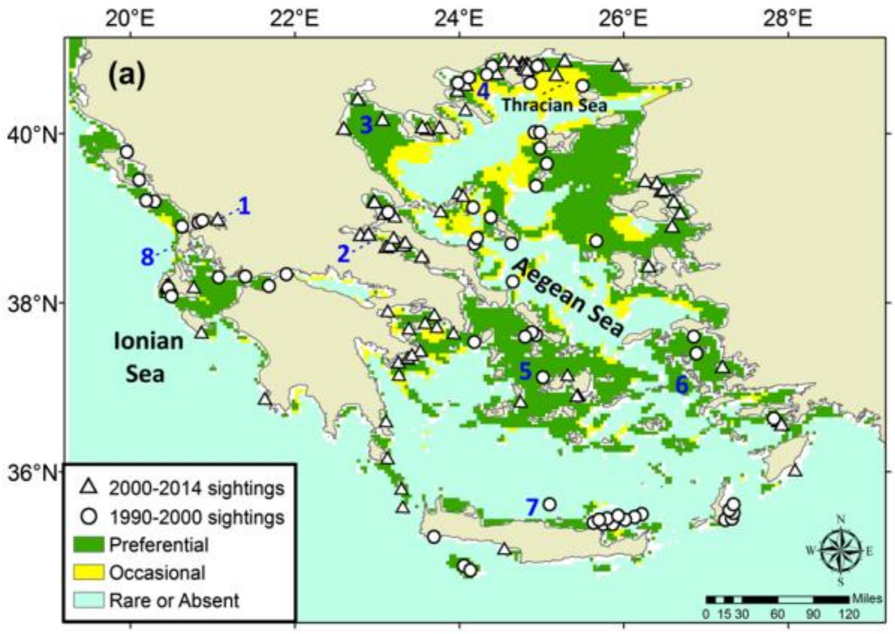


Figure 8. Predicted distribution of bottlenose dolphins in the Mediterranean Sea according to the data of ACCOBAMS observed during the aerial survey of the Survey Initiative (ASI) (ACCOBAMS et al., 2021).

In the Aegean Sea (Figure 9), the main hot spot areas for bottlenose dolphins include the north part (around the Thracian Sea, the Thermaikos Gulf), the northern Sporades, the Cyclades, the eastern part and the north Dodecanese (from the southern part of the island of Samos, through Patmos, Lipsi, Leros to Kos) (Giannoulaki et al., 2017).



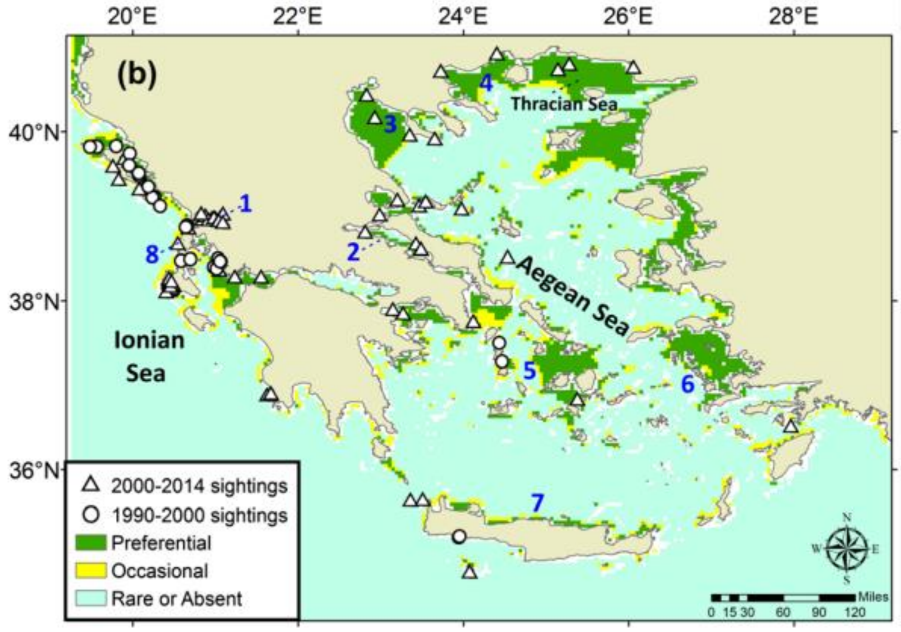


Figure 9. Habitat preference of bottlenose dolphins during (a) early (May, June, July) and (b) late (August, September) summer in the period 1990-2014 in the Aegean Sea (Giannoulaki et al., 2017).

2.3.5 Threats

The subpopulations of bottlenose dolphins have declined by at least 30% since 1946, but the numbers may vary with a region of the Mediterranean Sea (Bearzi & Fortuna, 2006). Some subpopulations in the Mediterranean, such as in the Adriatic Sea, have decreased by 50% since the 1960s (Bearzi et al., 2004). The total population size of Mediterranean bottlenose dolphins is estimated at 10,000 individuals (Bearzi & Fortuna, 2006).

In the past, bottlenose dolphins have been subject to extensive direct killing (Bearzi et al., 2004). Bottlenose dolphins and common dolphins were the primary targets of the culling campaigns in Mediterranean waters (Hammond et al., 2012). State-supported systematic extermination campaigns of dolphins happened until the early 1960s (e.g. the Adriatic Sea) (Bearzi et al., 2004; Tonay & Notarbartolo di Sciara G, 2021) and until the 1970s in the Aegean Sea (Tonay et al., 2015). For instance, records showed that a total of 84.9t of dolphins were caught in the Turkish Aegean Sea in 1968 (Tonay et al., 2015).

Nowadays, due to their coastal habitats, the Mediterranean subpopulations are endangered by cumulative stressors and other escalating pressure (Bearzi & Fortuna, 2006; Lejeusne et al., 2010; Schipper et al., 2008). The anthropogenic impacts are enormous in the Mediterranean basins, like nowhere else in the coastal zone globally (Coll et al., 2010). The major threats include prey depletion caused by overfishing and environmental degradation, habitat loss, pollution (plastic, chemical and noise pollution), epizootic outbreaks, climate changes and other human-induced threats (Bearzi & Fortuna, 2006; Schipper et al., 2008; Bearzi et al., 2009). Other adverse effects include marine traffic, illegal fishing practices, bycatch, and accidental mortality in fishing gear (particularly in bottom trawlers, driftnet, gillnets, purse seines) (Tonay & Notarbartolo di Sciara G, 2021). Therefore, based on these direct and indirect impacts, the subpopulations of bottlenose dolphins have declined in the Mediterranean in recent decades (Bearzi et al., 2009; Bearzi & Fortuna, 2006; Cañadas et al., 2002; Tonay & Notarbartolo di Sciara G, 2021). Nevertheless, the species seems to be more resilient, opportunistic and adaptable to human threats than other cetacean species (Bearzi et al., 2009).

2.3.6 Conservation status

Dolphins can be protected based on national or international protection status. Legislative instruments of international protection provide a crucial framework for protecting their environment, but conservation of bottlenose dolphins and the enforcement of illegal practices are scant (Bearzi et al., 2009). National protection is managed by the states and can support the conservation of bottlenose dolphins in a specific area, such as the Dolphin Conservation Reserve on the islands of Cres and Losinj (Croatia) or around the island of Kalamos (Greece) (Bearzi et al., 2009).

The bottlenose dolphin subpopulations are protected in the Mediterranean Sea protected under the EU Habitats Directive (92/43/CEE) in Annex II and Annex IV, the Barcelona Convention on Specially Protected Areas of Mediterranean Importance (SPAMI) in Annex II, the Agreement on the Conservation of Cetaceans in the Black Sea, the Mediterranean Sea and Contiguous Atlantic Areas (ACCOBAMS) in Appendix II, the Washington Convention (CITES) in Annex II, Bern Convention in Appendix II, the Convention on Migratory Species (CMS) in Appendix II (Tonay & Notarbartolo di Sciara G, 2021). Protected areas under the ACCOBAMS for bottlenose dolphins are the Amvrakikos Gulf (Greece), north-western Sardinia and the Tuscany archipelago (Italy), the coast of the Cres-Lošinj (Croatia) and the Turkish Straits (ACCOBAMS, 2021).

2.4 Photo-identification

Photo-identification (photo-ID) is a method that uses photographs of cetacean species' dorsal fins or flukes, whereby the identification of individuals is according to their natural markings such as scars, nicks and variable pigments (Wüersig & Jefferson, 1990). Different identifying features are applied to recognise the individual depending on the species. Each fin or fluke of cetacean species has its unique shape, natural marking, notches, and various scars, allowing for long-term identification and creating a photo catalogue (Wüersig & Jefferson, 1990; Smith et al., 2013). This method has been used since the 1970s to study group structure, population size, movement patterns and site fidelity (Wüersig & Jefferson, 1990). Simultaneously, a photo-ID study allows researchers to find information about cetaceans' age and population size (Wüersig & Jefferson, 1990). However, this method is less generally used for gregarious species (e.g. pilot whales) that aggregate in large groups because they are often broadly dispersed (Hupman et al., 2018). It is often impossible to photograph each individual, which may thus result in imprecise estimates (Hupman et al., 2018).

This method has been widely applied for bottlenose dolphins due to their occurrence at the coast water, relatively high mark ratio, and manageable population size (Smith et al., 2013; Hupman et al., 2018). However, some natural marks, especially on the surface of the body and fin, may change, heal or disappear over time

(Wüersig & Jefferson, 1990; Smith et al., 2013). The distinguishing changes must be recorded and adjustments monitored during resightings to avoid misidentification (Yoshizaki et al., 2011). For bottlenose dolphins, the distinguishing features are mainly nicks and notches on the dorsal fins; and temporary marks (e.g. scars, scratches, and uneven pigment spots) (Wüersig & Jefferson, 1990). Nevertheless, not all spotted animals have the same probability of being identified, which can result from different factors: different quality of photographs, variability of dolphin's distinctiveness (i.e. notches and nicks on the dorsal fin) and different behaviour (e.g. some dolphins are less likely to approach vessels) (Hupman et al., 2018). Photo-ID catalogues of bottlenose dolphins allow tracking and identifying individuals over extended areas and years, helping improve knowledge of movement patterns and protection within a managed area (Bearzi et al., 2011; Zanardo et al., 2016).

3 Methodology and data used

3.1 The study area

The boat surveys were concentrated in a core study area in the eastern Aegean Sea, also known as the Dodecanese area (Figure 10), around the islands Leros, Lipsi, Patmos, Agathonisi, Marathons, Fourni, Ikaria, and Samos, which covers approximately 7,000 km^2 of the sea surface. This study area is a vast archipelago with dozens of small islands and bays with different depths (can reach up to 1500 m in the Ikaria basin).

The study area is a biodiversity hotspot for marine mammals with high habitat diversity, including seagrass meadows with the endemic *Posidonia oceanica*, coralligenous bottom, and shallow bottom of sand (IUCN-MMPATF, 2017). This study area is part of the marine protected areas (MPAs) proposed habitat for dolphins and whales by ACCOMABS (Alexopoulos, 2013), Central Aegean Important Marine Mammal Area (IMMA) (IUCN-MMPATF, 2017) and Marine Natura 2000 sites with Special Protection Area (SPA) or Special Conservation Interest (SCI) (MAPAMED, 2020).

However, this study area is highly impacted by anthropogenic activities, including prey depletion caused by overfishing (including illegal fishing), shipping traffic and habitat degradation (Coll et al., 2010; Vlachopoulou et al., 2013; Pietroluongo et al., 2020). The fish stock in this area has decreased significantly over the last decades (Vlachopoulou et al., 2013). The main impact of reducing fish stocks was trawling within 2.8 km from the coast (Vlachopoulou et al., 2013). The study from Vlachopoulou et al. (2013) focused on fishing in the coastal areas of the islands of Samos and Fourni, as shown in Appendix 1. Additionally, significant shipping traffic has been noted during the surveys, which causes noise and plastic pollution (Vlachopoulou et al., 2013). Along the study area, small-scale artisanal fisheries are active with high fishing efforts (Sini et al., 2019) that use trammel net, gillnet, longline (Vlachopoulou et al., 2013; Pietroluongo et al., 2020). In this area, there is little fisheries regulation, despite the legislation and directives of the European Union (Vlachopoulou et al., 2013).

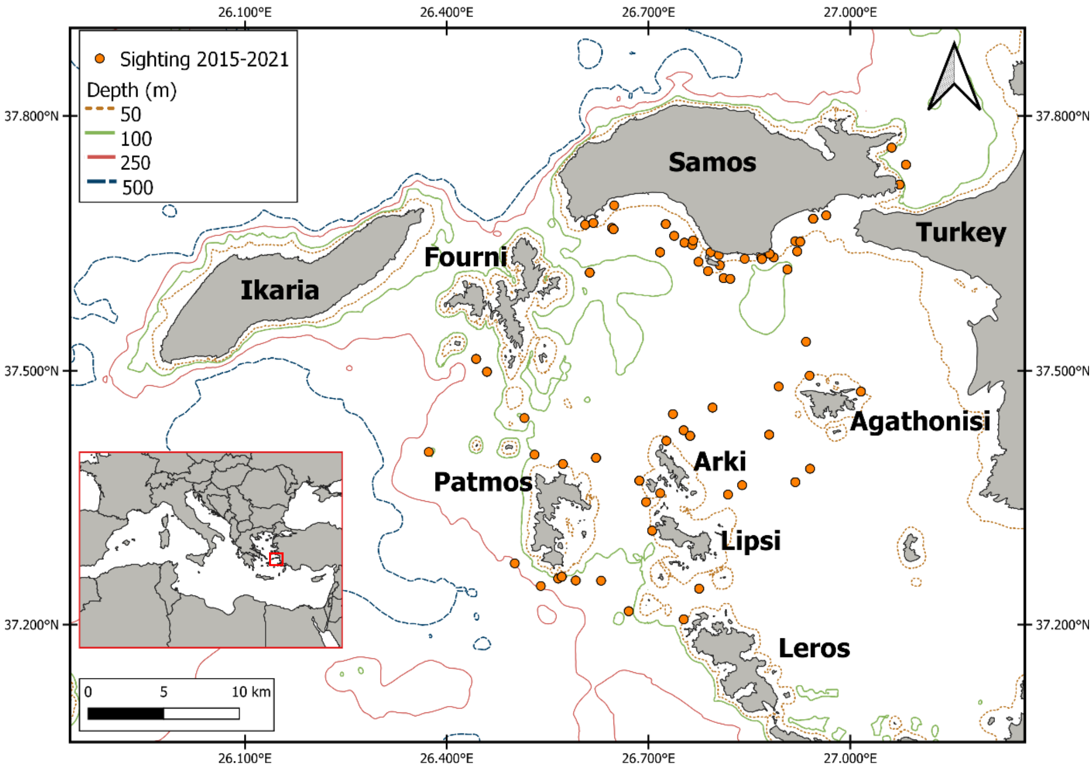


Figure 10. Study area within the Mediterranean Sea in the eastern Aegean Sea with bathymetry and sightings of bottlenose dolphins from January 2015-August 2021.

Six cetacean species with bottlenose dolphins are common and year-round in the study area: short-beaked common dolphin (*Delphinus delphis)*, striped dolphin (*Stenella coeruleoalba)*, Risso's dolphin (*Grampus griseus)*, Cuvier's beaked whale (*Ziphius cavirostris)*, sperm whale (*Physeter macrocephalus).* This study area also provides a vulnerable habitat for the most endangered pinniped species, the Mediterranean monk seal (Karamanlidis & Dendrinos, 2015). Due to the lack of information, the total number of seals remains unclear; however, it is estimated that approximately 150 individuals live in the central Aegean Sea (IUCN-MMPATF, 2017). Moreover, in this study area occur two turtle species: loggerhead sea turtle (*Caretta caretta)* and green sea turtle (*Chelonia mydas)* (Casale & Margaritoulis, 2010). In addition, different species of sea birds have been recorded in this study area interacting with cetacean species, such as Audouin's gull (*Ichthyaetus audouinii)*, great cormorant (*Phalacrocorax carbo)*, Mediterranean shag (*Phalacrocorax aristotelis)*, storm petrel (*Hydrobates pelagicus)*, Mediterranean shearwater (*Puffinus yelkouan)* and, Scopoli's shearwater (*Calonectris diomedea).*

3.2 Data collection

The data were collected by the Archipelagos Institute of Marine Conservation during boat surveys conducted year-round from January 2015 to August 2021 in the eastern Aegean Sea (Figure 11). During the 4-month internship from June to September 2021, the author of this thesis participated in data collection and data processing during the study period. Surveys were conducted onboard three Archipelagos' research vessels Pinelopi, Naftilos and Aegean Explorer. Pinelopi is a 16 m steel dual-mast sailing boat with a 140 hp engine, Naftilos is a 15.24 m twomast sailing boat with a 90 hp engine, and the Aegean Explorer is a 21.74 m motorboat with two 450 hp engines (Archipelagos Institute of Marine Conservation, 2021). The boat's average speed was around 7-8 knots during the survey.

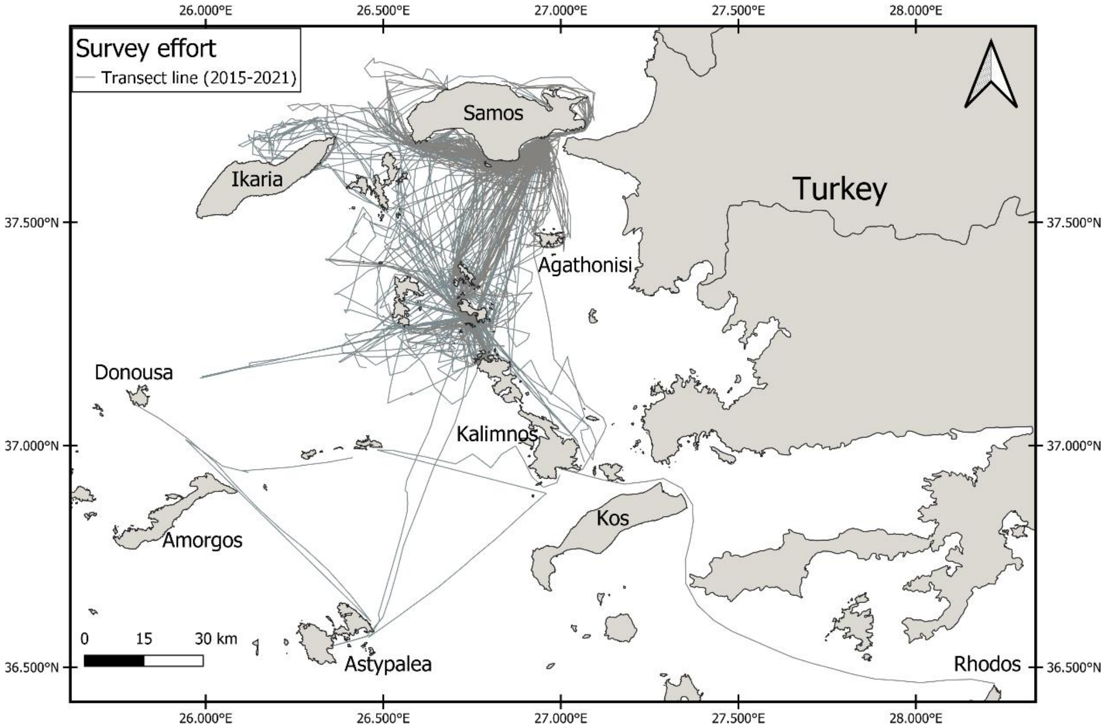


Figure 11. Boat survey transects were carried out from January 2015 to August 2021.

The following environmental parameters were recorded during a survey every 30 minutes:

- Boat Survey Number: Starts from 1 every year
- Crew Members: Observers' Names
- Boat Name: Pinelopi, Naftilos, Aegean Explorer
- Date and Time
- Coordinates (GPS)
- Sea state
    - 0 = mirror-like, smooth sea surface
    - 1 = scaly ripples, no foam crests, 0-0.2 m
    - 2 = small wavelets, glassy crests, no breaking, 0.2-0.5 m
    - 3 = large wavelets, crests begin to break, scattered white caps, 0.5-1 m
    - 4 = small waves becoming longer, numerous whitecaps, 2-3 m
    - 5 = moderate waves taking longer to form, many whitecaps, some spray
- Effort:
    - Off Effort – X
    - Positive Visual – PV (observers are working, and the sea state is ≤ 2)

- Negative Visual – NV (observers are working, and the sea state is >3)
- Positive Visual/Acoustic – PVA (observers are working and using the HP, and the sea state is ≤ 2)
- Negative Visual/Acoustic – NVA (observers are working and using the HP and sea state is >3)
- With Cetaceans – W (during a sighting)

- Boat Speed (knots)
- Wind speed (knots) and direction
- Cloud coverage (percentage)
- Boats: types of other boats spotted: Sailing (S), Sailing with Engine (SE), Motorboat (MB), Fishing Boat (FB) and Vessel (V)
- Sighting N: of Cetaceans, Turtles and Seals (starts from 1 every survey)
- Species
- Total Number: Total N of individuals
- Adults: Number of adults
- Juvenile: Number of Juveniles
- Calf: Number of Calves
- Notes

In addition to cetacean sightings, data were collected on seabird species, marine litter, acoustic checks, and marine traffic. The GPS constantly tracked the boat's route. The following equipment was used during the boat survey:

- Garmin GPS,
- Binoculars,
- Cameras (Canon 1300D, Canon 70D, Nikon D5200) and lenses (18-155mm, 70-300 mm),
- Bioacoustics Laptop (with Pamguard 64 Software),
- Aquarian AS-1 hydrophone,
- Zoom PodTrak P4,
- PA-4 hydrophone preamplifier,
- Film Camera for Boris Video Camera (Sony Zeiss Vario-Tessar with 54x image zoom).

For crew safety, boat surveys were carried out only under the following weather conditions: daylight and no fog, wind intensity and sea state not exceeding ≤ 3 on the Douglas scale and Beaufort scale ≤ 4 with good visibility. During the survey, at least five trained observers consistently scanned the sea surface with the naked eye and binoculars and divided it into different spots to visually cover the whole boat. The boat was viewed as the center of a clock. Two to four people (depending on the boat) observed cetaceans and marine litter (covered the 12-5 trine and the 7-12 trine), one person observed seabirds (covered the 3-9 trine), and one person entered data for the environment, marine litter, and bioacoustics on the laptop. One or more crew members were off. Observers changed their positions after 30 minutes of observation to limit the effects of bias and fatigue.

When an individual or a group of dolphins were sighted, the research vessel approached the dolphin(s). At the same time, the team began collecting data on date, location (GPS coordinates), the angle of first contact measured via a compass, species, group size (number of dolphins), and dolphins' behaviour. In addition, the sea and weather conditions with depth were also documented. During the sighting, tasks were divided among the observers: filming the behaviour of the dolphins, taking pictures of dolphins and their dorsal fins, filling in the datasheet about an ethological behaviour on 3 min intervals, and updating the environmental data on the laptop using PAMGuard every 15 minutes.

3.2.1 The environmental variables

The data for the environmental variables concerning habitat preference was obtained from outside resources (Table 1). The depth (DEP in m), distance from the coast (m), sea surface temperature (SST in °C), salinity (SAL in psu), chlorophyll concentration (CHL in mg m^{-3}) were calculated and assessed for each dolphin encounter. Table 1 shows all environmental variables with information on units of measure, temporal resolution, spatial resolutions, sensor, and source. Environmental data were used for statistical tests and map outputs.

Table 1. Environmental variables used for analysis in R and QGIS programmes.

Variable	Units	Temporal resolution	Spatial resolution	Sensor	Source
Depth (DEP)	m	Static	1 m	Merged Satellites	EMODnet DTM
Sea Surface Temperature (SST)	°C	Monthly average	4 km	MODIS AQUA	NASA Ocean Color
Surface Chlorophyll (CHL)	mg/m^3	Monthly average	1 km	Merged Satellites	Copernicus
Sea Surface Salinity (SAL)	psu	Monthly average	4 km	Merged Satellites	Copernicus
Distance from Coast (DISTANCE)	m	Static	-	QGIS analyst tool	QGIS calculations

3.2.2 Photo-collection

The following cameras were used to collect the photograph for photo-ID: Canon 1300D, Canon 70D, Nikon D5200 with lenses: 18-155mm, 70-300mm. Photos were taken perpendicular to the dolphin's body axis (90° angle) by focusing on the dorsal fin for the nicks and notches along the trailing edge of the fin. Burst mode (8 fps) was used to take multiple pictures of the surfacing dolphin, allowing for a higher probability of capturing the best moment for a good quality picture. Autofocus, preferably with a specific mode for fast-moving objects if available on the used camera (e.g., sports mode with specialised tracking autofocus), was used to ensure that the pictures are well focused. Both sides of the boat were covered to have a higher chance to get photos of both sides of the dorsal fin. The dorsal fin photographs were later used to identify individual dolphins for photo-ID. Once all the dolphins were photographed, and the necessary information was obtained, the vessel's search for other dolphin groups resumed. Sightings lasted for a maximum of 30 minutes.

3.3 Data analyses

The software QGIS 3.20 (QGIS Development Team, 2022) was used to create predicted distribution maps and identify the types of habitat preferences that individuals and the subpopulation commonly found to inhabit in the study area. The software R studio (RStudio Team, 2020) was used to analyse the results of the correlations between the variables of interest (Table 2 and Table 3). Seasons were defined as summer (April – September) and winter (October – March) (Xoplaki et al., 2016).

3.3.1 Photo-identification

Picture quality (PQ) and distinctiveness (D) are the two most commonly used scoring systems for photo-ID pictures (Wüersig & Jefferson, 1990; Zanardo et al., 2016). PQ relates directly to the quality of the photo, while D relates to the individual in the photo. The PQ is based on an evaluation focus/clarity (Figure 12), angle (Figure 13) and contrast (Figure 14) (Wüersig & Jefferson, 1990).

Figure 12. Three different ranges of focus on dorsal fins: (a) score 1: excellent focus; (b) score 4: moderate focus; (c) score 8: poor focus. Focus/Clarity relates to the crispness or sharpness of the picture. Lack of clarity is caused by poor focus, excessive enlargement, motion blur, or poor resolution. The range of the score for the focus is from 1 (the best) to 8 (the worst).

Figure 13. The different angles of dorsal fins (a) score 1: 90° angle (perpendicular); (b) score 4: slight angle; (c) score 6: oblique angel. The angle is ideally at 90°. The range of the score for the angle is from 1 (the best) to 8 (the worst).

Figure 14. The range of contract dorsal fins (a) score 1: ideal contrast; (b) score 2: moderate contrast; (c) score 3: excessive contrast. High contrast causes the picture to lose detail, and low-contrast pictures lose the fin into the background and lack definition. The range of the score for the contract is from 1 (the best) to 3 (the worst).

Each category was given an individual rating. PQ does not consider the marks/patterns on the fins. The sum of each of the characteristics determined the PQ score as follows:

○ PQ1 = 3-5,
○ PQ2 = 6-9,
○ PQ3 ≥ 10.

The D relates (Figure 15) to how distinctive the markings on the individual's fin are and how easily the individual can be identified in varying levels of PQ. Dolphins were primarily identified according to the notches on the dorsal fin's trailing edges. Additional marks on the surface of the fin, such as pigmentation, scars and fin shape, were also applied for classification.

Figure 15. Categories of the distinctiveness (D) of four different dorsal fins (a) highly distinctive (D1) (highly distinctive primary nicks/notches and have an excellent chance of being identified from all photographs, no matter the dorsal side); (b) highly distinctive (D1) (highly distinctive primary nicks/notches); (c) distinctive (D2) (less distinctive minor nick/notches with discernible pigmentation patterns); (d) non-distinctive (D3) (no distinctive nicks/notches).

Each photo was assessed and given a PQ and D score. Only the highly distinctive (D1) and distinctive (D2) photos with PQ1 or PQ2 were used for the photo-ID catalogue and further analysis of this study. Photographs that did not meet the quality parameters were discarded (Wüersig & Jefferson, 1990). Each photo was then cropped around the focal point of the dorsal fin and any visible part of the body (Bearzi et al., 2008).

The cropped photos were first compared and matched to discover how many individual dolphins were seen on a given day (internal matching). The photos from the internal matching were then sorted into a catalogue of identifiable individuals (external matching).

The photo-ID catalogue contains photographs of bottlenose dolphins previously observed and identified during boat surveys. At the same time, this catalogue is constantly updated with dolphin individuals who are newly discovered in the study area. The new picture of the dolphin was renamed with the code for the catalogue as the following example (Figure 16):

CCC-YYL_X_EEE_DD.MM.YYYY_BOAT_NN_###.

- CCC = code of individual, progressive number (three digits) running from the first animal sighted in 2015.
- YY = the year (two digits) when the dolphin was first identified.
- L = letter denoting how distinguishable the individual is:
    - W - well-marked with ≥3 distinct, long-lasting marks can be identified from either side and bad quality pictures),
    - S - slightly marked individuals with ≤2 distinct, can be identified from either side, but not bad quality pictures).
- X = side of the dorsal fins of dolphins, L marked for the left side and R marked for the right side.
- EEE = encounter number (three-digit)
- DD.MM.YYYY = day (two-digit), month (two-digit), year (four-digit) where the individual was first sighted
- BOAT = name of the boat the survey was conducted on, all uppercase letters
- NN = first and last initial of the photographer (e.g. Beatriz Tintoré = BT)
- ### = consecutive number of the picture (three digits), starting at 001 for the first picture, then 002, then 003, et cetera. Consecutive numbers start again from 001 for each encounter but not for each new photographer within an encounter.

ID individuals	Photographs
063_19W_L_008_17-10-2019_PINELOPI_AD_019	
097_20W_L_003_13-03-2021_PINELOPI_BT_072	

Figure 16. Examples of different individuals of bottlenose dolphins from the photo-ID catalogue.

The programme Zoner Photo Studio X (Zoner Photo Studio, 2022) was used for the matching process and assembling catalogues.

3.3.2 Habitat preference

A nonparametric Mann-Whitney U test was used to test the distribution of identified dolphins (from the photo-ID catalogue) between environmental variables (bathymetry, distance from shore, sea surface temperature, salinity, and chlorophyll concentration) and seasons (winter and summer) (Table 2). Data were not normally distributed based on the Shapiro-Wilk test. For analysis, these packages were applied in R studio: ggplot2, car, ggpubr, foreign, coin, tidyverse, psych, psycho.

Table 2. Response and explanatory variables and statistical tests for habitat preference performed in R.

Response variable	Explanatory variable	Statistical test
Season	Bathymetry	Mann-Whitney U test
Season	Distance from the shore	Mann-Whitney U test
Season	SST	Mann-Whitney U test
Season	SAL	Mann-Whitney U test
Season	CHL	Mann-Whitney U test
Year	Encounter rate	Kruskal-Wallis rank test

The encounter rate was calculated as the total number of dolphins sighted per kilometres spent on effort and the total number of re-identified dolphins from the photo-ID catalogue per kilometres for each year (Bearzi et al., 2008). Kruskal–Wallis rank test was used to test encounter rate among years (Table 2). Density was measured as dolphins/km^2 of the study area (Bearzi et al., 2009).

3.3.3 Subgroup association

A nonparametric Mann-Whitney U test was used to test the group size distribution between seasons (winter and summer). Data were not normally distributed. To test the correlation between the group size of identified bottlenose dolphins with the distance from the coast and the depth, a general linear model (GLM) with an assumed Poisson distribution was used (Table 3). For analyses, these packages were used in R studio: ggplot2, car, lattice, ggpubr, tidyverse, psych.

Table 3. Response and explanatory variables and statistical tests for subgroup association performed in R.

Response variable	Explanatory variable	Statistical test
Group size of identified individuals	Season	Mann-Whitney U test
Group size of identified individuals	Bathymetry	GLM, Poisson distribution
Group size of identified individuals	Distance from the shore	GLM, Poisson distribution

4 Results

4.1 Survey and photo-ID effort

Five hundred boat surveys with 22,300 km of active search effort were conducted in the eastern Aegean Sea at different seasons between January 2015 and August 2021. A total of 477 bottlenose dolphins were sighted during the seven-year study period. A total of 131 bottlenose dolphins were photo-identified in the photo-ID catalogue and repeatedly resighted with 250 resightings in the study area based on dorsal fin markings (Table 4).

Table 4. Summary of the survey efforts, number of surveys, number of bottlenose dolphins sighted, photo-identified and re-identified dolphins from the catalogue (2015-2021).

Year	Effort (km)	Number of surveys	Number of dolphins sighted	Number of dolphins catalogued	Number resightings of photo-ID dolphins
2015	1262.9	69	63	29	39
2016	3121.5	88	68	9	27
2017	2957.5	71	44	4	6
2018	1587.1	58	49	15	28
2019	6496.2	92	81	28	50
2020	2828.7	48	60	24	49
2021	4046.1	74	112	22	51
Total	**22300**	**500**	**477**	**131**	**250**

Encounter rates in 2015-2021 were calculated for sighted dolphins and photo-identified dolphins. The mean value of the encounter rate of sighted bottlenose dolphins was computed on 2.6 dolphins per 100 km and 1.4 photo-identified bottlenose dolphins per 100 km and the variation among years was not statistically significant (Kruskal–Wallis rank test $H = 6$, $df = 6$, $p > 0.05$) (Table 5).

Table 5. Encounter rates of the total number of dolphins sighted (not photo-identified) and photo-identified bottlenose dolphins over the years (2015-2021).

Year	Encounter rate of sighted bottlenose dolphins (not photo-identified)	Encounter rate of photo-identified bottlenose dolphins
2015	0.050	0.031
2016	0.022	0.009
2017	0.015	0.002
2018	0.031	0.018
2019	0.012	0.008
2020	0.021	0.017
2021	0.028	0.013
Total	**0.026**	**0.014**

The mean density of 477 sighted bottlenose dolphins in the study area with approximately 7000 km^2 was 0.068 dolphins km^2, and 250 resightings of identified dolphins were estimated to have an average density of 0.0357 dolphins km^2.

Figure 17 displays the cumulative number of photo-identified bottlenose dolphins over the years with the number of surveys.

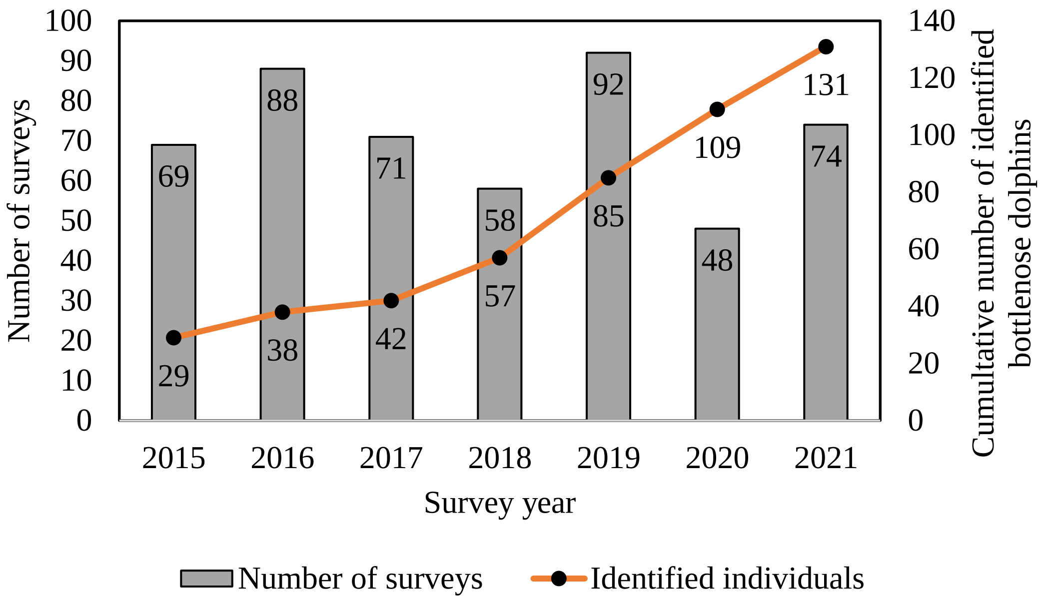


Figure 17. Annual cumulative number of photo-identified bottlenose dolphins over the years (2015-2021).

During 500 boat surveys, 250 photo-ID resightings were measured within seven years of the study period, with a high number of 69 resightings in October. Other photo-ID resightings of bottlenose dolphins over the months are shown in Figure 18. Due to bad weather conditions, the number of resightings was low in December - February.

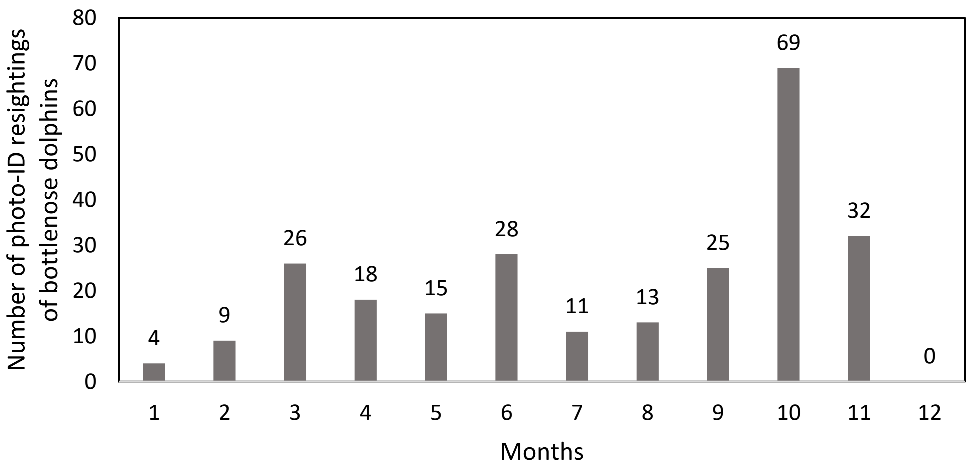


Figure 18. The number of bottlenose dolphins re-identified over the months (2015-2021).

The 131 distinctive individuals were resighted from 1 to 7 times. Among these, 67 individuals (51%) were photographically captured only once, 31 individuals (24%) were sighted twice, 28 individuals (21%) three or four times and 5 individuals (4%) were resighted up to 4 times (Figure 19).

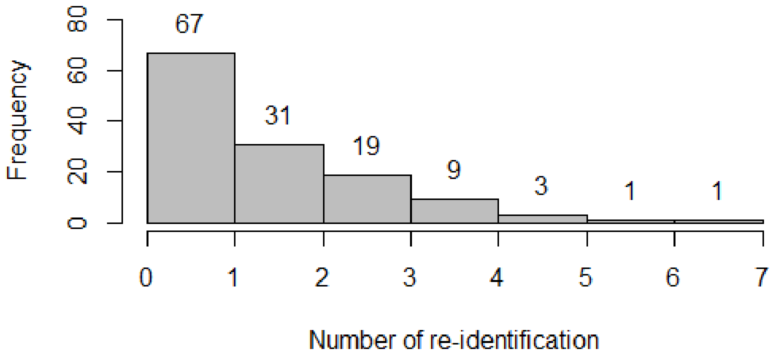


Figure 19. Frequency of photo-identified resighting of bottlenose dolphins in the eastern Aegean Sea (2015-2021).

The 89 bottlenose dolphins were sighted in one year (68%), 26 individuals in two different years (20%), 13 dolphins in three different years (10%) and 3 bottlenose dolphins (016_15S; 029_15S; 041_17S) were resighted in four different years (2%), no bottlenose dolphin was resighted in every year from 2015-2021 (Appendix 2). Table 6 shows the annual sighting histories of bottlenose dolphins (photo-ID records across years) in three or four different years.

Table 6. Sighting histories of bottlenose dolphins resighted in three or four different years.

ID	2015	2016	2017	2018	2019	2020	2021
001_15S	2	1		1			
008_15W	1	1					2
016_15S	2	1		1	1		
022_15W	1	3					2
023_15S	1			1	1		
026_15S	1			1			1
028_15S	2	4		1			
029_15S	1		1	1		2	
041_17S			1	1	1	1	
044_17W			2	1			1
052_18W				1	1	2	
057_18W				1	1	1	
058_18S				1		2	1
065_18S				1	1		1
072_19W					1	2	1
076_19S					1	1	1

From the photo-ID catalogue, 80 individuals were slightly marked (marks with ≤ 2 distinct), and 51 dolphins were well-marked (marks with ≥ 3 distinct). Figure 20 shows the well-marked dorsal fin of the bottlenose dolphin designated as ID number 022_15W from the photo-ID catalogue. Dolphin 022_15 was first sighted in 2015 in the southern part of Samos, then resighted three times in 2016 in the southern part of Samos and two times in 2021 in the eastern part of Samos.

Figure 20. Well-marked dorsal fin of the bottlenose dolphin with ID number 022_15W, photo-identified in different years in the southern and eastern part of Samos.

4.2 Habitat preference

The topography of the study area from the southern side of the island of Samos through the island of Patmos, Agathonisi, Lipsi to the northern side of the island of Leros is represented by shallow waters with depths between 60-90 m that rarely exceed depths of more than 100 m with slope ranges between 0% - 10% (Figure 21). The study area is characterised by shallow waters, with an average depth of 332 metres. However, the deepest point of 1590 m is around Ikaria. Figure 21 presents sightings of photo-identified bottlenose during the summer and winter seasons with bathymetry in the eastern Aegean Sea between 2015 to 2021.

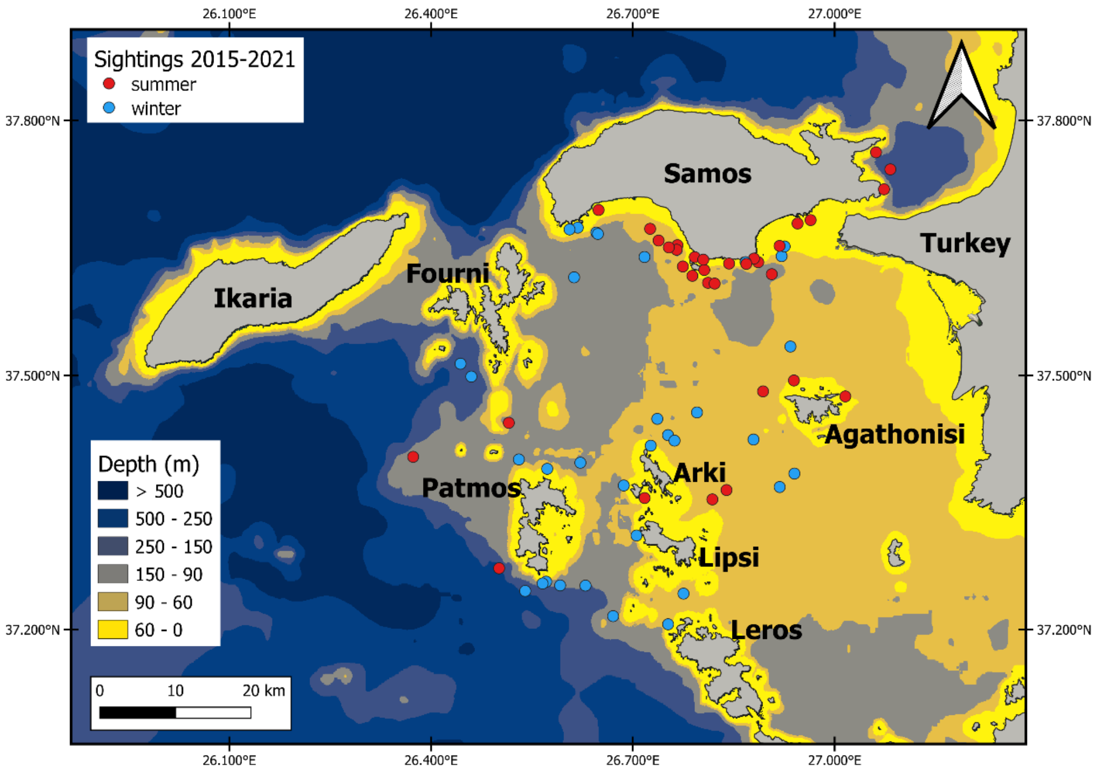


Figure 21. Map of bathymetry and sightings of photo-identified bottlenose dolphins in the study area during summer and winter seasons from 2015 to 2021.

Distribution of identified bottlenose dolphins varied from a depth ranging between 2.76 m and 251.42 m (mean = 78.12 m, SD (standard deviation) = 46.15 m), distance from the coast range from 288.7 m to 8,255.7 m (mean = 2,452.6 m, SD = 1826.63 m). The sea surface temperature where individuals were identified was measured ranging between 16.4 °C to 25.9 °C (mean = 21.36 °C, SD = 2.82 °C), the chlorophyll concentration between 0.042 to 0.32 mg/m^3 (mean = 0.093 mg/m^3, SD = 0.057 mg/m^3), and the salinity ranging between 39.04 psu to 39.58 psu (mean = 39.24 psu, SD = 0.092 psu) (Table 7).

Table 7. The minimum (Min.), first quartile (1st Qu.), median (Mdn), mean (with standard deviation), third quartile (3rd Qu.), and maximum (Max.) of environmental variables of identified bottlenose dolphins (n=250) from the photo-ID catalogue.

Environmental variables	Min.	1st Qu.	Mdn	Mean (SD)	3rd Qu.	Max.
DEP (m)	2.76	41.51	83.14	78.12 (46.15)	93.27	251.42
DISTANCE (m)	288.7	949.0	2209.4	2452.6 (1826.63)	3422.7	8255.7
SST (°C)	16.40	19.40	22.40	21.36 (2.82)	23.60	25.90
CHL (mg/m^3)	0.042	0.063	0.072	0.093 (0.057)	0.094	0.32
SAL (psu)	39.04	39.18	39.24	39.24 (0.092)	39.31	39.58

4.2.1 Seasonal habitat preference

A Mann-Whitney U test showed that the bathymetry distribution of identified bottlenose dolphins was significantly higher during the winter seasons (Mdn = 84.5, n = 140), compared to the summer seasons (Mdn = 70.81, n = 107) ($z = -4.8493$, $p < 0.001$, 95% CI [-33.58, -10.16]) (Figure 22).

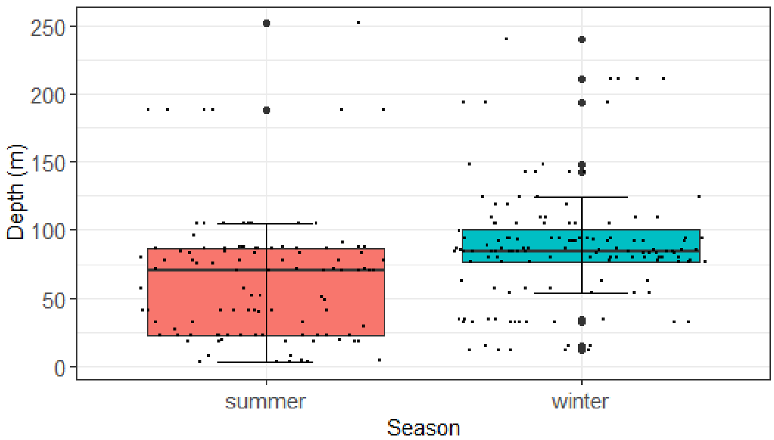


Figure 22. Bathymetry of photo-identified bottlenose dolphins compared to the seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

Distribution of bottlenose dolphins within distance from the coast was statistically significantly higher in winter seasons (Mdn = 2833.62, n = 140) than in summer seasons (Mdn = 1025.4, n = 107) ($z = -5.5108$, $p < 0.001$, 95% CI [-1579.854, -501.332]) (Figure 23).

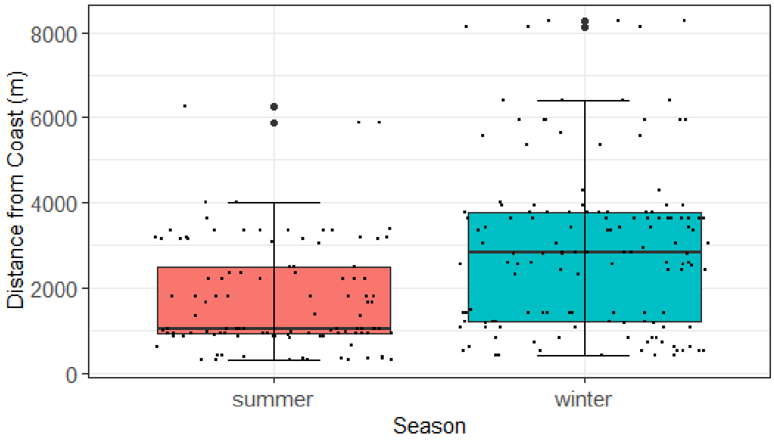


Figure 23. Distance from the coast of photo-identified bottlenose dolphins compared to the seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

A Mann-Whitney test indicated that the sea surface temperature for the location of photo-identified bottlenose dolphins was statistically significantly higher in summer (Mdn = 22.9, n = 105) than in winter seasons (Mdn = 21.1, n = 140) ($z = -5.02$, $p < 0.001$, 95% CI [-0.9999, -2.1]) (Figure 24).

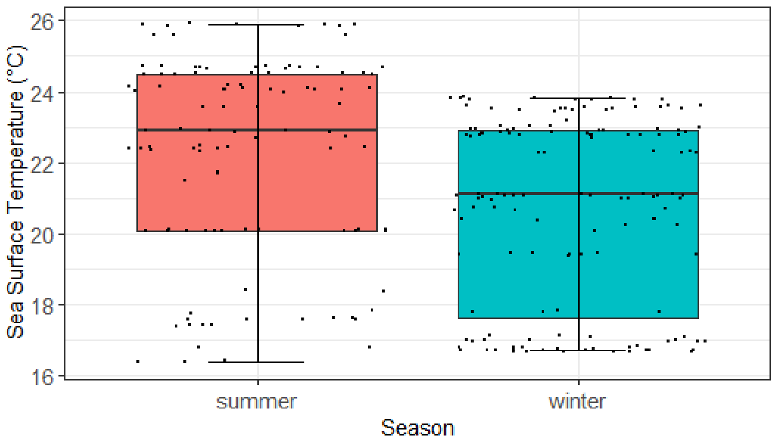


Figure 24. Sea surface temperature of photo-identified bottlenose dolphins compared to the seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

The salinity of the photo-identified bottlenose dolphins' distribution was significantly higher in the winter seasons (Mdn = 39.24, n = 140) compared to the summer seasons (Mdn = 39.18, n = 71) ($z = -3.971$, $p < 0.001$, 95% CI [-0.09995, -0.03993]) (Figure 25).

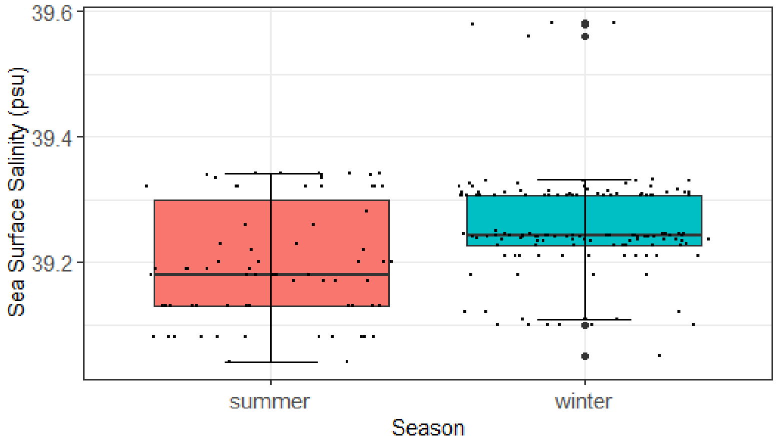


Figure 25. Sea surface salinity of photo-identified bottlenose dolphins compared to the seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

A Mann-Whitney U test showed no statistical difference between the surface chlorophyll concentration of the location for the photo-identified bottlenose dolphins and the seasons (summer seasons: Mdn = 0.08, n = 102; winter seasons: Mdn = 0.07, n = 140) (W = 8068, Z = 1.7262, p = 0.08448, 95% CI [-0.0013, 0.014]) (Figure 26).

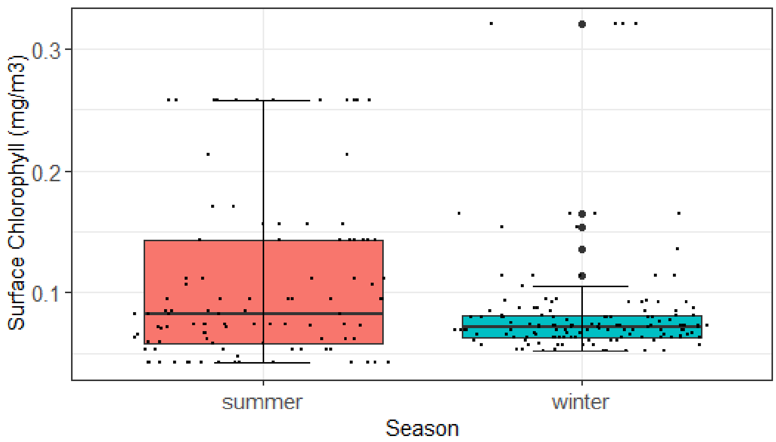


Figure 26. Sea surface chlorophyll of photo-identified bottlenose dolphins compared to the seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

Table 8 summarises environmental variables with the mean value, min, max, SD during seasons.

Table 8. The mean value, min, max, SD of environmental variables during seasons (2015-2021).

Env. variables	Summer season				Winter season			
	Mean	**Min**	**Max**	**SD**	**Mean**	**Min**	**Max**	**SD**
DEP	65.5	2.8	251.4	46.9	87.8	12.4	239.5	43.3
DISTANCE	1751	288.7	6249.8	1275.8	2988.8	417	8255.7	1999.2
SST	22.27	16.4	25.9	2.84	20.68	16.7	23.8	2.62
SAL	39.2	39.04	39.34	0.09	39.26	39.1	39.6	0.08
CHL	0.11	0.04	0.26	0.07	0.08	0.05	0.32	0.05

4.3 Subgroup association

The group size of photo-identified bottlenose dolphins, being the number of dolphins in the group, varied between 1 and 18 dolphins, although mostly in small groups from 1 to 6 dolphins in the group (Figure 27). The mean group size value was 6.227 individuals (SD = 3.25) and a median of 6 individuals. The table shows different ranges, mean values, median and standard deviation over the years (Table 9).

Table 9. The number (n), range, mean, median and standard deviation (SD) of a group size of photo-identified dolphins over the years (2015-2021).

Year	n	Range	Mean	Median	SD
2015	8	2-14	7	6.5	4.24
2016	6	8-18	10.5	8.5	3.99
2017	4	2-4	3.25	3.5	0.96
2018	7	3-9	5.71	5	2.56
2019	12	5-10	7.17	6	1.80
2020	9	3-15	7.11	6	3.89
2021	20	1-10	4.45	4	2.50
Total	**66**	**1-18**	**6.227**	**6**	**3.409**

Figure 27 illustrates the observed group size of identified bottlenose dolphins in the eastern Aegean Sea study area.

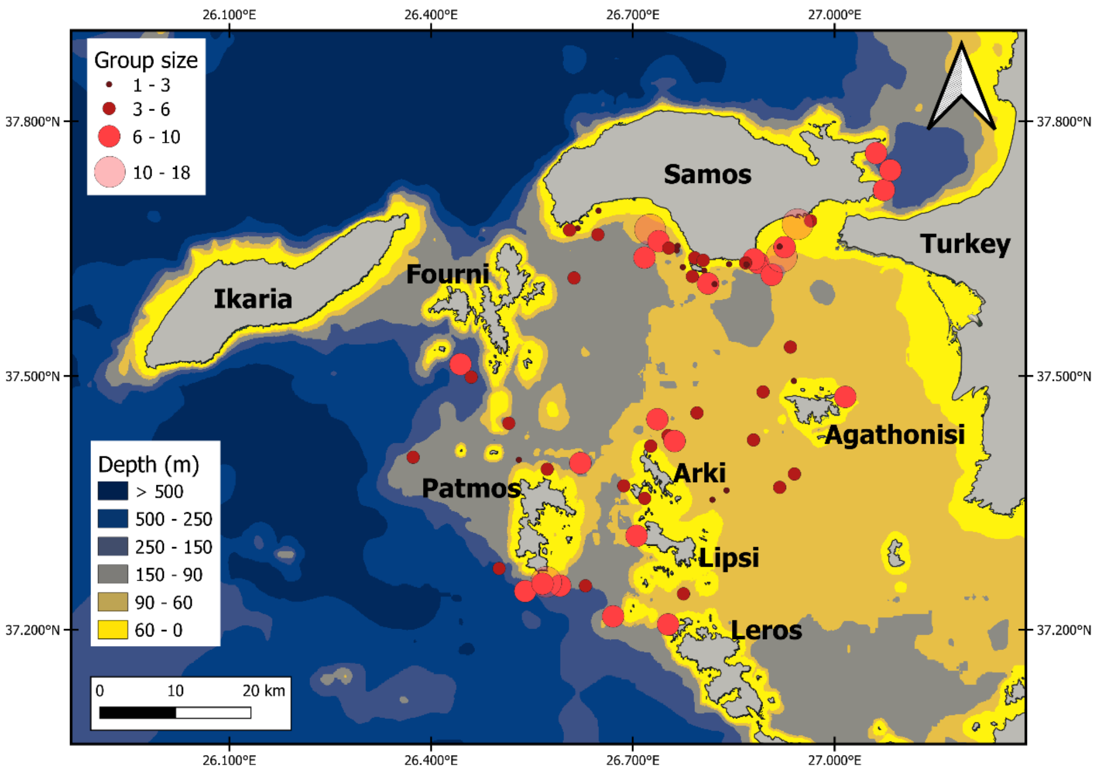


Figure 27. Group size of photo-identified bottlenose dolphins in the eastern Aegean Sea (2015-2021). The size of the circles symbolizes the number of dolphins in the group.

A Mann-Whitney U test was used to test the group size of identified bottlenose dolphins between seasons. The test revealed that the group size of photo-identified bottlenose dolphins was statistically significantly larger during the winter seasons (Median = 6, n = 33) compared to the summer seasons (Median = 4, n = 33) (z = -2.1104, p = 0.03482, 95% CI [-3, 0.001]) (Figure 28, 29).

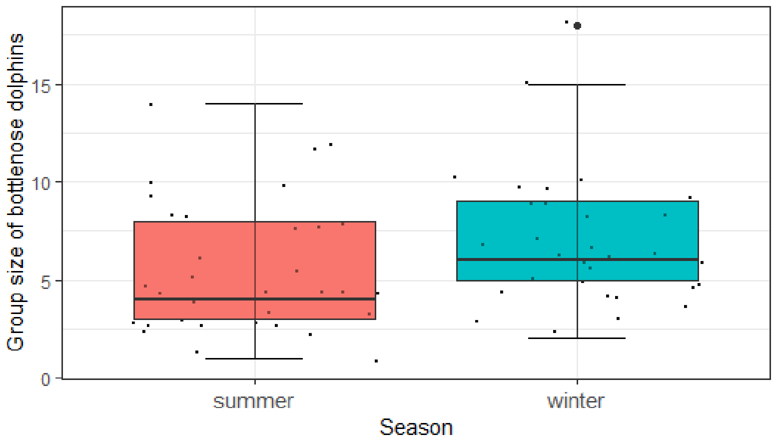


Figure 28. Group size distribution (n=66) of identified bottlenose dolphins between seasons (summer and winter 2015-2021). Boxplot shows minimum value in the data, interquartile range (25th percentile, median, 75th percentile), maximum value in the data and outliers.

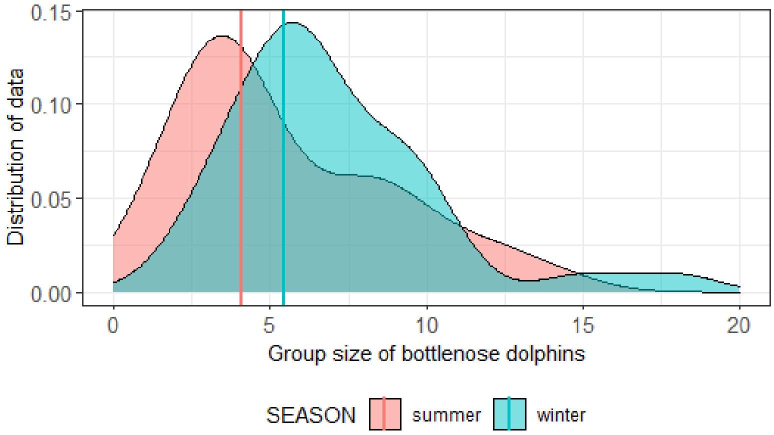


Figure 29. The density of group size (n=66) between seasons (summer and winter 2015-2021). The vertical lines demonstrate the mean value between the summer and winter seasons.

Table 10 displays the number, range median, mean, SD of a group size of identified bottlenose dolphins between seasons (summer and winter 2015-2021).

Table 10. Range, number, median, mean, SD of a group size of identified bottlenose dolphins between seasons (summer and winter 2015-2021).

Season	n	Range	Median	Mean	SD
Summer	33	1-14	4	5.52	3.4
Winter	33	2-18	6	6.94	3.32

A GLM tested the relationship between group size of identified bottlenose dolphins and distance from the coast and depth. The test results showed a non-significant correlation between the distance from the coast, depth, and the group size of identified bottlenose dolphins (Table 11) and (Figure 30).

Table 11. GLM numerical results about the relationship between the group size of identified bottlenose dolphins and the distance from the coast and the depth.

GLM results	Std. Error	z-value	p-value
Distance from the coast	2.558e-05	0.151	0.88
Depth	0.0005636	0.547	0.584

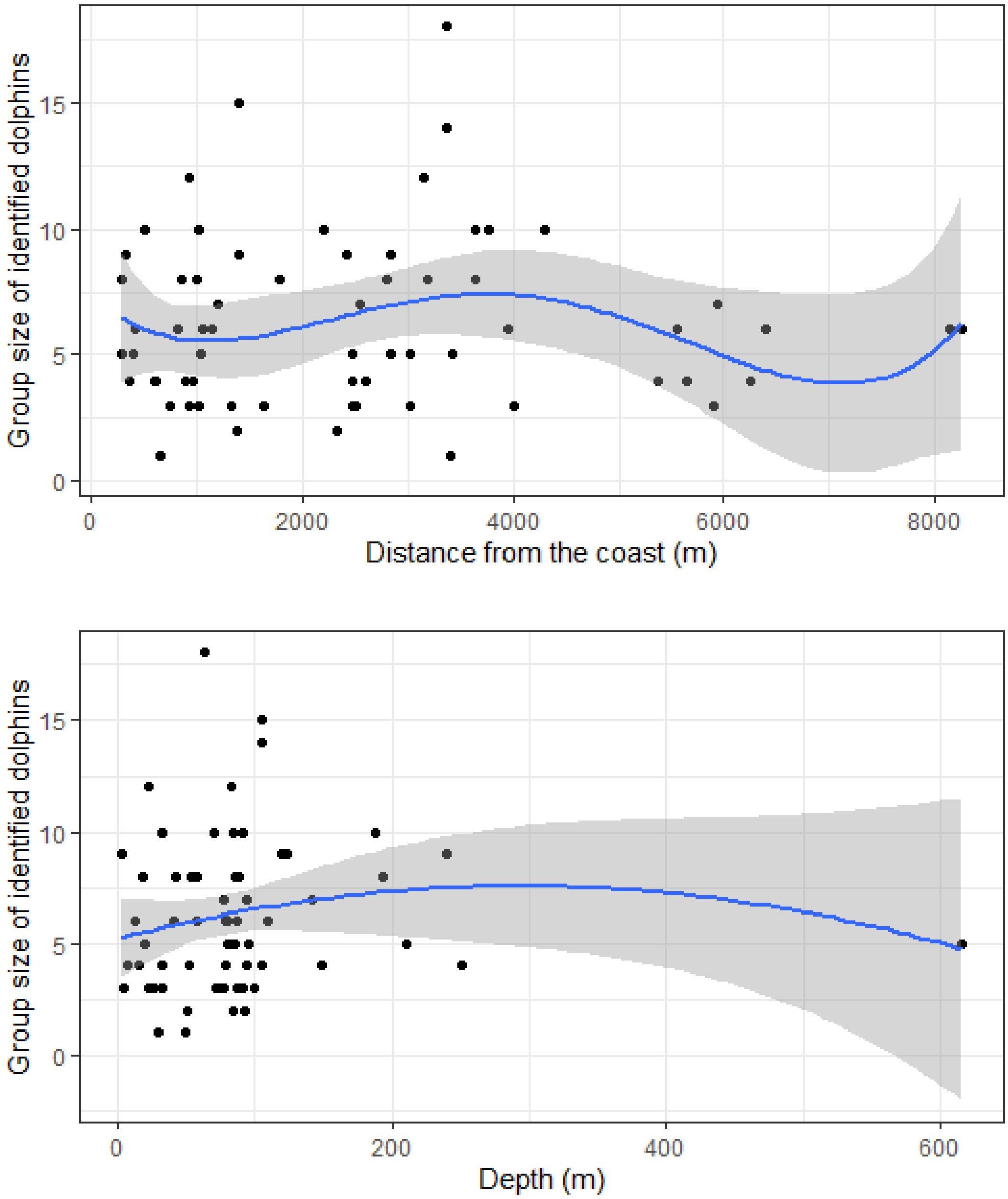


Figure 30. Effect of distance from the coast (a) and depth (b) on group size with 95% CI, (n = 66) of identified bottlenose dolphins (black circles).

5 Discussion

5.1 The abundance of bottlenose dolphins

During the surveys, this study recorded 477 sightings of bottlenose dolphins, 131 dolphins were identified in the photo-ID catalogue, and 250 times were repeatedly resighted in different sightings. From the 131 identified dolphins, 89 dolphins (68%) were sighted in one year (sometimes several times a year), and 42 were resighted (32%) in 2-4 different years. No bottlenose dolphin was resighted every year (Appendix 2). The results show that some dolphins appear to be residents with site fidelity within the area, which was also confirmed in the study by Bearzi et al. (2011).

The encounter rate of sighted dolphins was 2.6 dolphins/100 km and 1.4 photo-identified dolphins/100 km. The lower value for identified dolphins from the photo-ID catalogue was due to the quality and distinctiveness of pictures of dorsal fins. The overall encounter rate was similar in the North Aegean Sea with a value of 2.5 dolphins/100 km (Milani et al., 2018) but was higher in the Amvrakikos Gulf (Greece) with the value of 72.5 individuals/100 km (Bearzi et al., 2008) (the mentioned encounter rate values are for sighted individuals, not photo-identified). According to previous studies conducted in the Mediterranean Sea, densities of bottlenose dolphins ranged between 4 to 37 dolphins/100 km^2 (Bearzi et al., 2008, 2009). The study in the Alborán Sea (Spain) measured a density of 4.9 dolphins/100 km^2 and in the Almería (Spain) of 6.6 dolphins/100 km^2 (Cañadas & Hammond, 2006), while in the Amvrakikos Gulf (Greece) of 37 animals/100 km^2 (Bearzi et al., 2008). This study calculated the mean density of 6.8 dolphins/100 km^2 for sighted dolphins and 3.57 dolphins/100 km^2 for identified dolphins. The comparison of encounter rates and densities values should be taken with circumspection, as it depends on the different characteristics of the environment, prey abundance, method of data collection and other environmental variables (Bearzi et al., 2009).

5.2 Distribution of identified bottlenose dolphins over seven years

The habitat preference for bottlenose dolphins is related to the study area, which rarely exceeds depths of more than 100 m (Figure 21). Identified bottlenose dolphins occurred in the study area with a mean depth of 78 m. There was no record that the identified dolphins in the study area exceeded the depth of more than 260 m, which confirms that dolphins distribute mainly in the shallow water of coastal areas, continental shelf waters and close to the 100 m of isobath (Cañadas et al., 2002; Giannoulaki et al., 2017; Karamitros et al., 2020; La Manna et al., 2020). The results were also in accordance with previous findings, such as Bearzi et al. (2008) in Amvrakikos Gulf and Bonizzoni et al. (2019) in central Greece of Gulf of Corinth, who identified that prefer coastal waters within 300 m. Bottlenose dolphins were most often observed in shallow waters, and the number of dolphins gradually decreased with increasing depth. In this study area, unsuitable habitat for bottlenose dolphins would be in the northern and southern basins of Ikaria, where the depth exceeds 1500 metres. Based on different studies (Giannoulaki et al., 2017; Karamitros et al., 2020; La Manna et al., 2020), bathymetry and distance from the coast were the main substantial predictors that displayed the habitat suitability for bottlenose dolphins.

Identified dolphins occurred close to the shore with an average of 2.45 km from the coast. These findings are in agreement with other studies such as Marini et al. (2015) in the north of the Aegean Sea, who demonstrated a mean value of the distance from the shore of 2.4 km and Giannoulaki et al. (2017) in the Greek Seas (eastern Mediterranean), where a distance from the coast up to 7 km was identified. The distribution of dolphins near the coast within shallow water may be related to prey abundance near-shore (Bearzi et al., 2008; Marini et al., 2015; Karamitros et al., 2020). According to a study by Fernández et al. (2009), stomach samples based on dolphins suggested that the species feeds mainly in coastal waters. For instance, the distribution of European hake, a common prey for bottlenose dolphins, was recorded to a depth of 250 m (Marini et al., 2015). However, dolphins' foraging behaviour and prey preference differ with habitat, season, and other variables (Bearzi et al., 2009).

Salinity did not change substantially in this area during the study period, with an average of 39.24 psu. The measured salinity in the north of the Aegean Sea (Marini et al., 2015) was 37.70 psu with an SST of 17.29 °C. The evaluated data of dolphins in this study, together with a study in the northern part of the Aegean Sea (Marini et al., 2015), agrees with the study of Skliris et al. (2011) that the southern part of the Aegean Sea has a higher salinity and sea surface temperature as well as a higher warming trend than the northern part. In Greek Seas, Giannoulaki et al. (2017) demonstrated the habitat preference of bottlenose dolphins with chlorophyll concentration > 0.135 mg/m^3. In this study of the eastern Aegean Sea, the mean value of identified bottlenose dolphins within chlorophyll concentration was 0.093 mg/m^3. According to the study by Colella et al. (2016), the southern part of the Aegean Sea, in contrast to the northern part, has a lower concentration of chlorophyll.

5.3 Seasonal habitat preference

To better understand marine ecological processes and improve the conservation of cetacean species, it is essential to define which environmental variables are significant in the distribution of habitat preference. The environmental variables analysis focused on habitat selection to help identify potential threats to cetacean species and further conservation plans and management (Marini et al., 2015). This study used bathymetry, distance from the coast, salinity, sea surface temperature, and chlorophyll concentration to investigate the different distribution of identified dolphins between the summer and winter seasons. The results showed a significant influence of bathymetry, distance from the coast, salinity, and sea surface temperature on the distribution of identified bottlenose dolphins between summer and winter. However, the distribution of identified bottlenose dolphins occurrence was not significantly correlated to chlorophyll over seasons, which could be due to the lower concentration of chlorophyll and less seasonal effect in the study area (Colella et al., 2016).

During the study period in the summer seasons, photo-identified dolphins occurred at an average depth of 65.5 m with an average distance from the coast of 1.75 km. Dolphins were observed in waters with higher temperatures with a mean value of 22.3 °C, salinity of 39.2 psu and chlorophyll concentration of 0.11 mg/m^3. Higher chlorophyll concentration is in nearshore waters, near rivers related to nutrients (Colella et al., 2016) and has an adequate role in fish abundance in coastal waters (Cañadas & Hammond, 2006). The seasonal distribution of dolphins near the coast and shallow depths may be related to prey availability and abundance (Fernández et al., 2009; Marini et al., 2015; Inch et al., 2018) or due to the period when dolphins give birth to find the sheltered in coastal waters for calves (Robinson et al., 2007; Wells & Scott, 2009; Giannoulaki et al., 2017). The main calving season of bottlenose dolphins is most in the spring/summer months, with more significant increases during July and August (Bearzi et al., 1997; Wells & Scott, 2009). During the calving season, warmer water is essential for mothers during the lactation period, along with enough food for their calves to grow faster (Mann et al., 2021; Wells & Scott, 2009). The warmer SST may correlate with the calving season as an important area for breeding grounds. SST may also be associated with prey availability as a valuable part of the food web (Bearzi et al., 2008; La Manna et al., 2016).

Compared to the winter season, dolphins' distribution was in deeper depths with a mean value of 88 m and far from the coast with a mean value of 2.98 km. Identified dolphins occurred in waters with an average sea SST of 20.7 °C, SAL of 39.1 psu and CHL of 0.08 mg/m^3. Based on literature (Wells & Scott, 2009), lactating females with their calves feed in the area near the coast, while adolescent, adult males, and resting females forage far from the coast in larger group sizes. These findings could demonstrate the seasonal changes in the bottlenose dolphins' habitat (Giannoulaki et al., 2017). However, further investigation is needed to resolve whether the distribution of dolphins is related to prey availability and calving season.

5.4 Group association

The group size of identified dolphins ranged from 1 to 18 individuals but was most often observed in smaller groups of 6 dolphins (Figure 27). The average group size in the Mediterranean coastal waters is mostly around 7 individuals (Bearzi et al., 1997; Bearzi & Fortuna, 2006), confirming that the mean group size was 6.23 individuals in the study area. Compared to other Mediterranean areas, in the North-eastern Adriatic Sea (Kvarneric, Croatia) and Eastern Ionian Sea (Greece), the mean size was 6.8 individuals (Bearzi et al., 1997), whilst in the Aeolian Islands (Italy), the mean size was 12 dolphins (Bearzi et al., 2009). However, these results cannot be clearly compared, as they may differ in the method of data collection. In addition, the group size may vary according to other factors such as biogeographic area, prey availability, and feeding behaviour (Bearzi et al., 2009; Wells & Scott, 2009).

The group size of identified dolphins was statistically significantly larger during the winter season (mean size of 6.92 individuals) than in the summer season (mean size of 5.52 individuals). The changing average group size during the years and seasons could be related to the area, distribution and abundance of prey (Shane et al., 1986; Bearzi et al., 1997; Wells & Scott, 2009) or the composition of groups with calves, juveniles (Bearzi et al., 1997). Based on the study, Bearzi et al. (1997) observed that the group sizes with calves or juveniles were larger than when only the adult groups were present.

According to studies about fisheries from this study area (Inch et al., 2018; Vlachopoulou et al., 2013), prey may be scattered with limited resources due to fishing activities. The small size of a group may be related to foraging behaviour, where each individual in the group may have a better chance of catching limited prey (Bearzi et al., 1997). Habitat fragmentation may also affect changes in group sizes (Bearzi & Fortuna, 2006; Boisseau et al., 2010; Hammond et al., 2012; Tonay et al., 2015). In contrast, more prominent groups may benefit from cooperation in feeding where more prey is formed or travelling or for social causes for strengthening the social relations of the community, including mating (Bearzi et al., 1997). The GLM showed no significant relationship between group size of identified bottlenose dolphins with depth and distance from the coast. As confirmed by the literature (Wells & Scott, 2009), the group size of dolphins may not increase linearly with distance from the coast and depth.

5.5 Threats

The eastern area of the Aegean Sea is a hot spot area for the bottlenose dolphins population (Giannoulaki et al., 2017). However, human threats and cumulative pressure highly impact coastal waters in this study area (Coll et al., 2010). The Mediterranean Sea, especially the coastal zone (Coll et al., 2010), are currently exposed to many cumulative human-environmental stressors, such as the depletion of prey deportation caused by overfishing, habitat loss, environmental degradation, and pollution of different sorts and fishery-related mortality (Bearzi & Fortuna, 2006; Bearzi et al., 2008; Schipper et al., 2008; Bearzi et al., 2009; Lejeusne et al., 2010).

Additionally, the study from Samos by Vlachopoulou et al. (2013) identified the destructive fishing methods of trawling within a 1.8 - 2.7 km distance from the coast, which directly impacts the decline of fish stocks in this study area and the associated consequences for dolphins distribution. The main route of the trawler and abundance of fishing boats were observed in the southern part of Samos and in the northeastern part of the island of Archi (Appendix 1) (Vlachopoulou et al., 2013; Inch et al., 2018), where the significant number of identified bottlenose dolphins was observed (Appendix 3). The identified dolphins in this study occurred on average 2.5 km from the coast. Trawling up to 1.8 km from the shore is legally prohibited under both the EU and Greek law, but fishing activity was also identified in the immediate vicinity of the coast (Vlachopoulou et al., 2013). These practices have a cumulative impact on the marine ecosystem and may lead to functional and structural changes and degradation (Coll et al., 2010; Vlachopoulou et al., 2013).

In the last few years, other impacts that threaten dolphins in the area are pollution, anthropogenic noise, and increased tourist flow (Pietroluongo et al., 2020). According to Inch et al. (2018) and La Manna et al. (2013), vessels depending on size and type with different noise pollution may change the dolphins' distribution. The same fact confirmed a study in Italy by La Manna et al. (2013) that dolphins preferred to leave and avoid locations with intense disturbance by motorboats. Global warming, which is very pronounced in the Aegean Sea with heatwaves during summer seasons, may lead to poorer health for dolphins, habitat loss, differences in prey abundance and distribution, changes in behaviour, and may lead them to migrate to higher latitudes (Cañadas et al., 2002; Bearzi & Fortuna, 2006; Simmonds, 2017; Kebke et al., 2021). For instance, a recent study (Mann et al., 2021) has published significant calf mortality of the Indo-Pacific bottlenose dolphins due to solid heatwaves caused by climate change. These facts. Thus, based on this information, it is not surprising that the subpopulation of bottlenose dolphins in the Mediterranean has been declining in recent decades. Nevertheless, due to its opportunistic and flexible behaviour, the bottlenose dolphin appears to be more resilient to negative human influences, such as habitat degradation and overfishing, than other coastal cetacean species, which can adapt to these changes (Bearzi et al., 2009).

5.6 Conservation

Based on national protection, there is a need to implement direct measures and focus more on the threats, such as overfishing, prey depletion, disruption by boats, habitat degradation, reduction of vessels or pollutants to conserve bottlenose dolphins (Bearzi et al., 2009). As top predators, bottlenose dolphins indicate the health of marine ecosystems, and their population decline can significantly impact local ecosystems and local people (Carlucci et al., 2018; Karamitros et al., 2020). MPAs in the Aegean Sea are underrepresented, particularly in the epipelagic waters close to the shore (Micheli et al., 2013). In the Greek waters by the ACCOBAMS, there is only one region in the Ionian Sea (Amvrakikos Gulf) under protection for bottlenose dolphins. This study area is part of the Central Aegean IMMA (IUCN-MMPATF, 2017) due to the critically endangered Mediterranean monk seal. Unfortunately, the bottlenose dolphin was identified as a secondary species that did not meet the selection criteria for IMMA protection. In the Aegean Sea, bottlenose dolphins' habitat often includes the international waters of more than one country (Giannoulaki et al., 2017), which makes regulations challenging to enforce between countries. Fisheries laws are not actively implemented, and harmful and illegal fishing practices are not enforced sufficiently by the Greek government (Vlachopoulou et al., 2013; Marini et al., 2015; Inch et al., 2018). There is a need to set up comprehensive management and conservation strategies. In addition to control and enforcement, cooperation with artisanal fishermen and support of local fishing communities for sustainable fishing is suggested (Vlachopoulou et al., 2013).

5.7 Limitations

The study area was not evenly surveyed, and the boat survey efforts were primarily around the islands of Samos and Lipsi. During the study period (2015 - 2021), the same number of survey efforts has not been maintained over the years, due to which the number of sightings of bottlenose dolphins is also different. Additionally, data were not equally collected each month of each year, mainly due to bad weather conditions. During December, no individuals were identified in the database due to low boat survey effort. Moreover, few of the identified resightings were in January and February. In 2017, few individuals in the catalogue were identified because not all dolphins could be identified due to distinguishing features or picture quality. Limitations of the photo-ID method included insufficient quality of pictures with contrast, focus, and angles. Future improvements of PQ would enhance the interpretation and analysis of data.

6 Conclusion

This thesis studied the seasonal habitat preference and group association of identified bottlenose dolphins over seven years in the eastern Aegean Sea. The photo-identification method achieved these objectives, an effective tool for identifying bottlenose dolphins in this study area. The dataset from the photo-ID catalogue was used to examine the influence of environmental variables (bathymetry, distance from the coast, salinity, sea surface temperature, chlorophyll concentration) and the group size on the distribution of dolphins between summer and winter. Furthermore, this study analysed the correlation between group size of identified dolphins with depth and distance from the coast.

In the eastern Aegean Sea, it was catalogued 131 photo-identified bottlenose dolphins between January 2015 to August 2021. Photo-identified and resighted bottlenose dolphins were observed in coastal areas and waters surrounding the islands, shallow waters < 250 m, close to the shore < 8.2 km, sea surface temperature > 16 °C, with high salinity > 39 psu and with a mean value concentration of 0.93 mg/m^3 during the study period. The four environmental variables (bathymetry, distance from the coast, salinity, sea surface temperature) significantly influenced the distribution of identified bottlenose dolphins between seasons, except for chlorophyll concentration. During summer seasons, bottlenose dolphins showed habitat preference in shallower waters, closer to the shore and with higher sea surface temperature compared with the winter season. These findings of significant environmental variables and a seasonal effect showed potential habitat preference for bottlenose dolphins.

The mean group size of identified bottlenose dolphins was 6.27 individuals, and the group size ranged between 1 and 18 depending on the seasons. A Mann-Whitney U test compared group size between summer, winter and showed a significant difference in these variables. The group size of identified bottlenose dolphins was larger during the winter than in the summer. Thus, the second hypothesis, a difference in the group size of identified bottlenose dolphins related to the seasons, was accepted. GLM demonstrated no correlation in group size of identified bottlenose dolphins with depth and distance from the coast. As a result, the third hypothesis was rejected.

The photo-ID catalogue from 2015 to 2021 provides crucial data for future research studies about bottlenose dolphins for the Archipelagos Institute of Marine Conservation. The knowledge gained about habitat preferences and subgroup associations may also enhance the ability to respond to environmental and anthropogenic impacts and improve conservation planning initiatives in this study area. Effective conservation plans should consider the relationships between environmental variables and seasons, especially climate change and negative anthropogenic influences. Additionally, the results of this work can contribute to suitable habitat modelling and management strategies for cetacean species in the future, which can help establish MPAs. In conclusion, it is necessary to choose a proactive approach to reduce future environmental and human-caused threats to maintain the status of bottlenose dolphins, mainly since the population is vulnerable in the Mediterranean Sea.

Future research is needed to understand bottlenose dolphins' habitat preferences adequately. Consequently, future studies should focus on critical areas for bottlenose dolphins (shallow waters, close to the coast) and concentrate on their conservation and strategies to reduce anthropogenic threats. In addition to the studied environmental variables and the seasonal and year-on-year fluctuations, it would be interesting to examine other variables, including sardine presence along with productivity, prey distribution, the interaction of prey and cetaceans with crucial fishing grounds, diet composition, responses to anthropogenic influences, suitable conditions for calving, interactions with other cetacean species, or the impact of fish farms. Findings on the interaction of prey and cetaceans with crucial fishing grounds may help to investigate habitat preferences further and to ensure potential conservation and fishing restriction measures in areas with significant dolphin populations. In addition, spatio-temporal analyses or prediction models could contribute to the conservation measures of coastal species used in other studies.

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8 List of pictures and tables

List of pictures

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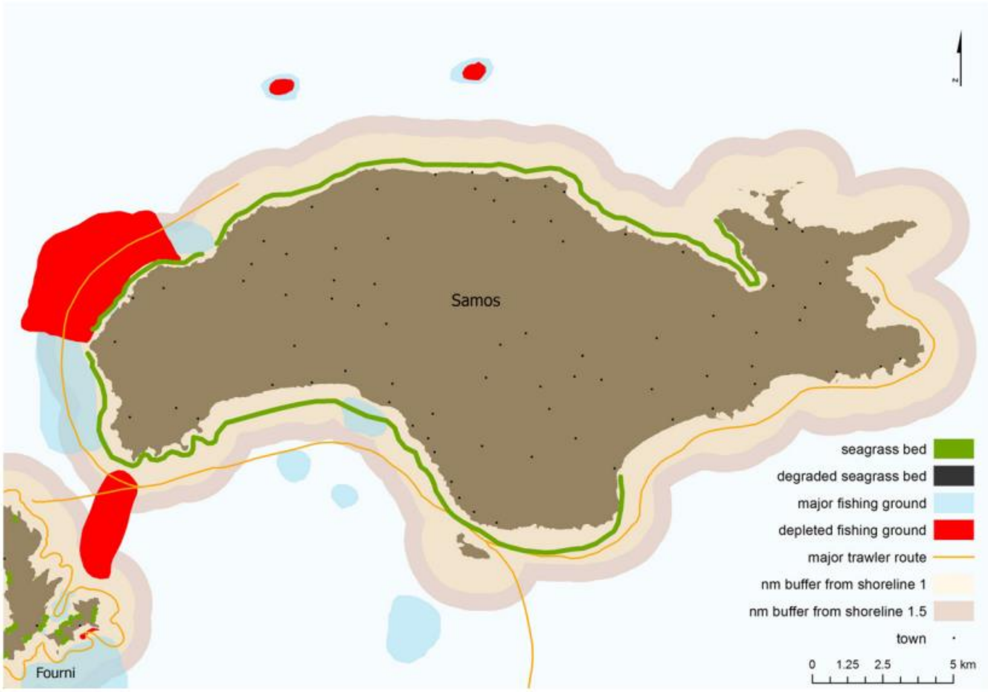
9 Appendices

Appendix 1. The map shows trawlers' routes, depleted fish stocks for one species and current and destroyed seagrass beds around Samos (Tammi, 2011; Vlachopoulou et al., 2013).



Appendix 2. Sighting histories of bottlenose dolphins that were photo-identified in the catalogue from 2015 to 2021. Numbers in the grey columns demonstrate the total resighting of bottlenose dolphins.

ID	2015	2016	2017	2018	2019	2020	2021
001_15S	2	1		1			
002_15S	1	1					
003_15W	3						
004_15S	1						1
005_15W	1						2
006_15S	1	1					
007_15S	1						
008_15W	1	1					2
009_15W	1						
010_15W	3						
011_15S	3	1					
012_15W	1						
013_15W	2						3
014_15W	1				1		
015_15S	1						
016_15S	2	1		1	1		
017_15W	1						
018_15S	1						
019_15S	1						
020_15S	1						
021_15S	1						

ID	2015	2016	2017	2018	2019	2020	2021
022_15W	1	3					2
023_15S	1			1	1		
024_15W	1	1					
025_15W	1						
026_15S	1			1			1
027_15W	1	2					
028_15S	2	4		1			
029_15S	1		1	1		2	
030_16S		1					1
031_16S		1		2			
032_16S		1					
033_16S		1		1			
034_16W		1					
035_16W		1					
036_16W		1					
037_16W		2					
038_16W		1					
039_17S			1	2			
040_17S			1		2		
041_17S			1	1	1	1	
042_16S		1			2		
043_21S							2
044_17W			2	1			1
045_18S				1	1		
046_18S				1			
047_18S				1			
048_18S				1			
049_18W				1			
050_18S				1			
051_18S				1			
052_18W				1	1	2	
053_18S				1			
054_18W				1			
055_18S				1		1	
056_18S				1			
057_18W				1	1	1	
058_18S				1		2	1
059_19S					1		
060_19W					2		
061_19S					2		
062_19W					1		
063_19W					2	1	
064_19S					4		
065_18S				1	1		1
066_19S					1		
067_19W					3	1	
068_19S					2		
069_19W					1		
070_19S					1		1

ID	2015	2016	2017	2018	2019	2020	2021
071_19S					1		
072_19W					1	2	1
073_19W					1	1	
074_19S					1		
075_19W					1		
076_19S					1	1	1
077_19S					1		
078_19W					2		
079_19S					1	1	
080_19S					2		
081_19W					1		
082_19W					1		
083_19S					1		
084_19S					1		
085_19S					1		
086_19W					1		
087_20W						1	
088_20W						1	
089_20S						2	
090_20S						1	
091_20W						1	
092_20S						3	
093_18S						1	
094_20W						1	
095_20S						1	1
096_20S						1	
097_20W						1	1
098_20S						1	2
099_20S						1	
100_20S						2	1
101_20S						1	
102_20W						1	
103_20S						1	
104_20S						1	
105_20S						2	
106_20W						2	
107_20S						2	
108_20S						2	
109_20S						1	
110_20S						1	
111_20S						1	
112_21S							2
113_21S							1
114_21S							1
115_21W							1
116_21W							1
117_21S							1
118_21S							1
119_21S							1

ID	2015	2016	2017	2018	2019	2020	2021
120_21W							3
121_21S							1
122_21S							1
123_21W							3
124_21S							1
125_21S							1
126_21W							1
127_21S							1
128_21W							1
129_21W							2
130_21S							1
131_21S							1

Appendix 3. Transect lines of boat surveys and sighting of identified bottlenose dolphins carried out from January 2015 to August 2021.