A peek into harbour porpoise strandings

Necropsy findings of Dutch stranded harbour porpoises (Phocoena phocoena) in periods of high stranding frequency





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Picture front page: Arnold Gronert, stranded adult harbour porpoise near Callantsoog, 2013

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This report was completed as a shared assignment of Utrecht University in Utrecht and Van Hall Larenstein University of Applied Sciences in Leeuwarden. We both follow a Bachelor's degree in Animal Management with a specialisation in Wildlife Management. To complete our study a thesis was conducted at the Faculty of Veterinary Medicine, the Department of Pathobiology of Utrecht University.

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ABSTRACT

The harbour porpoise (*Phocoena phocoena*) is a small toothed whale and one of the most abundant small cetaceans in the North Sea. The harbour porpoise used to be a common sight in Dutch waters. However, in the mid-20th century population numbers dropped and sightings became a rarity. The population increased again since 1980, and recent years suggests a shift in the population from northwest to southwest of the North Sea. This is also an explanation of the increase in sightings in the Netherlands. With this increase in sightings, simultaneously an increase in stranding numbers occurred. Whereas in 1970 17 harbour porpoises got stranded, this number increased to 873 in 2013, an exponential growth. The Ministry of Economic Affairs (MEA) has appointed the Department of Pathobiology of Utrecht University to conduct post mortem investigations on stranded dead harbour porpoises. This study combines stranding and necropsy data and aims towards a better insight in necropsy findings of harbour porpoises that were stranded during periods of high stranding frequency. Periods of high stranding frequency were identified by investigating year to year variation in monthly changes in stranding frequency. Three periods were identified as high periods (H) of stranding frequency; H1: Aug '08-Sep '09, H2: Apr '11-May '12 and H3: Feb '13-Jul '13. The intermediate periods (I) between the high periods were used as s reference. When high periods of stranding frequency were compared to the intermediate periods, it showed that stranded porpoises were shorter, lighter, younger and in a poor body condition. Every high stranding period had its own characteristics. In H1, harbour porpoises were relatively young and in a poor body condition compared to the intermediate period. During H1 more porpoises died due to trauma and most animals stranded in a specific geographic area, namely on the Wadden Isles. In H2, neonates weighed significantly less, stranded porpoises were relatively young, in a more putrefied state and in a poor body condition, compared to the two surrounding intermediate periods. During H2 the most common cause of death appeared bycatch. Most animals were stranded in Noord-Holland. In H3, neonates were relatively smaller in length and they weighed less. Also, porpoises were fresher compared to the intermediate periods. Most of the investigated harbour porpoises in this period stranded in Zuid-Holland. The results showed some distinctive features in all high stranding periods, suggesting that each high period of stranding frequency is unique and should be regarded as such. This means periods of high stranding frequencies cannot be predicted and the continuation of this research is important in order to preserve this indigenous marine mammal in the Netherlands.

Key words: Harbour porpoise, *Phocoena phocoena*, strandings, North Sea, necropsy, post-mortem research.

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1. INTRODUCTION

The harbour porpoise (Phocoena phocoena) is an indigenous cetacean living in the North Sea, and often seen around the coastal areas of the Netherlands (Geelhoed & van Polanen Petel, 2011). The harbour porpoise was very common along the Dutch coast until the mid-20th century and especially in the (in 1932 closed off) Zuiderzee (Deinse, 1946; Smeenk, 1987). In 1920, van Deinse started to collect data (no absolute figures) on stranded harbour porpoises, making the Dutch stranding records of harbour porpoise one of the longest stranding record schemes ever known (Addink & Smeenk, 1999). After the Second World War (WWII), this species gradually disappeared along the Dutch coast. However, it should be emphasised that data from that time was likely not accurate (citizens were not allowed on the beach) and the exact decline was therefore not documented. Although not documented, the decline was noted and van Deinse started to yearly record strandings from 1951 onwards. In the early 1960's a decline in the number of field observations as well as stranded harbour porpoises was documented (Smeenk, 1987). As an example of how rare the species had become, Dutch sea watchers recorded only 20 harbour porpoises during 40,000 hours of observations between 1970-1985 (Haelters & Camphuysen, 2009). Reasons for this decline are difficult to identify, but a possible explanation could be pollution caused by WWII. Other factors that could have played a role were the depletion of herring stocks (which already started in 1932), and an increased mortality due to fishing gear (Smeenk, 1987).

The recording of stranded cetaceans stopped completely in the Netherlands in 1965 due to the death of van Deinse. In 1970, a new cetacean recording scheme was set up by the zoological museums of Leiden and Amsterdam. In the mid-1980s to early 1990s, numbers of harbour porpoise sightings gradually increased again. This period was then followed by a large increase in sightings of 42% per year during the following 15 years (Camphuysen, 2004). An estimation in 2013 showed that abundance in the Dutch part of the North Sea is season dependent, and varies from approximately 85,000 around March to 26,000 individuals around July (Geelhoed et al., 2013).

With the increase in sighting numbers in Dutch waters over the last decades, the number of stranded harbour porpoises has increased simultaneously. In 1970, an average of 17 stranded harbour porpoise were reported, these numbers increased to 400-500 individuals annually between 2005 and 2010 to approximately 700-800 individuals per year between 2011-2013 (Fig. 1) (Camphuysen & Siemensma, 2011; Walvistrandingen.nl, 2014).

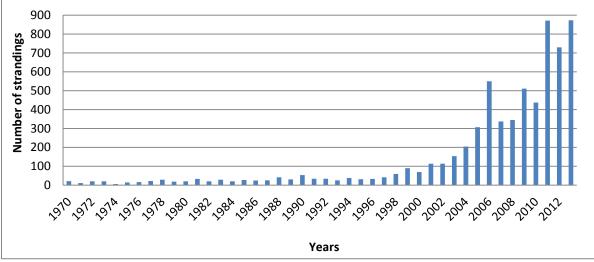


Figure 1 Numbers of stranded harbour porpoises in the Netherlands from 1970-2014 (*n*=6,480) (Walvistrandingen.nl, 2014)

1.1 PROBLEM DESCRIPTION

The trend of stranded harbour porpoises is exponential (Fig.1) and corresponds to the previously mentioned increase in sightings of harbour porpoises along the Dutch coast. Due to the increase in stranded harbour porpoises, the former Ministry of Economic Affairs (MEA) commissioned several researchers to investigate the causes of death of Dutch stranded harbour porpoises since 2006. Necropsies were conducted by experienced biologists and pathologists who collected general information like body measurements, sex, age class, body condition etc. Further, it was aimed to determine the cause of death of the stranded harbour porpoises and to collect tissue samples for future research. Frequent found causes of death included e.g. bycatch, emaciation and infectious diseases (Leopold & Camphuysen, 2006; ASCOBANS, 2009; Camphuysen & Siemensma, 2011). Recently, anthropogenic matters were more and more seen as the cause for cetacean strandings (Wright et al., 2013).

Stranded marine mammals are an important source of information since they represent a valuable sample of the living community (Pyenson, 2011). Wild harbour porpoises are difficult to detect due to their small size and elusive behaviour, which makes collection of information of stranded individuals even more important (Camphuysen, 2004). Certain reservations must be considered though, since the ecological relevance of stranding data is unknown. The geographical origin of a stranded individual is usually not possible to determine and the statistical credibility can be disputed (Peltier et al., 2011). The collection and research on stranded harbour porpoises on the Dutch coast can reveal possible changes in the population structure (Osinga et al., 2007). It is important to try to understand the causes of stranding, in order to determine risks for the population and to exclude zoonosis (Ministry of Economic Affairs, 2013). Post-mortem research is hereby vital, since the exterior of an animal often does not reveal the cause of stranding. A marine mammal stranding in a populated area, like on a Dutch beach, can raise public concerns and can have economic impacts. Public health could be at risk indirectly, since the carcass could affect the water quality, or directly, due to transmissions of zoonosis (Boness & Wieting, 2013), like Brucellosis (B. ceti) (Jacobs, 2012). Research on stranded harbour porpoises does not only fulfil a scientific purpose, it is also obligated by government policy, since the species is listed in several international, European and national legislations (Reijnders et al., 2009). As of December 2008 the Department of Pathobiology of Utrecht University has been commissioned by MEA to conduct necropsies, which provides a standardized database with valuable information including necropsy findings.

Several studies have been conducted on harbour porpoise strandings and necropsy findings. However, most of the studies examined general findings during a certain period. When fig. 1 is examined closely, some notably high stranding numbers can be found, such as the years 2006, 2009, 2011 and 2013. There is also a seasonal fluctuation, with March and August revealing higher stranding numbers (Camphuysen & Siemensma, 2011). No division of necropsy findings into periods of high and low stranding frequency was done before. This report refers to 'periods of high and low stranding frequency' rather than peaks and troughs to prevent confusion.

It is plausible that high frequency strandings have similar causes. For instance, in 2005 a mass stranding of harbour porpoises occurred on the Danish coast (Wright et al., 2013). That study concluded that a possible exposure to naval sonar led to an interaction with fisheries, which resulted in increased bycatch. A Dutch study from 2008 showed that similarities in pathology during different months were found. Results here showed that the overall health status of stranded harbour porpoises was generally good in winter, but not during summer. In the summer months they appeared to have empty stomachs, small blubber layers and more diseases (ASCOBANS, 2009).

This study combines stranding and necropsy data, in order to discover possibly significant features in periods of high and low stranding frequency. Differences in pathology findings which may exist between high stranding frequency and the long term trend line were investigated.

1.2 AIMS AND OBJECTIVES

This study aimed towards a better insight in the necropsy findings of harbour porpoises that stranded during periods of high stranding frequency. To achieve this aim, the study was divided into two areas of research. The first area of this study focussed on stranding data. Firstly, it was necessary to detect a general trend. Once the trend was known, periods of high stranding frequency were detected. The second area of this study examined the necropsy dataset. This part focussed on finding differences and/or relations in necropsy findings between periods of high stranding frequency and the general trend.

The results of this study acts as an information source for Utrecht University and other research institutions and helps with the understanding of necropsy findings during periods of high stranding frequency. Once it is known what happens in these periods of high stranding frequency regarding to necropsy findings, it might be possible to predict a future high stranding frequency. If, for instance, many porpoises have net marks (bycatch) in certain periods, and in the future a similar event happens, management actions can be taken to limit an possible period of high stranding frequency. This data might be useful for monitoring the health status of wild populations. Eventually, the results could contribute to the conservation of the harbour porpoise along the Dutch coast.

The main question of this study was:

Which necropsy findings characterize periods of high stranding frequency of Dutch harbour porpoises?

In order to answer the above main question, the following sub questions were formulated:

- 1. What is the trend in stranding numbers of Dutch harbour porpoises in different seasons and years between 1970 2013?
- 2. Which periods show a significantly higher stranding frequency of Dutch harbour porpoises compared to the trend between 1970 2013?
- 3. What necropsy findings are present during periods of high stranding frequency between 2008 2013?
- 4. What differences in necropsy findings are present in periods of high stranding frequency compared to the trend?
- 5. A.) What aspects of necropsy findings are distinctive for different periods of high stranding frequency?

B.) What aspects of necropsy findings are distinctive for different periods of low stranding frequency?

2. MATERIALS AND METHODS

This chapter describes what materials and methods were used in this study. The first two sections describe background information of the study area and study species. The following sections depict what data was used and how this was prepared for the analysis.

2.1 DUTCH COASTAL ZONE

The Dutch coastal zone is part of the North Sea, which is a large marginal sea of the Atlantic Ocean on the European continental shelf. It borders the coastlines of Norway, Denmark, the United Kingdom, Germany, Belgium, France and the Netherlands (Fig2.1) (Walday & Kroglund, 2002). The coastal waters of the North Sea are rich in nutrients due to the mixing of seawater with river waters (Jickells, 1998). The North Sea meets the Norwegian Sea and the Atlantic Ocean in the north above the Shetland Islands, the Baltic Sea in the west, between the borders of Sweden and Denmark and the English Channel in the south through the Street of Dover (Worldatlas, 2014). The total surface area of the North Sea is around 750,000m² and is rather shallow with an average depth of 90m and a maximum depth in the north of 725m (Walday & Kroglund, 2002). The surface temperature varies between 12°C and 20°C in summer and between 0°C and 8°C in winter. The salinity ranges from 25‰ to 35‰, this varies with the temperature of the water and increases towards the north. The North Sea experiences a semidiurnal tidal cycle, which consists of two high and two low tides of approximately equal size every lunar day (NOAA, 2008).

The United Kingdom, Denmark, Norway and the Netherlands are the major fishing countries. The fishing amount of these countries creates severe pressure on the marine ecosystems of the North Sea (OSPAR, 1999). Over 230 different fish species are found in the North Sea, of which 145 occur in the Dutch area. The highest diversity in fish species is found around the coastal zone. The most important commercial fish species for the Dutch fisheries are plaice, sole, cod and herring, whereas the most important prey species for marine mammals and birds are cod and other gadoids, herring, sandeel and gobies (Teal, 2011; Ecomare, n.d.).



Figure 2.1 The North Sea (Worldatlas, 2014)

The North Sea is a crowded sea; with tourists, fisherman, oyster and algae farms, offshore drilling rigs, tidal power stations, shipping and wind farms. Like most seas the North Sea is considered to be polluted to some extent. There are two types of pollution affecting the ecosystem; noise pollution from ships, oil and gas exploration and mining, and chemical pollution such as industrial waste, domestic sewage, atmospheric fallout, domestic and agricultural run-off and operational or accidental discharges (ASCOBANS, 2014).

The study area is the Dutch coastline (marked red in Fig 2.2). The length of this area is 353 km and in 254 km of this area dunes are present. The Dutch coast consists of broad sandy beaches and extensive dune ridges. It can be divided in three parts; *the Wadden region* is an area that consists of five dune islands and because of the bird species and seals present in this area it belongs to one of the most important nature areas in Europe; *the mainland coast* (provinces Noord-Holland and Zuid-Holland) has large dune areas that protect the low coastal plain, which consist of polders and peat meadows; *the South-western coastline* (the Delta) consists of a complex estuary of the rivers Rhine, Meuse and Scheldt and developed a wide variety of salt and brackish ecosystems. The beaches from the provinces Noord-Holland, Zuid-Holland and Zeeland are relatively busy compared to the beaches in the provinces Friesland and Groningen, this due to the large cities and the beach resorts that are situated in these provinces which attract tourists (EUCC, 2014).



The conservation of the coastal landscape fortunately has a high priority in the Dutch government's nature policy, primarily because of the flooding threat for two-thirds of the Netherlands (EUCC, 2014). The actions of the government consist of the strengthening of weak dunes and improvement of the quality of the environment, e.g. the recovering of dune vegetation (Rijksoverheid, 2014). Four sites are identified as marine areas in the Dutch Continental Shelf and coastal waters: the 'Doggersbank', 'Klaverbank' and two parts of the coastal zone, the 'Noordzeekustzone' in the north and 'Vlakte van de Raan' in the south. These areas are proposed as Special Areas of Conservation (SACs) under the European Habitat directive (ASCOBANS, 2009).

Figure 2.2 The study area (red line) in the Netherlands (Yurls.net, 2014)

The harbour porpoise shares the North Sea with a large number of other species, varying from zooplankton to birds and marine mammals. Besides the harbour porpoise, the most common marine mammals in the Southern part of the North Sea are the grey seal (*Halichoerus grypus*), the harbour seal (*Phoca vitulina*) and the white-beaked dolphin (Lagenorhynchus albirostris). Other species, such as the white-sided dolphin (*Leucopterus acutus*), the hooded seal (*Cystophora cristata*), the sperm whale (*Physeter macrocephalus*) the fin whale (*Balaenoptera physalus*) and the humpback whale (*Megaptera novaeangliae*) are occasionally seen (Bouquegneau et al., 2002; Ecomare, n.d.).

2.2 HARBOUR PORPOISE

The harbour porpoise (*Phocoena phocoena*) (Linneaus, 1758) is a small toothed whale and the most widely distributed species of all the cetaceans. Worldwide, four subspecies of harbour porpoises are recognized: *P.p. phocoena* in the North Atlantic Ocean, *P.p. vomerina* in the eastern North Pacific Ocean, *P.p. relicta* in the Black Sea and an un-named subspecies in the Western North Pacific Ocean (Rice, 1998). This study focusses on the subspecies *P.p. phocoena*, also known as the common or the harbour porpoise.

Description

The harbour porpoise belongs to the family *Phocoenidae*. Harbour porpoises are classified as Odontocetes (toothed whales) and have spade-shaped teeth which distinguishes them from dolphins (Camphuysen & Siemensma, 2011). Characteristics of the harbour porpoise are their robust, plump body with a rounded head and a small beak (Fig. 2.3). The harbour porpoise is a rather small cetacean and females are slightly bigger than males. On average females grow up to 150-160 cm and 55-65 kilograms, while males tend to grow up to 140-150 cm and 45-50 kg (Lockyer, 2003). Their exact life expectancy in the wild is unknown, but likely between 6-20 years (Masi, 2000) with a maximum recording of 24 years (Lockyer, 1995).



Figuur 2.3 Harbour porpoise (WDC, 2014)

Biology

Harbour porpoises live solitary, or in small groups of two to three individuals (Camphuysen & Siemensma, 2011). Communication with other individuals occurs acoustically, by using a specific pattern of clicks. Compared to other cetaceans, harbour porpoises must remain relatively close to each other in order to communicate, since they use high frequency sonar in a narrow sound beam. This is possibly an adaptation to avoid predation and harassment. Mother and calf always remain close to each other since the calf is dependent on its mother in the first year (Clausen et al., 2010; Miller & Wahlberg, 2013).

The harbour porpoise is one of the top predators in the North Sea and plays an important role in the entire ecosystem (Santos & Pierce, 2003; Christensen & Richardson, 2008). When top predators decline in numbers, it has an impact on the structure and functioning of entire marine communities. Top predators have a direct impact on prey, but also indirectly on prey behaviour, like foraging (Heithaus et al., 2008). Harbour porpoises are considered to be opportunistic generalist feeders (Christensen & Richardson, 2008) and echolocation is used to hunt prey (Miller & Wahlberg, 2013). In Dutch coastal waters the diet of the harbour porpoise mainly consists of coastal species, such as gobies, smelt and dragonet, as well as pelagic, schooling species such as mackerel and herring. Variation in diet in relation to age, sex, location and seasons has been reported (Jansen, 2013). Due to their small size little energy can be stored, which makes them more dependent on staying near food sources (Santos & Pierce, 2003). The daily feeding rate of a wild non-lactating adult is estimated to be 3.5% of the total body weight (Yasui & Gaskin, 1986).

Adult females produce offspring on average every two years. The gestation period is about 10.5 months and calves are 70-75 cm long at birth. Calves are weaned before they reach their first year. The harbour porpoise becomes sexually mature between three to four years of age, but are only physically mature at about five (females) and seven (males) years of age. Their mating season takes place after approximately one and a half month after calving and they have a promiscuous mating system (Perrin et al., 2009; Camphuysen & Siemensma, 2011). The calving season differs per region. In the Dutch part of the North Sea the calving season starts in May, extending to August. A peak in births is seen in July (Addink et al., 1995).

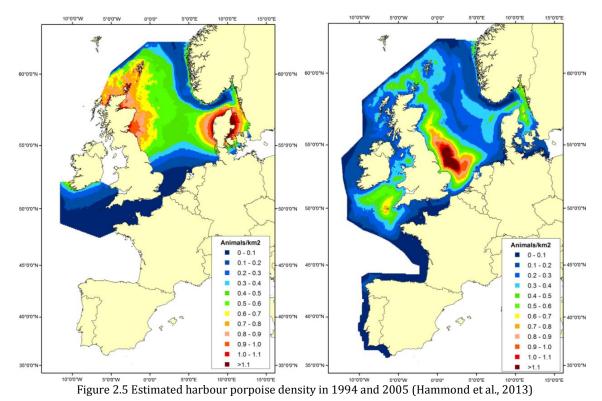
Distribution and abundance

The harbour porpoise is found throughout temperate waters of the northern hemisphere (Fig 2.4) and is rarely found in waters warmer than 17°C (Ridgway & Harrison, 1999). Harbour porpoises are mainly found in continental shelf waters, and frequently visit shallow bays, estuaries and tidal channels (Hammond et al., 2008). Distribution is thought to be linked to their prey, which is subsequently linked to environmental constraints such as bathymetry and hydrography (Sveegaard, 2011). It is estimated that the global abundance of the harbour porpoise consists of at least 700,000 individuals (Hammond et al., 2008).



Figure 2.4 Global distribution of the harbour porpoise, which occurs around the sub-Arctic and cool temperate waters of the North Atlantic, North Pacific and the Black Sea (IUCN, 2008)

The harbour porpoise is, together with the white beaked dolphin, the most abundant cetacean in the North Sea (Hammond, 2001). Results of two surveys on harbour porpoises in the North Sea in 1994 (SCANS-I) and 2005 (SCANS-II) showed respectively an estimated abundance of approximately 340,000 and 375,000 individuals. No statistically significant difference was found in the abundance of harbour porpoises between 1994-2005, however a difference in distribution was found. The main concentration of harbour porpoises in the North Sea shifted from northwest to southwest, where high densities around Denmark disappeared and densities in the Celtic Sea increased (Fig. 2.5) (Hammond et al., 2013).



Exact reasons for this shift are unknown, but is likely explained by a change in prey distribution and availability (Hammond et al., 2013). Over the last 150 years the food web structure of the North Sea changed, particularly since the mid-20th century. Changes occurred in the pelagic food web, whereby animals from lower tropic levels are more abundant nowadays. Since harbour porpoises are known

to be generalist feeders, more research is required in this field (Christensen & Richardson, 2008).

Status and threats

The harbour porpoise has gone from being listed as 'Vulnerable' in 1996 to currently being listed as 'Least Concern' on the IUCN Red List. The harbour porpoise is listed in several international, European and national legislations, conventions and agreements like the EU Habitats and Species Directive, Bern Convention, Bonn Convention, CITES, OSPAR, the Dutch Flora and Fauna legislation and the Natuurbeschermingswet (Ministry of Economic Affairs, 2014). The main objective in the protection of harbour porpoise in the Netherlands is to investigate the threats that this species is facing. Implemented measures to investigate these threats are e.g. aerial surveys, pathological research and research on bycatch and underwater sounds (Dijksma, 2013).

Despite the fact that the harbour porpoise is not close to being endangered, it does face several threats. One of the major threats is bycatch in fishing gear, especially in gill nets. Several studies have been conducted on bycatch in the Netherlands, where stranded individuals were necropsied. When the results of these studies are combined, it shows that in 12-14% of the cases there is evidence for possible bycatch and for 38% evidence of probable bycatch (*n*=681) (Camphuysen & Siemensma, 2011). Other threats that the harbour porpoise in the North Sea face are overfishing, climate change, underwater noise and pollution (Haelters & Camphuysen, 2009).

2.3 DATA COLLECTION

This section explains how the data was gathered. For this study two types of existing data were used: stranding- and necropsy data. The stranding dataset was obtained from the National Museum of Natural History Naturalis in Leiden and the necropsy dataset from Utrecht University. Not all harbour porpoises that stranded were necropsied. Stranded individuals that were too far along their in their decomposition were often brought to destruction immediately. Besides, funding allowed Utrecht University to investigate only a part (100-150 animals a year) of the stranded animals, which varied from very fresh to very decomposed individuals. In total 7,896 stranded harbour porpoises were reported from November 1848 until mid-April 2014, whereof 1,323 carcasses were necropsied on Texel and in Utrecht between March 2005 and March 2014.

2.3.1 STRANDING DATA

When a stranded cetacean was found along the Dutch coast, the Dutch stranding network was most of the time immediately notified. This network mainly consists out of the EHBZ (Eerste hulp bij zeezoogdieren, the stranding network of Pieterburen); an organisation which runs on volunteers and provides first aid to marine mammals, and IMARES (Institute for Marine Resources & Ecosystem Studies); a research institute which focusses on marine ecology. Besides them, Ecomare (a nature museum and rehabilitation centre) and animal ambulances were sometimes involved in marine mammal strandings. Fig 2.6 depicts harbour porpoise strandings in the Netherlands per municipality.

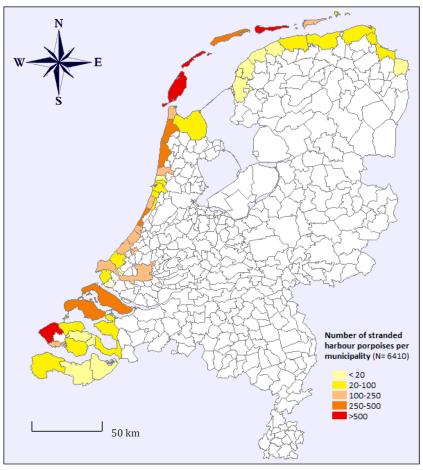


Figure 2.6 Spatial pattern in total harbour porpoise stranding reports between 1970-2013, walvistrandingen.nl, 2014 (*n*=6,410). Colour shadings indicate lower (pale yellow) and higher (red) stranding densities (see legend)

When cetaceans stranded alive, the SOS Dolfijn Foundation was notified; a rehabilitation centre for small cetaceans based in the Netherlands. In case of a dead harbour porpoise, members of the stranding network assessed in what state the carcass was, and whether it was useful for necropsy. This assessment was based on i.a. the state of the carcass and logistical considerations. When the harbour porpoise was useful for necropsy, it got either a tag or a note, where at least the stranding date and location was written on. In case of a really fresh cetacean, the ambition was to obtain a necropsy within 18 hours after death in order to collect very fresh and valuable samples. When a carcass seemed less fresh (dead >24h), the carcass was first temporarily stored in a freezer before necropsy took place. All Dutch stranded harbour porpoises, regardless of their state of decomposition, were entered in a database, which is kept by the National Museum of Natural History Naturalis in Leiden. Table 2.1 depicts what data is gathered in the stranding database.

Variable	Explanation	Variable	Explanation
ID	Harbour porpoise ID	Ст	Length of harbour porpoise in cm
Stranding ID	Stranding number	Length determination	Method of measuring length
Site	Place of finding	Sex	Sex of harbour porpoise
Beach post	Place of finding	Depot name	Name of depot
Species	Harbour porpoise	Publication	If individual was used for publication
Day	Day of finding	NSO tract	Area of stranding
Month	Month of finding	Particularities	Any special observations
Year	Year of finding	Name	Name of finder
Date	Date of finding		

Table 2.1 Stranding dataset variables. Bold variables were used for this study.

2.3.2 NECROPSY DATA

Necropsies on cetaceans were performed in Utrecht since 2008 mainly by A. Gröne, L. Wiersma, L. Begeman, L.L. IJsseldijk, S. Hiemstra and several students and volunteers. Necropsies were conducted according to the protocol of T. Kuiken and M. Garcia Hartmann (1991).

A record form was filled in for each necropsy (see Appendix II). Each individual got two numbers: an UT number which stands for Utrecht, indicating that the harbour porpoises were necropsied at Utrecht, and a GLIMS number, which indicates the individual in the entire pathology department database. Before the necropsy, the harbour porpoises were rinsed and weighed. All carcasses were checked for a chip, because when harbour porpoises from SOS Dolfijn are released back into the wild, a chip is implanted.

External observations and lesions

Firstly, the harbour porpoises were externally inspected and photographed. The decomposition code (DCC) (Appendix III) of the carcasses were estimated, and confirmed after examining the inside of the body. When the carcass was fresh (DCC 1-2), overview photos were taken from both sides and the ventral side. Then detailed photos of the head, torso, tail, fluke, dorsal fin, pectoral fins, teeth and genital split were taken. For putrefied individuals (DCC 3-5), only overview photos of lateral sides, ventral side, fluke and teeth were taken. Particularities like wounds, scars, net marks, skin lesions and amputations were photographed in detail for all animals. The gender was determined by the position of the genital split and the absence/presence of mammary gland openings. The external nutritive condition was examined and length and girth measurements were taken according to fig. 2.7.



Figure 2.7 Body length measurements

The age class was mainly based on the total length:

- < 90 cm Neonate;
- 91-130 cm Juvenile;
- >130 cm Adult.

Besides total length, the reproductive organs (e.g. pregnancies (seen by the corpus luteum on the ovaries) or lactation in females, and sperm production and testicle size in males), but also the condition of teeth was used to determine the age class.

Lastly, external signs of bycatch were examined and findings were divided into five categories, which can be found on page 18.

Subcutaneous observations and lesions

Blubber from the body was removed to observe the underlying tissues. To determine the nutritive condition (NCC) of the carcass (Appendix III), blubber and skin thickness were measured on the left lateral side:

- L13: Dorsal blubber and skin thickness in mm;
- L14: Lateral blubber and skin thickness in mm;
- L15: Ventral blubber and skin thickness in mm.

The presence or absence of subcutaneous- and pleural fat was also noted since this gave another indication of the nutritive condition. Muscularity, as well as any particularities like bone fractures and subcutaneous haemorrhage were also examined.

Internal observations and lesions

After the removal of the ribs (and the collection of the fifth rib for stable isotope analysis) the internal organs and structures became visible. Now the final decomposition code was determined. The intestines with the mesenteric lymph nodes were removed firstly. Then the organs could be removed one by one, usually starting with the stomachs¹, liver, kidneys and gonads. From the stomachs, the pancreas and spleen(s) were removed. Then the tongue, larynx, thyroid, oesophagus, lungs and heart were removed. The stomach, lungs, heart and intestines were cut open to examine the inside for parasites, contents and abnormalities. For the liver, kidneys, spleen, pancreas, gonads and lymph nodes the cut surface was examined. The head was then removed from the body. Eyes, ears (could be infested with parasites) and eight teeth from the mandible were removed and examined. The skull was serrated into two halves, in order to remove and examine the cerebellum and cerebrum. Depending on the state of the carcass, samples were taken. For DCC 3-5 individuals, only the stomachs, fifth rib, eight teeth and DNA were collected. For DCC 1-2 individuals, samples were taken for histology, toxicology, bacteriology, virology and parasitology. Table 2.2 depicts what data was gathered for the necropsy database used in this study.

¹ Harbour porpoises have four stomach chambers; a fore-stomach, a main stomach, a third chamber or pyloric stomach and a fourth chamber or duodenal ampulla (Tinker, 1988).

Variable	Explanation	Variable	Explanation
Serie	Place of necropsy	NCC	Nutritive Condition Code (1-6)
Carcass	Carcass number (UT)	Mass	Mass (weight in kg) of harbour porpoise
GLIMS	Individual database number for the database of the entire Pathology Department	Total length	Total length (in cm), from the tip of the nose to the fluke notch (L1)
EHBZ/IMARES	Tag code	L2	Front length (in cm) from tip of snout to tip of dorsal fin (L2)
Dd	Day of finding	L11	Girth, measured (in cm) right behind flippers (L11)
Mm	Month of finding	TL, L2, L11 real	If the measurements represent the reality or was an estimation due to incomplete carcass
Yy	Year of finding	L13	Dorsal blubber thickness (in cm)
Stranding location	Location of stranding	L14	Lateral blubber thickness (in cm)
Received via	Who provided the animal	L15	Ventral blubber thickness (in cm)
Name of finder	Name of person who found stranded porpoise	L13, L14, L15 real	If the measurements represent the reality
Age	Age class of harbour porpoise	Subcutaneous fat	If fat underneath the blubber layer was present yes or no and how many mm approximately
Sex	Sex of harbour porpoise	Pleural fat	If fat around the lungs was present yes or no
DCC	Decomposition code (1-5)	Bycatch based on external observations only	If bycatch was suspected due to external signs (certain, highly probable, probable, possible, no evidence)
Frozen	If carcass was frozen yes or no	Macro conclusion	Conclusion after the necropsy
Body sharp edged cuts	If body had any sharp edged cuts externally	Histology	If histology samples were taken yes or no
Head sharp edged cuts	If head had any sharp edged cuts	Probable cause of death	End conclusion, after histology
State of carcass	In what state the carcass is in	Comments	Any comments, particularities
Scavenging	Scavenging marks found		

Probable cause of death

After the necropsy, the record form was filled in and a preliminary conclusion was given. For DCC 3-5 individuals the probable cause of death was given immediately and no further histology was done. The probable cause of death for DCC 1-2 individuals was confirmed or adjusted after all the histology samples were processed and examined. For some animals the cause of death could not be determined, and were classified as unknown. The following causes of death were defined:

Infectious

Infections are the infiltrations of the host's body by viruses, bacteria and/or parasites, severe enough to cause death. The immune system of the host's body fights these infections. Infections in the animal can be seen by inflammations of the organs (by colouration, size and structure), and in the histology afterwards.

Trauma

Trauma is a serious injury or shock to the body, from violence or an accident and is a situation that could cause great distress, shock and also often immediate death. Causes could be ship propellers,

fishing gear or predators. Trauma can be caused pre-mortem, as by being hit by a propeller, or postmortem by scavenging. Division can be made by the finding of red-discoloration of the underlying tissue where the trauma occurred, and this can be histological confirmed as haemorrhages. Porpoises can also survive trauma, but then die due to e.g. sepsis. This often shows a reaction of the wound, like thickening and discoloration of the blubber and skin suggesting healing of the area.

Bycatch

Bycatch is the capture of non-target marine species in fishing nets. A harbour porpoise is considered a victim of bycatch when net marks were present on the carcass. Some other factors contribute to this presumption, like the presence of lung oedema (suggesting suffocation), a good body condition (suggesting a healthy animal) and a full stomach with undigested fish present (suggesting recent feeding prior to death). Bycatch is subdivided by the following categories which indicate the certainty that the animal could be a victim of bycatch:

- Possible bycatch: allocated when there is a visual conformation on the external and internal carcass, like unhealed notches in the extremities and net marks;
- Probable bycatch: allocated when there are visual conformations as net marks externally, the porpoise is in good body condition, the porpoise has recently fed, the porpoise suffered from lung oedema and the porpoise has macroscopically no other abnormalities;
- Highly probable bycatch: allocated for porpoises who firstly were assigned to the 'probable bycatch' category. After macroscopic analysis, together with the exclusion of diseases or other abnormalities, the bycatch category was either upgraded to highly probable, or another cause of death was allocated;
- Certain bycatch: only allocated to the animals that were received from fishermen or found entangled in nets and in which the necropsy confirmed that these animals were highly likely caught when still alive according to above mentioned characteristics.

If there was no evidence of bycatch or the possibility of bycatch remained unknown due to the state of the carcass, this was noted.

Emaciation

Emaciation is abnormal thinness, and can be caused by e.g. a lack of nutrition, parasites, trauma, due to a disease, due to a lack of hunting experience (in juveniles) or by a low fish stock. Emaciation is characterized by a thin blubber layer, lack of internal fat and by empty stomachs.

Starvation

Starvation as cause of death is only allocated to neonates. These animals starve after losing their mother, which is usually a quick process (hours to a day) because in this age class almost constant feeding is necessary. Starvation in juveniles and adults is believed to be a longer process (days and months) and therefore classified as emaciation due to visible signs as mentioned above in the emaciation category.

Birth defects

All the harbour porpoises that died with problems during pregnancy and birth, e.g. dystocia as well as dead foetuses were allocated in this category.

Other

This category includes all the cases which did not fit in any of the above categories, e.g. liver failure or live strandings as the cause of death.

2.4 DATA PREPARATION

This section describes how the obtained data was arranged in order to make analyses possible.

2.4.1 STRANDING DATA

The acquired stranding database ran from November 1848 till mid-April 2014, and consisted of a total of 7,896 stranded harbour porpoises. Data before 1970 was considered not reliable and was therefore not used (Keijl, 2014, pers. comm., 16 April). Data from 2014 was not used either for this study since this was not a complete year. Entries without a stranding location were also excluded. This meant 6,480 Excel entries were available for this study.

Not all variables from the dataset, shown in table 2.1, were needed for this study since they were not relevant. Variables which were used are displayed in table IV.1 in Appendix IV. Data was entered in SPSS and a number was allocated to the values of the variables 'sex' and 'province'. Other variables were ID, day, month, year and length.

2.4.2 NECROPSY DATA

The original dataset had 1,323 Excel entries and ran from March 2005 until March 2014. Data before December 2008 was considered as not reliable, because the necropsies were not standardized before that time. Therefore, this data was excluded from this study. Data from 2014 was also excluded, since it was not a complete year. Data from 2009 until the end of 2013 was used, which meant 1,122 Excel entries were available for this study. Fig. 2.8 depicts the percentage of necropsies per year.

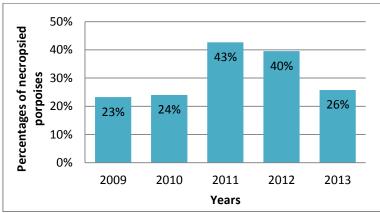


Figure 2.8 Percentage of necropsied harbour porpoises (n=1,122) relative to the total number of Dutch stranded harbour porpoises (n=6,480) between 2009-2013

To analyse the necropsy dataset, again not all variables were needed. Table IV.2 in Appendix IV depicts the ones which were used for this study. For the necropsy dataset the categories in the variables 'age', 'sex', 'DCC', 'state of carcass', 'scavenging', 'NCC' and 'cause of death' were for the statistical program SPSS transformed into numbers. In agreement with L.L. IJsseldijk, the data of the variables 'DCC' and 'NCC' were rounded up to make analysis in SPSS possible.

Two variables; 'Stranding code' and 'Period code' were created. 'Stranding code' covered if a harbour porpoise stranded in a period of high stranding frequency or not. The 'Period code' was made after the high/low/trend periods were known. More information can be found in chapter 3.2.

Since the variable 'Cause of death' had many different values, it was divided into eight categories as prior mentioned on page 17. These categories all include different causes of death and are presented in table IV.3 in Appendix IV. In the category 'unknown' the term 'pending' can be found. This means the cause of death is not known yet since the histology results were not examined during the time of this study.

2.5 DATA ANALYSIS

This section describes how the acquired data was analysed. Used programs were IBM SPSS Statistics 20 software and Microsoft Office Excel 2013.

With the analysis of the stranding dataset the first two sub questions were answered, namely:

- 1. What is the trend in stranding numbers of Dutch harbour porpoises in different seasons and years between 1970 2013?
- 2. Which periods show a significantly higher stranding frequency of Dutch harbour porpoises compared to the trend between 1970 2013?

In order to answer the first question, an overview of what occurred in the stranding data was made in Excel. Firstly the stranding trend over years and months between 1970-2013 was created. Then the sex, age classes and location of strandings were examined.

After an overview of stranding aspects was made, the presence of seasonal/monthly variance in the number of strandings was examined more closely as this had an influence on selecting the method/model of analysing the data. In fig. 2.9 the monthly variance in harbour porpoise strandings is depicted for each decade. As seen in the figure, there is a seasonal variance over recent decades, with high stranding frequencies in the months March and August.

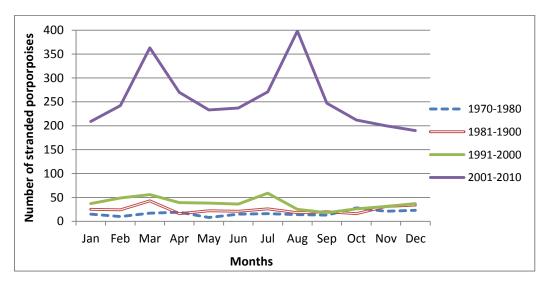


Figure 2.9 Number of harbour porpoise strandings along the Dutch coast per month over decades between 1970-2010 (*n*=4,081)

Due to the monthly variance, a derivative method was used for the determination of the high/low/trend stranding frequency. The derivative measured the sensitivity of the change of stranding numbers per month which was in turn determined by previous stranding numbers per month. The derivative for each month over the years 2000 to 2013 was determined (Fig. 2.10). The years 1970-1999 could not be used in this method due to the low numbers of strandings per month, which caused the derivative to fluctuate too severe for interpretation.

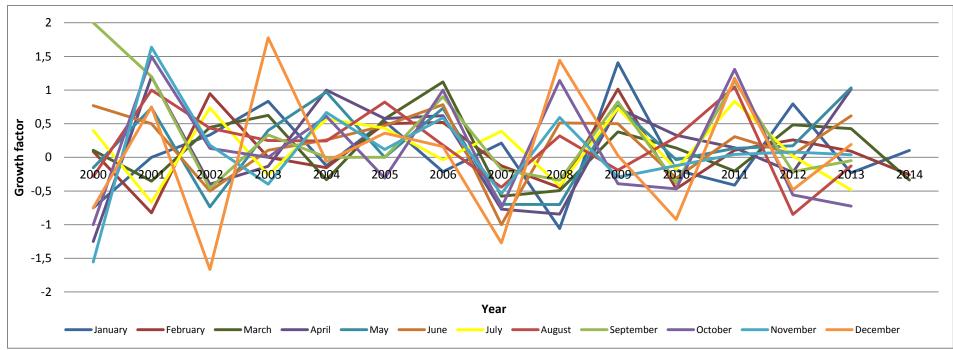


Figure 2.10 Derivative values of harbour porpoise strandings per month over the years 2000-2013

As shown in fig. 2.10, the derivative of the months show high stranding frequency (>1) and low stranding frequency (<-1). There are months that show larger fluctuations such as December, and smaller fluctuations such as November. Also a large fluctuation in the years 2000-2005 can be seen, however this is probably due to the lower stranding numbers per month which make these years less reliable.

For interpretation of the time period of the high stranding frequencies (H), a similar graph was created, only then in chronological order. Also the moving average (MA) over five points was determined to prevent noise and create a better image to review. The MA was determined over five points, since this is an odd number and to reduce the chance of exceeding the boundaries of different periods of high and low stranding frequency. Since the word 'trend' was not exact enough, the word 'intermediate' (I) was used instead from now on. This graph (Fig. 3.7) is displayed in chapter 3.2.

The definition for the identification of the high stranding frequencies, based on fig. 2.10, was:

A period of high stranding frequency has an ongoing positive value for at least six months with a minimum of two consecutive points above 50% of the average number of derivatives.

When the different periods (intermediate, high and low stranding frequency) were known between 2000-2013, the third question was answered. This question was as follow:

3. What necropsy findings are present during periods of high stranding frequency between 2008 – 2013?

In order to analyse this question, Excel was used. At first two periods were assigned; a high standing period or not (coded with 0 and 1 respectively). Since there were hardly any differences visible, it was decided to break down these periods into six different periods since each period of high stranding frequency could be unique. The variables 'Cause of Death', 'NCC', 'DCC', 'Sex' and 'Age class' from the necropsy dataset were subdivided per period between 2009-2013. Since periods were defined from 2000 (Fig. 2.10), the variable 'Stranding location' from the stranding dataset was subdivided from 2000. Based on these results, stacked bar charts were made to emphasize the contribution of each variable per period. The variables 'Mass' and 'Total length' were entered in SPSS, in order to obtain boxplots. Since the figures showed outliers, both were subdivided per age class as well. For all variables the value 'unknown' was excluded from the analysis.

Once the intermediate and high stranding frequency periods were identified, together with the accompanying necropsy findings, these findings were compared between the periods. These questions were as follow:

- 4. What differences in necropsy findings are present in periods of high stranding frequency compared to the trend?
- 5 A.) What aspects of necropsy findings are distinctive for different periods of high stranding frequency?
 B.) What aspects of necropsy findings are distinctive for different periods of low stranding frequency?

In order to answer the last set of questions the following steps were made in the statistical program SPSS. Foetuses in the dataset influenced the results as these are found in different stages (very young to nearly juvenile), it was decided to remove these animals from the dataset (n=17). Also, for harbour porpoises in an advanced state of decomposition of DCC >3 (4: very putrefied, 5: remains) (n=556), or a state of carcass above 4 (5: incomplete, 6: remains, 7: blubber parts, 8: skeletal parts) (n=297) the probable cause of death was more difficult to examine. Therefore these results were less reliable and thus taken out of some of the analyses in which this was found necessary. During answering the second question, it appeared that no low stranding frequency was present between 2009-2013. Therefore question 5b could not be answered. For all variables it applied that when a period of high frequency was compared to two periods of intermediate stranding frequencies, these intermediate periods were combined to one group.

Test for normality

Prior to the actual analysis to answer the research questions the variables were tested by using the Kolmogorov-Smirnov test for a normal distribution. The outcome of this test was that most of the variables were not normally distributed (p<0.001 for these variables), with the exception of the variables length and mass. Therefore non-parametric statistical tests were used for further analysis of the variables that were not normally distributed. The variables length and mass were normally distributed. The variables length and mass were normally distributed.

Length and mass analysis

An ANOVA test was used to test for the variation in length and mass of the porpoises in the six different periods of stranding frequency. To measure the homogeneity of the groups, a Levene's test was used. Additionally, a Generalized Linear Model (GLM) - univariate model was made to get an indication of the group size (if n>30), mean and upper and lower bound of the mean. When the requirements for the ANOVA test were not met, a Kruskall Wallis was used in order to test for

significant associations. To rule out the possible differentiation in age, the data was also tested for each age group with a Kruskall Wallis test. Also, a new variable 'Body condition score' (BCS) was calculated from the variables 'Length (*L*)', 'Mass' (*M*), and the mean of the blubber thickness (*b*):'BCS = $V(L \times M) \times b'$ according to Heide-Jørgensen *et al.*, (2011). A boxplot was created and an ANOVA test was used to test the variation in this variable between six different periods.

When a period of high frequency was compared to two periods of intermediate stranding frequency, these intermediate periods were combined to one group. All the animals with a DCC >3, were excluded from this analysis, also the animals with a state of carcass above 4 were removed.

Sex

A cross tabulation with a Pearson Chi-Square test was used to test for differences in male-female division between the different periods. All the animals of which the sex was unknown, were excluded from this analysis (n=46).

Age

A Mann-Whitney U test was used to test for differences in the mean age between the different periods, all animals with the age noted as unknown were removed from the data (n=31).

NCC/DCC

A Mann-Whitney U test was used to test for differences in the mean NCC/DCC between the different periods. For NCC, all animals with an unknown NCC (n=286), animals with a state of carcass above 4 (n=297) and animals with DCC 4 or 5 (n=556) were excluded. For DCC, the animals with the unknown DCC were removed (n=13).

Cause of death

A cross tabulation with a Pearson Chi-Square test and Adjusted Standardized Residuals was made to test for the differences in causes of death between the different periods. If the two variables had no relation, the adjusted residuals had a standard normal distribution, a mean of 0 and a standard deviation of 1. An adjusted residual which is larger than 1.96 indicated that the number of stranded harbour porpoises in that cell was significantly larger than would be expected (significance level of 0.05). An adjusted residual which was less than -1.96 indicates that the number of cases in that cell was significantly smaller. All the animals with an unknown cause of death (n=596), animals with a state of carcass above 4 (n=297) and animals with DCC 3, 4 or 5 (n=814) were excluded.

An additional analysis of the variable cause of death was performed where the differences in age and sex were taken into account. A cross tabulation with a Pearson Chi-Square test and Adjusted Standardized Residuals was made for the whole period (2009-2013). However due to the low number per cause of death in the different age and sex classes the analyses per period could not be made.

Stranding location

A cross tabulation with a Pearson Chi-Square test and Adjusted Standardized Residuals was made to test for the differences in stranding location between the different periods from 2000 to 2013. The animals with an unknown stranding location (n=1) were excluded from this analysis.

Additional analysis

The harbour porpoises in periods of high stranding frequency were mutually tested to indicate any differences between these periods (H-H). Also all the animals in the periods of high standing frequency were compared with all the animals in the intermediate periods to indicate if there were generally differences between high and intermediate stranding periods (H-I).

3. RESULTS

In this chapter the results are depicted. The first two sections describe the results of the stranding data, the latter sections consist out of the necropsy results.

3.1 TRENDS IN STRANDING

This section contains descriptive information of harbour porpoise strandings between 1970-2013.

Trend in stranding numbers over years and in seasons

Fig. 1, displayed in the introduction, showed the stranding numbers between 1970-2013 with an increase in stranding numbers over the years. The monthly stranding frequency, shown in fig. 3.1, depicts a seasonal stranding pattern, whereas the months March and August show a higher number of strandings compared to the surrounding months. An overview of the seasonal variation in harbour porpoise strandings per month per year is displayed in Appendix V.

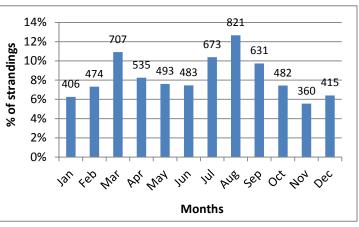


Figure 3.1 Percentages of seasonal pattern in harbour porpoise strandings between 1970-2013 (*n*=6,480) Numbers above represent absolute numbers.

Age class

Fig. 3.2 shows the abundance in stranding numbers per age class per year. The emersion pattern is that there is an overall increase in strandings, but the increase has been most pronounced in juveniles. Of the total number of strandings (*n*=6,480) 9% was neonate, 41% juvenile, 18% adult and for 32% the age remained unknown, this difference is tested significant (Pearson Chi Square<0.001). In fig. 3.3 the seasonality per month of stranded harbour porpoises is shown, where it can be seen that most neonates stranded during the summer months. For juveniles, two higher stranding periods around March and August were seen. These differences were tested significant (Pearson Chi Square<0.001) in March and August for juveniles and in June, July and August for neonates.

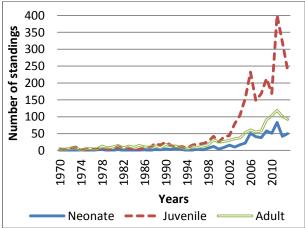


Figure 3.2 Number of stranded adults (TL>130), juveniles (TL 91-130) and neonates (TL<90) from reported harbour porpoises with length information between 1970 and 2013(*n*=4,411). Tl was either estimated or measured.

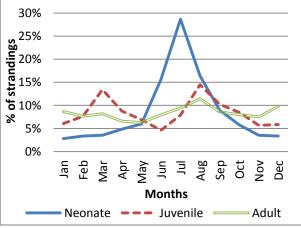
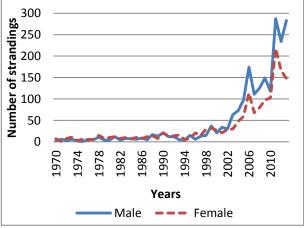


Figure 3.3 Seasonality in patterns in percentages of numbers of stranded adults (TL>130), juveniles (TL 91-130) and neonates (TL<90) from reported harbour porpoises with length information between 1970 and 2013 (n=4,411). Tl was either estimated or measured.

Sex

Fig. 3.4 shows the number of males and females that stranded between 1970-2013. The overall pattern shows that at least in recent years, from 2000 onwards, males seemed to be more prevalent. Of the total number of strandings between 1970 and 2013 32% was male, 24% female and for 44% the sex was not determined. A significant difference was found in sex over the years (Pearson Chi Square: p<0.001). Especially in 2012 and 2013 there was a notable difference, where numbers of males were respectively 234 and 283, and females 166 and 148. Fig. 3.5 depicts the percentages of identified sexes per month. Compared to males, a higher percentage of females stranded in March and in the winter months (October to January). In the other months, males stranded more often, with a notable high stranding period in August. This difference in sex over months was tested significant (Pearson Chi Square: p=0.031).



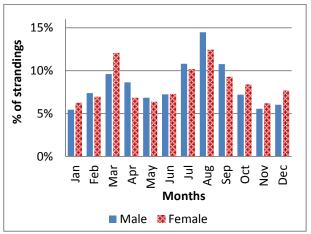


Figure 3.4 Number of stranded harbour porpoises divided in male and female between 1970-2013 (n=3,622). The animals of which sex is unknown are excluded (n=2,858)

Figure 3.5 Seasonality in percentages of stranded harbour porpoises divided in male and female between 1970-2013 (n=3,622) The animals of which sex is unknown are excluded (n=2,858)

In order to find out if there were differences between stranded males or females in the different age classes, fig. 3.6 was created. This figure showed a particular pattern. For both neonates and juveniles males were most frequent, whereas for adults females are most prominent. Males were more prevalent at neonatal age and in the juvenile age class and females were more prevalent in the adult age class. These differences between sex and age classes were tested significant (Pearson Chi Square: p<0.001).

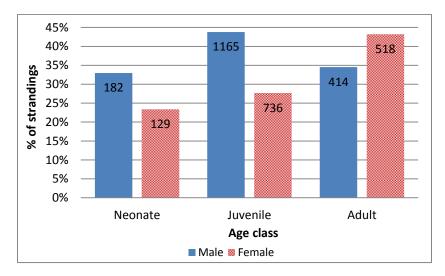


Figure 3.6 Percentages of age classes broken down by sex (n=3,144). 1,267 individuals were excluded since the sex was unknown.

In summary for the period of 1970-2013, significantly differences between the age classes were found, with juveniles showing two periods of high stranding frequency and neonates only one high stranding period per year. There was also a significantly different trend for the sexes, with females stranding more in March and winter, while male strandings tend to be more frequent in August. In general significantly more males than females stranded, but when only considering adults, there were more females than males.

3.2 PERIODICITY IN STRANDINGS

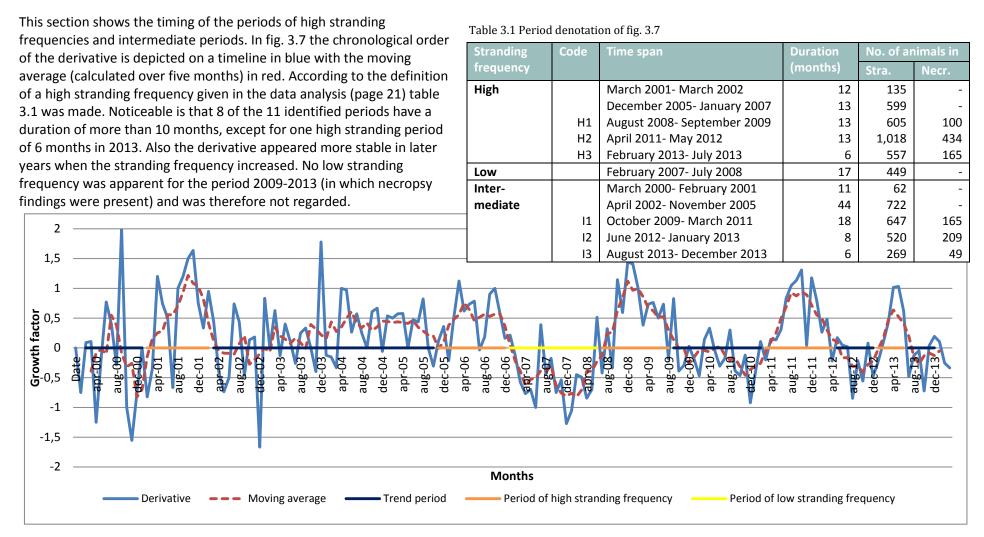


Figure 3.7 Denotation of periods of high/low/trend frequencies by the derivative between January 2000 - December 2013

3.3 NECROPSY FINDINGS IN HIGH AND INTERMEDIATE STRANDING PERIODS

In this section an overview of necropsy findings per defined period is given. Definitions of these periods were stated in table 3.1. H stands for a high period, I for an intermediate period, while the number behind states the chronological period (table 3.1). The variables mass, total length, body condition score, sex, age class, NCC, DCC and cause of death, were examined.

Mass

The mass of necropsied harbour porpoises is shown in figure 3.8. The median weight was relatively equally dispersed over the periods between 15-25 kg. The interquartile range of the first and the fourth quartile over all periods was large, except in I3. In the last four periods several outliers can be seen. For figure 3.9 the mass was divided by age class. This showed that the outliers from H3 in figure 3.8 were explained by the high number of juveniles (and thus low number of adults, which then were seen as outliers by SPSS). The average weight (outliers included) for neonates was 8.7 kg (range: 4.0-17.3 kg), for juveniles 19.4 kg (range: 7.5-38.0 kg) and for adults 40.7 kg (range: 21.7-62.0 kg). Table 3.2 depicts the average weights per period, showing that neonates were heaviest in I3 and lightest in H3. Juveniles were heaviest in H1 and lightest in I3. Adults were heaviest in I2 and lightest in I3.

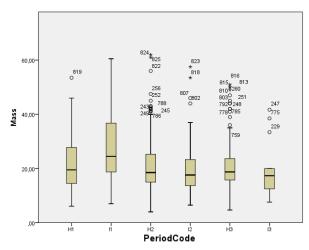


Figure 3.8 Boxplot showing the mass per period (*n*=444). Unknowns, DCC 4/5 animals, animals with a state of carcass >4 and foetuses were excluded (*n*=678)

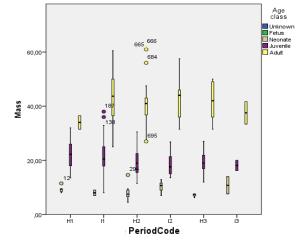


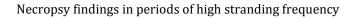
Figure 3.9 Boxplot showing the mass of adults (n=55), juveniles (n=290) and neonates (n=99). Unknowns, DCC 4/5 animals, animals with a state of carcass >4 and foetuses were excluded (n=678)

Table 3.2 Average weights in kg per age class per period ($n=444$). Unknowns, DCC 4/5 animals, animals
with a state of carcass >4 and foetuses were excluded $(n=678)$

	H1	11	H2	12	H3	13
Neonate	9,8	8,0	7,5	10,0	7,3	11,7
Juvenile	21,5	20,8	18,7	18,3	18,7	17,4
Adult	39,2	41,8	39,6	41,0	41,4	37,9

Length

Figure 3.10 shows a boxplot with the total length of the harbour porpoises per period. It shows that the median of the length does not differ greatly between the periods. The range in the last period (I3) is smaller than in other periods. In H1 and H2 several outlines are visible. For figure 3.11 the length was divided by age class. Most of the outliers seen in figure 3.10 were explained by the age class. The average total length for neonates was 82.3 cm (range: 62-91 cm), for juveniles 111.5 cm (range: 91-131 cm) and for adults 146.9 cm (range: 132-168.5 cm). Table 3.3 depicts the averages



lengths per period, showing that neonates were longest in I3 and shortest in H3. Juveniles were longest in H2 and shortest in H1. Adults were longest in H1 and shortest in H2.

PeriodCode Figure 3.10 Boxplot showing the total length (TL) per period (*n*=350). Animals with an unknown length, animals with a state of carcass >4 and foetuses were excluded (*n*=772)

Figure 3.11 Boxplot showing the total length (TL) of adults (n=94), juveniles (n=211) and neonates (n=45). Animals with an unknown length, animals with a state of carcass >4 and foetuses were excluded (n=772)

Table 3.3 Average total lengths in cm per age class per period (n=350). Animals with a unknown length, animals with a state of carcass >4 and foetuses were excluded (n=772)

	H1	11	H2	12	H3	13
Neonate	85,2	76,4	83,6	85,3	74,1	90,4
Juvenile	109,7	111,4	112,2	111,3	110,4	110,2
Adult	151,3	148,2	145,3	147,0	145,8	147,9

Body condition score

200.00

150,00

100,00

50,00

.00

щ

Total length

Table 3.4 depicts the average, minimum and maximum of the body condition score (BCS) per period. Period I1 had the highest average BCS, while I3 had the lowest. Figure 3.12 shows a boxplot with the BCS per period. It shows that the median shifts during the different periods. The range in period I2 is high, whereas the range in I3 is much lower. Period I3 also shows a notable small Q3.

Table 3.4 Body Condition Score data

	Average	Minimum	Maximum	n
H1	2,444.4	384.0	5,008.6	53
11	3,108.2	317.1	7,617.9	96
H2	2,024.9	241.5	7,078.1	111
12	2,363.6	487.7	5,864.1	20
H3	2,354.2	316.0	5,633.5	51
13	2,012.2	667.1	3,902.9	9

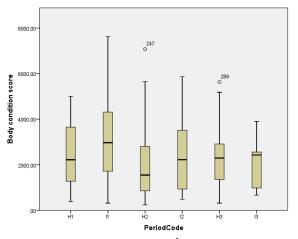
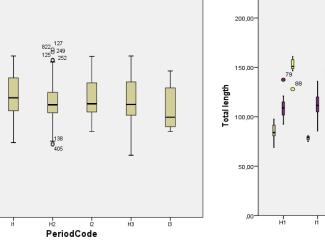
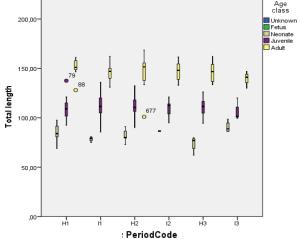


Figure 3.12 Boxplot showing the body condition score 'BCS = $\sqrt{(L \times M) \times b}$ ' per period (*n*=340) Animals with a unknown length, mass and blubber thickness, DCC 4/5 animals, animals with a state of carcass >4 and foetuses were excluded (n=782)





Sex

When figure 3.13 is examined, it seems that no visible differences in males and females per period are present. As was shown earlier (Fig. 3.4), males were more prevalent than females. Percentages of males ranged between 64.5% in H1 to 53.1% in I3, percentages of females ranged from 35.5% in H1 to 46.9% in I3.

Age class

In figure 3.14 the percentages of age class divided by period is depicted. Juveniles represent the majority of the stranded animals. Percentages of juveniles ranged between 74.6% in I2 to 50.0% in I3. Overall the number of foetuses was low (n=17). In H1 no foetuses were found, in I1 foetuses represented 2.5% of the total number. In the remainder periods percentages ranged between 1.4% in H2 to 2.1% in I3. The percentages of neonates was fairly constant (range: 10.2%-17.2%), except for period I1 (3.1%). The range of percentages of adults was 13.7% in I2 and 33.3% in I3.

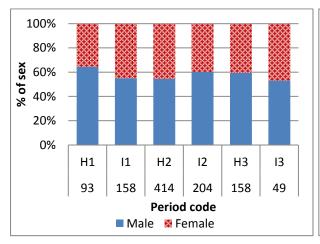


Figure 3.13 Percentages of sex per period (n=1,076) Numbers below the period code represent the total number of males and females per period. The numbers of unknowns are not depicted (n=46)

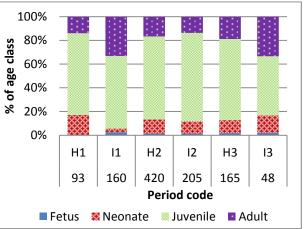


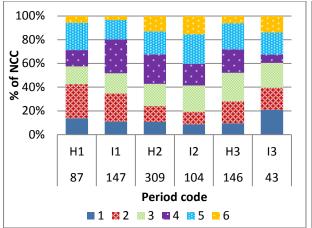
Figure 3.14 Percentages of age class per period (n=1,091) Numbers below the period code represent the total number of individuals per period. The number of unknowns are not depicted (n=31)

NCC

The percentages of assigned NCC codes are depicted in figure 3.15. Few differences over the periods were visible. In the first period NCC2 individuals were most prevalent. In the periods 11 and H2 the majority were NCC4 animals, whereas in I2 most of the animals were even in a worse nutritive condition; NCC5. Then in the last two periods the NCC increased again. In H3 most of the animals were classified as NCC3, in the last period most of the animals were classified as NCC1 and NCC3. Not displayed in this graph are the 25% of individuals of which the NCC could not be determined. Especially in H2 and I2 a high number (respectively n=125 and 105) of unknown NCC's were present.

DCC

Figure 3.16 shows the percentages of the assigned DCC per period. Period H1 shows no DCC1 (very fresh) animals, while I1 and I3 show percentages of respectively 14.8% and 16.3%. Most of the fresh DCC2 animals were found in H1 (46.0%), I1 (33.9%) and H3 (35.7%). Periods H2, I2 and I3 show with percentages of 48.4%, 62.6% and 59.2% more putrefied DCC4 animals. Most of the DCC5 animals were present in H2 and I2 (both 10%).



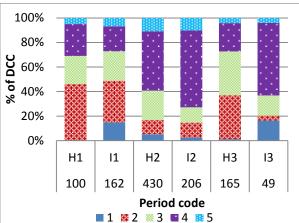


Figure 3.15 Percentages of NCC per period (n=836) Numbers below the period code represent the total number of individuals per period. The number of unknowns are not depicted (n=286)

Figure 3.16 Percentages of DCC per period (n=1,112)Numbers below the period code represent the total number of individuals per period. The number of unknowns are not depicted (n=10)

Cause of death

Figure 3.17 depicts the percentages of causes of death over the different periods. In three periods infectious diseases as a cause of death were most common; H2: 29.8%, I2:28.6% and H3 38.5%. In I1 bycatch was most prevalent, however in H1 (29.3%) and I3 (30.0%) bycatch was also relatively high. In the periods H1 (34.1%) and I3 (50.0%) trauma was the most common cause of death. Emaciation was highest in H2 (21.1%), the remainder range is 2.4% in H1 to 15.4% in H3. Starvation ranged from 14.6% in H1 to 0.0% in I3. Period I2 showed a high percentage of birth defects (7.1%) compared to other periods, which ranged from 0.0% in I3 to 3.8% in H3.

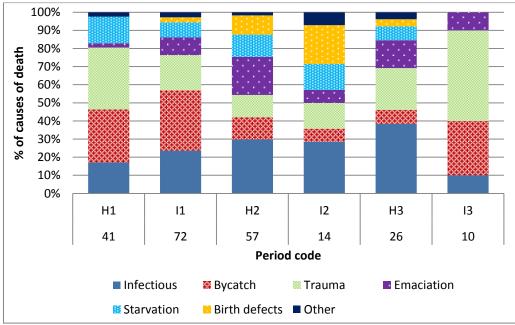


Figure 3.17 Percentages of causes of death per period (n=220). Numbers below the period code represent the total number of individuals per period. The animals with an unknown death cause, and DCC 3/4/5 animals (n=902) were excluded.

In general, the different periods did not show marked differences for mass, length, body condition, sex or NCC. The different periods appear to differ in age classes, carcass decomposition and cause of death. When separated by age class, mass and length also seem different over the assigned periods. What is evident tough, is that even the high stranding frequency periods do not seem to be caused by the same factors, but is different for the different periods of high stranding frequency.

In this section the necropsy findings from the previous section were analysed. All p values, adjusted standardized residuals and the *n* of the analysis are summarized in table 3.5 and 3.6.

Mass analysis

Analysis showed a significant difference in weight (kg) in two periods of high stranding frequency compared with the adjacent intermediate periods, where the animals weighed significantly less in high stranding periods (Kruskal-Wallis: p<0.05). For the first (H1) and second (H2) period of high stranding frequency, tests showed that in these periods significantly lighter animals stranded compared to the intermediate periods that came afterwards (H1: Kruskal-Wallis: p=0.003, H2: Kruskal-Wallis: p=0.002). For H3, tests showed that in this period there was no significant difference in weight of stranded animals compared to the intermediate period to the intermediate (Kruskal-Wallis: p=0.953).

The results of the weight analysis divided by the three age classes showed that there was no significant difference except for two cases, in H2 and H3 neonates were significantly lighter than in the intermediate periods that came before and afterwards (Kruskal-Wallis: p=0.037/p=0.010).

Length analysis

Analysis showed a significant difference in total length (cm) in two periods of high stranding frequency compared with the adjacent intermediate periods, where the length of harbour porpoises was significantly smaller in high stranding periods (ANOVA: p<0.05). For the first (H1) and second (H2) period of high stranding frequency, tests showed that in these periods significantly smaller animals stranded compared to the intermediate periods that came afterwards (H1: ANOVA: p=0.002, H2: ANOVA: p=0.026). For H3, tests showed that in this period there was no significant difference in length of stranded animals compared to the intermediate period that came before and afterwards (ANOVA: p=0.708).

The results of the length analysis divided by the three age classes showed that there was no significant difference except for one case, in H3. In this period neonates were significantly smaller than in I2 and I3 (Kruskal-Wallis: p=0.002).

Body condition score

Analysis showed a significant difference in BCS in two periods of high stranding frequency compared with the adjacent intermediate periods, where the stranded animals had a poorer BCS in high stranding periods. In H1, tests showed that in this period animals with significantly lower BCS stranded compared to the intermediate stranding period that came afterwards (ANOVA: p=0.014). For H2, tests showed that in this period animals with significantly lower BCS stranded compared to the intermediate and afterwards (ANOVA: p=0.014). For H2, tests showed that in this period animals with significantly lower BCS stranded compared to the intermediate periods that came before and afterwards (ANOVA<0.001). For H3, tests showed that in this period there was no significant difference in BCS of stranded animals compared to the intermediate period that came before and afterwards (ANOVA: p=0.744).

Sex

Analysis showed no significant difference in sex in the three periods of high frequency compared with the adjacent intermediate periods (Pearson Chi-Squared: p>0.05).

Age

Analysis showed a significant difference in age in two periods of high stranding frequency compared with the adjacent intermediate periods, where the animals were significantly younger in high stranding periods (Mann-Whitney U: p<0.05). For H1, tests showed that in this period significantly younger animals stranded compared to the intermediate that came afterwards (Mann-Whitney U:

p<0.001). For H2, tests showed that in this period significantly younger animals stranded compared to the intermediate period that came before and afterwards (Mann-Whitney U: p=0.014). For H3, tests showed that in this period there was no significant difference in age of stranded animals compared to the intermediate period that came before and afterwards (Mann-Whitney U: p=0.810).

NCC

Analysis showed no significant difference in nutritive condition in the three periods of high frequency compared with the adjacent intermediate periods (Mann-Whitney U: p>0.05).

DCC

Analysis showed a significant difference in decomposition in two periods of high stranding frequency compared with the adjacent intermediate periods (Mann-Whitney U: p<0.05). In H1, tests showed that in this period there was no significant difference in DCC of stranded animals compared to the intermediate that came afterwards (Mann-Whitney U: p=0.154). For H2, tests showed that in this period significantly more putrefied animals stranded compared to the intermediate period that came before and afterwards (Mann-Whitney U: p=0.004). For H3, tests showed that in this period significantly fresher animals stranded compared to the intermediate period that came before and afterwards (Mann-Whitney U: p=0.004). For H3, tests showed that in this period significantly fresher animals stranded compared to the intermediate period that came before and afterwards (Mann-Whitney U: p<0.001).

Cause of death

Analysis showed a significant difference in cause of death between the periods (Pearson Chi-Square=0.010). In H1, tests showed that a significant high number of animals with trauma stranded and a significant low number of animals with emaciation stranded in this period. For H2, tests showed that a significant high number of stranded animals which died due to emaciation and a significant low number of bycaught animals stranded in this period. For H3, tests showed no significant difference in all causes of death. In table 3.6 the adjusted standardized residuals are depicted. The analysis between the high stranding frequency and intermediate stranding frequency periods in common showed no significant difference (Pearson Chi-Square: p=0.644). The extended results of the adjusted standardized residuals of the causes of death are depicted in Appendix VI.

The additional analysis on the differences in causes of death compared to sex and age showed no significant differences between male and female (Pearson Chi-Square=0.052). In the different age classes there was a significant difference (Pearson Chi-Square: p=<0.001); neonates showed a high number of deaths due to starvation, juveniles died most often due to trauma and bycatch, and a significantly high number of adults died of infectious diseases.

Location

Figure 3.18 was based upon the stranding database and shows the percentages of harbour porpoises that stranded in the different provinces between 2000-2013. In the first nine periods the highest percentages of animals stranded on the Wadden Islands (range: 51.1% in H1 to 31.9% in I3). In the last two periods the numbers of stranded individuals dropped and the highest percentages were found in Zuid-Holland (period H3: 31.8%, I3: 27.1%). The Northern provinces Groningen and Friesland got the lowest number of strandings. However, in the fourth period, which runs from Dec '05 - Jan '07, relatively more individuals stranded in these Northern provinces, compared with other periods. Of the total number of that period, 3.5% stranded in Groningen, while 2.7% stranded in Friesland. In Noord-Holland the total number ranges between 23.3% in I2 to 11.4% in Feb '07 - Jul '08 (which was a low stranding period). The stranding numbers on Zeeland range from 26.5% in Feb '07 - Jul '08 and 26.4% in I3 to 13.9% in I1.

Analysis showed a significant difference in stranding location between the periods (Pearson Chi-Square=<0.001). The high stranding period between 2001-2002 showed no significant differences between the six locations. The high stranding period between 2005-2007 showed significant high

stranding numbers in the provinces Groningen and Friesland and low stranding numbers in Noord-Holland and Zeeland. The period of low stranding frequency (2007-2008) showed significant high stranding numbers in Zeeland and low stranding numbers in Noord-Holland. H1 showed significant high stranding numbers at the Wadden Islands and low stranding numbers in Zuid-Holland and Zeeland. H2 showed significant high stranding numbers in Zuid-Holland and low stranding numbers at the Wadden. H3 showed significant high stranding numbers in Zuid-Holland and low stranding numbers at the Wadden.

All locations with the exception of Zeeland showed a shift from negative to positive (or vice versa) in adjusted standardized residuals (between the periods I1 and H2) as shown in table 3.6. The provinces Groningen, Friesland and the Wadden Islands changed from positive to negative and the provinces Noord-Holland and Zuid-Holland changed from negative to positive. The province Zeeland showed between the significant low stranding numbers a period (the only low period) where the stranding numbers were significant high, the two periods latter were significant low again. The extended results of the adjusted standardized residuals of the stranding locations are depicted in Appendix VII.

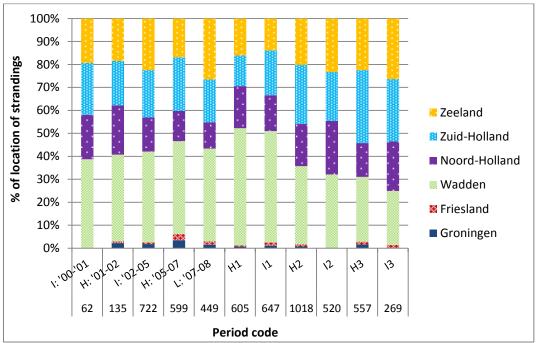


Figure 3.18 Percentages of location of strandings between 2000-2013 (*n*=5,583). Numbers below represent the total number of individuals per period.

Results of analysis between all periods of high stranding frequency and intermediate periods (H vs. I)

The analysis between H and I periods showed significant differences in the variables 'length' (total), 'mass' (total and neonates), 'body condition score' and 'age'. In the high stranding frequency periods, animals were younger and had a poorer body condition score than the porpoises in the intermediate stranding periods. The results of the analysis are depicted in table 3.5. The analysis of H vs. I in the variables 'cause of death' and 'location' showed no significant difference (Pearson Chi-Square >0.05).

Results of analysis between periods of high stranding frequency (H vs. H)

The analysis between the periods of high stranding frequency mutually showed significant differences in almost all the variables in all cases (p<0.05). The results of the analysis are depicted in table 3.5. Between H1 and H2, only the variables 'sex' and 'length' (total and juvenile) showed no significant difference. Between H1 and H3, the variables 'NCC', 'sex', 'age', 'mass' (total) and 'length' (total and juvenile) showed no significant difference. Between H2 and H3, the variables 'NCC', 'sex', 'age', 'mass' (total) and 'length' (total and juvenile) showed no significant difference. Between H2 and H3, the variables 'NCC', 'sex', 'age', 'mass' (total) and 'length' (total and juvenile) showed no significant difference.

In summary for 2009-2013, mass, length, body condition and age class showed significantly more younger and smaller porpoises in a bad body condition dying in H1 and H2 compared to the preceding and following intermediate periods. Lighter neonates (in H2 and H3) and shorter neonates (H3) stranded in the later high stranding periods. Sex and NCC were not significantly different between the different stranding periods, while H2 had more putrefied and H3 more fresh carcasses than in the intermediate periods. The periods of high stranding frequency differed significantly from each other in terms of cause of death with H1 significantly higher incidences of trauma and less to emaciation compared to the intermediate periods. H2 showed significantly more incidences of significantly in all factors from the intermediate periods. In short, the porpoises in the periods of high stranding frequency had a poorer body condition and were lighter, shorter and younger than those from the intermediate periods. For the period 2000-2013 there was also a significant shift in location of porpoise strandings from the Wadden Islands towards Noord- and Zuid-Holland.

Variable		H1 – I1	n	H2 – I1&I2	n	H3 – I2&I3	n	H1 - H2	H1 - H3	H2 - H3	H – I
Mass	Total	0.003	53-99	0.002	139-140	0.953	96-55	< 0.001	0.155	0.273	0.004
	Adult	0.401	7-36	0.420	29-44	0.621	16-11	< 0.001	< 0.001	< 0.001	0.470
	Juvenile	0.584	35-60	0.127	91-86	0.491	70-32	< 0.001	0.019	0.022	0.506
	Neonate	0.241	11-3	0.037	19-10	0.010	10-12	< 0.001	< 0.001	< 0.001	0.013
Length	Total	<0.001	53-93	0.026	119-116	0.708	52-33	0.502	0.272	0.590	0.026
	Adult	0.149	7-33	0.513	28-40	0.616	16-10	< 0.001	< 0.001	< 0.001	0.530
	Juvenile	0.171	35-75	0.823	74-71	0.699	28-17	0.191	0.197	0.329	0.753
	Neonate	0.102	11-3	0.969	17-5	0.002	8-6	< 0.001	< 0.001	< 0.001	0.121
Body cor	ndition score	0.014	53-96	<0.001	111-116	0.744	51-29	0.200	0.724	0.345	<0.001
Sex		0.142	93-158	0.338	414-362	0.904	158-253	0.236	0.898	0.125	0.929
Age		<0.001	93-160	0.014	420-365	0.810	165-253	0.045	0.144	0.783	0.003
NCC		0.256	53-96	0.124	136-136	0.686	96-54	< 0.001	0.074	0.484	0.421
DCC		0.154	100-162	0.004	429-367	<0.001	164-254	< 0.001	0.022	< 0.001	0.718

Table 3.5 P values overview of statistical tests for differences in variables between periods. Yellow colours indicate significant values.

Table 3.6 Adjusted standardized residuals overview of statistical tests for differences in variables between periods. Green indicates that the number of harbour porpoises in that cell is significantly larger than would be expected, red indicates smaller than expected.

Variable		H1	n (H1-I1)	l1	H2	n (H2-I2)	12	H3	n (H3-I3)	13
Cause	Bycatch	1.2	12-24	2.8	-2.1	7-1	-1.4	-1.9	2-3	0.6
of death	Emaciation	-2.1	1-7	-0.7	2.5	12-1	-0.6	0.6	4-1	-0.2
	Infectious	-1.4	7-17	-0.4	0.9	17-4	0.3	1.6	10-1	-1.1
	Trauma	2.1	14-14	-0.6	-2.0	7-2	-0.7	0.2	6-5	2.2
	Birth defects	-1.7	0-2	-1.2	2.0	6-3	2.7	-0.4	1-0	-0.8
	Starvation	1.0	6-6	-0.7	0.5	7-2	0.5	-0.5	2-0	-1.1
Location	Groningen	-1.5	4-7	-0.5	-1.6	8-1	-2.4	0.7	9-0	-1.9
	Friesland	-1.5	3-9	0.8	-0.4	10-0	-2.5	0.4	7-4	0.6
	Wadden	6.8	309-315	5.7	-3.2	346-166	-3.2	-5.2	157-63	-5.2
	Noord-Holland	1.1	111-101	-0.9	4.2	186-121	4.2	-1.4	82-58	2.2
	Zuid-Holland	-5.6	81-126	-1.8	-0.5	263-111	0.5	5.7	177-73	2.0
	Zeeland	-2.7	97-90	-4.3	1.8	205-121	1.8	1.4	125-71	2.6

4. DISCUSSION

The main purpose of this report was to gain a better insight in the necropsy findings of harbour porpoises that stranded during periods of high stranding frequency. Low stranding periods were not apparent, but some interesting results were found in three periods of high stranding frequency. This chapter discusses how the used methods could have influenced the results, and interprets and compares the found results with available literature.

4.1 LIMITATIONS OF THE SET-UP

The definition of the high stranding frequency periods was based on own observations of the moving average. The definition was made on the smallest visual high stranding period. This means that this definition might only be suitable for this study and therefore cannot be extrapolated to other studies. The periods were chosen based upon monthly variances. With SPSS a trend analyses (Time Series Modeller) can be made, in order to predict a trend and make assumptions of what might happen in the (near) future. Although this is an often used method, it did not work for this study since there was no consistency in the number of strandings per month. Some high stranding frequencies already could be seen with the naked eye, namely years such as 2006, 2009, 2011 and 2013 and the months March and August. Between 2009-2013 no low stranding frequency was found, and thus the last sub question was therefore not answered. It must be kept in mind that period I3, which runs from August 2013-December 2013 in this study, is not finished, and presumably continues further. Therefore, data from this period might change when this study is continued with data of future years. The first period, H1, started in August 2008, and continued until September 2009. Necropsies started in 2009, meaning that no data was available for four months. This might have influenced the results, since the late summer months and early winter months were not taken into account due to this lack of data.

In order to analyse the cause of death and location of strandings, standardized adjusted residuals in SPSS were used. An adjusted residual which is > 1.96 indicates that a variable is significantly larger than would be expected, and vice versa for <-1.96. SPSS rounds these numbers to one digit behind the point. This means that when a variable is 1.95, this digit is rounded up to 2.0. For both trauma and birth defects in period H2 (Table VI.1 in Appendix VI/Table VII.1 in Appendix VII) this number is 2.0. It might be possible that the original digit was 1.95, and thus not significant. However, the chance that this might occur is not very high.

For the cause of death analysis per period DCC 3/4/5 animals were excluded (as well as the unknowns). For DCC 1/2 animals histological samples were taken and examined, resulting in a better and more reliable insight into their cause of death. By using only fresh animals for cause of death conclusions, some reservations must be made considering how representative this is for the whole population. It is likely that fresher animals die closer to the shore, and more putrefied animals die further at sea, resulting in a longer period to drift (depending on currents and wind) before they wash ashore. It is possible that porpoises further at sea die from other causes, but, this is a topic in which further research is needed. Besides, the majority of necropsied porpoises were frozen. When only DCC1/2 animals are taken into account, 79.5% of them were frozen prior to the necropsy. Freezing results into freeze artefacts, hampering histology. A study on the freezing and thawing effect on pinniped carcasses showed that these processes could cause artefacts which resemble e.g. traumatic lesions (Roe et al., 2012).

For both the stranding data and necropsy data inter-observer variability could be present. Although the stranding network has an increasing experience and gets better coordinated than in the early days, inaccuracies are easily made when dealing with multiple people who work on a voluntarily base. Inaccuracies and/or mistakes in e.g. length measurements and sex determination could arise, and have led to many 'unknowns' in the database. Regional differences in reporting of harbour porpoise strandings must be taken into consideration as well. Stranding reports suggest that the areas Rottum, Schiermonnikoog, Voorne-Maasvlakte and Zeeuws-Vlaaderen are underreported and that there is a substantial difference between reported and expected stranding reports in the Wadden Sea (Camphuysen et al., 2008).

Necropsies were conducted by several people. Especially DCC and NCC, but also cause of death can be interpreted differently by different people. By working with a protocol and close communication observer bias is minimized. By creating a new variable (the body condition score), the inter-observer variability problem was tackled for the NCC variable. The BCS was created according to Heide-Jørgensen et al. (2011), who adjusted the original formula of Ryg et al. (1990) for Phocid seals to harbour porpoises. In the study of Heide-Jørgensen blubber thickness was measured at three dorsal points along the body. In this study blubber thickness was measured along three ventral points cranial to the dorsal fin. These measurements were used as this was the only available data in this study, but it is unknown whether this affects the results of the BCS.

4.2 STRANDING DATA

Strandings occur year-round, however, usually in March and August higher numbers of strandings were reported. Harbour porpoises are migratory animals (Sveegaard, 2011), whether more or less harbour porpoises occur/strand on the Dutch coast is thus monthly depended. It is most likely that the stranding data gives a realistic representation of the overall seasonality in strandings, since the Dutch coast is generally well accessible and well surveyed throughout the year, reporting rates are therefore not considered to be season dependent (Camphuysen & Siemensma, 2011).

When numbers of strandings per province are examined (Table IX.1 in Appendix IX), most of the harbour porpoises strand on the Wadden Islands (1,261 between 2009-2013). The number of necropsied animals per province does not always correspond with the numbers that strand (Table IX.2 in Appendix IX). In 2011 and 2012 most of the necropsied animals came from Zeeland, while most harbour porpoises stranded on the Wadden Islands. This is due to logistical considerations, since it is more expensive and takes more efforts to transport porpoises from the Wadden Islands to Utrecht.

With 60% of the total number of stranded harbour porpoises between 1970-2013, juveniles are represented the most in the Netherlands. This is in concordance with other studies, such as Haelters & Camphuysen (2009) and Camphuysen & Siemensma (2011). A higher percentage of juvenile strandings could be the cause of a higher mortality rate of juvenile animals, or due to the fact that juveniles are more represented in Dutch coastal waters (Geelhoed & van Polanen Petel, 2011). The high frequency of neonate strandings in summer is expected, since the calving season starts around May, and extents to late August in Dutch waters (Addink et al., 1995). These results could indicate that coastal waters are used as nursery areas, however Geelhoed et al. (2013) observed calves also outside coastal areas during aerial surveys. For juveniles two higher stranding frequencies were found, during spring (February-early April) and in late summer (August-September). After the weaning period, a juvenile faces a difficult time in which they need to survive alone. Juveniles leave their mother before they reach their first year, depending on their exact moment of birth (May-August). This presumably results in a higher death rate.

Of the stranded harbour porpoises between 1970-2013 males appeared more prevalent, at least in recent years. This imbalance in sex division is also found in other harbour porpoise populations e.g. by Lockyer (2003) in the North Atlantic Ocean, by Ólafsdóttir et al. (2002) in Iceland, by Lockyer (1995) in British waters and by Lockyer & Kinze (2003) in Danish Waters. When sex was compared

with age class, it turned out that for neonates and juveniles males were most prevalent, while for adults females were most common. Females are mature at an earlier age and grow faster than males, so they might be classified earlier in a higher age class (Ólafsdóttir et al., 2002), which could explain the observed female bias in adults. Another explanation for a higher prevalence of stranded adult females is that they stay closer to shore during gestation and nursing periods, when compared to males (Das et al., 2004). Being closer to the Dutch shore means a higher change of actually stranding on these coasts.

Strandings in EU countries

In the Netherlands harbour porpoise strandings have increased in recent years. Stranding numbers were compared with other EU countries bordering the North Sea, showing a corresponding increase, although not to the extend which occurred in the Netherlands. Fig. 4.1 shows the number of strandings per year per country. In the UK strandings numbers were highest around 2004 (n=472), 2005 (n=446) and 2006 (n=419). Then numbers dropped until around 200 for the next four years and from 2011 stranding numbers increased again. Denmark reached the highest number of strandings in 2008 (n=146), where stranding numbers almost tripled compared to the surrounding years. Germany had a remarkably low number of strandings in 2010 (n=88) when compared with surrounding years. In Belgium stranding numbers increased as well, with the highest numbers in 2011 (n=116) and 2013 (n=148). The stranding numbers of France have increased much in recent years, from 135 in 2010, 205 in 2011,351 in 2012 to 498 in 2013. Stranding numbers of the UK and France are second and third in comparison with Dutch stranding numbers, however these numbers does not only include the North Sea coast, but also the Atlantic Ocean, Irish Sea, the Channel etc. When only the North Sea coast is regarded, these numbers will most likely drop, showing how high the stranding numbers in the Netherlands are. It must be noted that stranding numbers are influenced by many aspects, such as climate factors, population density and coastline length (Deaville et al., 2010). More information on this subject is given in Appendix X.

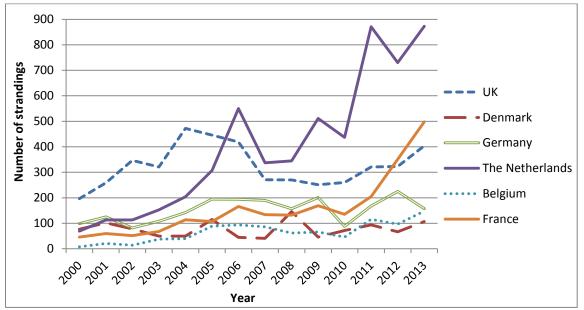


Figure 4.1 Number of harbour porpoise strandings in EU countries bordering the North Sea (stranding numbers reproduced from: Netherlands: (Walvistrandingen.nl, 2014), Belgium: (Haelters et al., 2011; ASCOBANS, 2012a; Haelters, 2014, pers. comm., 27 May), France: (Van Canneyt et al., 2013; Demaret, 2014, pers. comm., 1 July), UK: (Sabin et al., 2006; Deaville et al, 2009; Roel & Payne, 2012; Deaville, 2014, pers. comm., 20 June), Denmark: (Bie Thøstesen, 2014, pers. comm., 25 June), Germany: (Kock et al., 2011; Brtnik, 2014, pers. comm., 30 June))

A cross tabulation with a Pearson Chi-Square test and adjusted standardized residuals was created to test for the differences in numbers of harbour porpoise strandings between these six countries over the years (2000-2013). Analysis showed a significant difference between the different countries

(Pearson Chi-Square: p<0.001) with in the earlier years (±2000-2006), a significantly high number of strandings in the more Northern countries (UK, Denmark and Germany) and a significant low number in the Southern countries (the Netherlands, Belgium and France). Contrary to the later years (±2006-2013), where this was the opposite. In Appendix VIII the cross tabulation with the corresponding standardized adjusted residuals is depicted.

This increase in harbour porpoise strandings in the Netherlands could partly be explained by the shift in harbour porpoise density in 1994 and 2005 as stated in the study species. Observations of harbour porpoises showed greater densities along the Dutch coast in recent years (Camphuysen, 2011).

4.3 NECROPSY FINDINGS

High stranding periods vs. intermediate periods

When all high periods, and all intermediate periods were combined, it appeared that harbour porpoises were younger, lighter, shorter and had a poorer body condition score in the periods of high stranding frequencies in contrast to the intermediate periods. However, when the high periods were examined separately, it showed that these periods were not comparable to each other, and thus should not be combined since this gives a biased view on the results.

High stranding periods

When high stranding periods were compared with each other, many variables showed to be significant. An explanation for these results is that each period differ from the others. Almost each period started and ended in a different year and month and also the duration of the periods varied. Other factors such as environmental conditions, anthropogenic activities and variation in fish stocks also influence harbour porpoise strandings. It is likely that all high periods have different characteristics due to different factors, and are therefore not comparable.

Mass and length

When mass was examined, some outliers were observed. This could be explained by the possibility that these represent pregnant females; foetuses can weigh up to nine kg at birth (Lockyer, 1995). The results also showed that the necropsied harbour porpoises were smaller and weighed less in H1 and H2 than in I1, I2 and I3. Both mass and length were highly correlated to age class, which explains why these variables were significant in the first place, but not when they were divided by age class. Only for neonates the results differed. In the last high period, H3, neonates were smaller when compared to I2 and I3. Both in H2 and H3 neonates weighed less compared to the surrounding intermediate periods. However due to the low number of neonates (length: H3: 8, I2+I3: 6, mass: H2: 19, I1+I2: 10 and H3: 10, I2+I3: 12) the reliability of these results may be questionable and data should be complemented until analyses is possible.

Age

This study showed that necropsied harbour porpoises were relatively younger in H1 and H2 compared to the surrounding periods of I1 and I2. Possible causes could be bad weather conditions as well as inexperience of younger porpoises. An example are the young harbour porpoises that are cast away by their mother because of the birth of a new calf, these animals are unexperienced which makes them more vulnerable to bad weather conditions, low fish stocks and trauma attacks (mechanic or predators).

DCC

In H2 more putrefied individuals stranded compared to I1 and I2. For H3 more fresh individuals were found compared with I2 and I3. This seems to correlate with porpoise observations, as during part of H3 (April and May) relatively more porpoises were observed closer to shore than in other periods (Trektellen.org, 2014). Fresher dead porpoises are likely found very soon after death, meaning these

animals died close to shore. A possible explanation for these results is the fact that the spring of 2013 (H3 runs from Feb-Jul '13) was the coldest spring in 40 years (KNMI, 2013). The cold temperature might have caused a delay in the migration of the harbour porpoises (IJsseldijk & Begeman, 2013), so they subsequently stayed longer near the Dutch shore. Besides, it is likely that due to the cold temperatures the carcasses were preserved better.

NCC and body condition score

The nutritive condition (NCC) is difficult to classify and often arbitrary. Especially when animals are more putrefied, the NCC is harder to determine. The NCC was determined according to external observations, blubber thickness and presence/absence of pleural and subcutaneous fat. It must be taken into consideration that blubber thickness can be affected by decomposition (Haelters et al., 2012), is influenced by seasonal changes, individual variation, food quality and availability (Read, 1990). This was reflected in the results of this study; in the periods H2, I2 and I3 relatively more DCC4/5 animals stranded, and simultaneously more animals with a poor nutritive state (NCC5/6) stranded in these periods. Since NCC is debatable for the reasons mentioned above, a body condition score was created. For both the lowest averages of BCS and the most NCC5/6 animals were found in the same periods (H2, I2 and I3). The highest averages of BCS were found in 11 and H1. In H1 most of the animals were classified as NCC2, and also in I1 a high percentage showed to be NCC2. The results of this study showed that the BCS is a more objective measurement for condition, although the NCC classification did not differed much.

Causes of death

A higher percentage of emaciated harbour porpoises in H2 was found, a result which was also noted by Utrecht University, who assigned more animals in July-October 2011 as emaciated. Emaciation as a cause of death is debatable, since there were usually no other obvious factors for another cause of death. Emaciation is a common finding in marine mammals and can be the cause of many factors, e.g. the inability to find food, which could be due to an underlying disease, due to parasites, pneumonia, disturbance, hearing damage etc. (Dierauf & Gulland, 2001; Bouquegneau et al., 2007). In H2 the animals also significantly weighted less, and the body condition score was significantly lower. A possible explanation for these results could be a decline in fish stocks. A bachelor study conducted by Platel & Ransijn (2014) showed a decrease in herring catches in summer and early autumn in the northern part of the Netherlands from 2010 to 2012. Since summer and early autumn in 2011 this decrease is also shown in the southern part of the Netherlands. Sandeels and whiting also showed a decrease in numbers. The exact reason why more emaciated harbour porpoises stranded during H2 requires more additional research. Another interesting result is the fact that in the same period trauma was significantly lower. In H1 it was vice versa, as trauma was significantly higher and emaciation lower. This might be explained by a research which was conducted in British coastal waters, where harbour porpoises were pursued and killed by bottlenose dolphins (Tursiops truncatus). That study showed that in areas with high levels of bottlenose dolphins, harbour porpoises are significantly thinner, suggesting a trade-off decision between emaciation and predation. In this survival strategy the mass of an individual can reduce predation risk; a lower mass enhances escape time and increases agility when predators are encountered (MacLeod et al., 2007). Part of the trauma cases in this study were caused by grey seal attacks. This relatively new phenomenon was proven by a positive PCR result for grey seal DNA in wounds of harbour porpoises (van Bleijswijk et al., submitted). For this study a harbour porpoise was considered a grey seal victim when they suffered from similar but specific wounds. Besides, the exclusion of other causes of death together with the finding of undigested prey in the stomachs, a high NCC, and the exclusion of the presence of net marks contributed to this (Leopold et al., in preparation). Additional studies need to be undertaken to test this assumption.

Period I2 showed a relatively high number of birth defects, followed by period H3. In these periods birth defects occurred relatively more than in other periods. However, over all periods the total

number of birth defects is 12, making it less reliable for analysis, which should be considered while interpreting these results. Future data should be complemented until a reliable analysis is possible.

In the periods I1, H1 and I3 bycatch as a cause of death occurred relatively often. It is known that harbour porpoises are vulnerable to bycatch, and that these numbers have increased in recent years the Netherlands, Belgium and France (Hammond et al., 2013). In this study the bycatch rate was 22.3% (total n of 220 animals). In a Dutch bycatch study along the northern coast the bycatch rate was 7%-19% (n=153) (Osinga et al., 2008). In another bycatch research in the Netherlands in 2006 53-70% (n=64) of the examined harbour porpoises were suspected bycatch victims (Leopold & Camphuysen, 2006). These differences in bycatch rates between studies can be explained by interobserver variability and experiences. An increase in knowledge and experiences of the researchers involved also contributes to be able to define trauma and bycatch, but also other causes of death, better over the years. This might explain the high bycatch rate in 2006, in comparison with this study. To identify whether an animal was bycaught or not can be difficult and is often arbitrary. Whether an animal is bycaught is only certain if a fisherman hands the harbour porpoise over and the necropsy confirmed that it was highly likely that the animals was caught alive. In stranded porpoises, signs like the presence of net marks externally, together with the finding of undigested prey in the stomachs, a good NCC and lung oedema, gives a strong indication that an individual was the victim of bycatch, although this is never a certainty. When a harbour porpoise is in an advanced state of decomposition, external signs like net marks might go unnoticed. Therefore, it should be taken into account that bycatch numbers are probably an underestimation.

Neonates showed a high number of deaths due to starvation, which is an expected result. Only neonates are assigned to die of starvation, as individuals in this age class need constant feeding in order to stay alive. When they lose their mother, it is only a matter of hours before they die of this lack of food, referred to as starvation. The fact that juveniles most often died to due trauma and bycatch could be explained by their inexperience. Foraging along fishing nets seems like easy prey, but can cause entanglement when contact with the nets is made. Also things as the avoiding of boats (mechanic trauma) and seals (attacks) is something an animal will learn by getting older. The smaller size of juveniles probably will make them an easier target for seals than an adult porpoise. Results also showed that a high number of adult harbour porpoises died due to infectious diseases. Generally speaking; the older the individual, the slower the immune system responds and the ability to detect and response to diseases and parasites declines (Hall & Ahmed, 2007). More infections in adults could be the effect of the high levels of pollutants in the North Sea, like persistent organic pollutants (POPs), polychlorinated biphenyls (PCBs) and dichlorodiphenylethanes (e.g. DDT). An apex predator like the harbour porpoises suffers from bioaccumulation and -magnification, especially when the individual gets older. Higher concentrations of pollutants are thought to increase the susceptibility to diseases and parasites (Pierce et al., 2008). In addition, the older the animal, the more change of getting infested with parasites.

Location

The results of location per period suggest a shift in harbour porpoise strandings, since in H1 and I1 most porpoises stranded at the Wadden Islands, in H2 and I2 in Noord-Holland, whereas in H3 and I3 strandings in Zuid-Holland were most common. When stranding numbers in other countries are considered, it shows that France is getting more stranded harbour porpoises since 2010/2011 and that stranding numbers in Germany are actually decreasing. The results of this study suggest a shift southwards, but the extent to which this happens as well as possible reasons for such a shift needs additional research. Possible explanations for a shift could be a decline in prey availability (Camphuysen, 2004) or climate change (Evans et al., 2005) in the more northern part of the North Sea.

The primary objective of this study was to get a better insight in necropsy findings of harbour porpoises that stranded during periods of high and low stranding frequency. Stranding records from the Netherlands have shown an exponential increase in harbour porpoise strandings during the past years. Data from 1970-2013 shows two seasonal high stranding periods in strandings around March and August. Most of the harbour porpoises that strand in the Netherlands were juveniles. When the total number of strandings is examined, males slightly outnumber females, especially in neonates and juveniles. For adults, females were most prominent. Five high periods in stranding numbers were discovered between 2000-2013, of which three were present from 2009 and onwards, when necropsies were conducted. Low periods were not found between 2009-2013.

Some distinctive features were found in high stranding periods when compared to intermediate periods, the stranded porpoises were lighter, shorter, had a poorer body condition and were younger. Overall it can be said that no necropsy findings were characteristic for high or intermediate periods. When the different high periods were examined, the following characteristics per period of high stranding frequency were noted:

First high stranding frequency (H1) – Aug '08-Sep '09

In the first high period, harbour porpoises were relatively young and had a poorer body condition, compared to the intermediate period. Trauma, followed by bycatch as the cause of death was most prevalent. Emaciation was significantly low when compared with the intermediate period afterwards. Most of the strandings occurred at the Wadden Islands.

Second high stranding frequency (H2) – Apr '11-May '12

In the second high period neonates weighed less, porpoises were relatively young, more putrefied and had a poorer body condition than in the surrounding intermediate periods. Infectious as a cause of death was most common, but when compared with the intermediate periods emaciation showed to be significantly higher. Notable is that trauma was significantly lower. Most of the strandings occurred in Noord-Holland.

Third high stranding frequency (H3) – Feb '13-Jul '13

In the third high period neonates were smaller and weighed less than in the surrounding intermediate periods. Besides, the stranded porpoises were fresher regarding to their assigned decomposition code. Most of the porpoises stranded in Zuid-Holland.

Most of the findings in the high stranding periods could be explained by literature and findings of the Utrecht University. The results give a better insight of what characteristics occur during high stranding frequency periods. Even though periods of high stranding frequency were characterized by younger animals in poorer body condition, it appeared that high stranding periods are not comparable, meaning that every high period had different characteristics. This has complications for management, since this means that high stranding periods are most likely not predictable, and that each period needs to be seen and investigated as a unique event.

For this study mass, length, age, sex, NCC, DCC and cause of death were examined in high and intermediate stranding frequency periods. To get a more comprehensive understanding of what happens in these periods, other factors such as temperature, prey availability, construction activities, and fisheries should be taken into consideration as well.

Since the last period (I3) continues further in 2014, this study should be repeated in future years, in order to fully understand what happens in this period regarding necropsy findings. In addition, the stranding numbers are increasing by such a great extent in recent years, it is most likely that stranding numbers will drop in future years. Therefore, it is important to continue data collection for both the stranding database and the necropsy database, as the harbour porpoise North Sea population is bound to change to some extent.

Some of the results of this study raised some additional questions which could not be answered in the scope of this thesis. The results of this study showed a shift in strandings, so one of these questions is whether the Dutch harbour porpoise population is actually shifting from North to South. The SCANS studies executed by Hammond et al. date back to 2005, and the last study in Dutch harbour porpoise sightings dates back to 2011 (Camphuysen, 2011). It is highly interesting to know how the population distribution and abundance looks like nowadays, and if these results would confirm the results of this study. Another question raised by this study is due to the result of a high percentage of emaciated harbour porpoises, and a low percentage of trauma cases in H2. Further research should unravel whether emaciation and grey seal attacks correlate, similar to the cases on bottlenose dolphins and harbour porpoises interaction in British coastal waters. Additionally with stomach contents, fatty acid and stable isotopes analyses it might be possible to investigate whether stranded harbour porpoises foraged close to the shore or far out at sea before dying. For example, the proportion of stable isotopes of carbon found in bones and muscles give an indication of the foraging place of the harbour porpoise. In general it can be stated that when carbon values are higher, the porpoises foraged more in coastal areas (Jansen, 2013). When these results can be linked to the results of this study it is interesting to see whether porpoises close to the shore show different necropsy findings than the individuals far out at sea (e.g. differences in causes of death or the further at sea the more putrefied the porpoise).

As this study showed, interesting insights can be gained by performing necropsies. Since necropsies only started in 2009, it is of utmost important to continue with necropsies in order to obtain more knowledge and to enlarge the exiting database. Changes in climate (global warming) and anthropogenic effects (pollution, building activities, fisheries etc.) are ongoing and will have effect on animal populations and thus on the harbour porpoise as well. Continuing research on harbour porpoises might give an insight of the effects mankind has on marine mammals. Necropsies play a vital role in harbour porpoise conservation and thus aids to the protection of this species in order to preserve this Dutch indigenous cetacean for future generations.

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APPENDIX I: GLOSSARY

Adjusted	The ratio of the difference between the observed count and the expected count to the
standardized	standard deviation of the expected count. It assesses the significance for each variable
residuals	per cell.
ASCOBANS	International agreement on the Conservation of Small Cetaceans of the Baltic and North Seas (1994)
Bathymetry	The study of underwater depth of oceans or lakes
Bern Convention	International agreement which aims to conserve wild flora and fauna, natural habitats and to promote European co-operation (1979)
Bioaccumulation	The accumulation (build-up) of (chemical) substances in an organism
Bio magnification Bonn Convention	The increase of (chemical) substances as the animal is higher ranked in the food chain An agreement which is also known as Convention on the Conservation of Migratory Species of Wild Animals. Goal is to preserve migrating species (1979)
CITES	Convention on International Trade in Endangered Species of wild fauna and flora (1973)
Cetacean	A group which comprises all whales, dolphins and porpoises
Hydrography	A science which measures and describes physical features of oceans or lakes.
Longissimus dorsi	Muscle along the spinal cord
Moving average	The average of a fixed number of subsequent elements in a time series. Also abbreviated by MA.
Necropsy	Post-mortem examination
OSPAR	OSPAR is an agreement between Belgium, Denmark, Finland, France,
	Germany, Iceland, Ireland, Luxembourg, the Netherlands, Norway, Portugal, Spain,
	Sweden, Switzerland and United Kingdom to protect the marine environment of the North-East Atlantic
Dathalagy	
Pathology Promiscuous	Study and diagnosis of diseases A mating system where no strong pair-bonds are made
Profiliscuous P value	The estimated probability of rejection of the null hypothesis (H0), i.e.
r value	Determines the significance of the results
Stranding	Occurrence of a dead, ill or alive marine mammal immobilised ashore

Part 1 Identification	Number:	UT	UT					igs se nui	nbe	r:				
	GLIMS number:					E⊦	HBZ /	Hiele	cod	e:				
	Date stranded:			-				-		2	0			
	Date necropsied:			-				-		2	0			
Chip check ¹ :	Label location:													
□ yes / □ no	True location:													
negative / positive	Provided by:	EHBZ I Imares /Ecomare												
Date and time delivered:														
Notes:														
Part 2 Biometrics	Total length (tip-no Indicate size class carcass incomplete	when	L1 □ <9	0 🗆 91			cm >130		_2 _11.				.cm .cm	
Sex:	□∂ □♀ □ se	(certair x unkn	uncer OWN	tain)				3				Ŷ		
Body mass:							kg			rea	al / esti	mation		
Nutritive condition	code: 🗆 NCC1		C 2		C 3				T n		5 [יאיככ	6	
Expected age:	neonate i juve	nile 🗆	adult	🗆 un	kno	wn	, con	n \.,		delidelsens – Artus –	e	*/		
Carcass DCC:	□ very fresh DCC1 □ remains DCC5, r				•						• •			
State of carcass:	□ fully intact □ pe	ck or b	ite wou	unds		ncc	omple	te 🗆	ske	leta	l part	s, nam	ely:	
Storage:	□ direct delivery □	coole	d (ca.		nrs)		frozer	า 🗆 (othe	r:				
Bycatch:	🗆 certain 🗆 highly	probal	ole 🗆	proba	ble		poss	ible	□ n	o ev	videnc	ce , ma	arks:	
(based on external observation only)														

APPENDIX II: HARBOUR PORPOISE NECROPSY FORM

¹Harderwijk chips may be found directly underneath the dorsal fin on the left, approximately 5 cm off the mid-line

Part 2 Biometrics		Number	UT			
Organ	Side	Weight (grai	ms)	Organ	Side	Weight (grams)
Gonads	Left			Adrenals	Left	
	Right				Right	
Kidneys	Left			Lung	Left	
	Right				Right	
Spleen				Liver		
Mesenteric LN				Pulmonary LN		
Stomach				Heart		

Part 3 Photogra	phy	
Slides (digital, pr	iorities bold)	
Entire body		
Head only		
Blowhole		
Dorsal		
Pectoral fins		
Eyes		
Flank		
Tail		
Fluke		
Urogenital region		
Teeth		
Tongue		
External Observation	ons (Specify lesion and location)	
Internal observatio	ns (Specify organ)	

Vacroscopic evaluation summarised	Score						
Skin	□ ?	□ 0	□ 1		2		3
Muscular system	□?	□ 0	□ 1		2		3
Skeletal system	□ ?	□ 0	□ 1		2		3
Gastrointestinal tract	□ ?	□ 0	□ 1		2		3
Reproductive system	□ ?	□ 0	□ 1		2		3
Jrinary system	□ ?	□ 0	□ 1		2		3
Liver	□ ?	□ 0	□ 1		2		3
Spleen	□ ?	□ 0 0	□ 1		2		3
mmune system	□ ?	□ 0	□ 1 1		2		3
Respiratory system	□?	□ 0 □ 0	□ 1 □ 1		2		3
Central nervous system			□ 1 □ 1		2		3
Others			□ 1		2		3
Conclusion overall					-		0
I Provide a state of absent I or absent I or absent I or absent I or absent I moderately		ly affected	□ 3 macros		no ab	norm	aliti
Estimated significance of the presence/absence o	f criteria for the	e diagnos	is of byc	atch			
Criteria	Pr	esence	Abse	ence	Ob		
1. Health state					yes	?	n
A. Exclusion of other causes of death		+		•			
B. Good nutritional condition		+	-				
C. Evidence of recent feeding		+	0				
2. Contact with fishing gear							
A. Superficial skin lesions					yes	?	no
1. cuts in edge of mouth, fin or tail		++	0)			
2. encircling lesions around extremity		++	0				
2. encircling lesions around extremity		TT	0	·			
B. Bruises		+	0				
5 ,)			
B. Bruises		+	0)			C
B. Bruises C. Skull fractures		+	0				n
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia)		+	0		u yes	□ ?	n C
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs		+ + +	0		yes	□ ?	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae		+ + + +	0 0 - -			· · · · · · · · · · · · · · · · · · ·	[n [
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net		+ + + + +	- - 0			· · · · · · · · · · · · · · · · · · ·	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae		+ + + + +	- - 0			· · · · · · · · · · · · · · · · · · ·	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net		+ + + + + +	- - 0		· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net A. Amputated fin, fluke or tail B. Penetrating incision into body cavity C. Rope around tail stock		+ + + + + +	- - - 0 0		yes yes yes	· · · · · · · · · · · · · · · · · · ·	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net A. Amputated fin, fluke or tail B. Penetrating incision into body cavity		+ + + + + + +	- - - 0 0 0			· · · · · · · · · · · · · · · · · · ·	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net A. Amputated fin, fluke or tail B. Penetrating incision into body cavity C. Rope around tail stock		+ + + + + + + + ++	- - - 0 0 0 0 0 0 0 0			<pre></pre>	
B. Bruises C. Skull fractures 3. Lack of oxygen (hypoxia) A. Oedematous lungs B. Persistent froth in the airways C. Bullous emphysema in the lungs D. Epicardial and pleural petechiae 4. Damage during release of the net A. Amputated fin, fluke or tail B. Penetrating incision into body cavity C. Rope around tail stock D. Gaff mark		+ + + + + + + + ++	- - - 0 0 0 0 0 0 0 0		yes yes yes yes yes	<pre></pre>	

++ consistent with bycatch + bycatch possible **0** no significance for diagnosis - bycatch less likely - bycatch unlikely (Adapted from: Kuiken T. 1994. Review of the criteria for the diagnosis of by-catch in cetaceans. *In:* Kuiken T. (ed.) Diagnosis of By-Catch in Cetaceans. Proc. 2nd. ECS workshop on cetacean pathology, Montpellier, France, 2 March 1994. European Cetacean Society Newsletter 26: 38-43)

Part 5 Pathology		Nun	nber		U	Т						
Necropsy form - 1												
Date of necropsy:							Species:	Phocoen	a pho	coena		
Blubber thickness	s l	_13	r	mm (do	rsal)		L14 mn	n (lateral)	L 15		mm (v	ventral)
External observations & lesions - General observ.												
□ Skin lesions												
Net marks												
Cuts in mouth												
Cuts on fins												
Cuts on fluke												
Other cuts												
Blubber defects												
Encirc. lesions												
Denetr. incisions												
Amputations												
Rope marks												
Gaff marks												
Other												
Scavenging Subcutaneous	Seve	ere	□ Mod	erate	Mile	d	□ None					
observations & lesions												
Blubber												
Subcutis												
Musculature												
□ Skeleton												
□ Other □ Subcut. fat	Abser	nt ⊏	Preser	nt, approx	ximate tl	:hic	ckness:mr	n				

Part 5 Pathology	Number	UT
Necropsy form - 2	2	
Internal observa	tions & lesions	
Abdomen		
(tick if normal, describe if abnormal)		
Urinary bladder		
Mesenteric LN		
Intestine		
□ Stomachs: 1 st		
2 nd		
3 rd /4 th □ Spleen		
□ Pancreas		
□ Liver		
Adrenal		
□ Kidney		
Genital tract		
Genital tract LN		
	Sex 🗆 👌 🗆 🌳 🗆 Un	determined
	Age	le 🗆 Adult 🗆 Undetermined
Thorax		
(tick if normal, describe if abnormal)		
Trachea		
🗆 Lungs		
Pulmonary LN		
- Heart		
Oesophagus		
□ Thymus (present/absent)		

Part 5 Pathology	Number	UT
Necropsy form - 3		
Head and Neck		
(tick if normal, describe if abnormal)		
Blowhole		
🗆 Larynx		
Thyroid		
Oral cavity		
□ Teeth		
□ Eyes		
Auditory system		
□ Skull		
🗆 Brain		
Preliminary conclusions		
Probable cause of death		
Voorlopig resultaat voor de vinder (NL)		

Further comments:

Part 6 Sample Collection	Nur	nber		UT .							
Sample list											
	HP Forma- lin	Virolo Plasti bag / E -20°C	c F EP	acterio Plastic bag 20°C	Parasito Alcohol / Glycerin 70%/ 5%	/ A	lum	DPs ininium Jlass jar	HM Plastic bag	ALIM Plastic bag	Life History
Skin / huid	lesions	lesion	S								70% Alcohol
Blubber)J/ ex	MH	Tex	Tex	
Muscle / spier							ex		Tex	OJ/ Tex	
Rib (5 th)										OJ	
Genital split / genitale spleet		swab									
Mammary gland/ melkklier											
Gonads & reproductive tract											
Reproductive tract LN											
Placenta & umbilical cord											
Urinary bladder / urineblaas											
Intestine / darm											
Mesenteric LN											
Stomach / maag					parasites	S				ML	
Pancreas											
Spleen / milt											
Liver / lever					parasites		ex	MH	Tex	Tex	
Kidney / nier						T	ex		Tex	Tex	
Adrenal / bijnier											
Lung / long			pa	rasites	parasites	S					
Pulmonary LN											
Heart / hart											
Blood / bloed											
Thymus											
Thyroid / schildklier											
Auditory system / oor					parasites	S					
Eye / oog											
Teeth / tanden											4 teeth in water 4 in plastic bag
Brain / hersenen											
Other											
Collection/ DCC correlation	swab	DCC 1		D	CC 2		D	CC 3		DCC 4	and 5

APPENDIX III: DCC – NCC

DCC (Decomposition condition code):

The 'decomposition condition code' (DCC) is based on the external and internal decomposition signs of the carcass. It is the same as the 'condition code' (CC) defined in the ECS proceedings (Kuiken & García Hartmann, 1993)

DCC **1** Very fresh, dead less than 48 hours, may show signs of rigor mortis (< 24 h), blood still separates serum (24-48 h), rigidity of eyes is diminished but not very flaccid, cornea is not cloudy

DCC **2** Fresh, first signs of decomposition visible, eyes and surface quality of the skin reveal decomposition, otherwise good state, organs look intact, blood does not separate the serum, no smell of decomposition.

DCC **3** Putrefied, Skin peeling, moderate but clear signs of decomposition (changes in colour and consistency [flaccid]) of skin and organs, not suitable for bacteriology because of overgrowth, moderate smell of decomposition.

DCC **4 Very putrefied**, Advanced decomposition, skin and organs clearly altered, the loss of consistency changes the organ's shapes (especially liver), clear smell of decomposition, not suitable for any tissue analysis, even gross pathology is very unclear and can hardly be interpreted at all.

DCC **5 Remains**, Completely useless for pathological examination, organs are beyond clear recognition or absent, may be mummified, etc.

NCC (Nutritive condition code):

The nutritive state of the animal should be evaluated immediately before the necropsy, as a general impression gained from several details which may not be mentioned in the pathologists reports.

NCC **1** Very good nutritive condition, very well nourished, abundant blubber, significant other subcutaneous fat present in the dorsal neck and -sometimes- on the lateral thorax, Longissimus dorsi and neck are convex, the whole animal has a round, barrel-like body shape.

NCC **2** Good nutritive condition, well nourished, abundant blubber, some subcutaneous fat, Longissimus dorsi and neck are straight or slightly convex.

NCC **3** Normal nutritive condition, blubber is normal thickness, no subcutaneous fat present, neck and Longissimus dorsi are straight, upon movement of the animal sometimes slightly convex.

NCC **4 Poor nutritive condition**, blubber is on the thin side, sometimes skin thickness increased, neck and Longissimus dorsi visibly concave.

NCC **5 Very poor nutritive condition**, blubber is thin, skin thickness most often increased, Longissimus dorsi and neck clearly concave.

NCC **6** Extremely poor nutritive condition, severely emaciated, blubber is very thin, neck and Longissimus dorsi are severely concave, the contour of the scapula (especially the Spina scapulae) may be visible.

APPENDIX IV DATASET VARIABLES

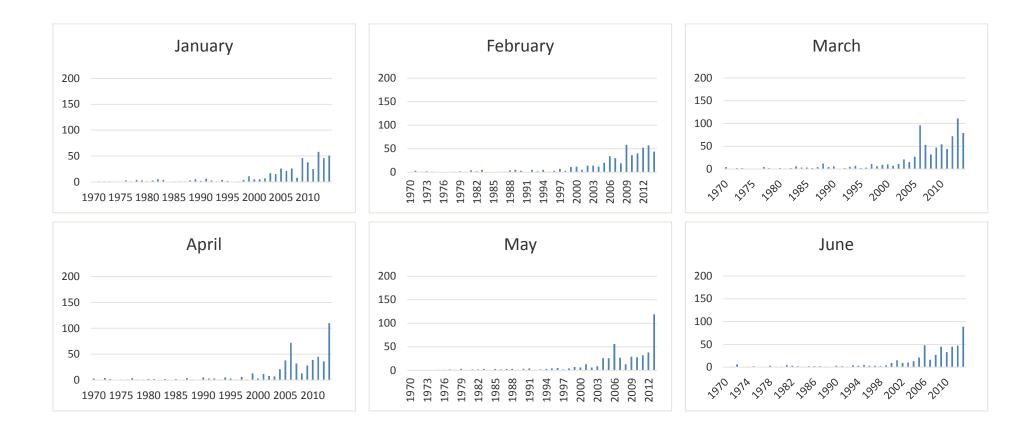
Variable	Values	Value name in SPSS
ID	Ordinal	-
Province	Groningen	1
	Friesland	2
	Wadden Islands	3
	Noord-Holland	4
	Zuid-Holland	5
	Zeeland	6
Day/Month/Year	Ordinal	-
Length	Metric (in cm)	-
Sex	Male	1
	Female	2
	Unknown	0

Table IV.1 Stranding dataset variables

Table IV.2 Necropsy dataset variables

Variable	Variable name in	Values	Value class	Value name		
Vallable	SPSS	values	value class	in SPSS		
IDcode	ID	-	Ordinal	-		
Day/Month/Year	Day/Month/Year	-	Ordinal	-		
Stranding Location	Location	-	Nominal	-		
Province	Province	See table 2.3	Nominal	See table 2.3		
Age	Age	Unknown	Ordinal	0		
0	0	Fetus		1		
		Neonate		2		
		Juvenile		3		
		Adult		4		
Sex	Sex	Unknown	Nominal	0		
		Male		1		
		Female		2		
DCC (decomposition	DCC	Unknown	Ordinal	0		
code)		Very fresh		1		
		Fresh		2		
		Putrefied		3		
		Very putrefied		4		
		Remains		5		
State of carcass	StateCarcass	Unknown	Nominal	0		
		Fully intact		1		
		Intact		2		
		Peck or bite wounds		3		
		Scavenged		4		
		Incomplete		5		
		Remains		6		
		Blubber parts		7		
		Skeletal parts		8		
Scavenging	Scavenging	Unknown	Ordinal	0		
		None		1		
		Peck marks		2		
		Mild		3		
		Mild-moderate		4		
		Moderate		5		
		Moderate-severe		6		
		Severe		7		
NCC (nutritive	NCC	Unknown	Ordinal	0		
condition code)		Very good nutritive condition		1		
		Good nutritive condition		2		
		Normal nutritive condition		3		
		Poor nutritive condition		4		
		Very poor nutritive condition		5		
		Extremely poor nutritive condition		6		
Mass	Mass in kg	-	Metric	-		
Cause of death code	DCode	See table IV.3	Nominal	0-6		
Stranding code	StrCode	Peak	Nominal	1		
		No peak		0		
Period code	PerCode	Subsequent periods	Ordinal	1-6		

Cause of death	Causes of death in necropsy dataset	Number of	Value	Value name
categories	Causes of death in netropsy dataset	individuals	class	in SPSS
Bycatch	 Certain bycatch 	150	Nominal	1
2 yearen	 (High) probable bycatch 	100	iterina	-
	 Highly probable bycatch, due to secondary 			
	infection after trauma due to bycatch			
	 Possible bycatch 			
Emaciation	• Emaciation	103	Nominal	2
	 Emaciation / blunt trauma 	100		-
	 Emaciation due to heart disease. 			
	 Emaciation due to infection 			
	 Emaciation due to tail lesion 			
	 Emaciation of unknown origin 			
	• Emaciation of unknown origin. Blunt trauma			
	 Emaciation. Blunt trauma 			
Infectious	 Infectious 	96	Nominal	3
	 Infectious / Trauma 			-
	 Infectious / Unknown 			
	 Infectious disease 			
	 Infectious? Viral? 			
	 Infectious disease. Live stranding 			
	• Sepsis due to infection - fish wire in epiglottis			
Trauma	• Acute trauma	90	Nominal	4
	o Trauma			
	 Trauma – grey seal 			
	 Trauma - not sharp edged! 			
	 Trauma - possible propeller strike 			
	 Trauma / Infectious 			
	 Trauma / Unknown? 			
	 Trauma, sharp edged 			
	 Trauma? Emaciation 			
	 Trauma? Starvation? 			
	 Possible trauma 			
	 Sharp edged trauma 			
	 Blunt trauma 			
	 Blunt trauma, acute 			
Birth defects	 Neonatal death (/other) 	27	Nominal	5
	 Perinatal problems / death 			
	• Still birth			
	• Still born			
a	o Dystocia			
Starvation	• Starvation	43	Nominal	6
	• Starvation due to infection			
	• Starvation. Blunt trauma			
Other	Starvation. Trauma.	40	No. 1	-
Other	• Live stranding / infectious	10	Nominal	7
	 1. life stranding 2. emaciation due to lung infaction 			
	infection			
	• Liver failure			
U.J	Suffocated by fish		No. 1	
Unknown	• Unknown	603	Nominal	0
	Unknown / acute death Dending			
	o Pending			



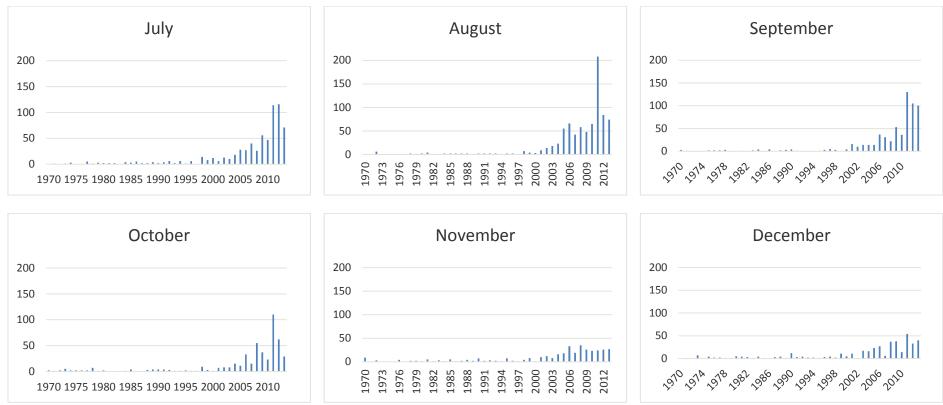


Figure V.1 Seasonal pattern in harbour porpoise strandings between 1970-2013

Table VI.1 Cause of death cross tabulation per period between 2009-2013. An adjusted residual of >1.96 (highlighted in green) shows that the number of harbour porpoises in that cell is significantly larger than would be expected, an adjusted residual of <-1.96 (highlighted in red) means smaller than expected.

				1	Period	dCode			Total
			H1	11	H2	12	H3	13	
	Ducatak	Count	12	24	7	1	2	3	49
	Bycatch	Adjusted Residual	1,2	<mark>2,8</mark>	<mark>-2,1</mark>	-1,4	-1,9	,6	
		Count	1	7	12	1	4	1	26
	Emaciation	Adjusted Residual	<mark>-2,1</mark>	-,7	<mark>2,5</mark>	-,6	,6	-,2	
	lafa ati a ua	Count	7	17	17	4	10	1	56
	Infectious	Adjusted Residual	-1,4	-,4	,9	,3	1,6	-1,1	
Cause Death Cada	Trouver	Count	14	14	7	2	6	5	48
CauseDeathCode	Trauma	Adjusted Residual	<mark>2,1</mark>	-,6	<mark>-2,0</mark>	-,7	,2	<mark>2,2</mark>	
	Dirth defecto	Count	0	2	6	3	1	0	12
	Birth defects	Adjusted Residual	-1,7	-1,2	2,0	2,7	-,4	-,8	
	Otom (otion	Count	6	6	7	2	2	0	23
	Starvation	Adjusted Residual	1,0	-,7	,5	,5	-,5	-1,1	
	Other	Count	1	2	1	1	1	0	6
	Other	Adjusted Residual	-,1	,0	-,5	1,0	,4	-,5	
Total		Count	41	72	57	14	26	10	220

CauseDeathCode * PeriodCode Crosstabulation

Table VII.1 Province/area cross tabulation per period between 2000-2013. An adjusted residual of >1.96 (highlighted in green) shows that the number of harbour porpoises in that cell is significantly larger than would be expected, an adjusted residual of <-1.96 (highlighted in red) means smaller than expected.

					TTOVING	e * Period (51055185018								
				Period											
			l: 00-01	H:01-02	I:02-05	H:05-07	L:07-08	H1	11	H2	12	НЗ	13		
		Count	0	3	13	21	7	4	7	8	1	9	0	73	
	Groningen	Adjusted Residual	-,9	,9	1,3	5,0	,5	-1,5	-,5	-1,6	-2,4	,7	-1,9		
	- · · ·	Count	0	1	5	16	6	3	9	10	0	7	4	61	
	Friesland	Adjusted Residual	-,8	-,4	-1,1	<mark>3,9</mark>	,5	-1,5	,8	-,4	-2,5	,4	,6		
		Count	24	51	286	242	182	309	315	346	166	157	63	2141	
Province	Wadden	Adjusted Residual	,1	-,1	,8	1,1	1,0	<mark>6,8</mark>	<mark>5,7</mark>	<mark>-3,2</mark>	<mark>-3,2</mark>	- <mark>5,2</mark>	<mark>-5,2</mark>		
Province		Count	12	29	107	79	51	111	101	186	121	82	58	937	
	Noord-Holland	Adjusted Residual	,5	1,5	-1,5	<mark>-2,5</mark>	<mark>-3,2</mark>	1,1	-,9	1,4	<mark>4,2</mark>	-1,4	<mark>2,2</mark>		
		Count	14	26	149	140	84	81	126	263	111	177	73	1244	
	Zuid-Holland	Adjusted Residual	,1	-,9	-1,1	,7	-1,9	<mark>-5,6</mark>	-1,8	<mark>3,0</mark>	-,5	<mark>5,7</mark>	<mark>2,0</mark>		
	Zeeland	Count	12	25	162	101	119	97	90	205	121	125	71	1128	
	Zeeland	Adjusted Residual	-,2	-,5	1,6	- <mark>2,2</mark>	<mark>3,5</mark>	<mark>-2,7</mark>	<mark>-4,3</mark>	-,1	1,8	1,4	<mark>2,6</mark>		
Total		Count	62	135	722	599	449	605	648	1018	520	557	269	5584	

Province * Period Crosstabulation

APPENDIX VIII: CROSSTABULATION COUNTRY COMPARISON STRANDING NUMBERS

Table VIII.1 EU stranding numbers cross tabulation per period between 2000-2013. An adjusted residual of >1.96 (highlighted in green) shows that the number of harbour porpoises in that cell is significantly larger than would be expected, an adjusted residual of <-1.96 (highlighted in red) means smaller than expected.

	country * year Crosstabulation														-			
	_	у	ear	y00	y01	y02	y03	y04	y05	y06	y07	y08	y09	y10	y11	y12	y13	Total
		Count		197	259	347	321	472	446	419	271	270	251	260	321	323	404	4561
	UK	Adjusted Residual		<mark>6,2</mark>	<mark>6,3</mark>	<mark>13,8</mark>	<mark>9,9</mark>	13,8	<mark>6,5</mark>	,9	-1,5	-2,5	-6,0	-1,9	-9,4	-9,6	-10,2	
		Count		77	102	78	50	50	115	45	41	146	46	72	94	67	107	1090
	Denmark Adjusted Residual			8,2	9,0	5,2	,2	-2,2	3,8	-5,7	-3,7	9,1	-4,3	,5	<mark>-2,3</mark>	-5,1	-3,4	
	Germany Adjust	Count		99	125	82	108	142	195	194	190	157	201	88	168	225	158	2132
		Adjusted Residual		4,8	4,4	-,7	1,5	1,0	2,9	,4	5,1	1,3	3,6	-4,4	-4,5	-,4	-8,5	
country	Count The	Count		69	113	113	153	204	307	550	337	345	511	437	871	730	873	5613
	Netherlands	Adjusted Residual		-9,5	-9,7	-9,8	-7,7	-9,7	-7,4	3,0	-1,5	-2,1	5,6	5,8	<mark>14,3</mark>	<mark>6,5</mark>	<mark>6,4</mark>	
		Count		8	21	14	38	40	89	94	86	62	66	46	116	97	148	925
	Belgium	Adjusted Residual		-3,9	-2,9	-4,1	-,5	-2,4	2,4	1,4	3,7	,0	-,4	-1,7	1,8	-,3	2,6	
		Count		46	60	52	68	114	107	166	134	132	169	135	205	351	498	2237
	France	Adjusted Residual		-2,8	-3,7	-4,6	-3,5	-2,3	-5,4	-2,6	-,8	-1,7	,1	-,5	-2,6	8,0	13,6	
Total		Count		496	680	686	738	1022	1259	1468	1059	1112	1244	1038	1775	1793	2188	16558

APPENDIX IX: DISTRIBUTION PER AREA OF SUPPLIED AND NECROPSIED HARBOUR PORPOISES

Table XI.1 Number of stranded harbour porpoises per province per year between 2009-2013 (n=3,410) Highest numbers per year are highlighted in blue.

	2009		201	.0	201	.1	2012 2013			3	Totals
	%	n	%	n	%	n	%	n	%	n	
Groningen	0,8%	4	0,5%	2	0,8%	7	0,7%	5	1,0%	9	27
Friesland	1,2%	6	0,5%	2	1,5%	13	0,1%	1	1,3%	11	33
Wadden	54,9%	274	47,1%	206	34,9%	304	32,1%	234	27,8%	243	1,261
Noord-Holland	16,8%	84	16,7%	73	18,8%	164	21,2%	155	16,8%	147	623
Zuid-Holland	13,0%	65	19,0%	83	23,3%	203	25,2%	184	30,2%	264	799
Zeeland	13,2%	66	16,2%	71	20,7%	180	20,7%	151	22,8%	199	667
Totals	100%	499	100%	437	100%	871	100%	730	100%	873	3,410

Table IX.2 Number of necropsied harbour porpoises per province per year between 2009-2013 (n=1,121) For one porpoise the location remained unknown and was thus excluded. Highest numbers per year are highlighted in blue.

	2009		201	.0	201	1	2012 2013			.3	Totals
	%	n	%	n	%	n	%	n	%	n	
Groningen	0.9%	1	0.0%	0	0.0%	1	0.0%	0	1.3%	3	5
Friesland	0.9%	1	1.0%	1	0.3%	1	0.3%	1	0.9%	2	6
Wadden	37.9%	44	26.7%	28	18.3%	70	12.5%	36	26.8%	61	239
Noord-Holland	25.9%	30	27.6%	29	17.0%	65	20.4%	59	24.1%	55	238
Zuid-Holland	8.6%	10	13.3%	14	25.1%	96	17.6%	51	22.8%	52	223
Zeeland	25.9%	30	31.4%	33	39.2%	150	49.1%	142	24.1%	55	410
Totals	100%	116	100%	105	100%	383	100%	289	100%	228	1,121

As it could be seen in figure 4.1, the stranding trend in the Netherlands increases more than in other EU countries. The following text gives more background information from these countries.

United Kingdom

In the UK the UK Cetacean Strandings Investigation Programme (CSIP) is a consortium of the organisations Institute of Zoology (IoZ), Scottish Agricultural College Veterinary Science Division (SACVSD), Natural History Museum (NHM) and Marine Environmental Monitoring (MEM). Their aim is to record information on all cetacean strandings in the UK and to necropsy +/- 100 cetacean per year (Roel & Payne, 2012).

The harbour porpoise is the most common and widely distributed cetacean in UK waters (Evans, 2012). The annual number of harbour porpoise strandings in the UK increased over the years. In the 1990's approximately 50-200 stranded, between 2002 and 2006 this number increased to 350-400 a year. After 2006 a decline in strandings was observed. Between 2005-2010 a total of 1,922 harbour porpoises stranded in the UK (England: 911, Scotland: 466, Wales: 507, Northern Ireland: 23, Isle of man: 14, Channel Islands:1). Of these 1,922 stranded harbour porpoises 512 were male, 466 female and 944 unknown. The months March and June reveal higher numbers of strandings (Deaville et al., 2010). During 2005-2010 478 individuals were necropsied and the three most observed probable causes of death were starvation (n=85), violent inter-specific interactions (bottlenose dolphin kills) (n=79) and bycatch (n=71) (Deaville et al., 2010).

Denmark

The most common cetacean in Danish waters is the harbour porpoise and the species occurs yearround (Evans, 2012). In 2008 223 harbour porpoises stranded along the Danish coast, in 2009 a total of 137 stranded individuals were reported. In 2010 140 harbour porpoises stranded, of which 28 harbour porpoises were necropsied. The probable cause of death for 25 individuals was bycatch, the remaining three were unknown (ASCOBANS, 2012b).

Germany

In Germany several authorities, organizations, research- and rescue centres are active in cetacean research and strandings (Schall, 2013). The vast majority of cetaceans strandings in Germany are harbour porpoises (Evans, 2012). Between 1990 and 2001, 996 harbour porpoises stranded along the North Sea coast. Male – female ratio was almost equal: 457 females and 466 males. Of 110 individuals the sex remained unknown. 17 of these stranded individuals were identified as bycatch. Strandings occurred year-round, but most stranded harbour porpoises were found in June, July and August. In the same period 229 harbour porpoises stranded on the German Baltic coast, whereof 105 had signs of bycatch (Siebert et al., 2006).

Belgium

In Belgium the Marine Animals Research and Intervention Network (MARIN) of the Royal Belgian Institute of Natural Sciences is responsible for stranded cetaceans. To a possible extent carcasses are collected and available for scientific research (Haelters et al., 2011).

The harbour porpoise is the most common cetacean in Belgium waters. Between 1970 and 2009 597 harbour porpoises stranded along the Belgium coast. From the late 1990's strandings increased, until 2008. After 2008, the number of strandings slowly decreased, but from 2010 an increase was observed again. A seasonal variation is also seen, whereas the months March to May and August show higher stranding numbers (Haelters et al., 2011). Between 1990 and 2000 40 stranded harbour

porpoises were necropsied. The three most common findings were emaciation, pneumonia and severe parasitosis (Jauniaux et al., 2002). In 2012 97 harbour porpoises stranded. A large proportion was too putrefied to conclude a possible cause of death, however, at least 15 harbour porpoises possibly died due to bycatch in fishing gear (Haelters, 2012).

France

Observatoire PELAGIS, (formerly the Centre de Recherche sur les Mammifères Marins, CRMM) and the University of La Rochelle coordinates the national marine mammal strandings recording programme. A increasing trend in stranded harbour porpoise along the French coast is seen, simultaneously an increase in sightings is observed.

Both in the Channel and Atlantic part of France year-round strandings are observed. In the Channel numbers of strandings are highest around March to May, in the Atlantic around January to March. Post-mortem research revealed that in 40% of the cases the harbour porpoise had signs of bycatch. (Van Canneyt et al., 2013).

Baltic Sea

In the Baltic Sea (located between Sweden, Finland, mainland of Europe and Denmark) the harbour porpoise is the only resident cetacean species (Scheidat et al., 2008). At least two subpopulations are recognized; the critically endangered Baltic Proper population and the more abundant Belt Sea population, both are distinct from the North Sea population. The decline in abundance is likely due to pollutants and more increasingly due to bycatch (Benke et al., 2014). In the German part of the Baltic Sea the number of stranded harbour porpoises increased from approximately 30 in 2000 to 150 in 2007. In 2007 the suspected bycatch rate was 47% (Herr et al., 2009). During the July and August most of the sightings of harbour porpoises occurred (Siebert et al., 2006).

Stranding numbers of Germany were divided between Schleswig-Holstein (SH) (North Sea and Baltic Sea), Mecklenburg – West Pomerania (MV) and Lower Saxony (LS). For this study only stranding numbers of Lower Saxony and the North Sea part of Schleswig-Holstein were used. For Denmark stranding numbers from only the North Sea coast were obtained. France divided the stranded harbour porpoise strandings between strandings on the coast bordering the Atlantic Ocean and the Channel, however, only a part of the Channel belongs to the North Sea area. For this study both numbers were combined. The UK stranding numbers were combined as well. Part of these numbers were not strandings along the North Sea (e.g. Wales, part of England and Scotland). Information about harbour porpoise strandings in Norway could not be found.