Temporary hearing threshold shifts and recovery in a harbor porpoise and two harbor seals after exposure to continuous noise and playbacks of pile driving sounds

Part of the Shortlist Masterplan Wind
'Monitoring the Ecological Impact of Offshore Wind Farms on the Dutch Continental Shelf'

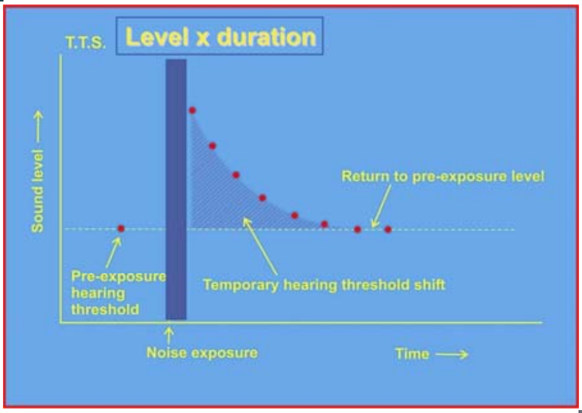

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Table of Contents

Nederlandse samenvatting

Tijdelijke gehoordrempelverschuivingen en gehoorherstel bij een bruinvis en twee gewone zeehonden na blootstelling aan ruisbanden en playbacks van heigeluiden

Veiligheidscriteria voor onderwatergeluid in de Noordzee zijn noodzakelijk om zeezoogdieren te beschermen tegen eventuele blootstelling aan gevaarlijke geluidniveaus gedurende hei-activiteiten. Echter, er is bij zeezoogdieren weinig bekend over de tijdelijke gehoordrempelverschuivingen (*Temporary Threshold Shift*; TTS) die veroorzaakt worden door niet- impulsieve en impulsieve geluiden. Om inzicht te krijgen in deze TTS en het daaropvolgend gehoorherstel, is de studie opgedeeld in twee delen, namelijk: blootstelling aan een ruisbanden (niet impulsief geluid) en blootstelling aan playbacks van opnames van heigeluiden (impulsieve geluiden).

De zeezoogdiersoorten die gebruikt zijn voor het onderzoek zijn de bruinvis (*Phocoena phocoena*) en de gewone zeehond (*Phoca vitulina*). Het gehoor van de bruinvis is het gevoeligst tussen 16 en 140 kHz en van de gewone zeehond *(Phoca vitulina)* tussen 0,5 en 40 kHz.

Om het effect van blootstelling aan **ruisbanden** op TTS te onderzoeken, zijn een jonge mannelijke bruinvis en twee jonge vrouwelijke gewone zeehonden blootgesteld aan een octaaf-band witte ruis (OBN) gecentreerd rond de 4 kHz. De TTS en het daaropvolgend gehoorherstel is door middel van een psychofysische methode gekwantificeerd (d.w.z. een dier dat getraind is op een bepaalde manier te reageren op geluiden). Elke diersoort is blootgesteld aan twee gemiddelde geluidsniveaus (SPLs:136 en 124 dB *re* 1 μPa voor de bruinvis en 148 en 136 dB *re* 1 μPa voor de zeehonden), die beide werden aangeboden gedurende een tijdsduur van 15, 30, 60 en 120 minuten. De ontvangen sound exposure levels (SELs; deze eenheid is een combinatie van geluidsniveau en de duur van het geluid) liggen in de range van 154 - 175 dB *re* 1 $\mu Pa^2 s$ voor de bruinvis, en in de range van 166 - 187 dB *re* 1 $\mu Pa^2 s$ voor de zeehonden. Zowel vóór als na de blootstelling aan de ruis zijn de gehoordrempels vastgesteld voor een smalbandige frequentie sweep (3.9-4.1 kHz; 1 s). De gehoordrempel van de bruinvis en zeehond 01 zijn vastgesteld tijdens de volgende tijdsintervallen na het uitzetten van de ruis: 1-4, 4-8, 8-12 en 48 minuten, bij zeehond 02 is deze vastgesteld op 12-16, 16-20, 20-24 en 60 minuten na de blootstelling (zeehond 02 werd namelijk altijd na zeehond 01 getest). TTS en gehoorherstel zijn gekwantificeerd door de gehoordrempels van *voor* de geluidsblootstelling af te trekken van de gehoordrempels *na* de blootstelling aan de ruis.

Gedurende de blootstelling aan de ruis zijn er geen veranderingen geobserveerd in het gedrag van de bruinvis. De hoogst gemeten TTS (~10 dB, d.w.z. dat het gehoor 10 dB minder gevoelig was dan normaal), is gemeten 1-4 minuten na blootstelling aan 120 minuten ruis met een SPL van 136 dB *re* 1 μPa (175 dB SEL). Gehoorherstel trad op, afhankelijk van de opgetreden TTS, binnen 4 - 48 minuten na de geluidsblootstelling (hoe groter de TTS hoe langer de herstelperiode duurde). Indien de geluidsblootstelling wordt uitgedrukt in sound exposure level (SEL) blijkt dat een verdubbeling van de geluidsduur meer effect heeft dan een verdubbeling van het geluidsdrukniveau (SPL) op de grote van de TTS (binnen het bestudeerde bereik van de huidige studie). Significante TTSs (in deze studie > 2,5 dB), traden op bij SELs van ~ 152 en 162 dB *re* 1 $\mu Pa^2 s$.

Bij de zeehonden is de maximale TTS (~10 dB, d.w.z. dat het gehoor 10 dB minder gevoelig was dan normaal) gemeten 1-4 minuten na blootstelling aan 120 minuten ruis met een SPL van 148 dB *re* 1 μPa (187 dB SEL). Het gehoor herstelde zich binnen een uur na blootstelling. Significante TTSs (in deze studie > 3 dB), zijn gevonden bij SELs van ~ 170 en 178 dB *re* 1 $\mu Pa^2 s$.

De studie laat zien dat SEL geen optimale voorspeller van TTS is voor continue geluiden met een lage SPL en van lange duur, omdat de duur en de SPL ongelijke rollen spelen in het veroorzaken van TTS. Indien SEL wordt gebruikt (hetgeen niet wordt aanbevolen) laten de huidige studie en de studie van Lucke *et al.* (2009) zien, dat bruinvissen kwetsbaarder zijn om lawaaigeïnduceerde TTS op te lopen dan was voorspeld door Southall *et al.* (2007). De huidige studie toont tevens aan dat de twee jonge zeehonden gevoeliger zijn om lawaaigeïnduceerde TTS op te lopen dan een oudere zeehond die gebruikt is in eerdere TTS studies (Kastak *et al*., 2005; gebruikmakend van een ruisband gecentreerd rond 2,5 kHz, trad TTS pas op bij een SEL van 183 dB *re* 1 μPa^{2}s).

Om de effecten van het **heien** op TTS te onderzoeken, zijn de bruinvis en de zeehonden blootgesteld aan laagfrequente pulsen met een piek bij 630 Hz (de meeste energie bevond zich tussen de 0,4 en 5 kHz) en met 90% van de energie in een periode van 124 ms. Dezelfde psychofysische methode als bij de ruisband studie is gebruikt om de gehoorgevoeligheid te meten vóór en na blootstelling. De gebruikte pulsherhalingsfrequentie van de afgespeelde hei-geluiden lag binnen de range die wordt gebruikt tijdens normale heiwerkzaamheden op zee (46 heiklappen per minuut, duty cycle 10%). Geen TTS werd gevonden na een blootstelling gedurende 120 minuten bij een gemiddeld piekniveau van 139 dB *re* 1 μPa (cumulatieve SEL: 152 dB *re* 1 μPa^{2}s) voor de bruinvis, en 164 dB *re* 1 μPa (cumulatieve SEL: 177 dB *re* 1 μPa^{2}s) voor de zeehonden.

Om de geluidsenergie per tijdseenheid te verhogen (om mogelijk toch TTS te veroorzaken) is het aantal heiklappen per minuut verhoogd van 46 naar 173 heiklappen per minuut (duty cycle 37%). Gehoordrempels zijn bepaald voor drie smalbandige sweeps (midden frequenties: 1, 4 en 8 kHz; bandbreedte: ± 2,5% van de midden frequentie) vóór en na blootstelling gedurende 120 minuten aan de afgespeelde heigeluiden bij een piek SPL van 139 dB *re* 1 Pa, hetgeen het maximale geluidsniveau was dat door de onderwaterluidspreker in het bruinvisbassin geproduceerd kon worden zonder dat er vervorming van het geluid optrad (SEL per puls: 115 dB *re* 1 μPa^{2}s; cumulatieve SEL: 158 dB *re* 1 μPa^{2}s), en een piek SPL van 164 dB *re* 1 μPa voor de zeehonden (cumulatieve SEL : 183 dB *re* 1 μPa^{2}s).

Zelfs met dit hoge aantal heiklappen per minuut werd geen meetbare TTS veroorzaakt, waarschijnlijk omdat het ontvangen niveau te laag was. Als TTS optrad was het zo klein dat het gehoor zich waarschijnlijk tijdens het interval tussen de pulsen kon herstellen. Uit gedragsobservaties bleek dat de bruinvis wegzwom van de geluidsbron. Eén van de zeehonden zwom weg van de geluidsbron gedurende de eerste twee sessies en ging uit het water bij een 2 dB hoger niveau. De andere zeehond zwom niet weg van de onderwaterluidspreker, hetgeen individuele variatie tussen de dieren laat zien met betrekking tot reacties op geluiden. In toekomstige gedragsstudies dienen zoveel mogelijk dieren te worden gebruikt om inzicht te krijgen in natuurlijke variatie in reacties op de geluiden.

Hoewel het aantal gebruikte hei-klappen per minuut tijdens de studie hoger lag dan in het algemeen gebruikt wordt bij de bouw van offshore windparken, suggereert de studie dat het gedrag van jonge, goed horende, bruinvissen en zeehonden op afstanden van tientallen km van offshore heilocaties beïnvloed kan worden (de mate van het effect is afhankelijk van de propagatie condities en het achtergrondgeluid). In hoeverre de geluiden de overleving en voortplanting van zeehonden en bruinvissen beïnvloed is afhankelijk van hun de tijdsbudgetten voor verschillende belangrijke ecologische gedragingen (zoals foerageren, zogen en rusten). Hierover is echter nog weinig bekend.

In de volgende fasen van dit onderzoeksprogramma zullen de heigeluiden luider worden afgespeeld om het geluidsniveau te bepalen waarop TTS begint op te treden. Tevens zullen er studies worden gedaan naar gedragsreacties van de 2 zeezoogdieren op heiklappen die worden afgespeeld met het op zee gangbare aantal per minuut. Hierdoor kunnen afstanden op zee waarop reacties optreden ten gevolge van heiklappen nauwkeuriger worden geschat.

Executive summary

Temporary hearing threshold shifts and recovery in a harbor porpoise and two harbor seals after exposure to continuous noise and playbacks of pile driving sounds

Safety criteria for underwater sound caused by offshore pile driving are needed to protect marine mammals from dangerous sound exposure. However, little is known about temporary hearing threshold shifts (TTS) induced by non-impulsive and impulsive sounds in marine mammals. To gain insight into the occurrence of and recovery from TTS in two species of marine mammal, the study was divided in two parts, namely exposure to a noise band (non-impulsive sound) and exposure to playbacks of recorded pile drive strike sounds (impulsive sounds).

The marine mammals species used in the present study were the harbor porpoise (*Phocoena phocoena*) which has its most sensitive hearing between 16 and 140 kHz, and the harbor seal (*Phoca vitulina*), which has its most sensitive hearing between 0.5 and 40 kHz.

To assess the effects of **noise bands** on TTS, a young male harbor porpoise and two young female harbor seals were exposed to fatiguing noise at eight combinations of sound pressure levels (SPL) and duration. Their temporary hearing threshold shifts (TTSs) and hearing recovery were quantified by using a psychophysical technique (they were trained to respond in a particular way when it detected a sound). Octave-band white noise (OBN) centered at 4 kHz was used as the fatiguing stimulus at two mean received SPLs per species (136 and 124 dB *re* 1 μPa for the porpoise, and 148 and 136 dB *re* 1 μPa for the seals), each offered at four durations (15, 30, 60 and 120 minutes). Received sound exposure levels (SELs) were in the range of 154 to 175 dB *re* 1 μPa^2s for the porpoise, and in the range of 166 to 187 dB *re* 1 μPa^2s for the seals. Hearing thresholds were determined for a narrow-band frequency-swept sine wave (3.9-4.1 kHz; 1 s) before exposure to the fatiguing noise, and at 1-4, 4-8, 8-12 and 48 minutes after exposure in the porpoise and in seal 01, and at 12-16, 16-20, 20-24 and 60 minutes after exposure in seal 02 (as seal 02 was always tested after 01). TTS and recovery of hearing were quantified by subtracting the pre-exposure hearing thresholds from the post-exposure hearing thresholds.

The porpoise did not respond behaviorally to the noise. The maximum TTS, measured 1-4 minutes after exposure for 120 minutes to the 136 dB *re* 1 μPa noise band (175 dB SEL), was around 10 dB (*i.e.* hearing was 10 dB less sensitive than normal). Recovery to the pre-exposure threshold was estimated to be complete within 48 minutes post exposure (the higher the hearing threshold shift, the longer the recovery). When the exposure inducing the TTS is quantified by means of the sound exposure level (SEL), this shows that a greater increase in SPL is needed than an increase in exposure duration to induce the same amount of TTS (within the range used in the present study). Significant TTSs (in this study of > 2.5 dB) occurred at SELs of ~152 and 162 dB *re* 1 μPa^2s.

In the seals, the maximum TTS, measured 1-4 minutes after exposure for 120 minutes to the 148 dB *re* 1 μPa noise band (187 dB SEL), was around 10 dB (*i.e.* hearing was 10 dB less sensitive than normal). Recovery to the pre-exposure threshold was estimated to be complete within one hour post-exposure. Significant TTSs (in this study of > 3 dB) occurred at SELs of ~170 and 178 dB *re* 1 μPa^2s.

The study suggests that SEL is not an optimal predictor of TTS for low SPL, long duration, continuous noise, as the duration and SPL play unequal roles in the determination of TTS by means of the SEL (*i.e.* doubling in duration has a greater effect than doubling in SPL). If SEL is used (which we do not advocate), the present study and the study of Lucke *et al*. (2009) show that harbor porpoises are more vulnerable to obtain noise-induced TTS than is predicted for high frequency echolocating odontocetes (Southall *et al*., 2007). It also shows that the two young harbor seals used in this study were more vulnerable to noise-induced TTS than another older animal (Kastak *et al.*, 2005; using a noise band centered at 2.5 kHz, found a TTS onset at a higher SEL of 183 dB *re* 1 μPa^{2}s).

To assess the effects of **pile driving sounds** on TTS, the porpoise and the seals were exposed to low-repetition rate pulses (playbacks of pile driving sounds) with an energy peak at 630 Hz (most energy was between 0.4 and 5 kHz) and with 90% of their energy within a 124 ms period. The same psychophysical technique was used to assess the hearing sensitivity before and after sound exposure, as was used for the noise band study. At a repetition rate within the range used in offshore pile driving (46 strikes/minute, duty cycle 10%) no TTS occurred after 120 minutes of exposure at an average received peak SPL of 139 dB *re* 1 μPa (cumulative SEL: 152 dB *re* 1 μPa^{2}s) for the porpoise, and 164 dB *re* 1 μPa (cumulative SEL: 177 dB *re* 1 μPa^{2}s) for the seals.

To increase the sound energy per time unit, the strike rate used during playbacks was increased to 173 strikes/minute (duty cycle 37%). Hearing thresholds were determined for three narrow-band frequency swept sine waves (center frequencies: 1, 4 and 8 kHz; bandwidth: ± 2.5 % of the centre frequency) before and after exposure to playbacks of pile driving sounds for 120 minutes at a peak SPL of 139 dB *re* 1 μPa, the maximum output that could be produced by the transducer in the porpoise pool without distortion (SEL per pulse 115 dB *re* 1 μPa^{2}s; cumulative SEL: 158 dB *re* 1 μPa^{2}s), and at a peak SPL of 164 dB *re* 1 μPa for the seals (cumulative SEL: 183 dB *re* 1 μPa^{2}s).

Even at this high strike rate, no measurable TTS was induced, probably because the received level was too low. If TTS did occur it was of such low magnitude that hearing probably recovered during the interval between the pulses. Behavioral observations showed that the porpoise swam away from the sound source. One of the seals swam away from the sound source during the first two sessions, and hauled out at a 2 dB higher level. The other seal did not swim away from the transducer when the pile driving sounds were played back, which demonstrates individual variation between animals in behavioral reactions to sounds. Behavioral response studies should involve as many animals as possible to gain insight into natural variation in responses to sounds.

Although the pile driving strike rate used in the present study was higher than that generally used in the construction of offshore wind farms, the study suggests that the behavior of young porpoises and seals, with good hearing, can be affected up to tens of km away from offshore pile driving sites (to what degree depends on the propagation conditions and background noise). How this would affect the seals' and porpoises' survival and reproduction depends on their time budgets for various ecologically important behaviors (such as foraging, suckling, and resting).

In the next phases of this research program, playbacks of pile driving sounds at a higher SPL could provide data on the TTS onset level, and behavioral response studies with playbacks of pile driving sounds at the normal strike rate could serve to make the estimates of deterring distances at sea more accurate.

1. Temporary hearing threshold shifts and recovery in a harbor porpoise and two harbor seals after exposure to continuous noise and playbacks of pile driving sounds

1.1. Background

Due to increasing levels of anthropogenic noise in the oceans, and growing concerns about its effects on the hearing and behavior of marine mammals (**Figs. 1 and 2**), there is a critical need for safe criteria for the exposure of marine mammals to noise. Some human activities, such as offshore pile driving, generate noise that is potentially harmful to marine mammals. The countries around the North Sea continue to build offshore wind turbine farms. So far, the wind turbines have been attached to the sea bed by means of large-diameter monopiles, which are driven into the sea bed with diesel pile drivers. Offshore pile driving produces high level impulsive sounds both in air and underwater. There is concern that at such high levels, sounds may cause hearing damage in marine mammals. Hearing impairment can be permanent (deafness) or temporary (later recovering to former sensitivity); both may have far-reaching effects on the survival and reproduction of the animals, and by extension, may have population-level impacts.

The recent European Marine Strategy Framework Directive states that underwater noise is a pollutant that, if harmful, needs mitigation, and for the European Birds and Habitats Directives, more information is needed about the effects of pile driving sounds on marine mammals. To determine if the pile driving sounds are harmful or not, and whether mitigation measures are needed, knowledge of the effects of pile driving sounds on marine mammal hearing is needed. In order to fill the knowledge gaps relating to the effects of underwater sound on marine mammals, a scientific research program was funded by the Netherlands Ministry of Infrastructure and the Environment, and the Ministry of Economic Affairs, Agriculture and Innovation. The outcomes of the research will form building blocks for understanding and mitigating any effects of underwater pile driving sounds on marine mammals. The Ministries commissioned SEAMARCO to carry out part of the research program: a project on the effects of high amplitude sounds on the hearing of marine mammals. The ultimate goals are to improve environmental risk assessments for the safe planning and execution of offshore pile driving activities, and to determine whether mitigation measures are needed.

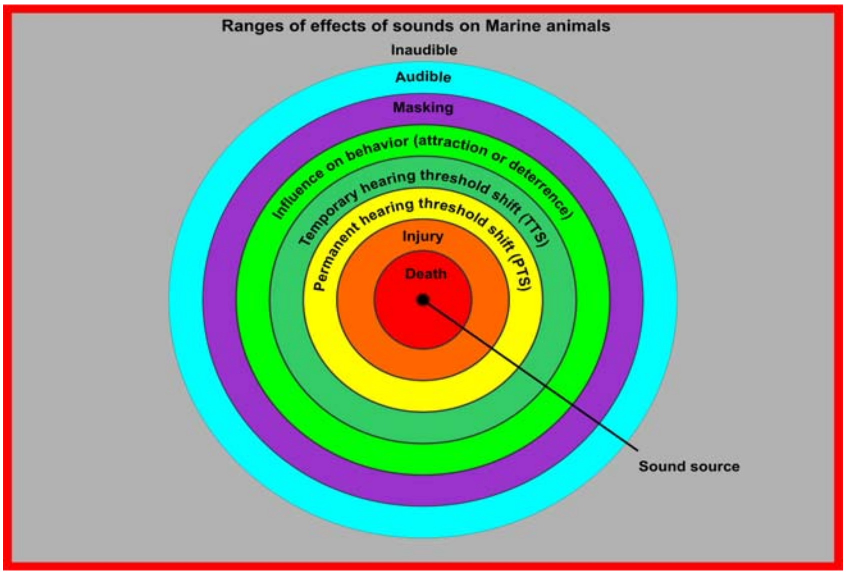

FIG. 1. A schematic presentation of the potential effects of a loud sound source on marine animals. As distance from the sound source increases, the received sound level decreases, and the severity of the effect decreases. The further away from the sound source, the wider the circle, so the lower the received sound level, but the higher the number of animals likely to be affected. In reality the circles often overlap (and may even change position, so for instance behavior may be influenced closer to the source than masking occurs), and may vary in width (they are not to scale) depending on characteristics of the sound source, propagation conditions, and characteristics of the receiver (the hearing of the animal).

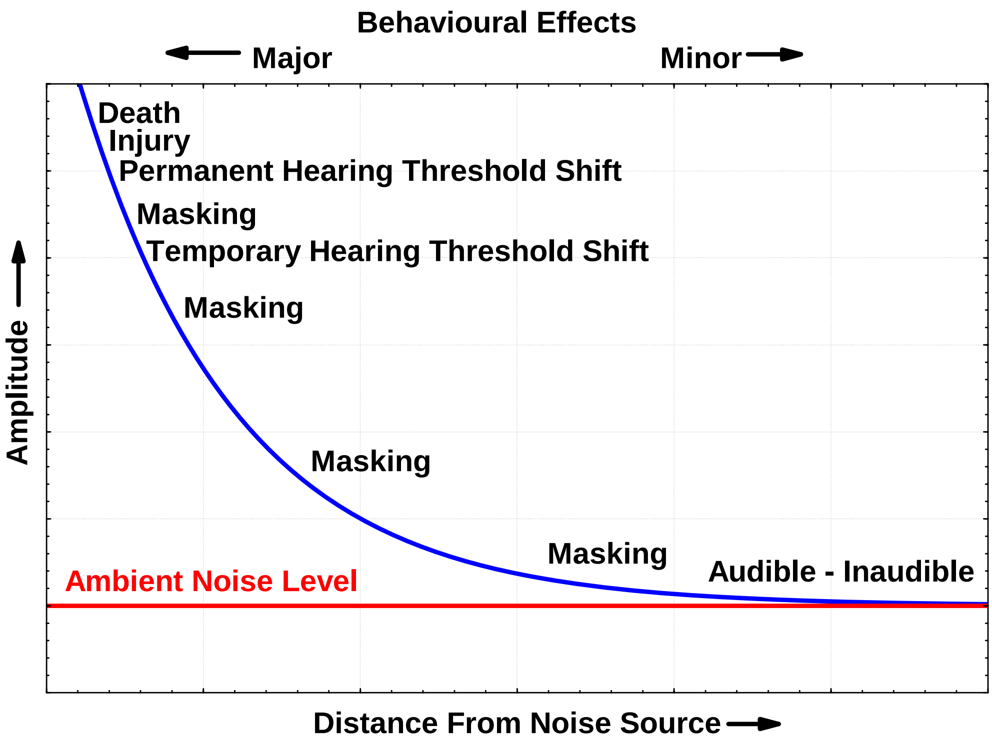

FIG. 2. The effects of sound as shown in Fig. 1, but with a more realistic linear scale showing that death and injury may only occur very close to a loud sound source, and that masking occurs in a wide range around the sound source. The sloping line depicts the gradual SPL drop as the distance increases (By John M. Terhune).

1.2. Introduction

High amplitude sounds can cause permanent hearing threshold shift (PTS), which means permanent loss of hearing. Exposure of marine mammals to sounds at such high levels should be avoided, and safety criteria to prevent this should be adopted. PTS studies have not been conducted with captive marine mammals for ethical reasons; instead, hearing damage is usually studied indirectly via studies on temporary hearing threshold shift (TTS; **Fig. 2**). This can be safely measured, and the data can be used to develop allowable exposure criteria.

TTS has been studied in chinchillas (*Chinchilla lanigera*), rodents with similar hearing to humans, to gain insight into hearing damage occurring in humans due to noise exposure (Clark, 1991). The occurrence and magnitude of TTS is determined by a variety of parameters, including:

1) Exposure level
2) Exposure duration
3) Spectral (frequency) content
4) Temporal pattern (continuous versus pulsed)
5) Kurtosis, or ‘peakedness’, of the signal (whether it is impulsive or non-impulsive)
6) Affected animal’s hearing range (frequency and sensitivity).

Studying TTS is complicated and time-consuming, as many combinations of parameters need to be studied in order to be able to predict the onset of TTS, the magnitude of TTS, and the timing of recovery after exposure to a particular sound for a particular period.

Not only PTS should be prevented, but also long duration TTS. Behavioral effects, resulting in, for example, animals temporarily or permanently abandoning ecologically important areas, may also be important results of exposure to pile driving sounds. For instance, porpoises have a high digestive rate and have to eat many times per day. If TTS influences its ability to catch fish for several hours, a porpoise’s physiological state may change. Its blubber layer may get thinner, causing the animal to lose more energy to the water, so that it requires even more food to maintain its body temperature at 37 °C in cold ocean water. If food is not readily available (due to the animal being deterred from optimal foraging areas), the porpoise may become hypothermic and die, or may have to spend energy searching for suitable food instead of carrying out other biologically important activities (e.g. breeding, lactation, etc.).

The most abundant marine mammal species in the Dutch part of the North Sea are the harbor porpoise (*Phocoena phocoena*) and the harbor seal (*Phoca vitulina*). Therefore, in this project we focus on the effect of sounds on the hearing of these species. Both species have similar, and very wide, distribution areas in the temperate coastal waters of the Northern hemisphere, so the results of the project are of value to the government regulators and policy makers of many countries.

In summary, the aims in the four studies making up the project were to determine:

1. The level/duration combinations of a noise band which cause TTS, the degree of TTS, and the rate of hearing recovery in porpoises.
2. The level/duration combinations of a noise band which cause TTS, the degree of TTS, and the rate of hearing recovery in harbor seals.
3. The level/duration combinations of playbacks of impulsive pile driving sounds which cause TTS, the degree of TTS, and the rate of hearing recovery in porpoises.
4. The level/duration combinations of playbacks of impulsive pile driving sounds which cause TTS, the degree of TTS, and the rate of hearing recovery in harbor seals.

1.3. Synopsis of methodology

We studied TTS in a captive harbor porpoise and two harbor seals, by using the behavioral psycho-acoustic method (*i.e.*, using trained animals) to determine hearing thresholds before and after exposure to loud sounds likely to cause hearing loss, known as fatiguing noises. The hearing thresholds after exposure and the recovery time were measured, and compared to baseline (pre-exposure) hearing thresholds to calculate the extent and duration of TTS (**Fig. 3**).

The SEAMARCO Research Institute was built specifically for underwater acoustic research, and so is very suitable for hearing studies (*i.e.* it is extremely quiet). The inter-session standard deviation of hearing thresholds recorded here is usually less than 2 dB, whereas in other facilities the standard deviation for hearing thresholds is often around 6 dB. Therefore, we are very confident that our hearing thresholds, and the TTS values calculated from them, are accurate.

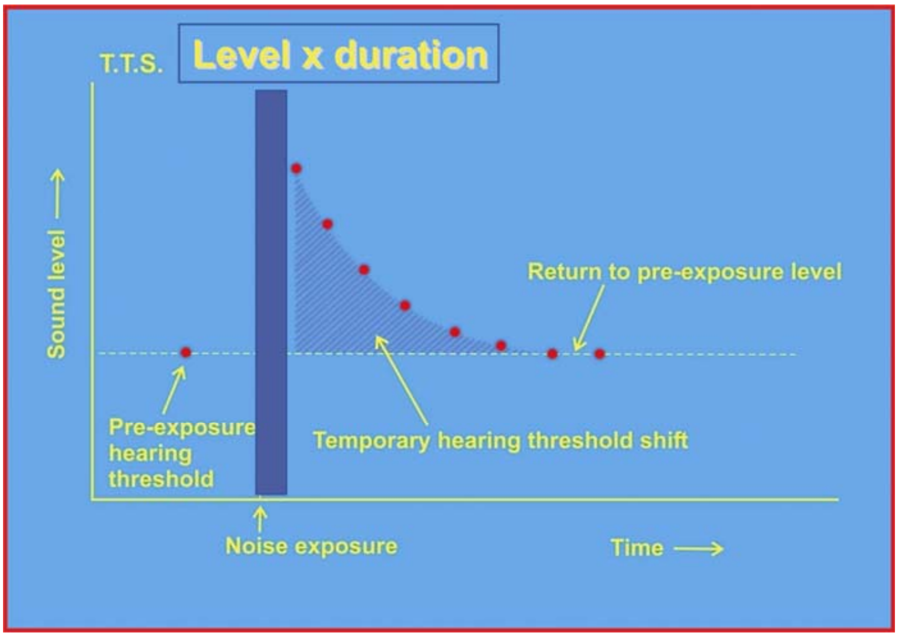

FIG. 3. The method used to measure TTS and recovery of hearing. First the pre-exposure, or baseline, hearing threshold is quantified. Then the animal is exposed to fatiguing noise of a certain level and duration (vertical blue bar). Thereafter, the hearing threshold is quantified several times, until it returns to the pre-exposure level (dashed horizontal line).

The potentially harmful fatiguing sounds of interest for this project were those produced during pile driving. Continuous broadband noises (similar to the sound of a vacuum cleaner) were first used as the fatiguing noise, so that the frequency band and level could be determined precisely, and because these noises were expected to cause less anxiety in the animals than pile driving sounds. Pile driving strike sounds have most (99%) of their energy in frequencies below around 5 kHz (**Fig. 4**). In order to produce high enough sound pressure levels with the transducers, the pile driving frequency bandwidth was divided into four octave-band white noise bands centered at 0.5, 1, 2 and 4 kHz. Higher frequency sounds cause TTS at lower fatiguing noise levels than lower frequency sounds in bottlenose dolphins (*Tursiops truncatus*), and the TTS increases at a higher rate for high frequency sounds (Finneran and Schlundt, 2010). Therefore the highest frequency noise band, with a centre frequency of 4 kHz (bandwidth: 2.8 – 5.7 kHz), was used in this project as the fatiguing noise tests (because porpoises have similar hearing characteristics as bottlenose dolphins). The three other noise bands may be carried out in later studies (**Fig. 4**). The noise band was offered at four durations and at two levels for each species. Also, control sessions were conducted during which the animals were only exposed to the normal (low) ambient noise (**Table 1**).

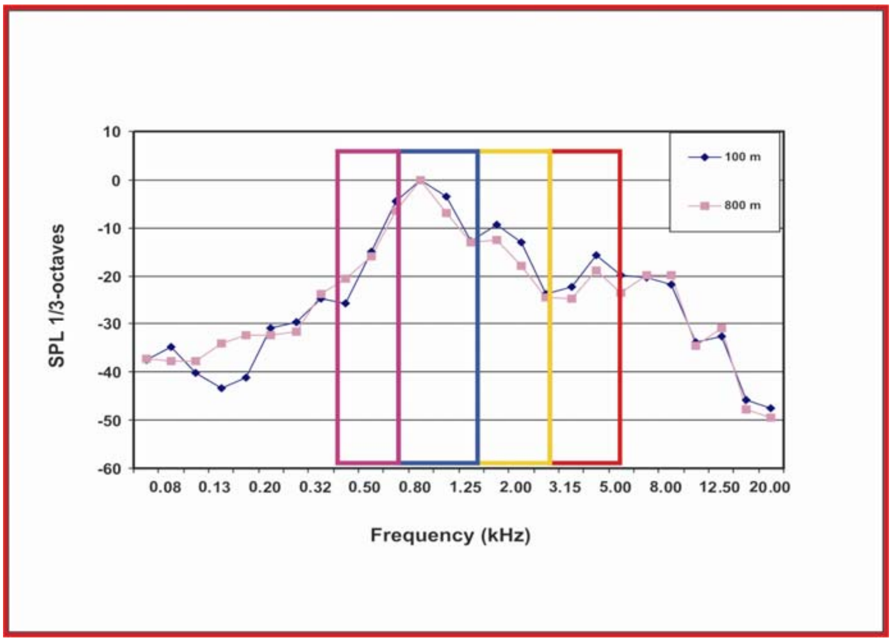

FIG. 4. The spectra of played back pile driving sounds recorded at 100 and 800 m from the pile driving site. Also shown are the four one-octave noise bands, of which the one on the far right (red box; centered around 4 kHz) was used as the fatiguing noise in the present project.

Table 1. The levels and durations at which the one-octave noise band centered at 4 kHz was offered to, and TTS was determined for, the porpoise (P) and seals (S). Also shown are the control sessions during which no fatiguing noise was offered.

4 kHz	Exposure time (minutes)			
Mean exposure SPL (dB 1 dB *re* 1 μPa)	15	30	60	120
148	S	S	S	S
136	S, P	S, P	S, P	S, P
124	P	P	P	P
Control	S, P	S, P	S, P	S, P

After determining the relationship between continuous broadband noise exposure and the occurrence of TTS in the two marine mammal species, we conducted similar tests in which the animals were exposed to playbacks of impulsive pile driving sounds.

1.4. History of the project

Because so little is known about TTS in harbor porpoises and harbor seals, and in order not to compromise the hearing and wellbeing of the animals, the study started cautiously; noise bands were used instead of impulsive sounds, and the received level and exposure time were increased in small increments. It took about a month to find safe levels and durations of the noise bands that could be tested in the main noise band TTS study. The sounds were shown to be safe and we exposed both the porpoise and the seals to the maximum levels the systems could produce (the studies were conducted in parallel due to the time pressure, and unfortunately the two new transducers each produced a different maximum output sound pressure level).

In the contract for the project between the Ministries and SEAMARCO, the following three studies were stipulated:

1. Test the effect of a noise band on the hearing of a harbor porpoise.
2. Test the effect of a noise band on the hearing of harbor seals.
3. Test the effect of pile driving sound on the hearing of harbor seals.

During the course of the project, the advisory committee showed an interest in testing the effects of pile driving sounds on harbor porpoises as well. Therefore, a fourth study was added to the project. Because the available time for the total TTS project remained the same, fewer noise band level-duration combinations could be tested, in order to have time available for the extra harbor porpoise pile driving study.

Although the goal of the project was to determine the TTS onset levels of noise bands and pile driving sounds, it became clear during the project that behavioral components were important as well. The studies were not designed as behavioral studies (the sound pressure levels were made as high as possible), but the animals' behavior during sound exposure was recorded on video and analyzed.

At the moment (June-August 2011) SEAMARCO is continuing to quantify TTS caused by 4 kHz noise bands in both species, in order to increase the number of available level-duration combinations. This, so far, self-funded research will allow us to determine more precisely the TTS onset levels, and to develop a model to predict TTS levels for other level-duration combinations.

1.5. Summary of results

Harbor porpoise noise bands

The maximum TTS, occurring 1-4 minutes after a 120 min exposure to 136 dB *re* 1 µPa, was around 10 dB (*i.e.* hearing was 10 dB less sensitive than normal) at a SEL of 175 dB *re* 1 $\mu Pa^2 s$ (**Fig. 5**). In one case, hearing recovered (*i.e.* returned to normal) at about 4 min post-exposure; in two cases, recovery occurred at about 8 min post-exposure; in the remaining five cases, it occurred between 12 and 48 minutes post-exposure. When TTS was induced it was relatively more affected by the increase in the exposure duration than by an increase in level (within the SPL range used in the present study). Significant TTSs, in this case of >2.5 dB, occurred at SELs of ~152 and 162 dB *re* 1 $\mu Pa^2 s$. The study suggests that SEL is not the optimal predictive parameter for TTS onset for long duration, low-level continuous noise. If SEL is used (which we do not advocate), the present study and the study of Lucke *et al.* (2009) show that harbor porpoises are more vulnerable to noise-induced TTS than cetaceans echolocating at high frequencies are predicted to be by Southall *et al.* (2007).

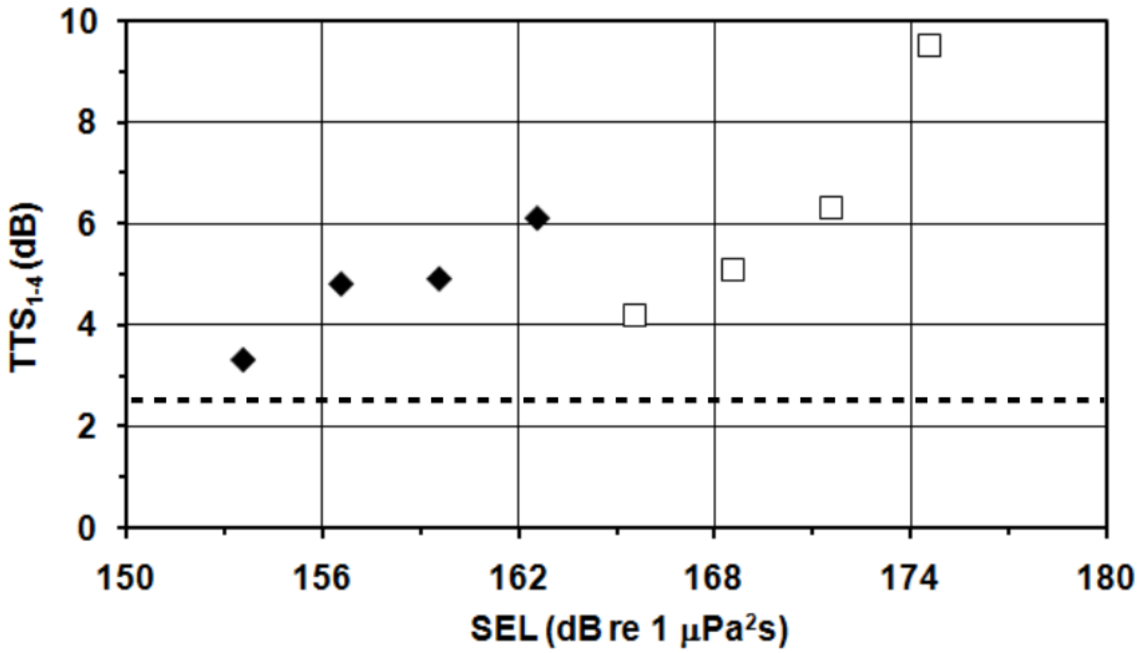

FIG. 5. The TTS_{1-4} (dB) induced in the first 4 mins after noise exposure in the harbor porpoise, as a function of the sound exposure level (SEL; in dB), for fatiguing sounds at 124 dB SPL (◆) and 136 dB SPL (□).Results are shown, from left to right, in order of increasing duration of fatiguing noise (15, 30, 60 and 120 min). The dashed line indicates the approximate level (2.5 dB) above which TTSs were found to be significant in the present study.

Harbor seal noise bands

The maximum TTS, measured 1-4 min after a 120 min noise exposure to the 148 dB *re* 1 µPa noise band (187 dB SEL), was around 10 dB (*i.e.* hearing was 10 dB less sensitive than normal; **Fig. 6**). Recovery to the pre-exposure threshold was estimated to be complete within one hour post-exposure. Significant TTSs, in this case of >3dB, occurred at SELs of ~170 and 178 dB *re* 1 $\mu Pa^2 s$. The study suggests that SEL is not an optimal predictor of TTS for long duration, low-level continuous noise, as the duration and level play unequal roles in the determination of the SEL. With SEL as a predictor of TTS, the present study shows that the two harbor seals used were more vulnerable to noise-induced TTS than another (older) animal (Kastak *et al*., 2005; using a 2.5 kHz centered noise band, found a TTS onset at an SEL of 183 dB *re* 1 μPa^{2}s).

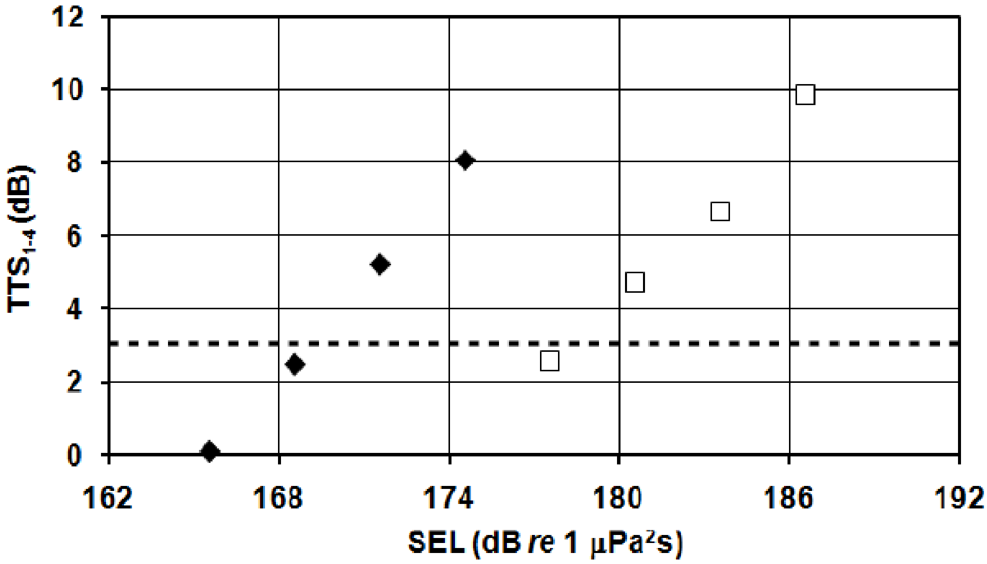

FIG. 6. The TTS (dB) induced in the first 4 mins after noise exposure in seal 01, as a function of the sound exposure level (SEL; in dB re1 μPa^{2}s), for fatiguing sounds at 136 dB SPL (◆) and 148 dB SPL (□). Results are shown, from left to right, in order of increasing duration of fatiguing noise (15, 30, 60 and 120 min), and only for TTS measured 1-4 min after exposure (so that seal 02 is not included). The dashed line indicates the approximate level (3 dB) above which TTSs were found to be significant in the present study.

Harbor porpoise pile driving sound playbacks

Hearing thresholds were determined for three narrow-band frequency swept sine waves (center frequencies: 1, 4 and 8 kHz; bandwidth: ± 2.5 % of the centre frequency) before and after exposure to playbacks of pile driving sounds for 120 min (173 strikes/min) at the maximum output that could be produced by the transducer without distortion (SEL per pulse: 115 dB *re* 1 μPa^{2}s, cumulative SEL: 158 dB *re* 1 μPa^{2}s). Even with this high strike rate, no measurable TTS was induced, probably because the received level was too low; if TTS did occur it was of such low magnitude that the porpoise's hearing probably recovered during the 220 ms interval between the pulses. Behavioral observations showed that the porpoise swam away from the sound source. Although the pile driving strike rate was higher than generally used in the construction of offshore wind farms, the study suggests that porpoises are affected tens of km away from offshore pile driving sites (the degree of effect depends on the propagation conditions and background noise).

Harbor seal pile driving sound playbacks

No TTS was measured immediately after exposure for 120 min (173 strikes/min at a peak SPL of 164 dB *re* 1 μPa; cumulative SEL: 183 dB *re* 1 μPa^{2}s). One seal swam away from the sound source during the first two sessions, and hauled out at a 2 dB higher level. No measurable TTS was observed in this seal, which had not tried to haul out to higher amplitude continuous octave-band noise in a previous study. The other seal did not swim away from the transducer when the pile driving sounds were played back, which demonstrates individual variation in behavioral reactions to sounds. Therefore, behavioral response studies should involve as many animals as possible to gain insight into the natural variation in behavioral responses. Although the pile driving strike rate was higher than generally used in the construction of offshore wind farms, the study suggests that harbor seals are affected tens of km away from offshore pile driving sites (the degree of effect depends on the propagation conditions and background noise).

1.6. Comparison of the two species and predictions by Prins *et al.* (2008)

The results from the two noise band studies (sections 2.1. and 2.2. of this report) show that, despite their high frequency specialization, harbor porpoises are more vulnerable to TTS than harbor seals (for the noise bands tested). For the porpoise, we found significant TTS above approximately 2.5 dB, at SELs of 152 and 162 dB *re* 1 μPa^2s, whereas significant TTS occurred for seal 01 at approximately 3 dB, at SELs of 170 and 178 dB *re* 1 μPa^2s (**Fig. 7**).

The harbor porpoise was deterred (in all sessions) by pile driving playback sounds with a peak SPL of 139 dB *re* 1 μPa, whereas seal 01 (not seal 02) swam away from the transducer only during the initial two sessions with pile driving playback sounds with a peak SPL of 164 dB *re* 1 μPa. Thus, it can be concluded that, though individual variation exists, the avoidance threshold level for seals is much higher than for porpoises, and that harbor porpoises are probably deterred further away from a pile driving site than harbor seals.

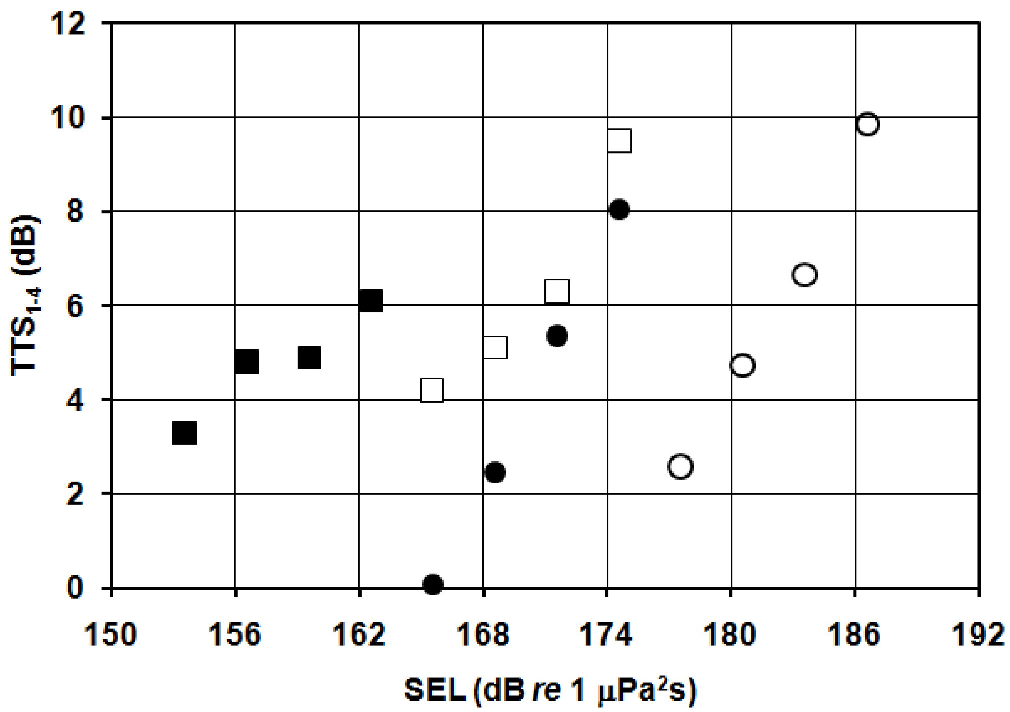

FIG. 7. The TTS (dB) in the first 4 min after sound exposure, for seal 01as a function of the sound exposure level (SEL), for fatiguing sounds at 136 dB SPL (●) and 148 dB SPL (○), and for the harbor porpoise as a function of the SEL for fatiguing sounds at 124 dB SPL (◆) and 136 dB SPL (□). Results are shown, from left to right, in order of increasing duration of fatiguing noise (15, 30, 60 and 120 min). TTSs were found to be significant above approximately 2.5 dB for the porpoise and 3 dB for the seals.

The results from the present project show that the predictions made by Prins *et al.* (2008) of distances from pile driving sites at which TTS starts to occur in harbor seals and harbor porpoises are not correct (the predictions were based on the best available knowledge at the time). The TTS onset SPLs of harbor seals for frequencies around 4 kHz are higher than for the harbor porpoise (counter to what was expected from their audiograms). Also, the avoidance threshold SPL (from which avoidance distances can be calculated) was predicted to be lower for harbor seals (resulting in avoidance at greater distances) than for harbor porpoises. Although the present project cannot provide specific distances, because the studies were designed to measure TTS and not behavioral responses, it appears that the avoidance threshold SPLs are higher for harbor seals than for harbor porpoises (i.e. that harbor porpoises are deterred further away from pile driving sites than harbor seals). However, controlled behavioral response studies should be performed to quantify and characterize behavioral reactions as a function of sound exposure type.

1.7. Ecological significance

For small TTS values (<10 dB), like those caused by the noise band exposures in the present studies, recovery in both seals and porpoises is very fast (around 30 min). In most cases, reduced hearing for such a short time period probably has little effect on the total foraging period of a porpoise or a seal. Higher exposure levels and longer exposure durations cause higher TTS values, and may cause PTS. If hearing is impaired, for periods of hours or even days, the impact is likely to be ecologically significant. Harbor porpoises forage by using echolocation, which requires acute hearing. To our knowledge, there have been no studies examining the effects of TTS on echolocation or foraging ability in odontocetes.

When TTS is induced by octave band noise, it is more affected by changes in exposure duration than by changes in level. This means that exposure to relatively low-level, long duration, anthropogenic noises may have an effect on the hearing of porpoises, and exposure to slightly louder sounds may affect the hearing of seals. However, the TTSs observed after such exposures are relatively small, and recovery is rapid.

During the noise band exposure periods, the swimming pattern of the seals was similar to that during the control periods: both underwater and aerial sound emissions did not cause the seals to swim away from the sound sources (probably because the noise was non-impulsive). In the wild, TTS may therefore be caused by sounds at levels that do not deter harbor seals away from the sound source, and it would be difficult to assess whether seals had habituated to a noise source without any adverse impacts, or whether they were experiencing a TTS (which would reduce their ability to hear sounds, and therefore make them less likely to avoid areas close to sound sources).

Although the seals did not avoid being under water, or the underwater source of the noise, they did spend more time with their heads at the water surface during fatiguing noise exposure. The study suggests that anthropogenic noise may affect the amount of time wild seals spend foraging underwater. How this would affect the seals' survival and reproduction depends on their time budgets for various ecologically important behaviors.

The most ecologically important frequency band for harbor porpoises is a 10 kHz band around ~125 kHz: the dominant frequency band of their echolocation signals. Small TTSs caused by noise bands centered at 4 kHz probably have little or no short-term effect on echolocation ability, and therefore have little ecological impact on harbor porpoises. However, little is known about the long-term effects of multiple and large TTSs, and on their effect on echolocation ability. Echolocation is a highly synchronized neurological mechanism, and TTS may lead to increased neural activity in the auditory pathways (as seen in chinchillas; Salvi *et al*., 2000), so TTS may affect the echolocation ability of the harbor porpoise. The overall effect of hearing disruption on echolocation (and therefore foraging) behavior may be critical. Research is needed to get insight in these effects.

By contrast, the frequency range of best hearing (within 10 dB of maximum sensitivity) of the harbor seals in the present study is from 0.5 to 40 kHz. This means that the TTS caused by the noise band centered at 4 kHz in the present study may have an ecologically important impact; it is likely to reduce the audibility of ecologically and socially important sounds for seals. For example, a TTS of 6 dB would halve the distance at which the seal, suffering that TTS, would be able to detect another seal, a prey item (e.g. a vociferous fish), or a predator acoustically.

Although no TTS occurred in the harbor porpoise or in the harbor seals as a result of pile driving playbacks, the information obtained can be used conservatively to calculate the distance from pile driving sites at which impacts to hearing are negligible. For this calculation, information is needed about the source level of the sound source (pile driving) and the propagation conditions, as well as the duty cycle and level and duration of the sounds the animal was exposed to in the present study.

A model, based on the relatively low playback levels used in the study, using assumed propagation conditions, and taking into account the fact that the playback strike rate the animals were exposed to was much higher than that normally used at sea, indicates that TTS is unlikely to occur outside a few hundred km of a pile driving site. However, behavioral effects of pile driving sounds may occur at distances of up to tens of km for harbor porpoises and harbor seals.

These estimates do not take into account potential habituation to the sounds or the effect of the proximity of the sound source, which was very close to the animals in this study. The behavioral effects warrant further study, as does the effect of the distance from the sound source of the animals. Despite the fact that harbor seals have more sensitive hearing than harbor porpoises in the frequency range in which most of the energy of pile driving sounds occur, the porpoise showed behavioral responses to the playbacks of pile driving sounds at lower received levels than the seals. It appears that not only the perceived level (i.e. the level relative to the hearing thresholds) plays a role in determining the effect of sounds on animals, but also the psychological perception (which is probably individual and species specific).

1.8. Suggestions for future research

The occurrence and magnitude of TTS depend on many parameters of the fatiguing sound, including exposure SPL, exposure duration, spectral content, temporal pattern (continuous or pulsed), and kurtosis (impulsive or not). Measureable TTS also depends on the hearing frequency that is tested to quantify it. Although the four studies making up the present project contribute significantly towards understanding TTS in harbor porpoises and harbor seals, not all the parameters could be tested in the 6 months (for the porpoise) and 8 months (for the seals) that were available for the project between the time the contract was signed (July 27, 2010) and the time the first draft report had to be submitted (April 1, 2011). In the Masterplan Wind 'Monitoring the Ecological Impact of Offshore Wind Farms on the Dutch Continental Shelf', three years were allocated for a comprehensive study on TTS in harbor porpoises. The studies on TTS caused by noise bands, with the addition of some data collected by SEAMARCO in self-funded research, will be submitted for publication. However, much more research on TTS caused by pile driving sounds is needed, and the studies in this report are considered to be phase 1 of a planned three-phase study. In the next 2 phases, the effect of pile driving playbacks on the behavior and hearing in the two marine mammal species will be studied in greater detail to increase our understanding.

It would be advantageous if funding for phases 2 and 3 became available soon, as the equipment set-up for pile driving sound emissions is in place, and the research team and study animals now have recent experience of research on TTS. The next phase of the Masterplan Wind 'Monitoring the Ecological Impact of Offshore Wind Farms on the Dutch Continental Shelf', could start at short notice if funding becomes available.

Suggestions resulting from harbor porpoise and harbor seal noise band studies

The degree of TTS is relatively more affected by the exposure duration than by the level, so low-level, long duration, anthropogenic noises may affect the hearing of harbor porpoises and harbor seals. In order to enhance our knowledge of TTS, and to relate it to environmental noises of concern, future research should include: finding the hearing frequency most affected by the 4 kHz noise band used in the present study (including, for porpoises, testing hearing for 125 kHz signals, similar to echolocation signals); carrying out more tests with 4 kHz bands, at shorter durations and higher and lower levels, so that sufficient data are available to model TTS development; evaluating the effect of fatiguing noise centre frequency on TTS (by testing one-octave noise bands with other centre frequencies); and quantifying the effect of intermittent noise (pulses) on TTS and recovery.

Suggestions resulting from harbor porpoise pile driving playback study

This study, part of phase 1 of a research program on the effects of pile driving sounds on harbor porpoises, was designed to demonstrate TTS in a harbor porpoise, but observations showed that the behavior of the porpoise was affected by the playbacks of pile driving sounds before TTS occurred. Therefore, we propose two follow up studies. As phase 2, we propose playbacks of pile driving at a higher SPL which could provide the TTS onset level. As phase 3, we propose a study of the behavioral response of porpoises to playbacks of pile driving sounds (normal duty cycle: ~10%) at four threshold levels which will be determined during a pre-test: 1) just no reaction, 2) increased respiration rate (increased anxiety and increased energy expenditure), 3) increased distance to the sound source (displacement from potential important habitats; decreased foraging time), 4) refusal to eat fish (this would reduce the food intake in the wild, leading to a reduced body condition).

The information which will be gained from this proposed study is important, because if porpoises swim away from pile driving sites, their received level will decrease, and thus the chances of them experiencing TTS will also decrease. On the other hand, TTS will probably only occur in a few porpoises which are close to the pile driving site. However, if a larger number of porpoises are deterred from a large area around the pile driving site (hundreds of square km), they may be driven from ecologically important areas, such as feeding areas. Such a behavioral response by many animals may have a greater effect on the population than TTS in a few individual animals.

Suggestions resulting from harbor seal pile driving playback study

The present study, part of phase 1 of a research program on effects of pile driving sounds on harbor seals, was designed to demonstrate TTS, but instead observations showed that the behavior of one of the seals was affected by the playbacks of pile driving sounds. As phase 2, we propose playbacks of pile driving at a higher SPL which could provide the TTS onset level. As phase 3, we propose a study of the behavioral response of harbor seals to playbacks of pile driving sounds (normal strike rate: duty cycle: ~10%), in which five levels will be determined during a pre-test: 1) just no reaction, 2) increased time spent with the head above the water surface (meaning reduced foraging time in the wild), 3) increased distance to the sound source (meaning displacement from potential foraging areas in the wild; decreased foraging time), 4) hauling out (meaning cessation of foraging in the wild, although this may be unrealistic as haul out areas are not often available in the vicinity), and 5) refusal to take fish (also meaning cessation of foraging in the wild). The information which will be gained from this proposed third phase is important, because if harbor seals swim away from pile driving sites, their received sound level will decrease, and thus the likelihood of them experiencing TTS will also decrease. On the other hand, TTS will probably only occur in a few harbor seals which are close to the pile driving site. However, if a larger number of seals are deterred from a large area around pile driving sites (hundreds of square km), they may be driven from ecologically important areas, such as those used for feeding. Such a behavioral response by many animals may have a greater effect on the population than TTS in a few individual animals.

1.9. Abbreviations and explanations

Duty cycle: The percentage of time that sound is produced.
kHz: Kilo Hertz (1000 oscillations per second)
PTS: Permanent hearing Threshold Shift (permanent reduction of hearing sensitivity; deafness)
SEL: Sound Exposure Level (integration of all acoustic energy in a certain event, related to 1 second).
SPL: Sound Pressure Level
TTS: Temporary hearing Threshold Shift (temporary reduction of hearing sensitivity)

1.10. References

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