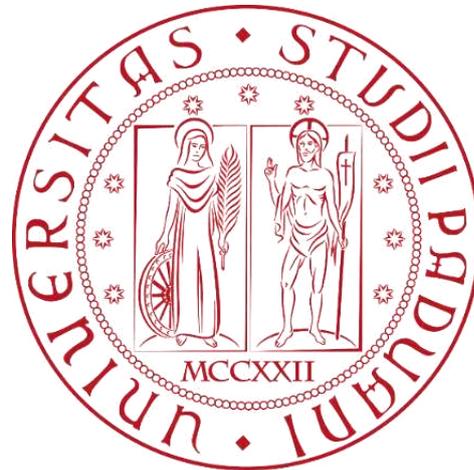


UNIVERSITÀ DEGLI STUDI DI PADOVA

DIPARTIMENTO DI BIOLOGIA

Corso di Laurea magistrale in Marine Biology



TESI DI LAUREA

Social structure of the
bottlenose dolphin (*Tursiops truncatus*)
in the Thermaikos Gulf

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ANNO ACCADEMICO 2024/2025

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ABSTRACT

This study represents the first attempt to investigate the social structure of the common bottlenose dolphin (*Tursiops truncatus*) inhabiting the Thermaikos Gulf, Greece. This area is characterized by high socio-economic and ecological value, but it is subject to intense anthropogenic pressure. Studying the ecology, social organization, and behaviour of the local bottlenose dolphin population is therefore crucial to assess the condition of the species and to build effective management and conservation strategies. Between 2021 and 2025, 85 boat-based surveys were conducted, resulting in 45 sightings and the photo-identification of 133 individuals. Association analyses using the Half-Weight Index (HWI) were performed in SOCPROG 2.3 on 26 well-marked and frequently re-sighted individuals. The overall mean HWI (0.21) was consistent with other values reported for other Mediterranean coastal populations, indicating a fluid fission–fusion society. Although permutation tests did not reveal statistically significant preferred associations, results were close to the threshold of significance and suggested a tendency to non-random relationships and the presence of a probable social pattern. Cluster and network analyses identified three loosely connected subgroups, showing variable degrees of sociability within the population. This study provides a valuable basis for future research on the species in the region and highlights the importance of consistent, long-term, and standardized monitoring to support effective conservation planning in one of the most anthropogenically impacted areas of the eastern Mediterranean.

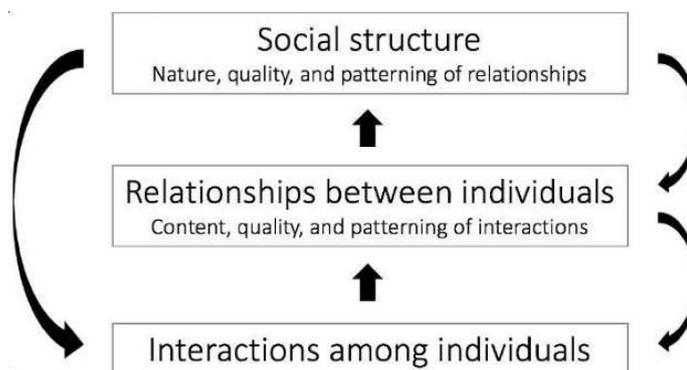
1. INTRODUCTION

1.1 Animal social structure

Social structure is defined as the pattern of social interactions and the resulting relationships that develop among conspecific individuals that live in proximity and regularly interact with one another (Kappeler & van Schaik, 2002). These relationships can include cooperation, competition in the search for mates or resources, collaboration for parental cares, and in some cases even cannibalism. For this reason, social structure plays a central role in the population biology, influencing processes such as gene flow, reproductive success or spatial distribution (Wilson, 1975).

The range of possible social structures is very wide, from simple and loosely connected systems, often constrained by anatomical, physiological, or ecological factors, to highly complex and dynamic societies, especially in cognitively advanced mammals such as primates and cetaceans. This variability makes the task of describing and classifying societies of mammal species especially challenging (Whitehead, 2008). The most commonly cited and recognized conceptual basis for defining social structures is the framework proposed by Robert A. Hinde (1976). This framework establishes a hierarchy of three levels of organization, which are in order interactions, relationships and finally social structure (Figure 1). The first level consists of direct active exchanges between individuals that can be measured and quantified in frequency, duration, and type. Relationships are then formed from repeated interactions and are distinguished by their stability over time and by the type of behaviours present. Finally, the last level is represented by the social structure of a population which includes the nature, quality, and structure of the relationships among its members (Hinde, 1976).

Figure 1. Hinde's framework for the analysis of animal social structures (Whitehead, 2008).



In order to analyse the social structure of a population in the way proposed by Hinde it is important to collect detailed information about the associations between individuals for a certain period (Whitehead, 2008). However, many animals with complex and interesting social structures live in environments that make it challenging to collect detailed information about their interactions. A clear example is represented by cetaceans, which inhabit aquatic environments.

1.2 Importance of studying the social structure of cetaceans

The study of cetaceans is of fundamental importance for both ecological research and conservation science. As they occupy a high trophic level, they are key biological indicators of the overall health status of marine environments. In fact, fluctuations in the abundance of these apex predators may have negative effects on the whole ecosystem (Hoyt, 2012). Moreover, the fact that several cetacean species are classified as vulnerable or endangered highlights the urgent need for their protection and preservation (Braulik et al., 2023). Their educational, scientific, and economic value makes them important flagship species for the protection of the aquatic environment and the creation of large marine conservation areas (Notarbartolo & Würsig, 2022).

Understanding in particular the social structure of cetaceans is crucial to study their behaviour, their ecology, and ultimately to assess the condition of the species and therefore implementing their management and building effective conservation strategies (Whitehead, 2008).

1.3 *Tursiops truncatus*

1.3.1 General information and distribution

The common bottlenose dolphin, *Tursiops truncatus* (Montagu, 1821), is one of the most intensively studied and widely known cetacean species globally (Bearzi et al., 2009). It is a medium-sized delphinid, characterized by a robust body with a typical counter-shaded coloration, going from various shades of grey to a noticeably lighter ventral area and a curved dorsal fin. Adults usually range from 1.9 to 3.8 meters in length, with an average weight of 200 kilograms and males are generally larger than females (Jefferson et al., 1993). The bottlenose dolphins have a very wide distribution, inhabiting temperate and tropical marine waters all around the world (Figure 2). The geographic range of the species appears to be limited by the temperature and prey availability (Wells & Scott, 2009).

Figure 2. Global distribution of *Tursiops truncatus* (IUCN Red List, 2019).



They can have different distribution patterns, exhibiting high site fidelity or, in the contrary, showing seasonal movements of hundreds of kilometres (Pace et al., 2022). In the Mediterranean Sea, it is one of the most frequently observed cetaceans (Notarbartolo & Demma, 2004). Within this region, bottlenose dolphins occupy different habitats, usually inshore and near the coast, occurring primarily in waters shallower than 100 metres, but can also be found in straits, gulfs, lagoons, semi-closed eutrophic waters, and steep coasts without a continental shelf (Bearzi et al., 2008a). In the case of Greek waters, this species is the most commonly found and the second most abundant cetacean after the striped dolphin (*Stenella coeruleoalba*) (Frantzis, 2009). Although bottlenose dolphins are generally associated with deeper coastal waters, they can exhibit high site fidelity, even under very particular and challenging environmental conditions, as observed in the Amvrakikos Gulf population, which is known to live in 30 metres deep waters (Bearzi et al., 2008a).

1.3.2 Social structure in the Mediterranean Sea

According to genetic studies, bottlenose dolphins living in the Mediterranean Sea are divided in two ecotypes, pelagic and coastal. “Ecotype” is defined as a genetically distinct population within a species, which is adapted to a specific environment, due to natural selection. The coastal ecotype is primarily found in shallow waters, has strong site fidelity and show tight social groups genetically distinct due to limited gene flow and habitat adaptation. The pelagic ecotype, on the other hand, inhabits deeper offshore waters and show a more fluid social structure and greater migration, with consequent higher gene flow. This last ecotype is considered genetically similar to the North Atlantic pelagic dolphins (Gaspari et al., 2015). The population is further divided into resident sub-units with a certain degree of mutual isolation and specialised to their environment, even across small geographic scales (Gnone et al., 2022).

Most of the information about the behaviour of the species comes from coastal waters’ groups, which reportedly interact with humans. This species

typically shows a very fluid social organization, in which the composition of the groups frequently changes in size and composition over small temporal and spatial scales, often also within a few hours on the same day. Individuals may associate with others or travel alone (Bearzi et al., 1997). Group size is usually correlated to the behavioural context. In fact, several types of bottlenose dolphin groups have been described. Smaller groups are generally observed during diving, foraging, or traveling activities, while larger aggregations are more common during social interactions (Affinito et al. 2019). This type of complex and flexible interaction between individuals is called by experts “fission-fusion society” (Wells et al., 1987; Wells, 2003). A “group” is considered when a number of dolphins are swimming together and interacting within an area of 100 metres radius while engaged in similar activities (Wells et al., 1987). Typically, they are found in groups of 2 to 15 individuals (Wells & Scott, 2009). However, despite this dynamic group structure, bottlenose dolphin societies are characterised by long-term associations and their social systems appear to be stable over time (Gowans et al., 2007).

In some areas this species has also been observed in associations with other cetacean species, like the common dolphins (*Delphinus delphis*) (Bearzi et al., 2003) and long-finned pilot whales (*Globicephala melas*) (de Stephanis et al., 2008).

Bottlenose dolphins can have different types of relationships, which are female-female, male-male, mixed-sex and mother-calf associations, creating basic social units that can include nursery groups, groups of mixed sex juveniles or strongly bonded pairs. They express affiliation by physical contacts, proximity, and synchronous swimming (Connor et al., 2000). Firstly, females tend to associate with other females of similar age and reproductive status, so with or without calf, or rarely can also be solitary. The reasons behind this bond can be summarized with cooperation, defence from predators or male harassment, and finally, competition over resources (Wells et al., 1987).

On the other hand, males travel alone or form pairs or trios to cooperate in the courtship of females and compete and defend themselves from other alliances attacks (Connor et al., 1992). Because of these interactions with other male dolphins, they usually show more scars on their bodies and notches on the dorsal fins than females (Wells & Scott, 2009). Associations between males and females occur much more often during the mating season, although mixed-sex groups have also been reported (Bearzi et al., 1997).

Finally, mother and calf spend most of their time together, swimming synchronously or within 10 metres from each other (Figure 3). The calf remains dependent during the first few years of life, usually 3 years (Connor et al., 2000).

Figure 3. A mother bottlenose dolphin (Tursiops truncatus) with her calf exhibiting typical mother–calf association behaviour.



1.3.3 Ecological drivers of social structure

The composition and size of bottlenose dolphin groups are potentially affected by several ecological factors, including prey distribution and availability (Gowans et al., 2007), habitat structure (Wiszniewski et al., 2010), cultural transmission (Sargeant et al., 2005), breeding success

(Möller & Harcourt, 2008), predation risk (Heithaus & Dill, 2002), male competition (Connor et al., 2001), and risk of infanticide (Dunn et al., 2002). The effects of past extermination campaigns, fishery activity and several different threats like noise pollution, have also contributed to the patchiness of their distribution (Bearzi et al., 2008b).

Similarly to the other species, the stability of relationships is primarily connected to the trade-offs between benefits of living in group, such as protection from predators, cooperation and calf care, and the costs, including competition for food and the risk of disease and parasites transmission (Krause & Ruxton, 2002). Some studies also recognised that there are individual variations, likely due to gender differences, life stage and site fidelity, even more complex given their behavioural plasticity (Arcangeli et al., 2017).

The key drivers that are cited the most in studies to influence the presence of bottlenose dolphins are seasonality and habitat characteristics. In fact, habitat topography and environmental factors like surface salinity, productivity, depth and temperature create barriers, restricting movement and dispersal. Furthermore, the seasonal changes of these abiotic factors influence the biotic ones (Wiszniewski et al., 2010). Different population structures and genetic differentiation have been found to coincide with different types of habitats (Natoli et al., 2005). However, the most important driver according to most studies is prey availability and its seasonal distribution pattern, with environmental variables generally playing a secondary role. In fact, fluctuations in local food resources can affect the size and stability of social groups (Connor et al. 2000). Bottlenose dolphins are more likely to form big groups when prey is abundant and widely distributed, while group size tend to be smaller when food is scarce, leading to more competition. Living in group offers various benefits, including a more efficient foraging, cooperative hunting minimizing search time and effort and defence of food resources from other species (Gowans et al., 2007). Moreover, prey preferences can also vary with the seasons, in response to both prey distribution fluctuation and changes in metabolic needs (Wells et

al., 1999a). As already stated, changes in abiotic factors such as salinity and temperature, indirectly influence the distribution of bottlenose dolphins, as they affect the abundance and distribution of their preys. For example, reduced cloud coverage leads to increased productivity, a greater concentration of zooplankton and consequent changes in the entire food web (Cubero-Pardo, 2007).

Sex-based differences were also found, as distribution of female dolphins is often related to habitats where food resources are more abundant, and the risk of predation is lower. The distribution and behaviour of males, on the other hand, is usually connected more to the female presence than to availability of preys (Cubero-Pardo, 2007).

Regarding predation, it is thought to be a minor variable in the Mediterranean Sea. Bottlenose dolphins are preyed occasionally by sharks, such as white sharks (*Carcharodon carcharias*), or by other cetaceans, like the false killer whale (*Pseudorca crassidens*). However, the numbers of large sharks have declined drastically over the past century in the Mediterranean Sea, and the false killer whales are rare in the basin (Bearzi et al., 2008b).

Additionally, anthropogenic activities can strongly affect their behaviour, causing changes in the habitat use and altering social structure and organization. In fact, this species unfortunately faces several significant threats. After experiencing a population decline caused by historical direct killing campaigns conducted until the 1960s, it is currently negatively impacted by rapid degradation of the marine environment, ongoing by-catch in fishing gears, and ship strikes (Bearzi et al., 2012). According to the IUCN Red List criteria, the Mediterranean subpopulation of bottlenose dolphins was classified as “Least concern” in 2021 (Natoli et al., 2021), with a previous assessment of “Vulnerable” in 2009. However, specific subunits that live in particular environments are considered more threatened. For example, the population living in the Amvrakikos Gulf was ultimately assessed as “Critically Endangered”, with a decreasing current population

trend, probably given by their geographical isolation, low genetic diversity, and the deteriorating quality of the waters of the gulf (Gonzalvo & Notarbartolo, 2021).

Moreover, the species is increasingly in conflict with small-scale fisheries, such as trammel nets, and trawlers. Dolphin–trammel net interactions have been reported to be impactful on movement patterns and habitat selection. In addition, in many areas, bottlenose dolphins have learned to follow trawlers, as they provide an opportunistic, easy, and reliable source of food. It has also been observed that individuals tend to associate with others that have similar foraging preferences and experience, needed to interact safely and efficiently with fishing boats (Pace et al., 2012). However, they directly damage the nets and “steal” the fish, reducing catches and causing negative reactions of fishers, and can also injure themselves (Bearzi et al., 2008b; Blasi et al., 2015).

Finally, climate change and global warming are causing dramatic effects on their behaviour and distribution, despite it is impossible at present to predict the future impacts (Cañadas & Hammond, 2008).

1.3.4 Methodologies and variables for studying social structures

There are a variety of different methods to study the social structure of cetaceans, each with strengths and limitations. For this reason, scientists usually combine multiple approaches to have a more comprehensive understanding.

The most used method is the systematic visual survey, which involves counting and observing animals using a boat moving along transects, with the goal of estimating species density and study their behaviour. One of the methods is using random transects to ensure homogeneous and consistent coverage of the area of interest. Navigation is conducted *ad libitum*, meaning the researchers sample different portions of the study area on different days and they choose routes based on weather conditions, ensuring diverse spatial representation and avoiding sampling bias (Bearzi et al., 2008a). During the survey, data on location using a GPS,

environmental conditions, sea state, group size, age and behaviour of the individuals, such as feeding, travelling or socializing, are also collected. This method provides direct observation, but it is weather-dependent and limited by observers' bias.

Photoidentification techniques is the mostly commonly used approach for studying social structure in cetaceans, as it is non-invasive and cost-effective, without the necessity of capturing the animals or applying external marks (Boyd et al., 2010). Individuals are identified based on unique features such as dorsal fin shape, scars, specific long-lasting marks and notches (Figure 4). Repeated sightings help researchers tracking population dynamics (Würsig & Würsig, 1977). Photoidentification data are typically collected during boat surveys and efforts are made to obtain high-quality images of both sides of each individual in the group. Subsequently, images are processed, and high-resolution ones are selected in order to create a catalogue, used to compare and match the individuals in the pictures (Boyd et al., 2010).

*Figure 4. Examples of dorsal fins of individuals of *Tursiops truncatus* showing natural markings such as scars and notches used for photo-identification.*



Other methods to study social structure are short-term capture and release, genetic sampling, systematic tagging, hydrophones to record vocalizations, or radiotracking, but these techniques are more invasive and costly (Bearzi et al., 2009).

The study of bottlenose dolphin social structure relies not only on observations of group composition and behaviour but also on the categorisation of individuals by sex and age, which is important to examine

more in detail the social roles and association patterns (Boyd et al., 2010). Usually, dolphins are categorized into hierarchical age classes, which are newborns, calves, juveniles and adults (Table 1). The sex can be determined opportunistically from photographs of the genital area during aerial behaviour or bow-riding. Females are identified by the presence of mammary slits and the urogenital slit close to the anus. In contrast, males are distinguished by the presence of an erect penis, the absence of mammary slits, or a visible gap between the urogenital slit and the anus. Additionally, adults that are consistently observed with a calf swimming in the infant position are classified as females (Díaz López & Methion, 2017).

Table 1. Age-class categorisation based on Díaz López & Methion (2017).

Age class	Definition
Calf	Dependent dolphin smaller than 1.5 metres, with foetal folds. Strictly associated with the mother. Uncoordinated surfacing behaviour and swimming in the infant position (underneath the mother, lightly touching her abdomen).
Juvenile	Independent dolphin about two-thirds the length of an adult. May swim in close association with an adult but never observed in the infant position. It has few rake marks and skin lesions.
Adult	An independent dolphin with a robust body and darker skin coloration. It may be marked or unmarked.

Over time, information on ages, sexes, familial relationships, habitat use and distribution, interactions with boats or other species are accumulated. All of these variables are important in shaping and understanding the social structure (Wells et al., 1987).

After the data collection, the common analyses in recent published papers (Blasi et al., 2015; Louis et al., 2015; Arcangeli et al., 2017; Papale et al., 2017; Cipriano et al., 2022) include assessment of the role of sex and age in the composition of the group, individual residency patterns, the temporal stability of associations and if fisheries affect such social patterns.

To measure association between individuals the half-weight association index (HWI) is used. It ranges from 0, indicating that the pair had never been

2. STUDY AIMS AND OBJECTIVES

This thesis aims to provide an initial insight into the social structure of the bottlenose dolphin (*Tursiops truncatus*) in the Thermaikos Gulf. This area represents one of the most important socio-economic regions in Greece, characterised by intense human activities, including tourism, fishing, shipping, aquaculture, as well as hosting several protected coastal areas that support a rich marine biodiversity. However, the gulf is characterised by significant anthropogenic pressures such as pollution, habitat degradation, and disturbance from maritime traffic. Studying the ecology, behaviour and social organisation of the local bottlenose dolphin population and understanding how they interact with the possible threats they face, is therefore crucial for effective management and conservation planning.

Despite the ecological importance of the Thermaikos Gulf, information on the bottlenose dolphin subunit is limited. Since 2021, a project has been ongoing to systematically monitor the abundance, distribution, and habitat use of marine mammals in the region. The present thesis focuses specifically on analysing for the first time the social structure of bottlenose dolphins in the area. To achieve this aim, photo-identification, behaviour, and location data collected between 2021 and 2025 were analysed to investigate patterns of association and describe the social organisation of the population. Understanding the social relationships within this population is essential, as social structure plays a key role in shaping population dynamics, reproductive success, genetic flow and the resilience of the species to environmental and anthropogenic pressures.

The main research questions addressed in this thesis are:

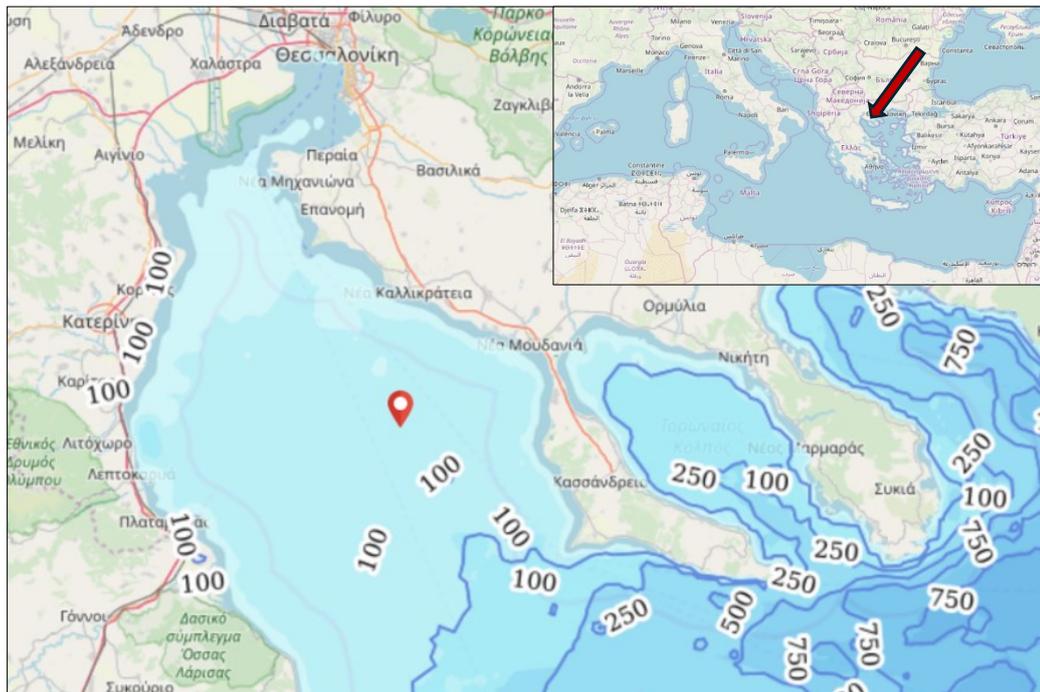
- Do bottlenose dolphins in the Thermaikos Gulf associate randomly, exhibiting no significant social structure?
- Are dolphin subgroups in the area defined by environmental variables such as depth and distance from shore?

3. MATERIALS AND METHODS

3.1 Study area

The study was conducted in the Thermaikos Gulf, a semi-enclosed coastal basin located in the mid-latitude east-central Mediterranean Sea (Figure 6).

Figure 6. Map of the Thermaikos Gulf with bathymetry (OpenSeaMap - The free nautical chart).



The Gulf covers an area of approximately 3,300 km², extending along the coast of the Greek Chalcidice Peninsula from the city of Thessaloniki to Cape Possidi in the south. It connects to the North Aegean Sea through the southern border and receives inflows from several local rivers. It can be divided into three major sub-basins, called the Inner Gulf in the north, the Central Gulf situated between Capes Megalo Emvolo and Epanomi, and the Outer Gulf extending southward, where the actual survey area of this study is located. Water depth ranges from 40 m in the Inner Gulf to approximately 200 m in the Outer Gulf, where the basin gradually deepens connecting with the Aegean Sea (Androulidakis et al., 2024).

The northernmost part of the gulf hosts the city of Thessaloniki, the second-largest metropolitan area in Greece with an estimated population of 1.5 million. Moreover, the coastal zone is densely populated and hosts a wide

range of socioeconomic activities, including agriculture, industry, tourism, maritime transport, extensive fisheries, aquaculture, and mussel farming. Despite the intense human presence, the region also presents several protected coastal areas, reflecting its high ecological value. For all these reasons, the gulf holds significant regional, national, and international importance (Kaberi et al., 2023).

This complex marine system is subject to strong anthropogenic impacts and pollution, particularly from the accumulation of heavy metals, hydrocarbon, nutrient over-enrichment, suspended particulate matter within the water column and marine litter (Price et al., 2005). However, the gulf also supports highly valuable marine habitats, including *Posidonia oceanica* seagrass meadows, coral assemblages and saltmarshes, which host high biodiversity and several endemic species. Numerous studies have documented a variety of populations of fish, birds, algae, invertebrates, and marine mammals, making the Thermaikos Gulf one of the most biologically productive marine regions in Greece (Androulidakis et al., 2024).

3.2 Data collection

Data were collected from July 2021 to September 2025 following the Thermaikos Dolphin Project Research Protocol developed by iSea (Giovos et al., 2022). Visual surveys were performed with a speedboat starting from Sani Marina in the Chalcidice Peninsula and following random routes, defined to ensure equal and balanced spatial and temporal coverage each month. Surveys were carried out only under favourable sea conditions (Beaufort Sea State ≤ 2), maintaining a speed of 20 knots (37 km/h) and observing the surrounding waters with a naked eye. During each survey data were systematically collected using the mobile application CyberTracker (Liebenberg, 2003), in which GPS position and time is automatically recorded and observational information can be manually input, such as sea state, dolphin behaviour, group size and composition, marine traffic, floating litter, fishing gears, and presence of other animal

species like seabirds, turtles and fish. Track coordinates and depth (m) were also obtained with a portable GPS (Garmin ECHOMAP-UHD2).

When a pod was detected, the boat would approach the dolphins carefully with an angle of at least 30° from the direction of travel of the animals, maintaining the speed below 4 knots, to minimize the disturbance and to not alter their behavior. The group was then followed for up to 30 minutes for behavioural observations and photo-identification. Behavioural data were collected every 5 minutes during each sighting and classified into six main categories, which are swimming, travelling, socializing, foraging, milling, and resting (Stockin et al., 2006). Photo identification data were collected focusing on the dorsal fin, the tail, and the face of each dolphin using a digital camera Canon EOS R7 equipped with an EF 70–200 mm zoom lens. Attempts were made to photograph both sides of the dorsal fin of each individual in the group.

3.3 Data processing

Digital photographs were sorted for quality and cropped focusing on the dorsal fin using XnView MP. Duplicates were generated when multiple individuals were visible in the same image. Then the photos of each sighting were organized in a folder and named according to a standardized code (Species_Area_Date_Sighting_PhotoNumber). Finally, matching of individuals was performed by comparing fin markings, scars and not with the existing catalogue. New individuals were assigned a unique ID to update the catalogue.

3.4 Data analysis

The data collected during the surveys and the individual IDs were organized in an excel file in *group mode*, where each row corresponds to a distinct group of individuals observed during the sightings. The obtained dataset was analysed using the compiled version of SOCPROG 2.3 software running on MATLAB version 9.5.0 (Whitehead, 2009). A map of the spatial distribution of the sightings of bottlenose dolphins within the study area was also created using QGIS 3.40.4.

The mean and maximum Half-Weight Index for each individual was calculated, along with the association matrix.

The Half-Weight association Index (HWI) quantifies the strength of associations between pairs of individuals (Cairns & Schwager, 1987). This method is widely used in behavioural ecology and in particular in bottlenose dolphin social structure studies (Pace et al., 2011; Papale et al., 2016; Cipriano et al., 2022, Corrias et al., 2023). The index is described by:

$$\text{HWI} = \frac{X}{X + 1/2(Ya + Yb)}$$

Where X = number of groups including both dolphins a and b;
Ya = number of groups including dolphin a but not dolphin b;
Yb = number of groups including dolphin b but not dolphin a.

It ranges between 0 (a and b never seen together) and 1 (a and b always observed together).

To minimize the bias, the association analyses were performed using some selection criteria. First, individuals that were re-sighted less than three times were excluded, in order to ensure data reliability and representativeness (Chilvers & Corkeron, 2002). Moreover, individuals with a smooth dorsal fin and low level of marking, like calves, were also excluded, because they are generally difficult to identify, present a unique dependent relationship with their mothers and do not provide additional information on the overall social structure (Figure 7). Only marked adults and juveniles were considered (Louis et al., 2015).

Figure 7. Examples of a calf (a), a juvenile with a smooth dorsal fin (b), and marked adults (c, d).



When an individual was sighted multiple times in a single day, only the first sighting was used to ensure independence of sampling and to provide independent evidence of association. Finally, larger groups (more than 8 dolphins) were included only if at least 80 % of the individuals are photo-identified, as otherwise the results may be underestimated (Blasi & Boitani, 2014).

Relationships among individuals were examined using hierarchical cluster analysis and principal coordinate analysis. Finally, the null hypothesis that the dolphins associate randomly, exhibiting no significant social structure, was tested through a permutation test for preferred associations. The hypothesis of non-random associations can be accepted if the value of the standard deviation (SD) and the coefficient of variation (CV) are significantly higher than those computed from the randomly permuted data. The association matrix of HWIs was randomly permuted 1,000 times with 1,000 trials per permutation.

4. RESULTS

4.1 Data collection

A total of 85 surveys were conducted from July 2021 to June 2025, with an effort of 235 hours, providing 45 sightings of bottlenose dolphins. Surveys were carried out all year around, with the highest proportion of effort occurring during spring (47%) and summer (35.5%). Furthermore, the year 2025 resulted to have the most intense sampling activity and highest number of sightings, with a total of 27 surveys performed during the first half of the year alone, corresponding to 31.8% of the total survey effort. Among these, 26 surveys were conducted with the direct participation of the author between March and August 2025, contributing to data recording and photo-identification. Table 2 summarizes the annual survey effort, number of total sightings per year, and group size statistics of the bottlenose dolphins. The total number of surveys increased progressively during the study period. Group sizes ranged from 1 to 20 individuals, with an overall mean of 6 ± 3 individuals per sighting. Throughout the five-year study, 133 dolphins were identified and added to the catalogue, including 27 new individuals recorded during the internship period.

*Table 2. Annual survey effort and sighting data for bottlenose dolphins (*Tursiops truncatus*), recorded from 2021 to 2025.*

Year	Total surveys	Total sightings	Effort (h)	Months	Group size (Range)	Group size (Mean \pm SD)
2021	10	6	29	5	1-8	4 ± 3
2022	9	3	20	4	5-8	6 ± 2
2023	15	2	63	3	8-9	8 ± 1
2024	24	14	62	4	1-20	8 ± 7
2025	27	20	61	7	2-10	5 ± 2
Total	85	45	235	23	3-11	6 ± 3

The total number of individuals observed every year increased from 27 in 2021 to 68 in 2025, with the highest count recorded in 2025, corresponding to 35% of the total number of sighted individuals (Figure 8). The catalogue size increased consistently over the study period. The cumulative number of identified individuals is shown in Figure 9. Moreover, to account for

differences in sampling effort, the number of individuals observed was divided by the number of surveys conducted each year, and a linear regression was performed (Figure 10). The linear regression showed a slightly positive relationship but a low R^2 value ($R^2 = 0.0189$), indicating that the increase in individuals observed per survey is not statistically significant. Therefore, also given the small dataset (5 years), the trend is likely due to an increase on survey effort rather than an increase in the population.

Figure 8. Total number of individuals sighted per year (2021-2025)

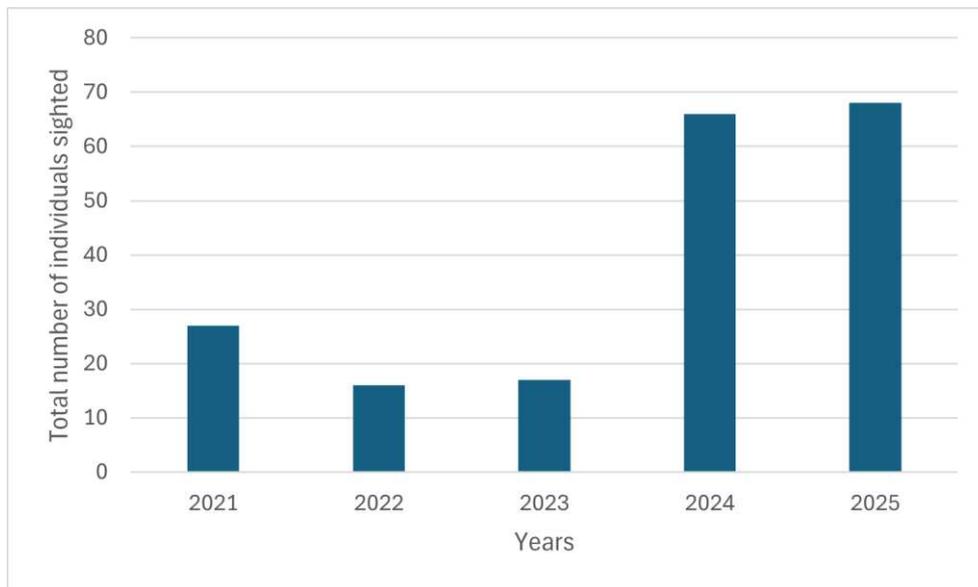


Figure 9. Cumulative number of identified individuals from 2021 to 2025

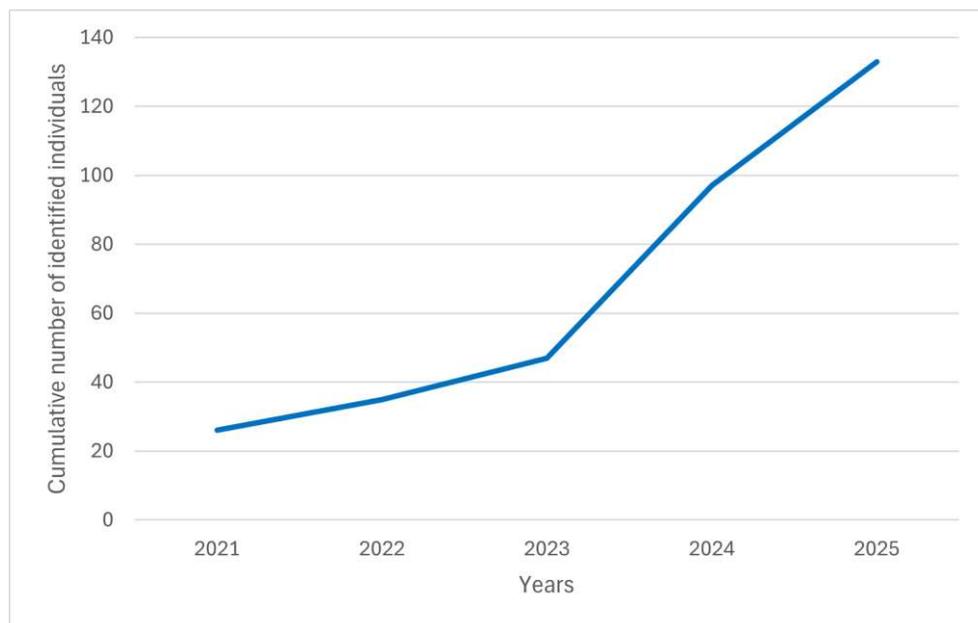
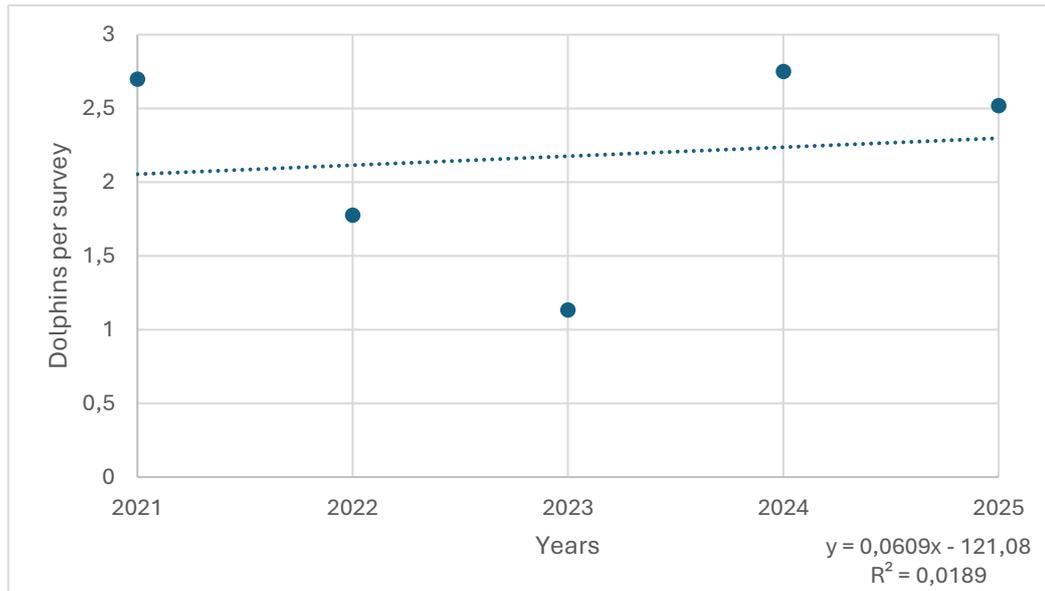
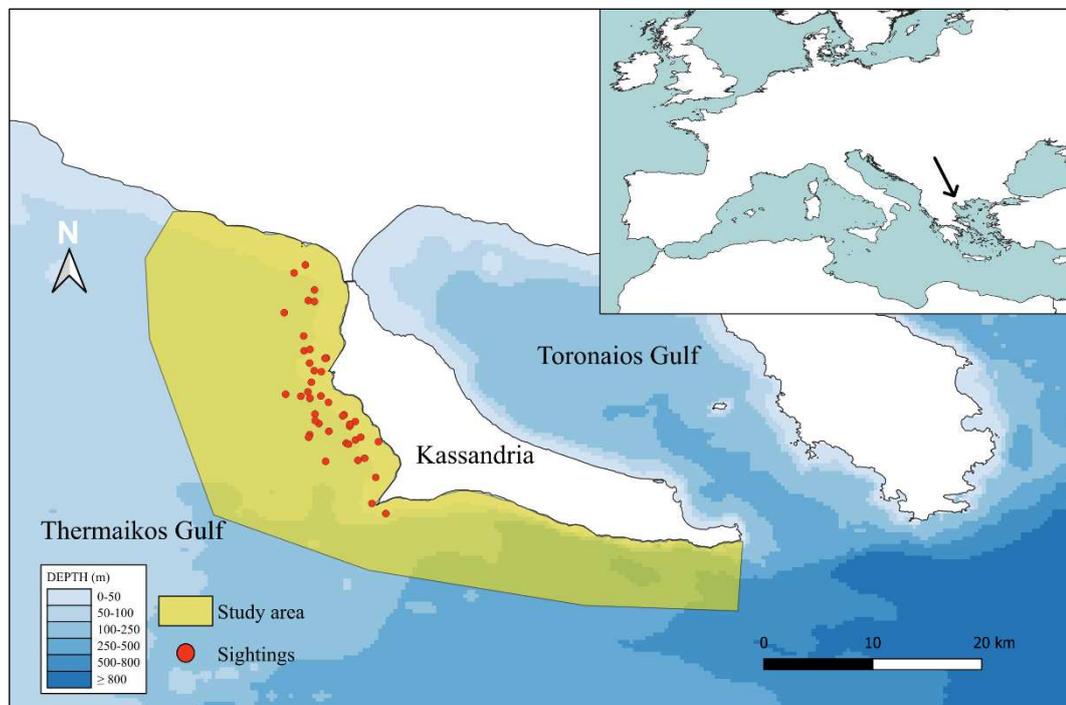


Figure 10. Linear regression showing the trend over time of mean number of dolphins per survey



The spatial distribution of the 45 sightings of bottlenose dolphins within the study area is represented in Figure 11. Sightings were mainly located in the coastal area, occurring at depths ranging from 6 to 83 metres and an average of 52 metres.

Figure 11. Study area and spatial distribution of *Tursiops truncatus* sightings from 2021 to 2025.



4.2 Association studies

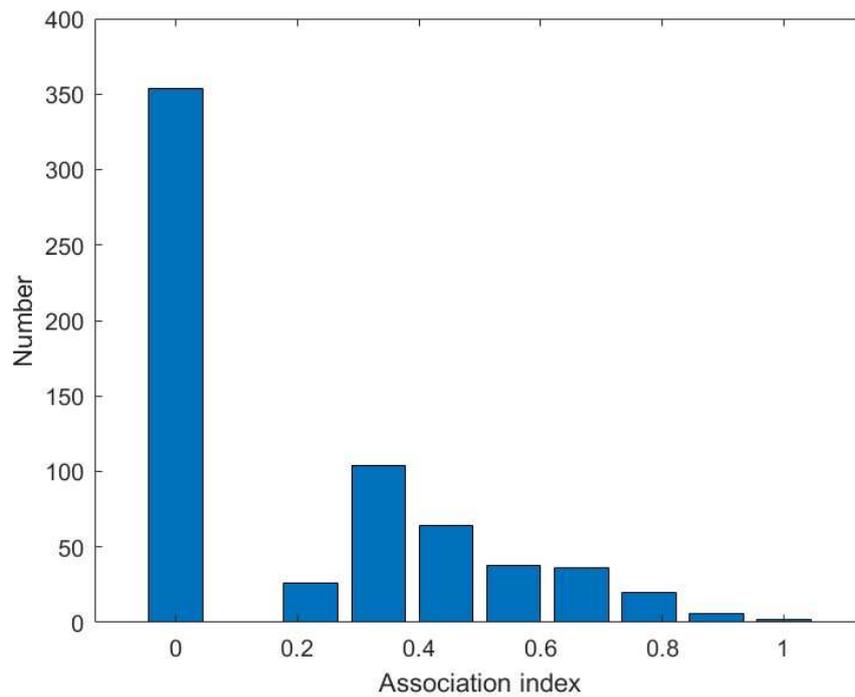
Applying the criteria reported in the material and methods section, the selection resulted in a final dataset comprising 34 sightings and 26 individuals. The overall mean value of the HWI across all individuals was 0.21, with an average of 6.16 associations per individual and an average maximum association index of 0.67. Individuals 21010, 21017, and 21018 displayed the highest mean association values (≥ 0.35), indicating frequent associations with other individuals during the study period. All three individuals are adults showing a high number of marks and notches. However, their sex could not be determined.

Table 3. List of included bottlenose dolphins (identified by their unique ID code) and the corresponding Half-Weight association Indices (mean association, sum of associations, and maximum association).

ID	Mean Assocs.	Sum of Assocs.	Max. Assocs.
21003	0.06	2.43	0.57
21004	0.10	3.59	0.67
21006	0.29	8.29	1.00
21009	0.14	4.57	0.67
21010	0.37	10.32	0.75
21011	0.27	7.82	0.86
21012	0.04	1.91	0.57
21017	0.37	10.31	1.00
21018	0.35	9.73	0.86
21019	0.21	6.20	0.67
21021	0.25	7.36	0.67
22005	0.19	5.79	0.67
23008-SON1	0.34	9.59	0.75
24011	0.09	3.31	0.40
24014	0.30	8.47	0.80
24015	0.30	8.38	0.86
24018	0.35	9.80	0.86
24021	0.32	8.95	0.75
24024	0.13	4.27	0.50
24025	0.07	2.79	0.50
24027	0.25	7.17	0.75
24031	0.09	3.21	0.50
24034	0.10	3.47	0.40
24038	0.18	5.43	0.50
25001	0.06	2.52	0.33
25009	0.14	4.58	0.57
Average	0.21	6.16	0.67

The histogram of HWI frequencies (Figure 12) shows the distribution of association strengths among all the individuals. The majority of associations were characterized by low or moderate values, whereas a limited number of dyads exhibited high HWI values (>0.8), indicating strong and consistent pairwise associations.

Figure 12. Histogram of Half-Weight Index (HWI) frequency distribution.



The dyads with the highest HWI value (> 0.80) are listed in Table 4. The highest value (HWI = 1.00), which corresponds to the maximum degree of association and implies that the pair was observed together throughout the entire sampling period, was observed between individuals 21017 and 21006, followed by several pairs with values between 0.80 and 0.86. These pairs represent stable and strong social bonds within the population.

Table 4. Dyads with the highest Half-Weight Index (HWI) values among individuals.

Dyad	Index
21017-21006	1.00
24015-24018	0.86
21011-24015	0.86
21018-24015	0.86
24015-24014	0.80

The permutation test assessing possible preferred or avoided associations in the population (Table 5) showed that the standard deviation (SD=0.2551, random SD=0.2493) and the coefficient of variation of association indices (CV=1.2351, random CV=1.2031) were higher in the observed data compared to random values. However, this difference was not statistically significant (p -value = 0.078 and p -value = 0.05). These results suggest that the null hypothesis, indicating that bottlenose dolphins in the Thermaikos Gulf associate randomly exhibiting no significant social structure, cannot be rejected.

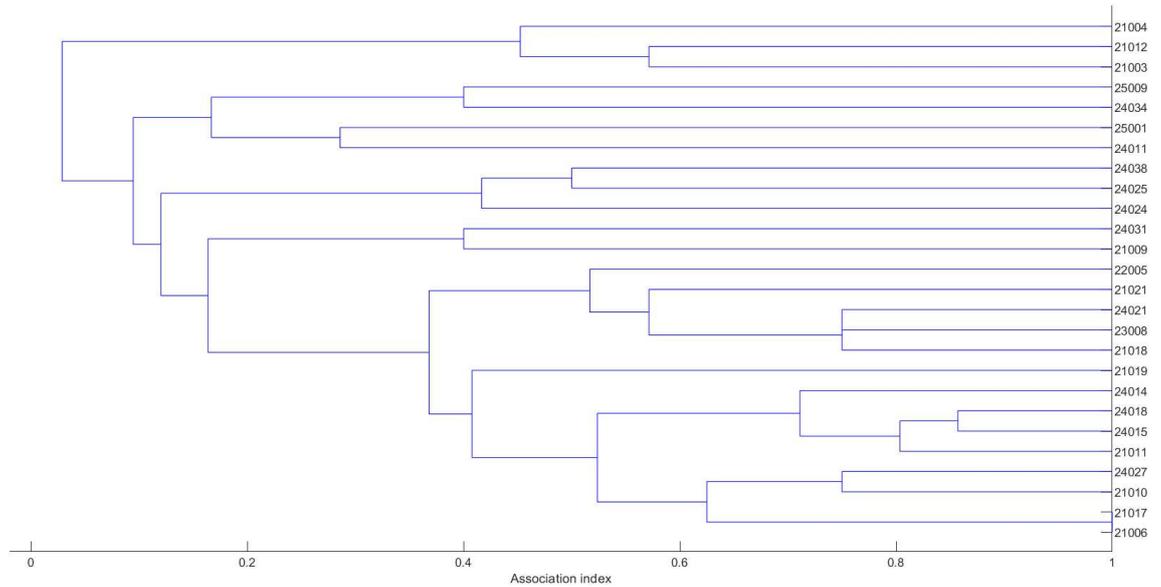
Table 5. Permutation test for possible preferred or avoided associations in the population.

Test statistic	Real	Mean (random)	P-value
Mean	0.20652	0.20724	
SD	0.25506	0.24933	0.0780
CV	1.23505	1.20309	0.0500

Although the permutation tests did not provide statistical support for preferred associations, this does not exclude the possibility of the presence of an underlying social structure. In fact, it is important to say that the values are close to the threshold of significance and thus suggest a probable tendency toward non-random associations. Therefore, additional network analyses were performed to investigate potential clustering patterns and the organization of social relationships.

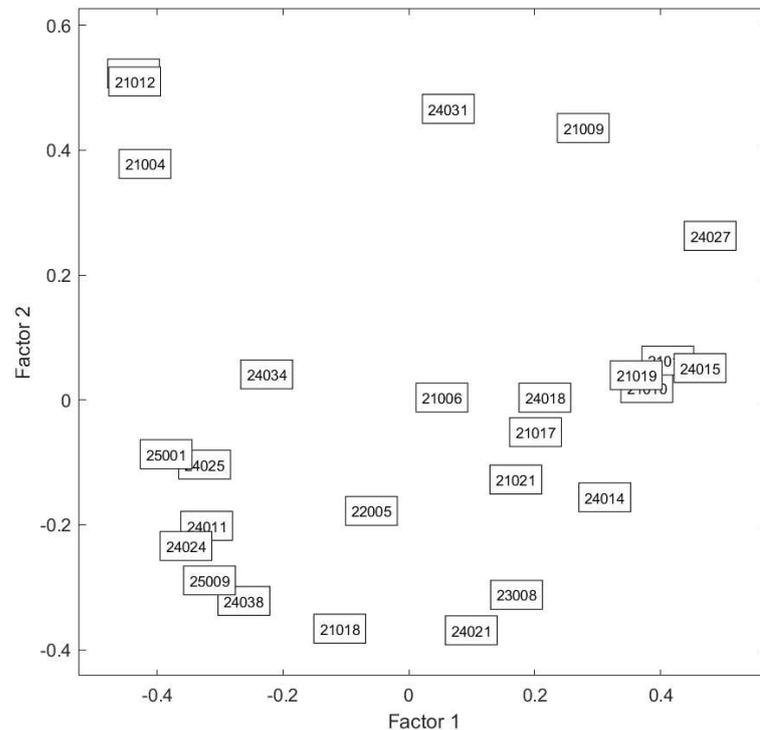
A hierarchical cluster analysis based on the association indices was carried out to examine the presence of groups or clusters within the population (Figure 13). The dendrogram reveals that some individuals are more closely associated than others, and three potential clusters might be observed. However, these subgroups are not clearly well-separated and distinct, suggesting a relatively fluid social organization.

Figure 13. Cluster analysis dendrogram based on Half-Weight Index values.



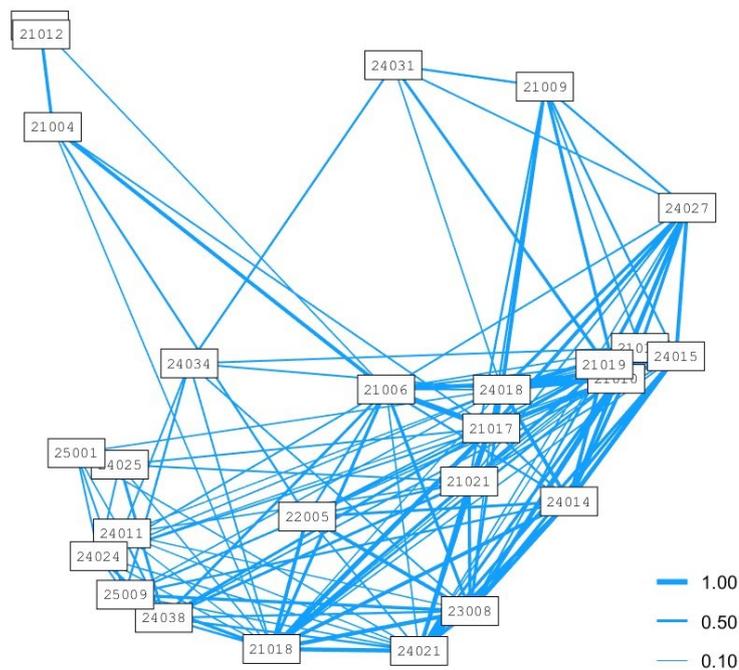
A Principal Component Analysis (PCA) was then performed to further analyse the association patterns among individuals (Figure 14). The first two principal components explained the majority of the variation, 33.1% and 25.2% respectively, showing relations similar to the ones detected in the previous analysis.

Figure 14. Principal Component Analysis (PCA) of individual association patterns.



Finally, the social network diagram based on the association indices (Figure 15) shows the social structure of the included bottlenose dolphins. Each node represents an individual, and the thickness of the connections between the nodes indicates the strength of their relationship. Moreover, the distance between nodes is directly proportional to their association. The network shows again three possible subgroups, a bigger group where tighter connections are present and two peripheral subgroups with fewer contacts, suggesting variable degrees of sociability within the population.

Figure 15. Social network diagram of the 26 included bottlenose dolphins. Each node represents a different individual.



5. DISCUSSION

This study represents the first attempt to describe the social structure of the bottlenose dolphin (*Tursiops truncatus*) population living in the Thermaikos Gulf. The knowledge of the social structure is crucial to develop specific measures for the protection of the species, especially in a basin affected by several anthropogenic pressures. For this reason, 85 boat surveys were conducted over the course of five years, resulting in an observational effort of 235 hours and the photo-identification of a total of 133 individuals, which were subsequently catalogued. During the study, bottlenose dolphins were sighted in groups of variable size (1-20 individuals) primarily in coastal areas, at depths ranging from 6 to 83 m, with an average depth of 52 m. The observed depth distribution is consistent with the literature, which reports that the Mediterranean subpopulation of *T. truncatus* is typically found in coastal habitats shallower than 100 m (Natoli et al., 2021).

The following association analyses, based on 26 well-marked and most frequently re-sighted individuals, resulted in a relatively low average Half-Weight Index (HWI = 0.21), typical of a fission-fusion society and coherent with what has been described in other studies about other coastal populations in the Mediterranean Sea (Papale et al. 2017, Cipriano et al., 2022, Corrias et al., 2023). Most associations resulted weak and only a few dyads showed strong and stable bonds with an HWI higher than 0.8. However, the permutation test revealed no statistically significant difference between observed and random association patterns, indicating that the null hypothesis of random association cannot be rejected. In other words, there is no sufficient evidence that individuals display strong or consistent social relationships, and their interactions may occur randomly rather than following a specific social pattern.

Despite the apparent lack of statistically significant preferred associations, further analyses were carried out anyway to investigate the presence of potential social relationships and hypothetical clusters within the population, as the results were close to the threshold of significance, suggesting a

possible tendency toward non-random associations. In fact, the cluster and principal coordinate analyses showed the possible occurrence of three subgroups, although they were not strongly separated. This type of clustering is typical in populations where individuals associate opportunistically, often influenced by ecological and social factors such as prey availability, reproductive competition, cooperation for calf care and familiarity (Connor et al. 2000; Krause & Ruxton, 2002; Gowans et al., 2007). The social network diagram further supported this result, showing variable degrees of sociability, with some individuals having tighter associations and others with fewer connections. However, these results should be interpreted with caution, as they are based on limited data. Further research including more temporally consistent and long-term monitoring is required to accurately describe the social structure of the population.

It is important to highlight that studying the social structure of species with dynamic and fluid organizations can be especially challenging, particularly in the case of marine mammals that inhabit complex environments, where direct observation is limited. Indeed, sample size and limited data may have influenced these results. Only individuals sighted at least three times were included, which reduced the dataset to 26 dolphins and 34 sightings. While this criterion improves the reliability of the analyses, it may have excluded individuals that play relevant roles in the population, underestimating the true complexity of social relationships.

Moreover, even if survey constraints like weather dependency are common in cetacean studies, the spatial and temporal distribution of the survey effort may have biased the detection of associations. In fact, surveys were more concentrated in spring and summer months and most importantly they were not consistent throughout the years.

Another challenge encountered was related to photo-identification quality. The sea conditions, visibility or light reflection on the water surface occasionally made it difficult to capture high-quality photographs. Moreover,

it was often challenging to take pictures of all the individuals present during the sighting, particularly of animals moving fast.

Furthermore, environmental and anthropogenic factors probably affected the observed association patterns. The Thermaikos Gulf is characterized by intense maritime traffic, fishing activities and tourism, which may influence dolphin behaviour and habitat use (Kaberi et al., 2023). Interactions with trawlers or touristic speed boats and cruises, for example, could lead to temporary aggregations for feeding opportunities or to the avoidance of specific areas, influencing group composition.

For all these different reasons, several aspects of the study could be improved in future research. Firstly, increasing the temporal and spatial consistency of surveys would allow a more balanced and reliable representation of associations. Secondly, increasing the number of re-sightings and the amount of individuals included in the analyses would improve statistical power and allow the identification of possible social patterns. Additionally, information on the sex and age class of individuals was not available in this study. Including these variables in the future could provide valuable insights on potential demographic drivers of association. Moreover, combining photo-identification with other methods, such as acoustic monitoring or genetic sampling, could provide a more comprehensive understanding of the population. Finally, the study of spatial variables such as depth, distance from coast, and marine traffic could offer valuable insights on how environmental and anthropogenic factors influence distribution and associations of bottlenose dolphins.

6. CONCLUSION

This master's thesis represents an important first step in understanding the social organization of the common bottlenose dolphin (*Tursiops truncatus*) population inhabiting the Thermaikos Gulf in Greece. Although the statistical analyses did not reveal the presence of a clearly defined or stable social structure and several limitations were encountered, the results offer valuable insights into the ecology and behaviour of this species. In fact, the research highlights to a certain extent the dynamic and fluid nature of the social organization of the species, which is coherent with the fission–fusion model typically observed in coastal dolphin populations. Understanding the mechanisms of social relationships is crucial as they play a major role in shaping genetics, reproductive success, and resilience to an environment which is constantly changing.

Overall, this study has contributed to increase the knowledge and understanding of bottlenose dolphins in a region that had not previously been investigated in relation to this topic. Surely, the results underline the importance of continuous and standardised monitoring to capture in a reliable way the complexity of relationships within the population. The data from this preliminary study need to be further developed in the coming years, in order to verify and deepen the conclusions obtained. Finally, as the Thermaikos Gulf represents an important area both in the ecological and socioeconomic aspects, these findings also carry important conservation implications. Indeed, the present research represents a basis for future studies with the aim of building effective conservation strategies for this local bottlenose dolphin subunit. In ecosystems like this basin, which is one of the most anthropogenically impacted of the eastern Mediterranean, where dolphins coexist with intense fishery, shipping traffic, tourism and pollution, understanding how individuals interact and use their habitat can help identify areas of ecological relevance. This is crucial for the establishment of Marine Protected Areas (MPAs), raise public awareness and implement appropriate mitigation measures.

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