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Short note

Age Determination of Harbour Porpoise (Phocoena phocoena relicta) from the Bulgarian Black Sea Coast

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Abstract. The following research presents the results of a pilot for Bulgaria age determination of one of the three cetacean species in the Black Sea, the harbour porpoise (*Phocoena phocoena relicta*), based on analysis of growth layers groups (GLGs). Knowledge of age structure and longevity (maximum age) are essential for modeling of marine mammal population dynamics. For small cetaceans, growth layers groups deposited in the teeth are most useful measure, because they indicate chronological age. Teeth were taken from 42 harbour porpoises (*Ph. ph. relicta*). The samples were collected from dead, stranded individuals from the Bulgarian Black Sea coast. The maximum duration of life found in this study is 10 years. The age structure shows dominance of individuals of two age groups - up to one year old and 5 years old. In the present study, no significant differences here found in the age structure in the different areas where the samples were taken, which leads to the conclusion that the age distribution of the stranded individuals *Ph. ph. relicta* in researched area is even.

Key words: Harbour porpoise, age determination, Black Sea, Bulgaria.

Introduction

One of the three cetacean species which are distributed in the Black Sea is the Harbour porpoise *Phocoena phocoena relicta* (Linnaeus, 1758). It is the second most abundant marine mammal species inhabiting the Black Sea and adjacent waters. But the Harbour porpoise is the most stranding cetacean on the Bulgarian Black Sea cost (Evtimova et al., 2016; 2018). A method of determining the age of sampled individuals is a necessity for the study of the population dynamics of any species. A previous study for age determination of the Harbour porpoise shows that 95% of specimens from the Black Sea are at the age of 11 or younger, from the Sea of Azov — 12 or younger (Gol'din, 2004). Other studies reported a life span of 10-12 years in the North Sea (Gaskin & Blair, 1977) and in the North Atlantic around the coasts of Greenland, the maximum age is 12 years in one animal and 17 years in another (Lockyer, 2001). This study aims to track the age determination of strandings harbour porpoises along the Bulgarian Black Sea Coast.

Material and Methods

A total of 42 animals, were obtained from coastal waters of the Bulgarian Black Sea

coast, between January 2015 and November 2018. The samples are collected from dead bodies of harbour porpoises found on the sea shore during covering of transects. The selected places were located along the whole Bulgarian coastline (Table 1). The focus was mainly on relatively wild, uninhabited sandy and rocky beaches.

Table 1. The sampling sites along the Bulgarian Black Sea Coast.

Place	GPS coordinates	Place	GPS coordinates	Place	GPS coordinates
Durankulak	N 43.6771 E 28.5640	Aheloy	N 42.6390 E 27.6608	Pomorie	N 42.6039 E 27.6306
Durankulak	N 43.6809 E 28.5632	Pomorie	N 42.5821 E 27.6325	Pomorie	N 42.6062 E 27.6308
Durankulak	N 43.6929 E. 28.5641	Pomorie	N 42.6160 E 27.6321	Pomorie	N 42.5907 E 27.6317
Krapets	N 43.6579 E 28.5676	Pomorie	N 42.6213 E 27.6336	Pomorie	N 42.6018 E 27.6307
Krapets	N 43.6179 E 28.5737	Pomorie	N 42.6246 E 27.6350	Krapets	N 43.6468 E 28.5717
Ezerets	N 43.6039 E 28.5681	Pomorie	N 42.6256 E 27.6354	Aheloy	N 42.6286 E 27.6370
Ezerets	N 43.5116 E 28.5676	Pomorie	N 42.6272 E 27.6362	Krapets	N 43.7290 E 28.5734
Ezerets	N 43.5843 E 28.5743	Pomorie	N 42.6104 E 27.6311	Shabla	N 43.5582 E 28.5974
Shabla	N 43.5553 E 28.6010	Pomorie	N 42.5928 E 27.6313	Shabla	N 43.5457 E 28.6041
Shabla	N 43.5456 E 28.6441	Pomorie	N 42.5992 E 27.6307	Ravda	N 42.6413 E 27.68.08
Shkorpilovtsi	N 42.9409 E 27.9027	Shkorpilovsi	N 42.9409 E 27.9027	Shabla	N 43.5702 E 28.5851
Shkorpilovtsi	N 42.9955 E 27.8905	Ravda	N 42.6375 E 27.6710	Shkorpilovtsi	N 42.9526 E 27.8994
Kamchia	N 43.0148 E 27.8894	Ravda	N 42.6373 E 27.6776	Krapets	N 43.6181 E 28.5739
Kamchia	E 43.0130 N 27.8895	Aheloy	N 42.6262 E 27.6356	Krapets	N 43.6341 E 28.5768

Several methods have been used to determine the age of cetaceans but the examination of the tooth is at present the most reliable criterion and the most used for the Odontoceti. The teeth are collected from dead, stranded cetaceans on the coastline. Samples of teeth are collected from the central maxilla or mandibles of *Ph. phocoena.* The teeth are stored in 70% ethanol solution. The selected teeth are embedded in a polyester inclusion resin, then subjected to microtome cutting; later they are glued to a microscope slide on thin layers, colored with toluidine blue and covered with Canadian balm. The age of the Harbour porpoises was determined using dentinal Growth Layer Group (GLG) method (Boutiba, 2012) (Fig. 1).

Histological section of a tooth Odontoceti shows the following structure:

- A thin outer layer of enamel formed in the foetal period (Myrick, 1983).
- The neonatal line is the first of a series of layers of neonatal dentin, denoted

Growth Layer Group (GLG), and lies within the pulp cavity. This line is formed by the change of nutrition during the transition from foetal life to postnatal life (Nishiwaki & Yagi, 1953; Ross, 1977).

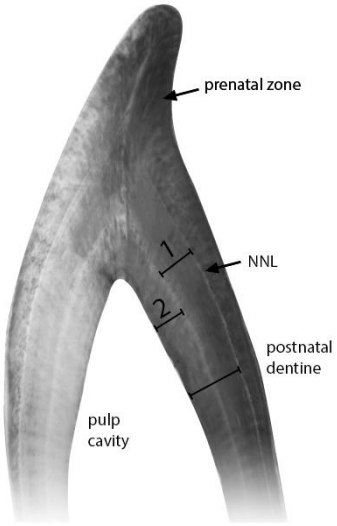

Fig. 1. Generalised longitudinal section of a porpoise tooth. *Legend:* NNL - neonatal line. 1 and 2 - Growth Layer Group (GLG).

The GLG in the secondary dentin correspond to one year in the Odontoceti.

The GLG consist of two sub-layers: a wide opaque area and a narrow translucent area.

Results and Discussion

The current study present the results of a pilot for Bulgaria age determination of Cetaceans. The study was made based on samples from a total of 42 harbour porpoises.

The life span of the Harbour porpoise is up to 24 years (Reeves, 2002), but less than 5% from the animals live beyond 12 years (Koschinski, 2001), because there is high mortality in the first years of age.

The maximum age determined for harbour porpoise in our study is 10 years. Age structure shows clear dominance of individuals up to one year and those between five and six years (Fig. 2).

At this stage, it is not possible to explain the high mortality of individuals in the fifth year and to describe the reasons leading to this occurrence.

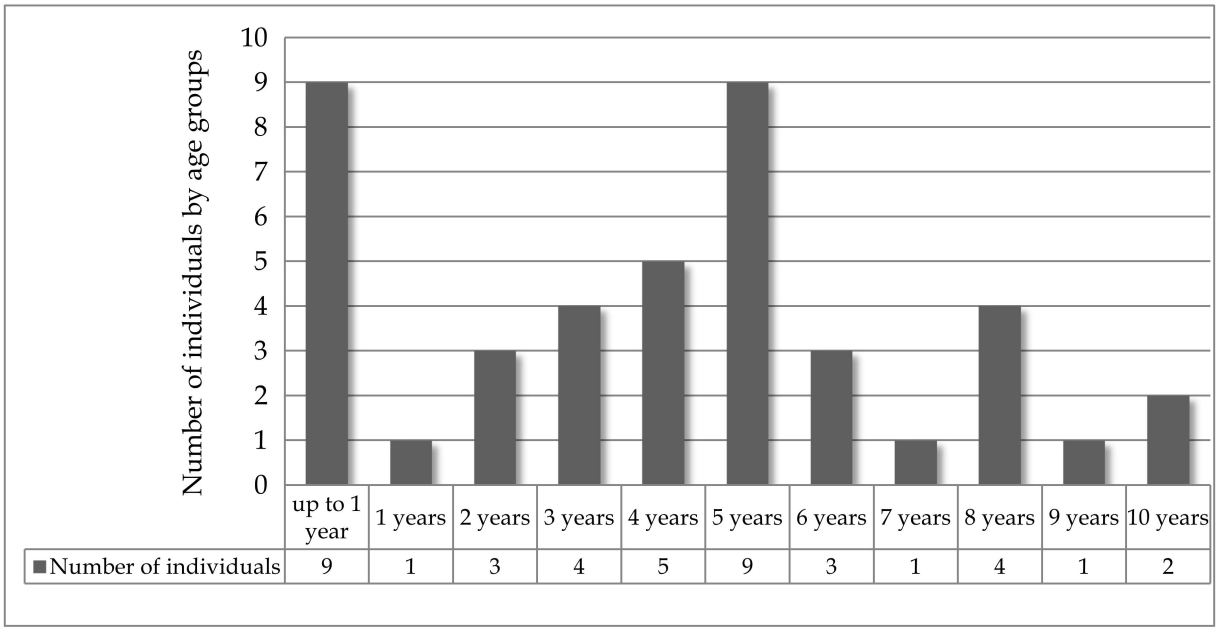

Fig. 2. Distribution of the number of the studied dead harbour porpoises by age groups.

It can only be noted that this peak coincides with the middle life cycle of the Harbour porpoise (in this study), which is ten years (from birth to death). Thus, the life

span of the Harbour porpoise in this study is comparable with the values reported from Gol'din (2004) - up to 11 years.

Conclusions

The maximum age determined for the Harbour porpoise in this study is 10 years.

The duration of life is studied in detail in the Harbour porpoise with two clear peaks of mortality rates at the beginning and mid-life periods.

In our study the age determination of *Phocoena phocoena* in the Black Sea does not show significant differences from previous studies on the same species in other seas or oceans.

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