



Behavioural response of grey seals to pile-driving

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Summary

Marine mammals rely on underwater sound for social interaction, communication, navigation, predator avoidance and foraging. Pile-driving during the construction of offshore windfarms produces high energy, broad spectrum sound that can be detected by marine mammals, causing injury or changes in behaviour that could reduce body condition and reproductive potential. The growth of offshore windfarm industry in the North Sea is cause for concern over impacts on marine mammals.

Grey seals (*Halichoerus grypus*) are abundant, highly mobile predators in the North Sea. They are potentially capable of detecting pile-driving activity at distances beyond 100 km, but their responses to anthropogenic sounds are still largely unknown.

In the Netherlands, grey seals haul-out in the Wadden Sea in the north, and the Delta region in the south, and forage in the adjacent North Sea. To examine if movement and behaviour could be influenced by pile-driving, we tracked grey seals during the construction of the Luchterduinen windfarm in 2014 and Gemini windfarm in 2015.

GSM-GPS tracking devices were attached to 20 grey seals in 2014 and 16 in 2015, at haul-out sites from where the seals were most likely to move near the respective windfarms. 20 seals provided location data during the individual pile-driving events, and were within 70 km of the windfarm construction site. This resulted in 175 exposure events that could be used for further analysis. A number of events were near pile-driving (i.e. 36 exposures were within 30 km of pile-driving.)

Reactions of the grey seals to the pile driving were diverse, and included: altered surfacing or diving behaviour, and changes in swim direction including swimming away from the source, heading into shore or travelling perpendicular to the incoming sound, or coming to a halt. Also, in some cases no apparent changes in their diving behaviour or movement was observed. Of the different behavioural changes observed a decline in descent speed occurred most frequent, which suggests a transition from foraging (diving to the bottom), to more horizontal movement. Our analysis showed that these changes in behaviour were on average larger and occurred more frequent at smaller distances from the pile driving events, and such changes were statistically significantly different at least up to 36km. For example, up to 36km, the exposed seals showed a decline in descent speed in 39 of the 58 exposures.

On 12 occasions, seals displayed a significant decline in the descent speed at larger distances, sometimes well beyond 36km. For example, during one instance, a grey seal at 48 km from pile-driving drastically reduced its descent speed and average dive depth when pile-driving started, and immediately after pile-driving ceased, continued to pre-piling behaviour. For individual exposures, however, it cannot be excluded with certainty that the change in behaviour incidentally coincides with the start of pile driving.

In addition to changes in dive behaviour, also changes in movement were recorded. There was evidence that *on average* grey seals within 33 km were more likely to swim away from the pile-driving. This was only the case for Gemini, where pile driving intensity was higher and GPS location estimates were more frequent than Luchterduinen. When the track of individual seals revealed a change in direction, this was not always away from pile-driving. This suggests that seals might not always be able to locate the location of pile-driving accurately.

Approximately half of the tracked seals (i.e. 16 of 36) were absent from the pile-driving area all together, the small sample size and constraints on interpretation of the data prevented us from proving long-term avoidance as a result of pile-driving. Seals that did not visit the area may have done so to avoid pile-driving activity or simply be drawn to other areas. In some cases, seals exposed to pile-driving at close range, even at distances shorter than 30 km, returned to the same area on subsequent trips. This suggests that some seals had an incentive to go to these areas, which was stronger than the potential deterring effect of the pile-driving.

This study defines a behavioural response in diving that can be linked to foraging, thus being an important consideration for understanding impacts of underwater sound on grey seals. The behavioural responses are very diverse and more detailed studies are needed to clarify these. The use

of accelerometers would enable a more detailed description of the seals' movement underwater including prey capture attempts, and therefore provide for a step forward in studying the influence of various anthropogenic sounds on seals. Also, the use of hierarchical state-space models would allow a more robust classification of the different behavioural states (e.g. transiting, searching, foraging, resting, etc.) and how sound exposure level influences the switching probability between these behavioural states, taking into account a multitude of behavioural response variables.

Samenvatting

Zeezoogdieren zijn sterk afhankelijk van onderwatergeluid voor sociaal contact, navigatie, ontwijken van predatoren en foerageren. Voor de aanleg van windparken op zee wordt geheid en het geluid wat hierbij vrijkomt kan vaak op tientallen kilometers worden waargenomen door zeezoogdieren. Dit kan leiden tot verandering in hun gedrag, en kan mogelijk leiden tot verminderde conditie van het dier en zelfs de hele populatie.

Ondanks dat de grijze zeehond (*Halichoerus grypus*) een veel voorkomend roofdier in de Noordzee is, zijn er nog weinig gegevens over hoe ze in het wild reageren op onderwatergeluid. In Nederland gebruiken grijze zeehonden ligplaatsen in de Waddenzee in het noorden, en de Delta in het zuiden, en foerageren met name in de aangrenzende Noordzee. Om te onderzoeken hoe hun beweging en gedrag beïnvloed kan worden door heien, werden grijze zeehonden uitgerust met zenders gedurende de aanleg van Luchterduinen windpark Luchterduinen in 2014 en Gemini windpark in 2015.

In totaal werden 20 GSM-GPS zenders aangebracht op grijze zeehonden in 2014, en 16 zenders in 2015. Van de in totaal 36 grijze zeehonden die een GSM-GPS zender hadden, kwamen er 20 zeehonden binnen een straal van 70 km van een hei-sessie, wat resulteerde in 175 blootstellingen. Een relatief klein deel van die blootstellingen (36) waren in de nabijheid (<30km) van het heien.

Waargenomen gedragsveranderingen van grijze zeehonden bij aanvang van het heien waren divers, en omvatten: een verandering van hun gedrag aan het oppervlak of gedurende het duiken, en een verandering in zwemrichting of snelheid, zoals het wegzwemmen van het hei-geluid. Ook werd regelmatig ogenschijnlijk geen gedragsverandering waargenomen tijdens het heien. De gedragsverandering die het vaakst werd waargenomen was een vermindering in de verticale duiksnelheid, wat een transitie suggereert van foerageren (recht naar beneden duiken) naar wegzwemmen (meer horizontale beweging). De statistische analyse liet zien dat een degelijke gedragsverandering duidelijker en gemiddeld vaker werd waargenomen op kleinere afstanden van het heien, en dergelijke verandering was statistisch significant tot ongeveer 36km.

In een tiental gevallen liet een gezenderde zeehond een afname in duiksnelheid zien op grotere afstand van het heien. Bijvoorbeeld, gedurende een hei-sessie liet een grijze zeehond die zich 48 km van het heien bevond een sterke reductie in duiksnelheid en gemiddelde duikdiepte zien bij aanvang van heien, en direct nadat het heien was gestopt, vertoonde de zeehond duikgedrag wat vergelijkbaar was met dat van voor het heien. Echter, voor individuele blootstellingen kan er niet met 100% zekerheid gesteld worden of gedragsveranderingen het gevolg zijn van het heien of andere vormen van verstoring

Naast veranderingen in duikgedrag, waren ook veranderingen in beweging zichtbaar. Er waren sterke aanwijzingen dat gemiddeld grijze zeehonden tot op 33km een grotere kans hadden van het heien weg te zwemmen. Dit was alleen zichtbaar voor Gemini windpark. Dit zou kunnen komen doordat de hei-intensiteit voor dat park hoger was, of omdat de gemiddelde tijdsinterval tussen de GPS locaties kleiner was vergeleken met Luchterduinen, en veranderingen in beweging daardoor makkelijker te registreren waren. Hoewel er veranderingen in zwemrichting werden waargenomen, waren deze niet altijd weg van het heien. Dit zou erop kunnen wijzen dat zeehonden misschien niet altijd in staat zijn precies te bepalen waar het hei-geluid vandaan komt.

Hoewel ongeveer de helft van de gezenderde zeehonden helemaal niet in de buurt van het heien kwamen, was de steekproef te klein en proefopzet ongeschikt om lange termijn vermijding van het heien te bepalen. Namelijk, een reden dat een zeehond niet in het buurt komt van de hei-locatie kan te maken hebben met het ontwijken van heien, of simpelweg omdat het een voorkeur heeft voor andere gebieden. Sommige zeehonden kwamen herhaaldelijk terug om te foerageren in het gebied nabij (<30km) de hei-locatie. Dit suggereert dat voor deze dieren de wens om in het gebied te foerageren groter was dan de eventuele nadelige effecten van het heien.

Deze studie heeft gekeken naar een verandering in duikgedrag wat gerelateerd is aan foerageren, en is daarmee een belangrijke graadmeter voor het begrijpen van het effect van onderwatergeluid op grijze zeehonden. Echter, de waargenomen gedragsveranderingen zijn heel divers, en om die reden zijn meer gedetailleerde studies nodig om deze variatie te begrijpen. Een oplossing is het gebruik van

versnellingsmeters die een meer gedetailleerde beschrijving van het gedrag van zeehonden onderwater kunnen opleveren. Met dergelijke versnellingsmeters kan bijvoorbeeld gekeken worden wanneer en hoe zeehonden hun prooi vangen, maar ook preciezer hoe zeehonden wegzwemmen van verstoring. Een andere oplossing is het gebruik van zogenaamde Hidden Markov Models die in staat zijn om beweging en duikgedrag te classificeren in verschillende typen gedragingen (zoals zwemmen, zoeken naar voedsel, rusten, etc.). Tevens kunnen dergelijke modellen onderzoeken hoe blootstelling aan geluid leidt tot een verandering in gedrag, zoals het stoppen met foerageren en wegzwemmen. Dit brengt het onderzoek weer een stap dichterbij het begrijpen wat de consequentie van menselijke verstoring is op zeezoogdieren.

1 Introduction

Grey seals (*Halichoerus grypus*) are among the most abundant marine mammals in the North Sea, along with harbour seals (*Phoca vitulina*) and harbour porpoises (*Phocoena phocoena*) (Reijnders & Lankester 1990). While several studies have studied the response of porpoises to anthropogenic underwater sound in the wild (Carstensen *et al.* 2006, Lucke *et al.* 2009, Tougaard *et al.* 2009, Hastie *et al.* 2015), relatively little is known on its effect on wild seals, especially grey seals. One prevalent source for underwater sound in the North Sea is pile-driving for offshore windfarm installation (Breton & Moe 2009). Prior to 2017, almost 2000 offshore wind turbines were installed in the North Sea and it is planned that by 2020, there will be >10,000 (information source <http://www.4coffshore.com/offshorewind/>, last accessed Jan 2018). Pile-driving of each turbine tower (monopile) into the sediment produces a series of high-impact, broad-band, pulses of noise at approximately one second intervals over 1-3 hours (Bailey *et al.* 2010). Madsen *et al.* (2006) suggest that pile-driving can be detected by marine mammals over hundreds of kilometres. However, immediate and longer-term responses to such sounds or impacts on the animals are still largely unknown. Some responses of harbour porpoise to pile-driving have been documented (Carstensen *et al.* 2006, Lucke *et al.* 2007, Lucke *et al.* 2009, Tougaard *et al.* 2009, Kastelein *et al.* 2012, Teilmann & Carstensen 2012, Dähne *et al.* 2013), but there is little data on harbour seals (Hastie *et al.* 2015, Russell *et al.* 2016) and virtually none available for grey seals. This study addresses the lack of such data by investigating the movement responses of individual grey seals tracked during the construction of two offshore windfarms in Dutch waters.

Since the 1970s, there has been increasing interest in how anthropogenic underwater sounds, including shipping noise, active sonar, seismic surveys, construction activity, explosions, and acoustic deterrents may alter the behaviour and survival of marine mammals (Payne & Webb 1971, Myrberg 1978, Southall *et al.* 2007, Richardson *et al.* 2013). Marine mammals rely on sound production and detection for communication, navigation, predator avoidance and foraging (Tyack 1997, Noad *et al.* 2000, Curé *et al.* 2013). Human sounds may mask their abilities to detect important sounds (Erbe *et al.* 2016), produce aversion responses that could restrict behavioural and movement options affecting reproduction and survival, or cause direct injury and even death (Ketten 1995, D'Amico *et al.* 2009, Kight & Swaddle 2011).

Restrictions on industrial underwater sound in the North Sea that aim to protect marine mammals have primarily focussed on minimising impacts on harbour porpoises (Teilmann & Carstensen 2012, Kastelein *et al.* 2013c, Thompson *et al.* 2013). Harbour porpoises are the most abundant species widely distributed, migratory and reliant on their echolocation to survive (Hammond *et al.* 2002, Carlström 2005). They are also highly responsive to anthropogenic sounds (Lucke *et al.* 2007, Lucke *et al.* 2009, Tougaard *et al.* 2009, Kastelein *et al.* 2012). Harbour porpoises detect and readily flee from the near vicinity of pile-driving activities (Carstensen *et al.* 2006, Tougaard *et al.* 2009, Dähne *et al.* 2013). Hearing is also important for seals as demonstrated by their acute underwater hearing (Bodson *et al.* 2006) and they rely on sound recognition to detect danger (Deecke *et al.* 2002), and potentially food, or to recognise conspecifics (Graham *et al.* 2009). They may therefore function sub-optimally if their hearing ability is masked or damaged. This could be an issue specifically in the case of pile-driving as seals hear in the range 0.1 to ~69 kHz, with maximum sensitivity at around 1 kHz (Kastelein *et al.* 2009, Stansbury *et al.* 2014, Cunningham & Reichmuth 2016), coinciding with sounds produced by pile-driving. These sounds peak at 0.5 kHz close to the source and 0.5 to 1 kHz at greater distance (de Jong *et al.* 2013). By comparison, harbour porpoises hear best in the range 16 to 140 kHz, with maximum sensitivity between 100 and 140 kHz (Kastelein *et al.* 2002), well above this peak. One field study provided pulsed noise at a frequency of ~1 kHz that displaced harbour seals while harbour porpoise seemed unaffected (Götz & Janik 2015).

Although seals are likely to detect pile-driving over great distances, some individuals may endure potential harm from the sound, to pursue other activities, such as accessing haul-out sites and feeding. Recently, harbour seals in the UK were reported to remain in the vicinity of pile-driving despite receiving multiple exposures to sound levels that were sufficient to cause shifts in their hearing thresholds (Hastie *et al.* 2015), still the overall harbour seal density was lower during pile-driving up to 40 km away (Russell *et al.* 2016).

Responses by seals to potential dangers are likely to vary greatly, depending on individual histories, size, sex, condition, and hearing ability (Lucke *et al.* 2016). This was observed in grey seals and harbour seals (Götz & Janik 2011, Kastelein *et al.* 2013b). The latter laboratory study provided unique information on behavioural variability by seals in response to potentially damaging sound levels. Due to a transducer malfunction, two captive harbour seals were exposed to a higher than anticipated sound source for 60 min: the mean received sound pressure level (SPL) was 163 dB re 1 μ Pa. One of the seals avoided exposure to this sound by hauling out; the other seal chose to remain in the water where it received a sound exposure level (SEL) of 199 dB re 1 μ Pa²s, causing severe TTS (44 dB, 12-16 min. post exposure). As survival might depend on it, wild seals may be more likely than a trained, captive seal, to tolerate potentially damaging levels of anthropogenic sound. A further motivation for the seals to tolerate potentially harmful sounds is that the sounds may mask their approach from prey, thus enhancing feeding opportunities (Chan *et al.* 2010).

As part of the environmental monitoring for windfarm construction in Dutch waters, we investigated grey seal movement and behaviour during periods of pile-driving for two windfarms: Luchterduinen and Gemini. Luchterduinen was constructed in water depths of 18-24 m, 23 km off the central Dutch coast in 2014, and Gemini was constructed in water depths of 32-34 m, 50 km north of the Dutch Wadden Sea, in 2015. The hypothesis is that seals will swim away from pile-driving and change their diving behaviour. When foraging, grey seals will spend a large proportion of time near the bottom (80-90%), and we expect this to be reduced when exposed to pile-driving. The probability of change is more likely to occur at smaller distances and higher SEL. The aim here is to primarily study changes in behaviour during pile driving, not to estimate PTS or TTS. Results provide insights specifically into impacts of pile-driving on grey seals, but in general, also shed light on potential impacts of anthropogenic underwater sound on pinnipeds.

2 Materials & Methods

2.1 Pile driving

Pile-driving for Luchterduinen was conducted in the period 31 July to 16 October 2014. In total 44 monopiles (each 4.5 m diameter) were pile-driven into the seabed: 43 for wind turbines and one for an offshore high voltage station (OHVS) (Figure 1, upper). Pile-driving for Gemini was conducted between 1 July and October 17, 2015, for 150 turbine towers (4.5 m diameter) and eight monopiles to support two OHVSs (Figure 1, lower).

Activities leading to each pile-driving event also produced underwater sound that may have been detected by seals. Before each monopile was pile-driven, the vessel was positioned using active sonar, jacked-up, and an acoustic harbour porpoise deterrent was switched on (Faunaguard, SEAMARCO Ltd, Netherlands). The porpoise deterrent produced sounds at ultra-sonic frequencies (60-150 kHz) anticipated to be at, or just above the seals top hearing range. The monopile was lowered to the sea floor and a pile-driving hammer was positioned over it.

Once pile-driving commenced, hammering was not continuous. It commenced with a 'soft-start', i.e. no or light (~200 kJ) power, to ensure the monopile seated correctly and penetrated the substrate in a controlled manner. Initial hammering consisted of one or several blows followed by pauses of up to several minutes for observation/adjustment. As the monopile penetrated further, the frequency, duration and power of hammering increased. In later stages, hammering was at a rate of 40-50 blows per minute for 30 minutes or longer at energy levels >700 kJ. Compared to *Luchterduinen* different pile-driving hammers were used for Gemini, with higher pile-driving energy (see Figure 1).

After a monopile reached its required depth, the acoustic deterrent was switched off. The vessel installed fixtures to the monopile, then jacked-down and moved to the next location. At Luchterduinen, one vessel performed all the pile-driving leaving periods of 2-3 days without any pile-driving while the vessel restocked. At Gemini, two vessels operated so time-gaps between pile-driving events were shorter and occasionally two monopiles were being installed simultaneously (see Figure 1). Often pile-driving ceased during winds above 15 m/s.

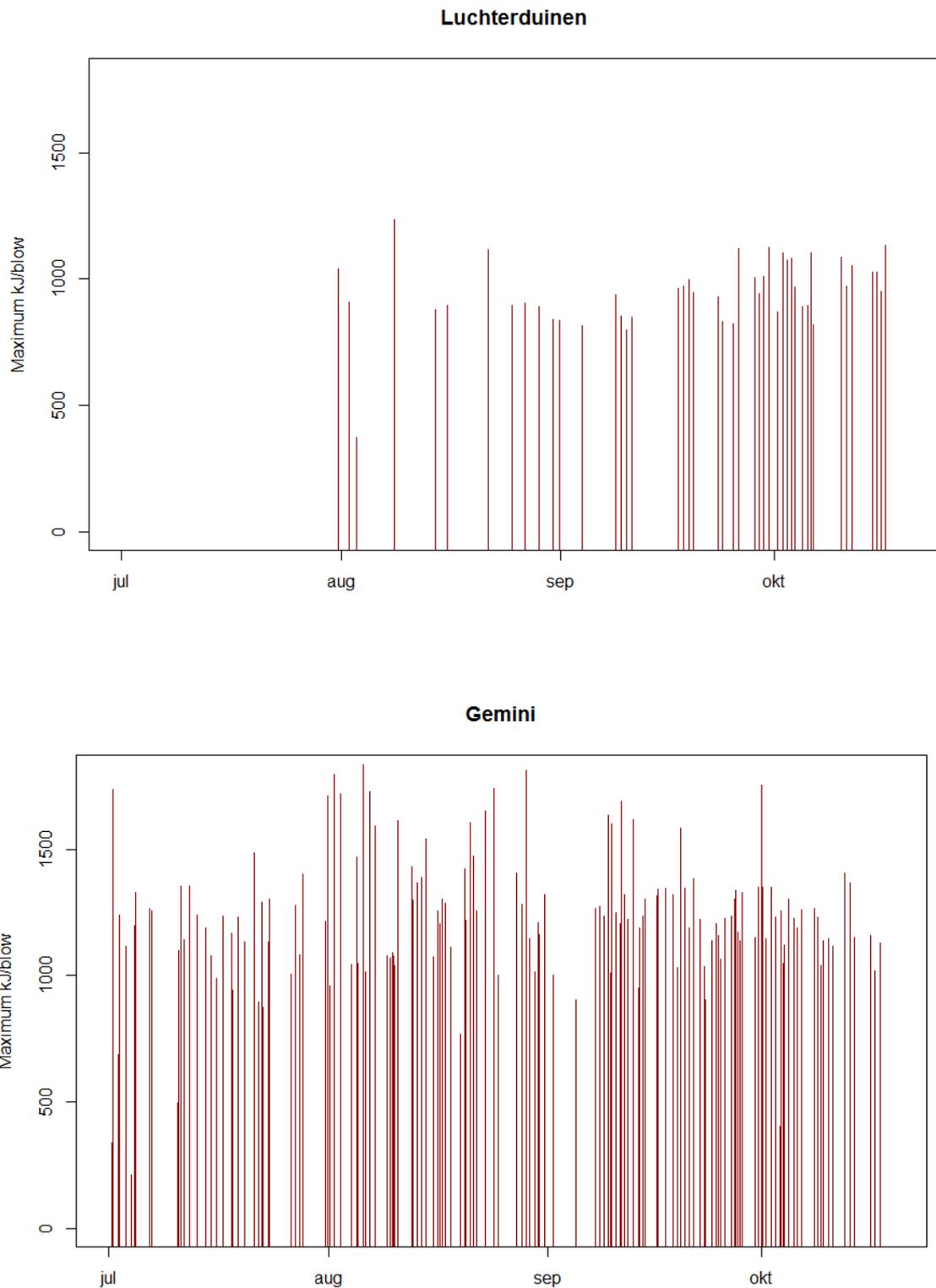


Figure 1. Pile-driving activities (vertical red lines) for Luchterduinen in 2014 (upper figure) and Gemini in 2015 (lower figure).

2.1.1 Underwater sound levels produced by pile-driving

The Aquarius 1.0 sound propagation model (Binnerts & de Jong 2016) was used to estimate the single strike Sound Exposure Level (SELs in dB re 1 $\mu\text{Pa}^2\text{s}$.) for the Luchterduinen windfarm only. Details

are described in (Binnerts & de Jong 2016). The source level (SL_E) was estimated based on the estimated propagation loss and sound measurements for the Princes Amalia Wind Park (PAWP, de Jong & Ainslie 2012). The model was used to make SEL_{SS} predictions (unweighted and Mpw weighted, Southall *et al.* 2007) for five representative monopile locations in the Luchterduinen windfarm (E01, E06, E21, E36 and E43), assuming a piling energy of 1000 kJ, for eight wind speeds (0, 2, 4, 6, 8, 10, 12 and 14 m/s) and two receiver depths: 1 m above the bottom and 1 m below the surface. At distances beyond 15 km, the SEL_{SS} were considered less precise due to the less well-known effects of wind (determining wave action) on sound propagation.

For the analysis in the present study, each pile-driving location was allocated to the nearest of the five pile-driving location for which SEL_{SS} estimates were available. For each grey seal exposed to pile-driving, the concurrent wind speed data (measured at the PAWP wind turbine WTG59 or, if unavailable, WTG60) were used to select the relevant map of SEL_{SS} predictions. For each exposure with known GPS location, the local estimate of SEL_{SS} was extracted. Finally, assuming a fixed percentage of blow energy will be transferred to sound energy, the SEL_{SS} could be rescaled for different pile-driving energy levels (B_E in kJ) by adding $10\log_{10}(B_E \text{ kJ}/1000\text{kJ})$ dB to the provided SEL_{SS} estimate for 1000kJ.

For Gemini, sound measurements were made (Remmers & Bellmann 2016), and the data were used to validate the Aquarius 1.0 model. Maps of SEL estimated for the different monopiles and wind speeds are currently unavailable for the Gemini study, and hence the changes in grey seal behaviour in response to pile-driving for Gemini, were only related to distance to the pile-driving site.

2.2 Seal movement

2.2.1 Field deployments

Seals were tracked using GPS-GSM transmitters (weight 330 g in air, volume 150 cm³) from the Sea Mammal Research Unit (SMRU, Scottish Oceans Institute, Scotland). These provide Fastloc® GPS location-determinations, dive depth, sea temperature and haul-out time measurements. Recovery of data was through the GSM mobile-phone network. Up to 3-months of data were stored in the memory of the transmitters and could be relayed via the GSM mobile-phone system when the seal hauled out within range of a network. The 3-month data storage facility was required in case seals remained for extended periods at sea or at haul-outs that were not covered by the GSM network.

The Fastloc® GPS in the transmitter attempted to determine a location after a pre-set interval and when the antenna was next exposed. To maintain battery life throughout an anticipated sample period of 10 months, the sample interval was set at 15-minutes. Seal location and dive data were transmitted from the tracking devices via the GSM-network to land-based computers (at SMRU, Scotland) and could be downloaded over the internet as Access files.

To get overlap with the Luchterduinen site, tracker deployment sites were selected to the north and south of the Dutch coastal zone. In the north, the deployment site was in the inlet of the Eierlandse Gat, between the islands of Texel and Vlieland (53.20°N, 4.91°E) (Figure 3). In the Delta region, grey seals were caught at the Aardappelbult sandbar south of Rotterdam (51.79°N, 3.78°E). For the Gemini site, areas chosen for seal captures were near the island of Ameland, Pinkegat to the east (54.44°N, 5.94°E) and the Blauwe Balg to the west (53.43°N, 5.60°E).

Field trips to deploy the transmitters were conducted in April 2014 for Luchterduinen, and April and September 2015 for Gemini. Seals were captured at low tide adjacent to sandbars using a purpose-built seine-net of approximately 100 m length and 8-m drop. Healthy individuals that had completed their moult were selected to carry transmitters. We attempted to get an even spread of males to females and sub adults to adults (adult nose-to-tail lengths were >140 cm females and >160 cm males). Selected seals were strapped into purpose-built cradles and had the transmitter glued (Permacol 2240 epoxy resin, Permacol BV, Netherlands) to their pelage at the mid-dorsal point behind the neck. While the glue set (approximately 10-15 minutes), the seals were sexed and measured (standard length and weight). Once the glue had set, each seal was released and, upon release all proceeded directly to the water. All seals were released within 90 min. of capture.

All required permits to enter protected areas and handle animals were obtained. These included permits under the Dutch Nature Protection Act (Natuurbeschermings Wet) from the provinces of Zeeland and North-Holland, a permit under the Flora and Fauna Act (Flora en Fauna Wet) from the Dutch government and protocols approved by an animal ethics committee (Dier Ethische Commissie, DEC) of the Royal Netherlands Academy of Science (Koninklijke Nederlandse Academie voor Wetenschappen, KNAW).

2.2.2 Processing tracking data

Location determinations and dive data were accessed via the internet from a data storage facility at SMRU, UK. For the dive data, several descriptive variables were calculated. The dive depth was recorded at 1%, 2.5%, 5%, 10%,, 90%, 95%, 97.5% and 99% of the duration of each dive. The dive starts when the depth sensor records a depth below 1.5m. The initial descent speed of the dive was estimated as the change in dive depth between the 1% and 2.5% quantile time-point of dive, divided by the corresponding dive time between those two points. The estimated speed between the start of the dive (1.5m depth) and the 1% quantile point could not be used, because the precise start time of the dive was recorded at 4 second resolution, which was often too imprecise, particularly for the shorter dives. The variable 'mean to maximum dive depth ratio' was calculated as the average dive depth of each dive, divided by the local water depth. Depth estimates were extracted from the harmonized EMODnet Digital Terrain Model (DTM, see <http://www.emodnet-hydrography.eu/>), which is based on regional DTMs, and gaps with no data coverage were completed by integrating the GEBCO Digital Bathymetry. If the dive depth exceeded the local depth, the maximum dive depth was used as local water depth instead. The 'speed to maximum dive depth' was calculated as depth of the first dive point beyond 90% of the maximum dive depth, divided by the time required to reach that depth.

GPS location estimates could in theory be obtained every 10 minutes, but in practice there were occasional long time gaps of several hours between the GPS locations. To estimate the seal position at each dive and other (regular) points in time, a continuous time correlated random walk model was fitted to entire track of each individual seal (CTCRW, function `crawlWrap` in R-package `momentuHMM` (Johnson 2017, McClintock & Michelot 2017)). This model was subsequently used to predict for each dive in between the GPS location fixes the seals' location (x and y-coordinates in UTM31N projection) and uncertainty of the location. Both the GPS and dive data were allocated to a specific period, in respect to the pile driving: 4 h to 5 min. prior to pile-driving (period 1), 5 to 0 min. prior to pile driving (period 2), during pile driving (period 3) and 0 to 4 h after pile driving (period 4). The period 2 (i.e. 5 to 0 min. prior to pile driving) was included because initial inspection of the dive data suggested that seals sometimes responded a few minutes to seconds prior to pile-driving, and it was assumed that this was due to some other pile-driving related sound which was not included in the pile driving data. Depending on the research question and corresponding analysis, specific data from the respective periods were selected.

All analyses were carried out in UTM 31N projection (EPSG code = 25831) except for the SELss maps, which were in National Rijkdriehoek projection. The estimated location of the seal during an exposure was based on the CTCRW location estimate of the last dive prior to pile-driving, or when not available, the location of the first dive after the start of pile-driving.

2.3 Seal responses to pile-driving – statistical analysis

2.3.1 Change in diving behaviour

When foraging, grey seals often dive to the sea floor, where they spend 80-90% of the total dive time. This type of foraging behaviour will lead to U-shaped dive, with a relatively long period of near-constant depth. When the seal was exposed to a loud sound, we expected this pattern to be disrupted. For example, seals may stop diving to feed on the bottom and attempt to flee from the sound source, leading to slower vertical descent and ascent rates, and also a lower percentage of time at maximum depth (i.e. more V-shaped dive). Even if they continue to forage, the disturbance and stress response may still lead to an earlier termination of the dive (i.e. shorter dives).

Since it was unknown *a-priori* which variable was likely to be most important, we explored three dive-related variables, and tested whether any of these variables changed when the seal was exposed to pile-driving:

1. Descent speed ($v_{descent}$ in m/s): The vertical speed measured between the 1% quantile of the dive and the point where the seal reached 80% of the maximum dive depth.
2. *Fraction of time near the bottom* (p_{bottom}): The fraction of each dive the seal spent near the bottom (here defined as >80% of local depth).
3. Average dive depth ($d_{\mu/max}$): The average dive depth (taking the surface duration with depth =0m, into account) expressed as fraction of the local depth.

For each grey seal present within 70 km of a pile-driving event (defined as an **exposure**), we selected the dive data from 4 h before until 2 h after the pile-driving event. For each dive, the response variables were calculated. For the proportions p_{bottom} and $d_{\mu/max}$, we assumed a beta distribution with $\mu = p/(p + q)$ and $\phi = p + q$, where p and q are the estimated parameters of the beta distribution, and μ and ϕ the derived parameters (Ferrari & Cribari-Neto 2004), and logistic link function (hence μ is the inverse of that link function; $\mu = \frac{e^\eta}{1+e^\eta}$)

$$p_{bottom}, d_{\mu/max} \sim Beta(\mu, \phi)$$

$$\mu = \frac{e^\eta}{1 + e^\eta}$$

For the descent speed $v_{descent}$, we assumed a gamma distribution

$$v_{descent} \sim Gamma(\mu, k)$$

$$\mu = e^\eta$$

In both cases, the linear predictor η was subsequently modelled as a function of period specific parameters ($\beta_{t_0}, \beta_{t_c}, \beta_{t_1}$) and a temporally correlated smooth

$$\eta = \beta_{t_0} + \beta_{t_c} + \beta_{t_1} + \nu$$

$$\nu = f(t) + \varepsilon$$

$$\varepsilon = Normal(0, \sigma) \quad (1.1)$$

The coefficient β_{t_0} is the average fraction of time at depth prior to pile-driving (t_0), and β_{t_c} and β_{t_1} quantify the relative *changes* in the fraction of time at depth during the pile-driving period (t_c) and 2 hours after the pile driving (t_1), respectively. ν is a temporally correlated auto-regressive term, which captures any correlation in the residuals. When pile-driving significantly reduces the fraction of time at depth during the pile-driving, the parameter β_{t_c} should be significantly smaller than zero.

This analysis produced for each seal and each pile-driving event, an estimate of the size of the effect (β_{t_c}), and corresponding uncertainty. Next, we modelled how these parameters varied as a function of the following covariates: distance to the pile-driving ($dist$), proportion of the trip (P_{trip}), estimated SEL (SEL), wind speed ($wind$; Wind speed correlates with wave action and, therefore, masking of sound propagation), individual-specific cumulative number of exposures (C_i) and individual (I):

$$\beta \sim Normal(\mu_\beta, \sigma_\beta)$$

$$\mu_\beta = s(dist)_i + s(SEL)_i + s(wind)_i + s(C)_i + s(P_{trip})_i + \pi_i$$

$$\pi_i = I_i + \epsilon$$

(1.2)

Where $s()$ are smooth functions of the variables. The size of the effect (i.e. β_{t_c}) was allowed to vary by individual using an individual-specific random effect π_i . Least-squared cross-validation was used to select the best model, where Eq. 1.2 specifies the full-model. SEL estimates were only available for Luchterduinen pile-driving, and hence the models using SEL as covariate were fitted to a subset of the

data. Although it would be theoretically possible to fit the models 1.1 and 1.2 in a single framework, given the large number of data points this was practically unfeasible.

2.3.2 Change in speed

In addition to a change in diving behaviour, the seals' response to pile-driving may include a change in horizontal speed (i.e. swim away from the perceived sound source). To estimate the movement of the seal relative to the pile-driving, its movement prior, during and after pile-driving was characterised by two vectors; one indicating the speed towards the pile-driving (v_1) and another perpendicular to pile-driving (v_2), which were estimated using standard matrix rotation functions:

$$v_1 = \sin(\alpha)v_x + \cos(\alpha)v_y$$

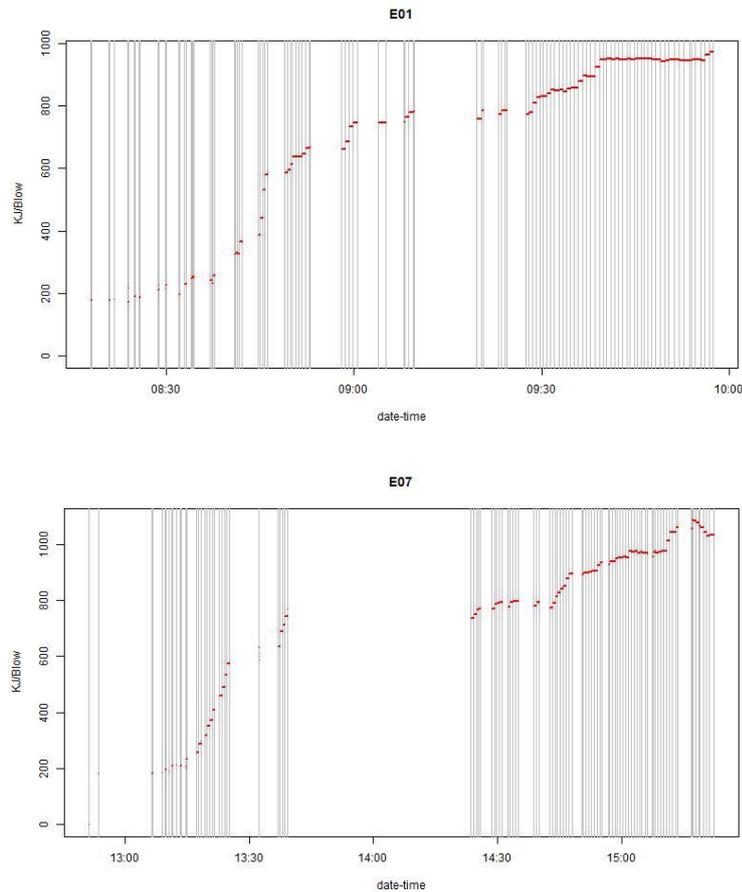
$$v_2 = \cos(\alpha)v_x - \sin(\alpha)v_y$$

v_x and v_y are the speed in x (longitudinal) and y (latitudinal) direction. The variables v_1 and v_2 were used to study if seals changed their behaviour in response to piling. The analysis was identical to the dive analysis, except that the model was fitted to the GPS location data (i.e. generally less observations) and the response variable was assumed to have a Gaussian distribution.

3 Results

3.1 Pile driving

Each monopile had a unique pile-driving record (Figure 2). Most monopiles were driven in during single periods, but some required two periods due to a malfunction in the first period. Also, several pile-driving events had intermediate breaks that exceeded 1 hour – sufficient time for an animal to consider the event had finished – so were considered to be two events. For Luchterduinen, this resulted in 45 pile-driving events, and for Gemini, there were 166 events. On average, each pile-driving event lasted 2 h with the maximum duration being 5.3 h. For Luchterduinen, the penetration depth of 40 (91%) monopiles ranged between 23 and 30 m into the sea floor (27 ± 4 m): four went deeper, the deepest to 47 m. For Gemini, the penetration depth varied between 19 and 34 m. Monopiles required on average 4100 blows to reach their required depth. The maximum hammer energy used in each monopile averaged 951 kJ (SD = 137 kJ) for Luchterduinen, and 1218 kJ (SD = 257 kJ) for Gemini (see also Figure 1).



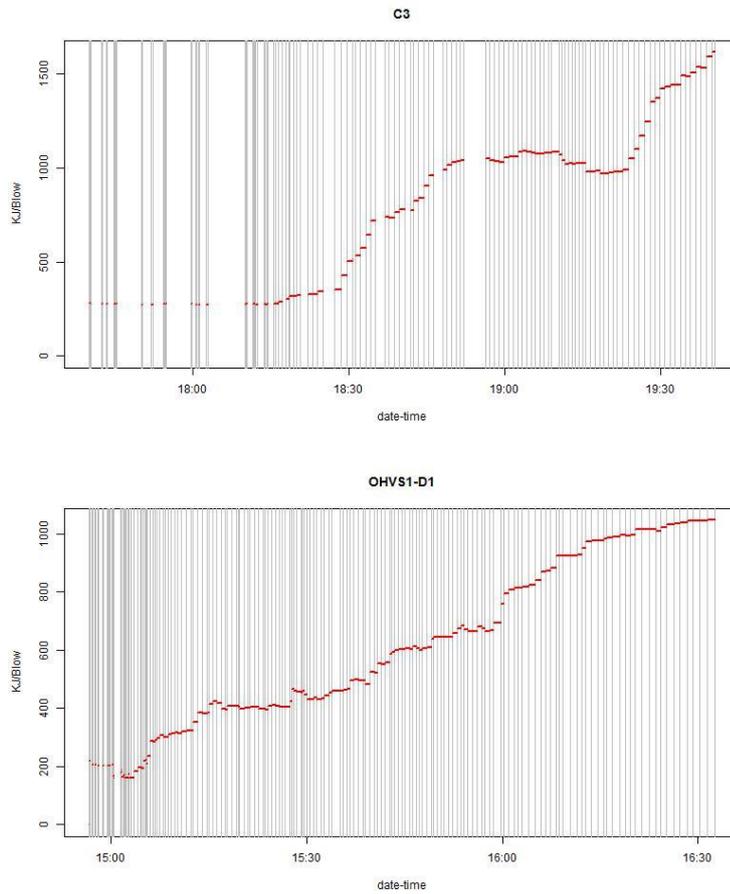


Figure 2. Examples of pile-driving logs from Luchterduinen (top two figures) and Gemini (bottom two figures). The red horizontal lines indicate energy level (kJ/blow). Vertical grey lines indicate start and stop times of a series of hammer blows with similar energy levels.

3.2 Seal movement

For overlap with pile-driving at Luchterduinen, 20 trackers were deployed in April 2014 – 10 north of Texel in the Wadden Sea and 10 at Aardappelbult in the Delta region (Table 1). Twelve of the grey seals were females and eight males; six were sub-adults and 14 adults. Nose to tail lengths averaged 155 ± 19 cm (range 117 to 187) and weights averaged 83 ± 30 kg (range 30 to 134). Periods of tracking averaged 213 ± 55 d (range 73 to 299). Nineteen of the trackers operated during part of the pile-driving period with 13 recording over the entire period.

For overlap with pile-driving at Gemini, seven trackers were deployed in April 2015, and nine in September 2015 (Table 1). Thirteen were deployed at Blauwe Balg, and three September deployments were at Pinkegat. Five of the seals were females and 11 were males, 12 were sub-adult and four were adult. Mean tracking durations were 171 ± 42 days ($n = 7$, range 98 to 208) for seals caught in April and 88 ± 50 days ($n = 9$, range 6 to 132) for seals caught in September. All seals were tracked during the pile-driving period, including four, that were tracked during the entire construction period.

Table 1. Grey seals tracked to investigate responses to pile-driving activities in the Netherlands in 2014 (at Luchterduinen) and 2015 (at Gemini).

Sample	Seal	Sex	Age group	Length	Weight	Date out	Last day	Duration
hg43L								
Delta	Z006	F	adult	170	121	4-Apr-14	10-Dec-14	250
Delta	Z007	F	sub adult	117	37	2-Apr-14	15-Oct-14	196

Sample	Seal	Sex	Age group	Length	Weight	Date out	Last day	Duration
Delta	Z018	F	adult	165	100	4-Apr-14	1-Oct-14	180
Delta	Z024	F	adult	149	82	3-Apr-14	4-Jan-15	276
Delta	Z037	M	adult	176	99	3-Apr-14	2-Sep-14	152
Delta	Z045	M	adult	187	126	4-Apr-14	22-Sep-14	171
Delta	Z046	F	adult	168	101	4-Apr-14	28-Jan-15	299
Delta	Z062	F	adult	165	86	4-Apr-14	4-Dec-14	244
Delta	Z063	F	sub adult	117	30	2-Apr-14	1-Aug-14	121
Delta	Z066	M	adult	152	70	2-Apr-14	4-Nov-14	216
Texel	T003	F	adult	155	74	16-Apr-14	10-Dec-14	238
Texel	T040	F	adult	152	97	15-Apr-14	5-Dec-14	234
Texel	T042	F	sub adult	136	50	15-Apr-14	9-Dec-14	238
Texel	T076	F	adult	179	101	16-Apr-14	11-Dec-14	239
Texel	T078	M	adult	158	80	16-Apr-14	30-Dec-14	258
Texel	T079	M	adult	172	134	15-Apr-14	10-Nov-14	209
Texel	T080	M	sub adult	142	60	16-Apr-14	4-Dec-14	232
Texel	T081	M	sub adult	136	47	15-Apr-14	27-Jun-14	73
Texel	T094	M	sub adult	140	59	16-Apr-14	28-Sep-14	165
Texel	T875	F	adult	159	108	15-Apr-14	5-Jan-15	265
hg46G								
Blauwe Balg	A077	M	sub adult	139	38	14-Apr-15	8-Nov-15	208
Blauwe Balg	A112	M	sub adult	146	63	15-Apr-15	12-Sep-15	150
Blauwe Balg	A113	M	sub adult	136	60	15-Apr-15	3-Sep-15	141
Blauwe Balg	A114	F	sub adult	112	32	14-Apr-15	8-Nov-15	208
Blauwe Balg	A116	M	sub adult	145	73	15-Apr-15	21-okt-15	189
Blauwe Balg	A119	F	adult	168	90	14-Apr-15	1-Nov-15	201
Blauwe Balg	A317	M	sub adult	111	35	14-Apr-15	21-Jul-15	98
hg51G								
Blauwe Balg	B110	F	adult	166	141	23-Sep-15	05-okt-15	12
Blauwe Balg	B112	M	sub adult	134	45	23-Sep-15	31-Jan-16	130

Sample	Seal	Sex	Age group	Length	Weight	Date out	Last day	Duration
Blauwe Balg	B113	M	adult	184	169	23-Sep-15	10-Feb-16	140
Blauwe Balg	B121	F	sub adult	123	41	23-Sep-15	2-Feb-16	132
Blauwe Balg	B130	M	sub adult	129	47	23-Sep-15	29-Sep-15	6
Pinkegat	B132	M	subadult	146	75	22-Sep-15	12-Jan-16	112
Pinkegat	B133	M	subadult	132	52	22-Sep-15	3-Dec-15	72
Pinkegat	B136	F	adult	157	85	22-Sep-15	31-Dec-15	100
Blauwe Balg	B144	M	subadult	135	47	23-Sep-15	24-Dec-15	92

The seals expressed considerable individual variation in movement with some remaining within Dutch waters and others crossing the North Sea (Figure 3 - Luchterduinen, Figure 4 - Gemini). The maximum distances achieved from catch locations ranged up to 861 km.

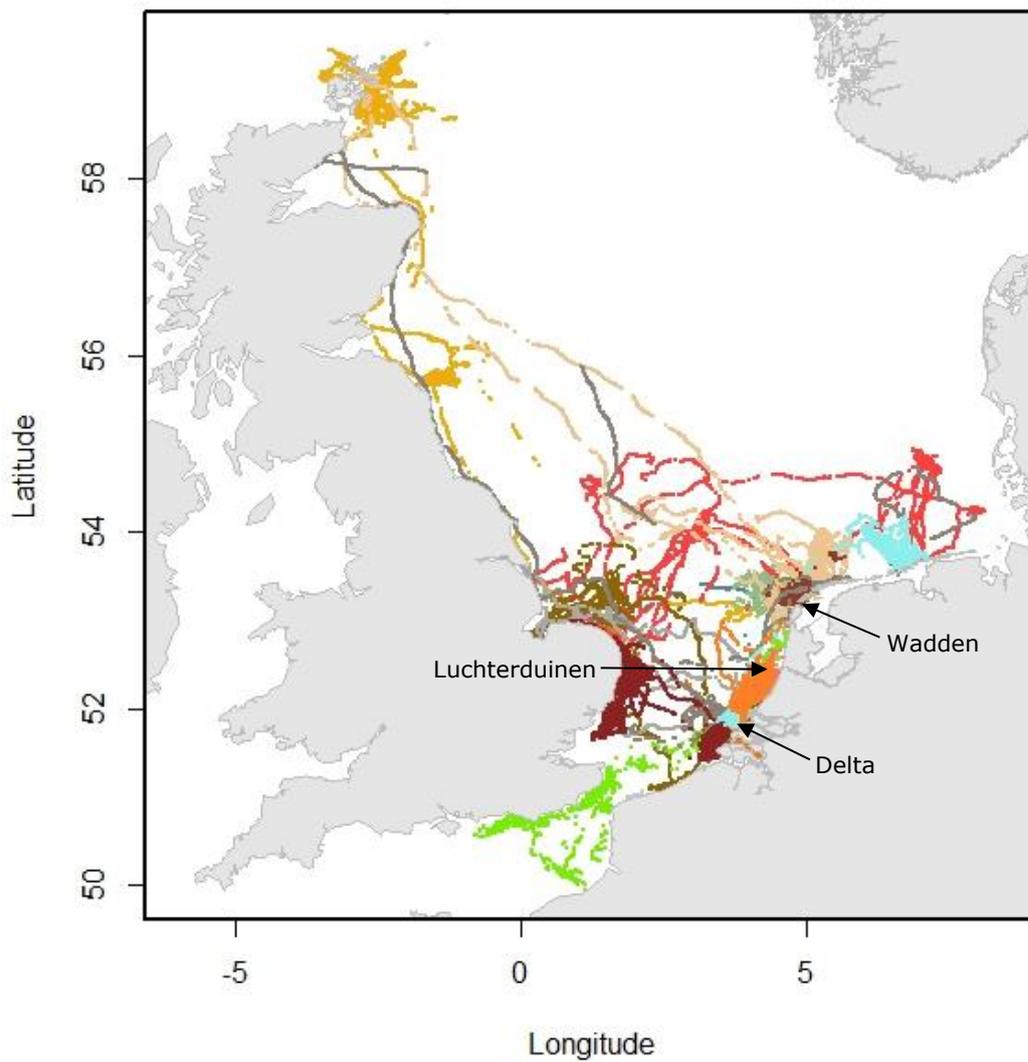


Figure 3. Luchterduinen 2014. Locations recorded for 20 grey seals tracked during 2014. Different colours are different seals. Deployment sites were in Wadden Sea, and Delta region, and the location of Luchterduinen windfarm is indicated.

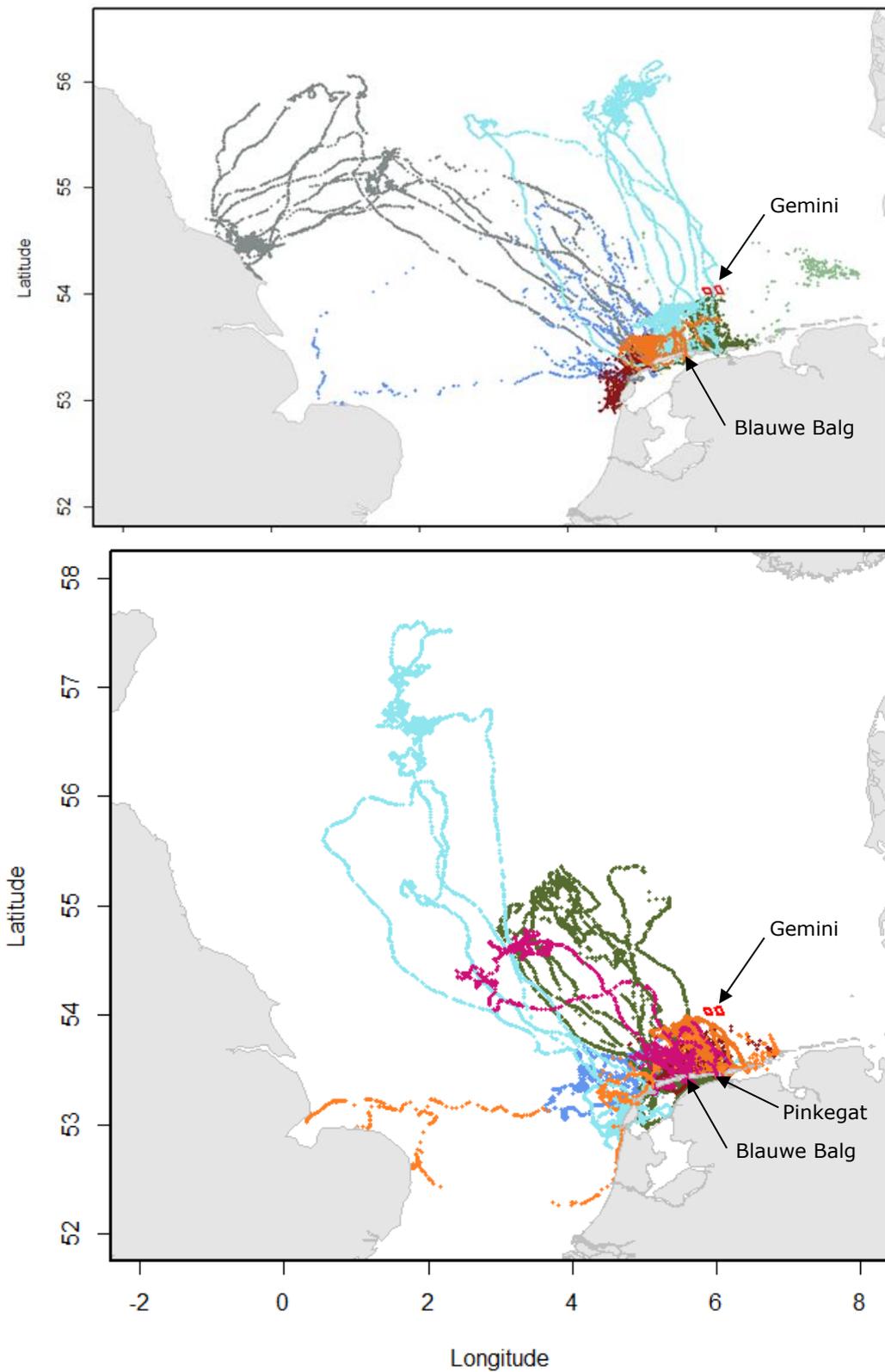


Figure 4. Gemini 2014. Locations recorded for grey seals tracked following tracker deployments in April (upper: seven seals) and September (lower: nine seals) 2015. Seal capture sites were Pinkegat and Blauwe Balg. The Gemini windfarm is indicated (red boxes).

3.3 Seal responses to pile-driving

3.3.1 Overlap between seals and pile-driving

Of the 36 grey seals tracked, movements of 20 grey seals were recorded during the pile-driving period of either Luchterduinen or Gemini, that were at least once within 70km of pile-driving and outside the shallow (<10m) waters of the Wadden and Delta coast (here defined as the shallow areas >50km from Pile-driving). The motivation for the exclusion was that received sound levels would be very unreliable. From these we identified 261 "exposures" (i.e. the seals' presence within 70 km of a pile-driving event). However, for several exposures there was no GPS location near the start of the pile-driving (i.e. within 2 hour), or dive data. Eventually, 175 exposures could be used for the analysis. At the start of pile-driving events, the smallest straight-line distances between tracked seals in the water and the pile location was 11km, and 36 were within 30km, the remaining 139 exposures were beyond 30km (Figure 6). Generally, the seals were inshore of the pile-driving events (i.e. in shallower waters), which was also the case during the pre-construct phase (Brasseur & Kirkwood 2015).

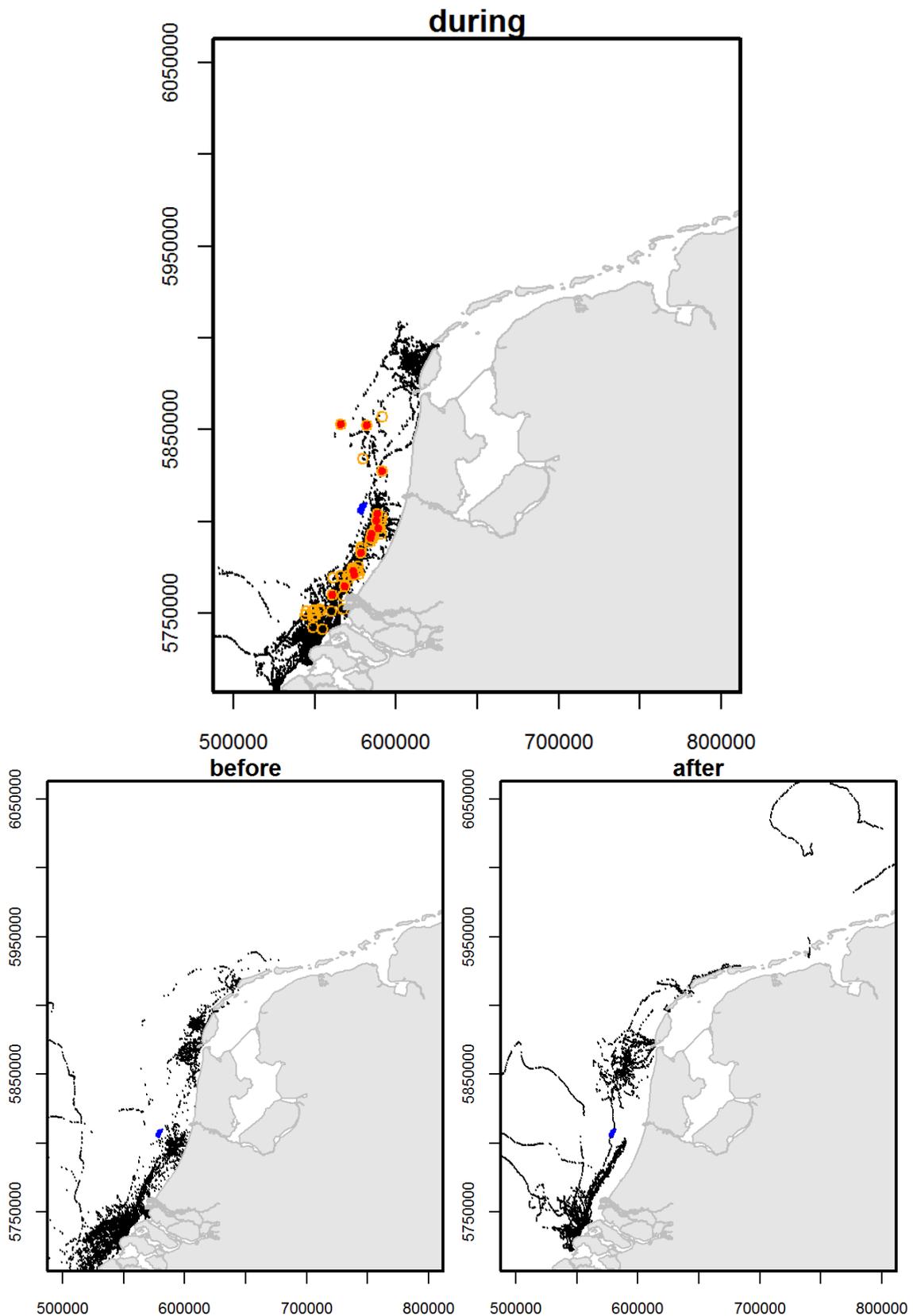


Figure 5. Top: Locations of seals exposed to Luchterduinen pile-driving 31 July to 16 October, 2014. Exposures are indicated by orange circles, red dots indicate exposures during which significant behavioural responses were detected, and black dots are all seal GPS locations during the respective periods. Blue areas are the windfarm areas. Below: seal distribution before and after the pile-driving

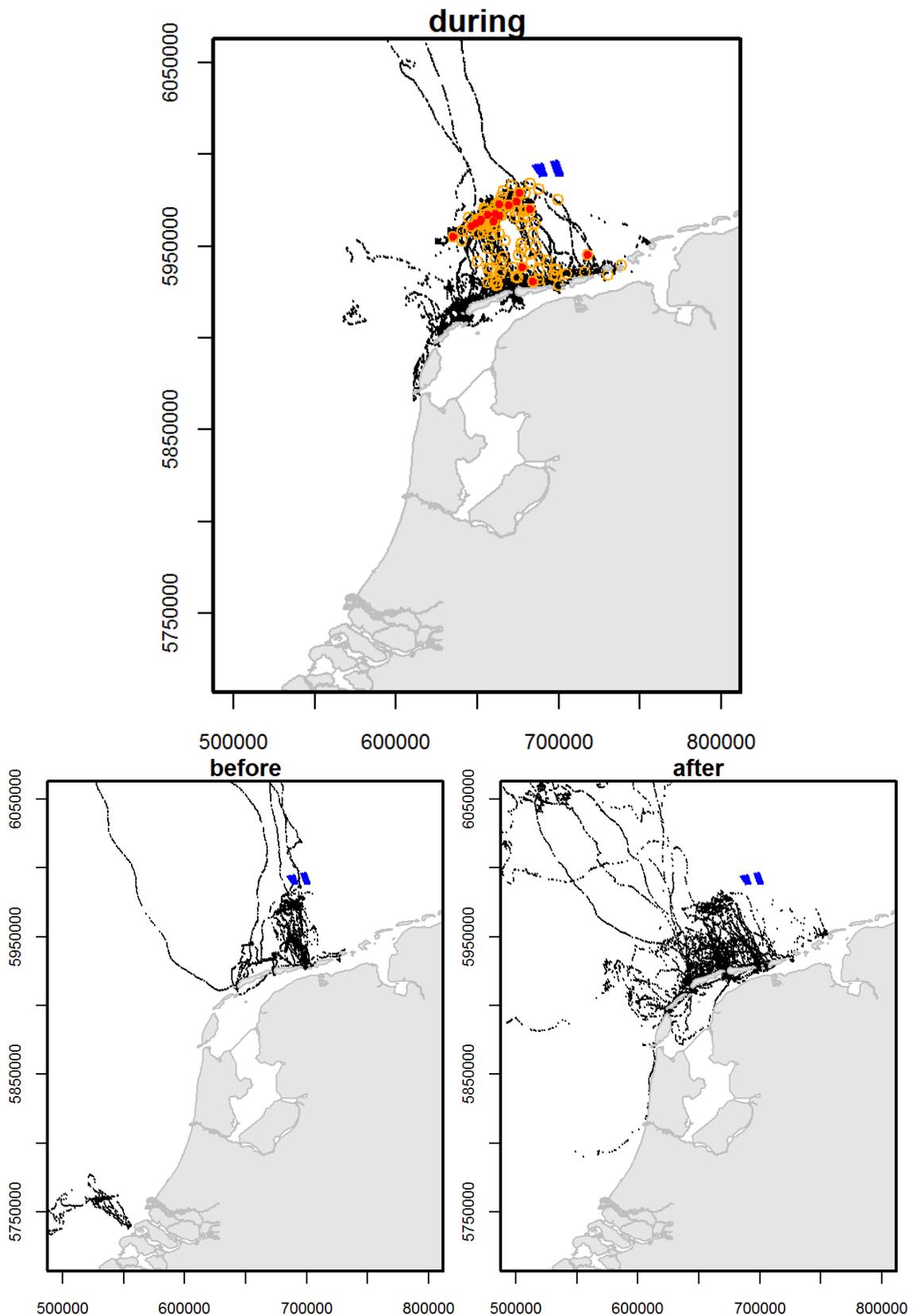


Figure 6. Locations of seals exposed to Gemini pile-driving 1 July to October 17, 2015. See Figure 5 for more details

3.3.2 Diving behaviour during pile-driving

Visually detectable changes to the dive patterns at the commencement of pile-driving were evident in a number of cases. The most typical responses observed were for a seal to lower its descent speed (presumably diving more diagonally and moving away) and when it was spending a high proportion of

its' time near the bottom (presumably foraging) prior to pile driving, to decrease its average dive depth (e.g. Figure 7). The diving pattern often became much more irregular; mixing long and short dives, with many dives not reaching the sea floor. Also, during pile-driving the seals performed occasional high speed descents and ascents.

Although there was a typical response pattern coinciding with pile-driving, exemplified by a reduction in bottom time, there was considerable variation within and between individuals. Seal hg46G-A119 provided an example of the variability in responses by an individual. On 25 September 2015, this seal was 30 km from a pile-driving event and responded by dramatically reducing its bottom time (Figure 8). One week later, on 4 October 2015, this seal was 45 km from a pile-driving event. The event coincided with the seal breaking from period at the surface, potentially at rest, and a resumption in bottom diving – hence, it showed an increase in bottom time (Figure 9).

The strength of a seals' response did correlate with distance from pile-driving, but there was large variability in the responses between exposures. On occasions a significant response occurred at large distances, e.g. based on the analysis one occurred at 48km away from the pile-driving (Figure 7). On other occasions, no clear response in diving was apparent, even at close range (i.e. 12km, Figure 10).

On several occasions, a change in movement direction or speed occurred. For example, seal hg46G-A119-14 (on August 24 2015, at 16km from pile-driving) showed a strong increase in swim speed, up to 1.8 m/s and persisted in swimming at such a high speed for more than an hour. Interestingly, the seal did not swim away in a single direction, but swam in a large circle during its exposure to pile-driving. On other occasions, pile-driving lead to a sudden change in movement direction, while no clear change in diving behaviour was apparent. This was for example the case for seal hg46G-A119-14, exposed to pile-driving 29km away, on August 4 2015. The seal did not swam away from the pile-driving, but made a 90-degree right turn (Figure 12). On several other occasions, exposure to pile-driving lead to a change in dive behaviour, and also a (temporary) change in movement away from the pile-driving (see e.g. Figure 14.)

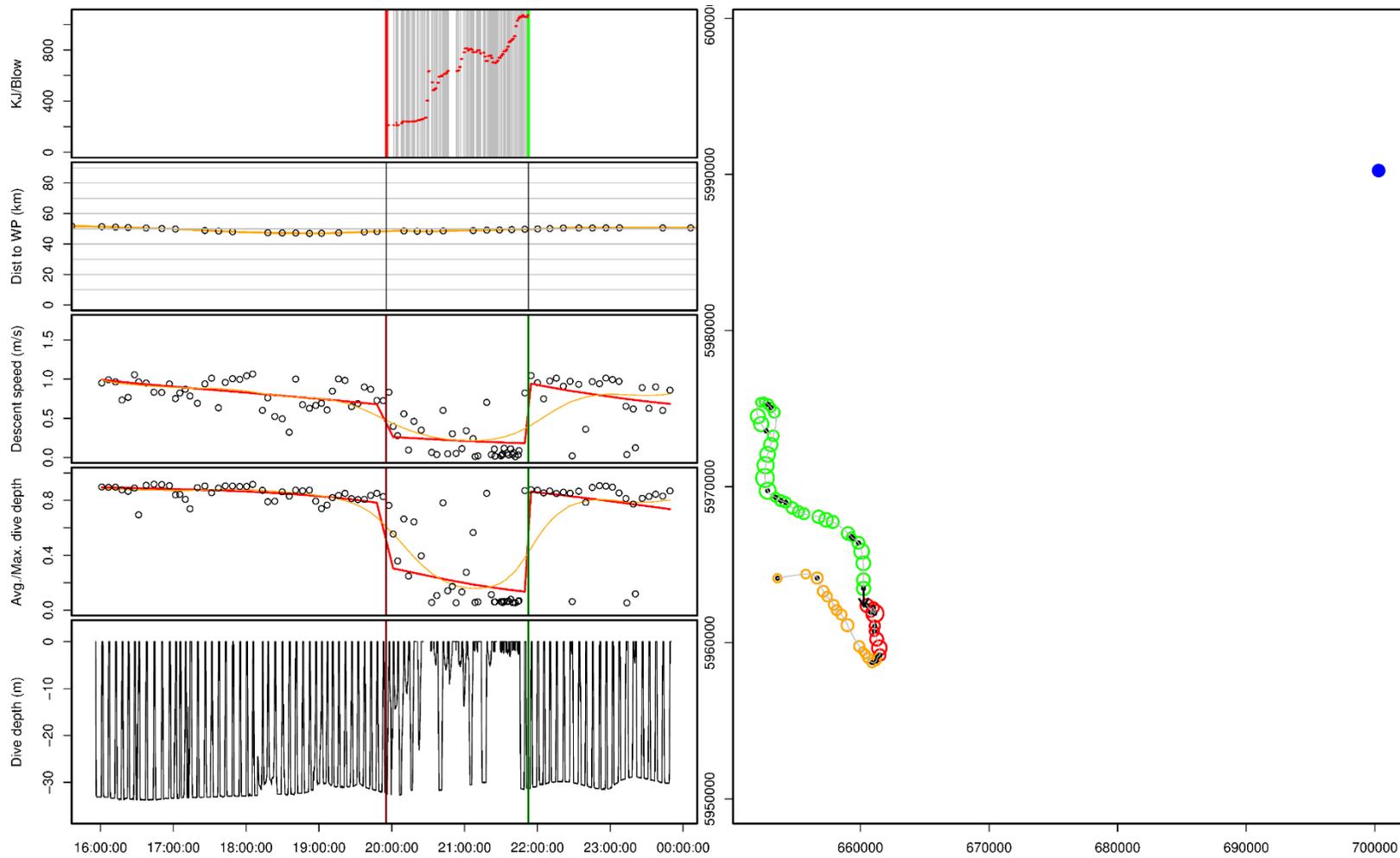


Figure 7. Example of a typical response to pile driving: seal hg46G-A119, 48 km away on 15 August 2015 (monopole X2). Coinciding with commencement of pile-driving (1st panel, left), the seals' diving behaviour became irregular (5th/bottom panel, left), descent speeds declined (3rd panel, left), and average dive depth (as fraction of the water depth) decreased (4th panel, left), implying that seals stayed more time near the surface or mid-water, and less time near the bottom. After pile-driving ceased, more routine dive patterns resumed. The 2nd panel on the left shows the distance to the wind park, with each dot representing a GPS fix. The right figure indicates the movement of the seal in relation to pile-driving site (blue dot). The colours represent locations prior (green), during (red) and after (orange) pile-driving. The end point of the black arrow indicates the expected location of the seal when the seal would

continue its normal track based on last direction and speed of the green, non-disturbed track. If the first red location (during pile driving) is far away from the end point of the arrow, this is indicative of a change in movement speed and/or direction.

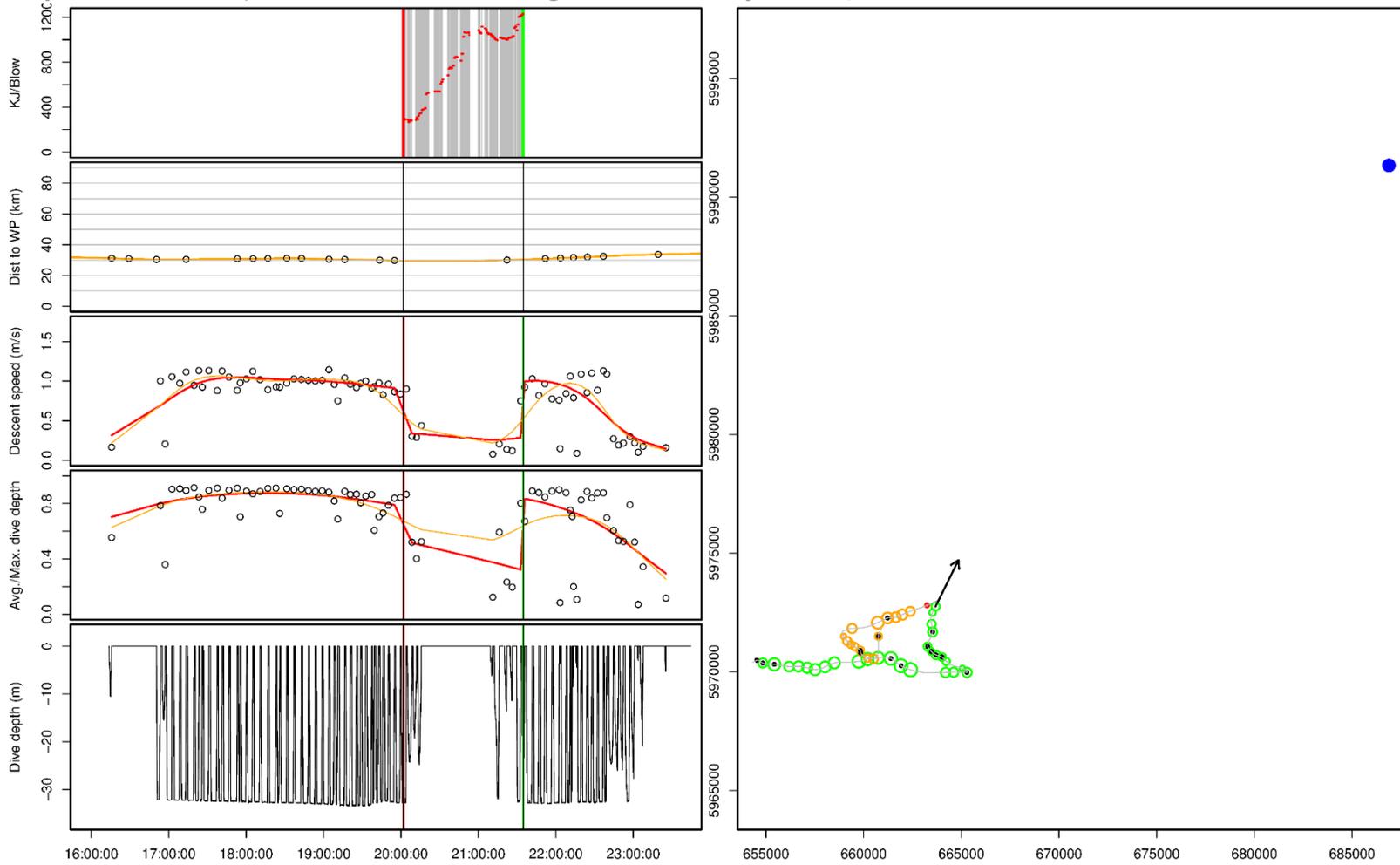


Figure 8. Example of seal reducing its bottom time and changing direction during a pile-driving event: seal hg46G-A119, 30 km from pile-driving on 25 September 2015 (monopile A2).

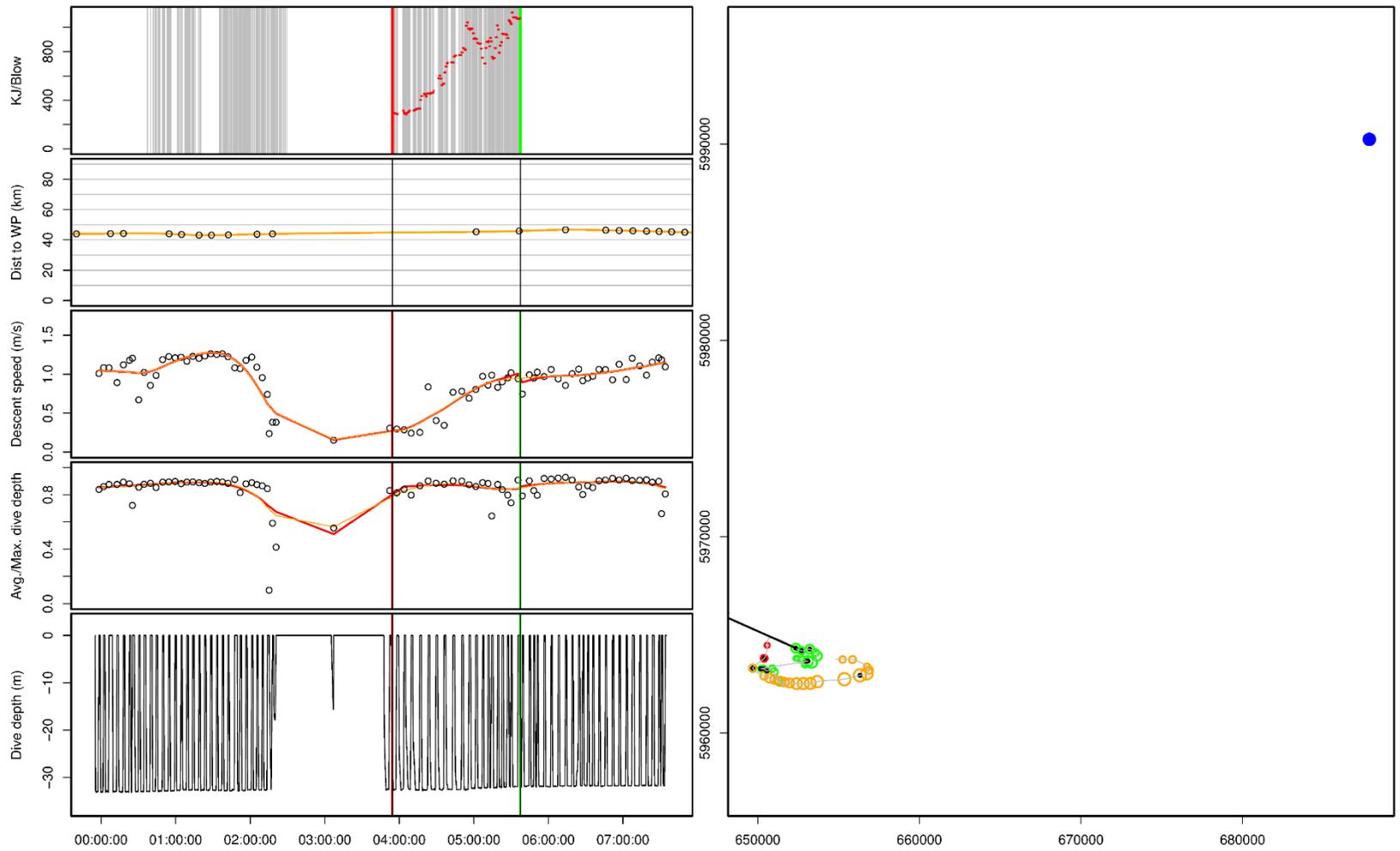


Figure 9. Example of seal increasing its bottom time during a pile-driving event: seal hg46G-A119, 45 km from a pile-driving on 4 October 2015 (Monopole J2).

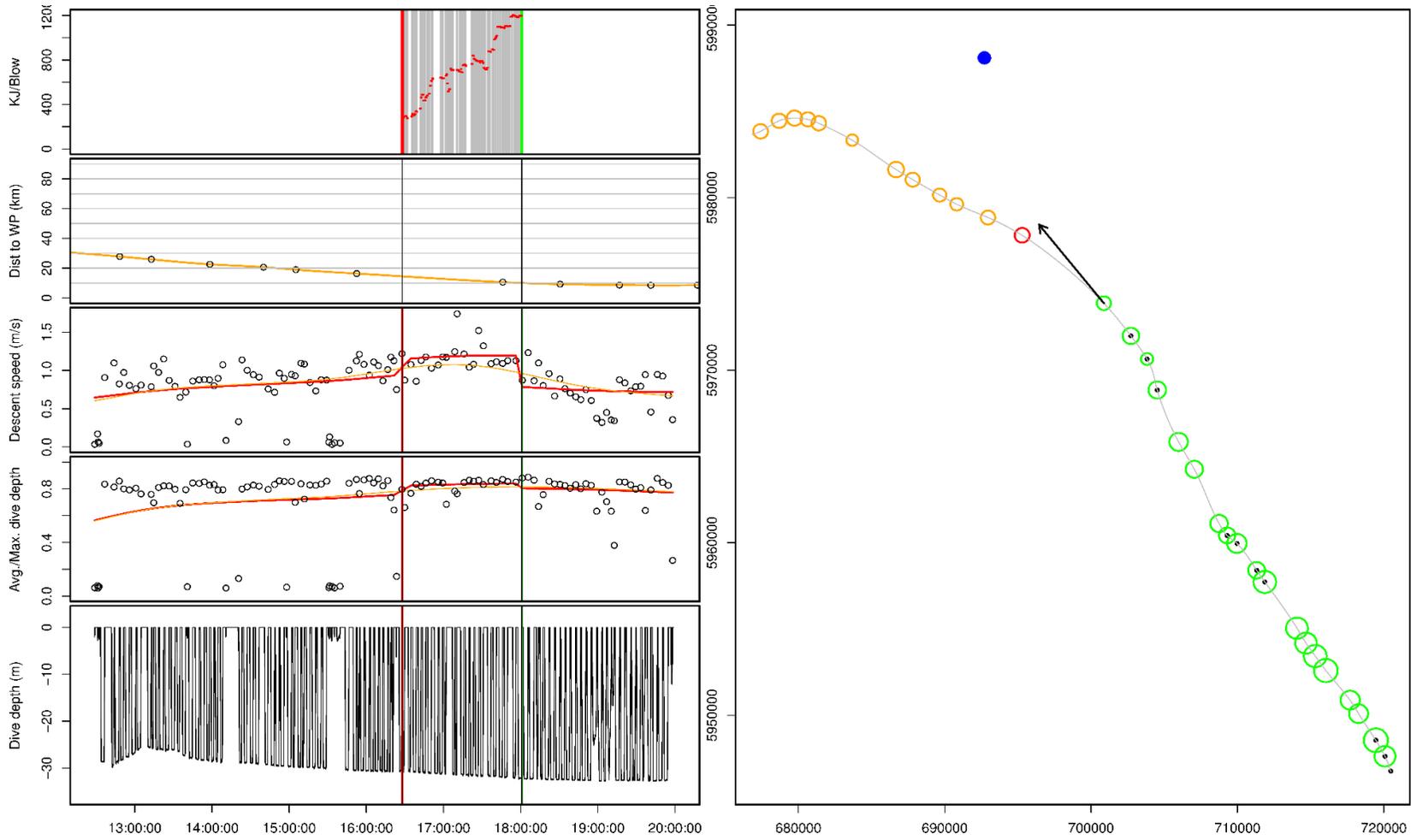


Figure 10. Example of seal showing only a slight response during a close pile-driving event: seal hg51G-B133, 12 km away on 24 September 2015.

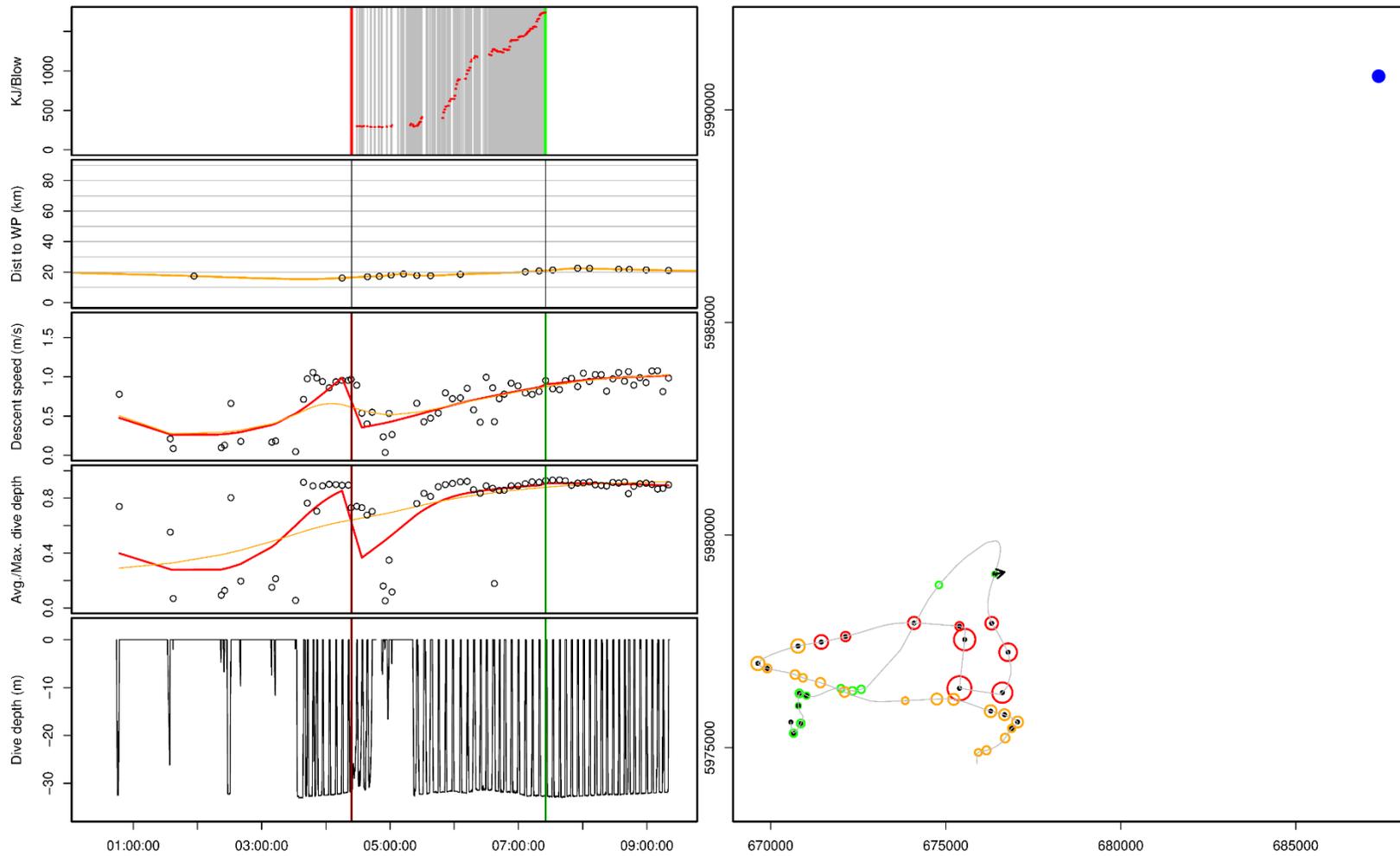


Figure 11. Example of seal hg46G-A119-14 (at 16km from pile-driving on August 24 2015, monopole K2) decreasing its dive depth and descent speed at the start of pile-driving event. Note the high horizontal movement speeds, shown by size of red dots on the map: up to 1.8 m/s during pile-driving. Such high speeds are at the 2% quantile of observed swim speeds for that seal.

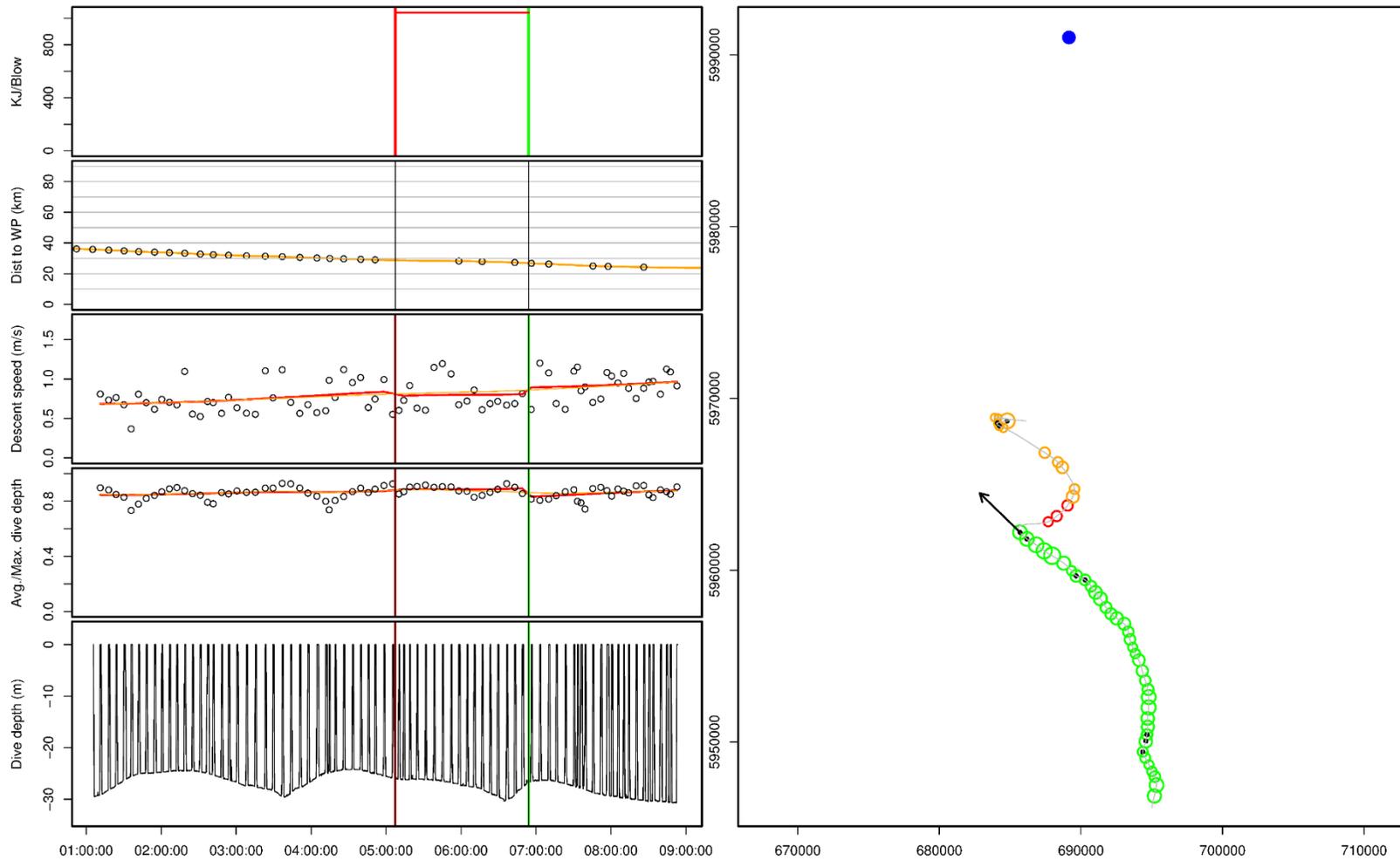


Figure 12. Example of seal hg46G-A119-14 at 29km from pile-driving on 4-8-2015 (monopole OHVS1-B3). Only a slight (non-significant) change in the average dive depth is apparent, but the seal has changed its movement course. If the seal had continued its path, the first red dot was expected to be located at the end of the black arrow, but clearly the seal did change course and speed once pile-driving commenced. Ultimately, it resumed its travel direction to pre-piling.

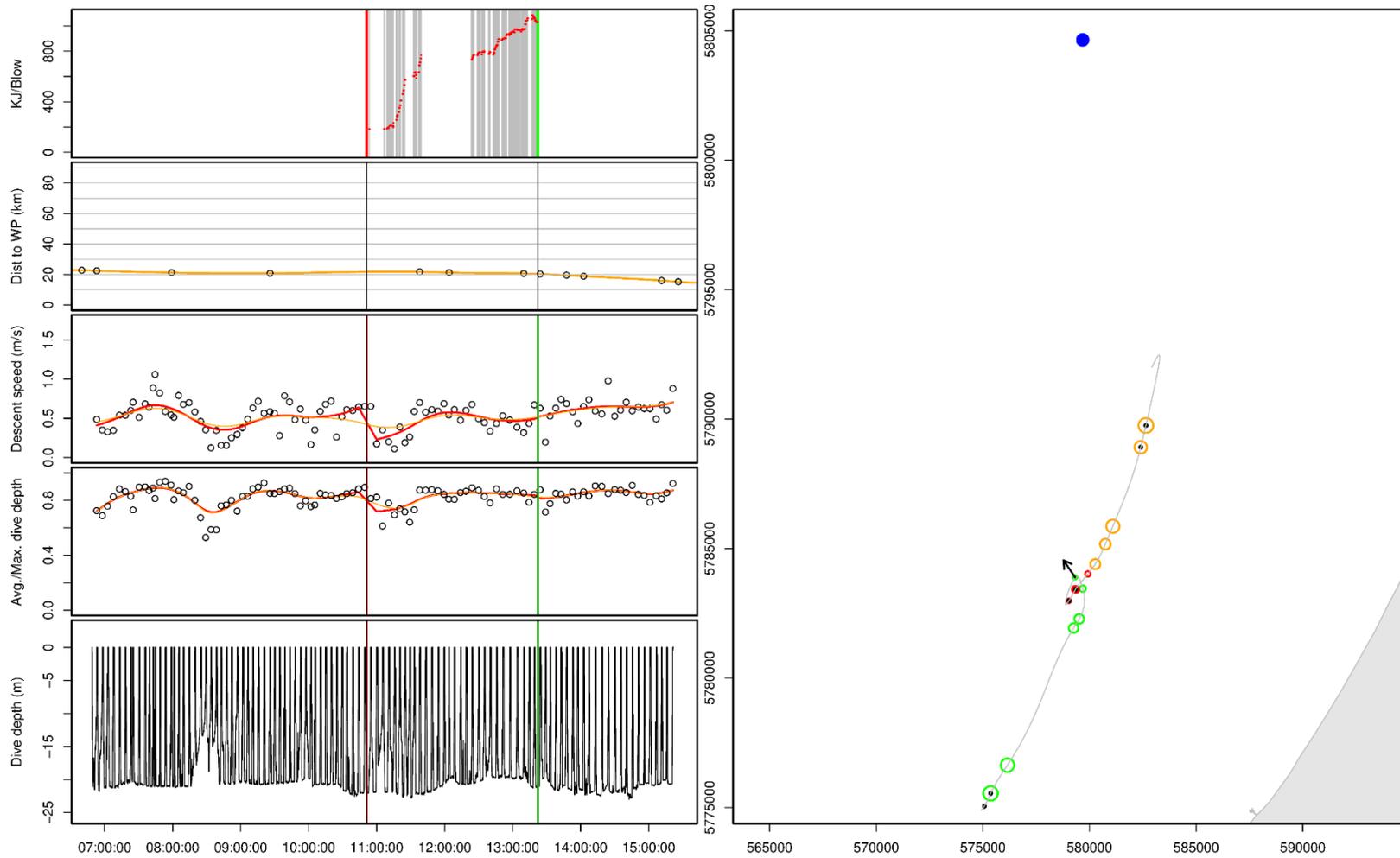


Figure 13 Example of change in movement direction away from pile-driving. Seal hg43LZ-Z024-14 was exposed to Luchterduinen pile driving (E07) 24km away on 10-10-2014. Just after the start of pile-driving, the seal decreased its descent speed (presumably swimming away), and decreased its average dive depth. Also it changed its swim direction: Prior to pile-driving it was heading north, but, after pile-driving had started, it was located further south, and must have changed its course. However, after approximately 30 minutes, it continued in northern direction again.

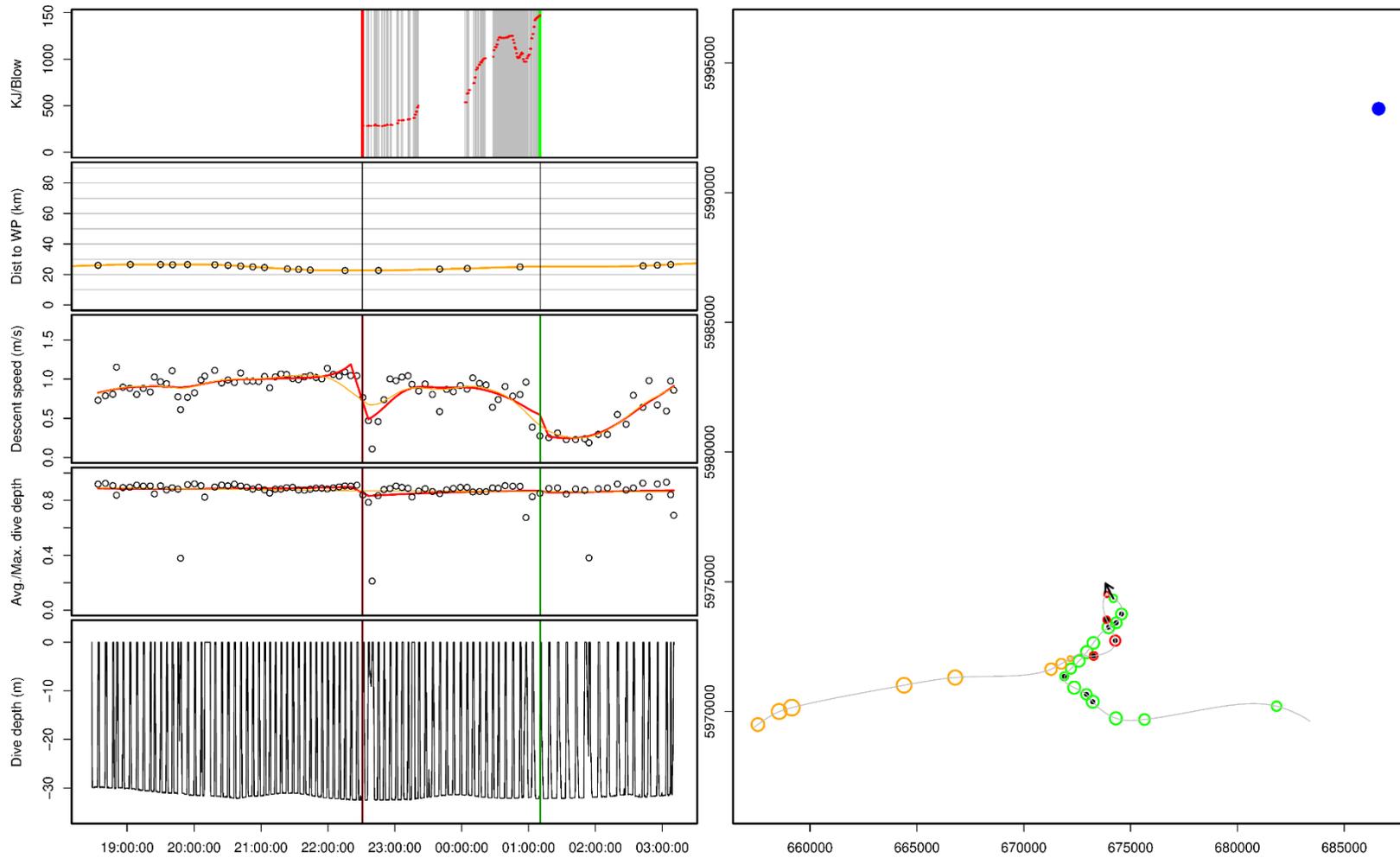


Figure 14. Another example of change in movement direction and dive behaviour. Seal hg46G-A119-14 was exposed to pile driving (B4) 23km away on 04-08-2015. Just after the start of pile-driving, the seal decreased its descent speed (presumably swimming away), and for a short duration, decreased its dive depth. Also, it changed its swim direction: Prior to pile-driving it was heading north, but, after pile-driving started, it moved first south, and eventually continued westward.

Despite the large variability in the behavioural response observed, some responses occurred consistently more often at smaller distances from the pile-driving, demonstrating the link with the pile-driving activity. From the behavioural response variables explored, the change in descent speed (measured from 1% of the dive to near the bottom), showed the strongest relation with distance to the pile-driving (p -value = 0.00014). Up to 36 km from the pile-driving the estimated average decrease in decent speed was significantly different from no change (i.e. 0). (Figure 15, top figure). The effect of distance to pile-driving on the change in descent speed was also observed when analysing the data from Luchterduinen en Gemini independently (Figure 15, bottom figures).

There was however large variability in the observed change in descent speed; Sometimes no change or even an increase in descent speed occurred at small distances from pile-driving (see also Figure 10). In other instances, as indicated by the thick circles in Figure 15 a significant decrease in descent speed was observed well beyond the 36km (e.g. at 48km, Figure 7). However, overall the largest changes in descent speed occurred at smaller distances, and such changes occurred more frequently.

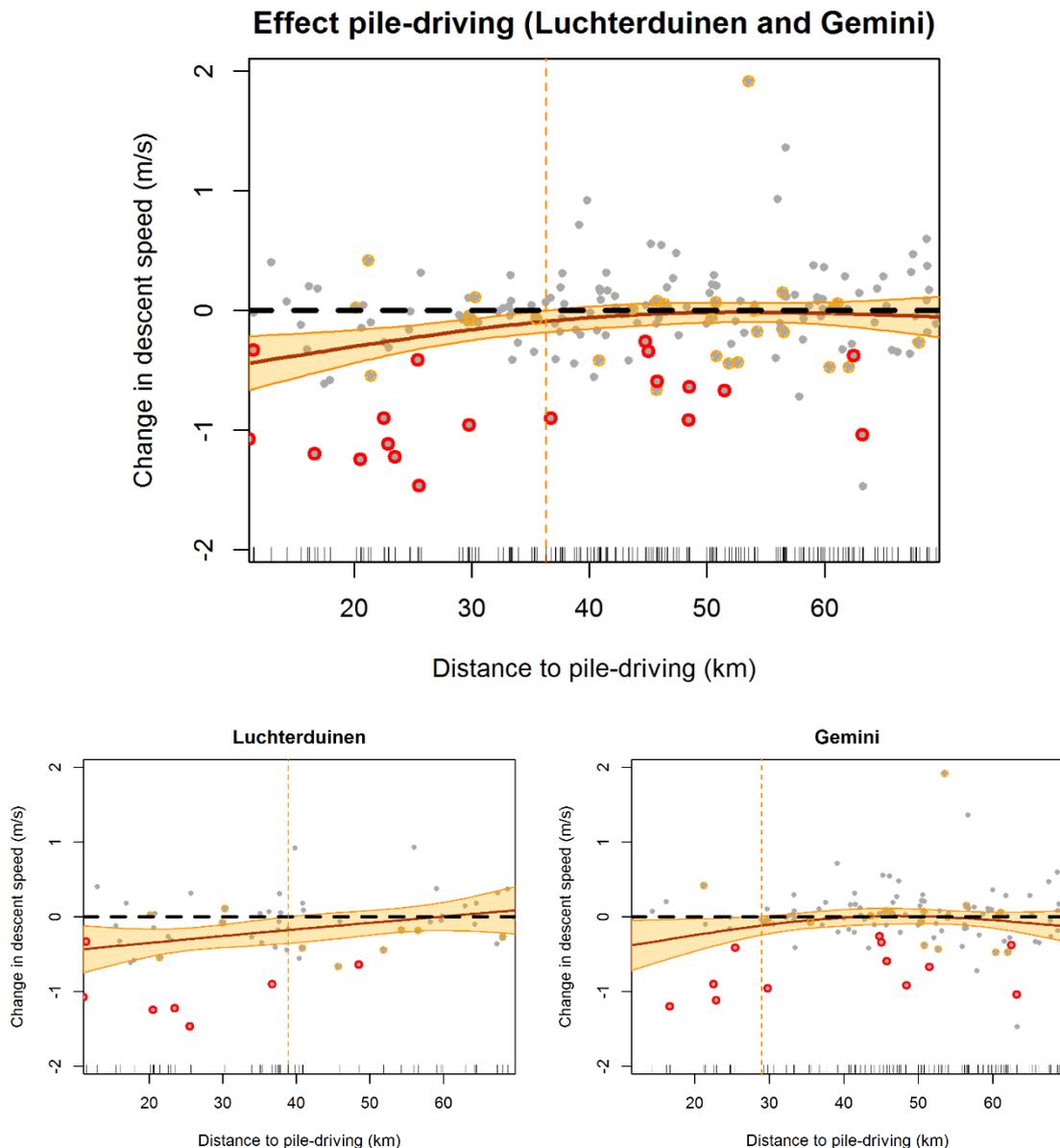


Figure 15. Change in descent speed (m/s) during pile-driving, as function of distance to the pile-driving. Each grey point represents an exposure. The solid black line represents the mean estimate, and the shaded orange area the 95% confidence interval (with 2.5% and 97.5% lower and upper limits, respectively). The orange vertical line (at 36km in top figure) indicates 97.5% certainty of a significant decrease in descent speed. Thick red circles are exposures where the descent speed drops significantly during piling. The lighter

orange circles are exposures where significant changes in other behavioural response variables were observed (i.e. average dive depth or change in (horizontal) movement). Analysis is presented for all data (top figure), and for Luchterduinen and Gemini separately (bottom figures).

We also studied whether other covariates influenced the size of the effect (i.e. change in descent speed). Seals may differ in their behavioural response to under water sound. However, adding seal-id as random effect, did not lead to a significant improvement ($p=0.43$) in explaining the observed variability in the responses. The number of animals, and hence the statistical power might be too low. Or the variability in response is perhaps more context-dependent, rather than individual dependent. The water depth influences the SELs, but may also affect the seal behaviour. However, adding a smooth of the depth as a covariate, did not lead to an improvement of the model (p -value = 0.35). However, since exposures occurring at shallow depths (>10 m depth) were not included in the analysis the effect of depth was possibly removed *a-priori*. Windspeed may increase the background noise, and as such mask the pile-driving sound. There were suggestions, based on single events, that this might occur (e.g. seal hg51G-B133 moving towards the pile-driving on 24 September, occurred at windspeeds of 6 Bft, Figure 10). Adding an interaction-term between distance to pile-driving and windspeed, did lead to a significant improvement in the model ($p= 0.0007$), though counterintuitive, the results suggest that the decline in descent speed is more severe at *higher* wind speed. Finally we tested whether an increased number of exposures for each individual led to an increase (sensitisation) or decrease (habituation) of the effect size. This was not observed in the data (p -value = 1.0). It should be noted that it was challenging to determine the number of times an individual was exposed to pile driving, since a seal might be capable of hearing pile-driving at large distances. It might however not consider this as 'disturbing' and might also not necessary know where pile-driving occurs.

3.3.3 Behavioural responses in relation to sound exposure levels (SELs)

Ultimately, seals are expected to increase their response relative to the loudness of sound they are exposed to, and distance to pile-driving is then merely a proxy for SEL (or Sound Pressure Level - SPL). However, wind-corrected maps of SEL for seals exposed to pile-driving near Gemini were not readily available at the time of this study. For Luchterduinen, the levels of sound at which seals were potentially exposed to pile-driving were estimated to range up to 150 dB re 1 μPa^2 (Figure 16). As expected, the SELs decreased with distance. However, there were considerable deviations, which were caused by the heterogeneous bottom topography and differences in wind speed. Overall, the SELs experienced near the surface was lower compared to close to the bottom, due to surface-specific attenuation of the pile-driving sound.

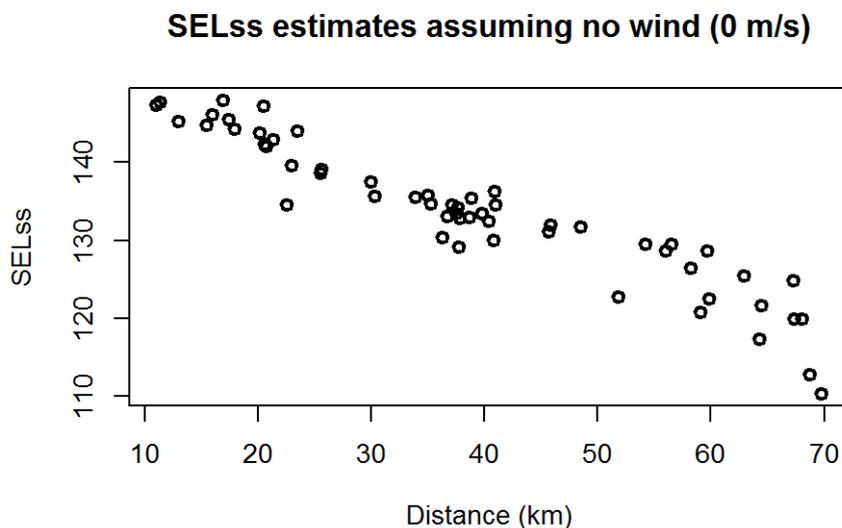


Figure 16. Correlation between Sound Exposure Level single strike (SELs, in dB re 1 μPa^2) near the bottom, and distance to the pile-driving site determined for the exposure events.

The figure is based on the SEL_{ss} estimates which assumes no effect of wind on attenuation (i.e. wind speed = 0 m/s).

To investigate the effect of SEL_{ss} on the change in descent speed, we used the SEL_{ss} estimates near the bottom, and assumed wind speed of 0 m/s (regardless of the actual wind conditions at the time of the exposure). In that case, a behavioural response was estimated to occur (with 97.5% certainty) up to 133 dB re 1 μPa^2 (Figure 17). For SEL_{ss} exceeding ~ 137 dB re 1 μPa^2 , the majority of exposures (i.e. 10 out of 18) did lead to a significant behavioural response in any of the dive or movement variables.

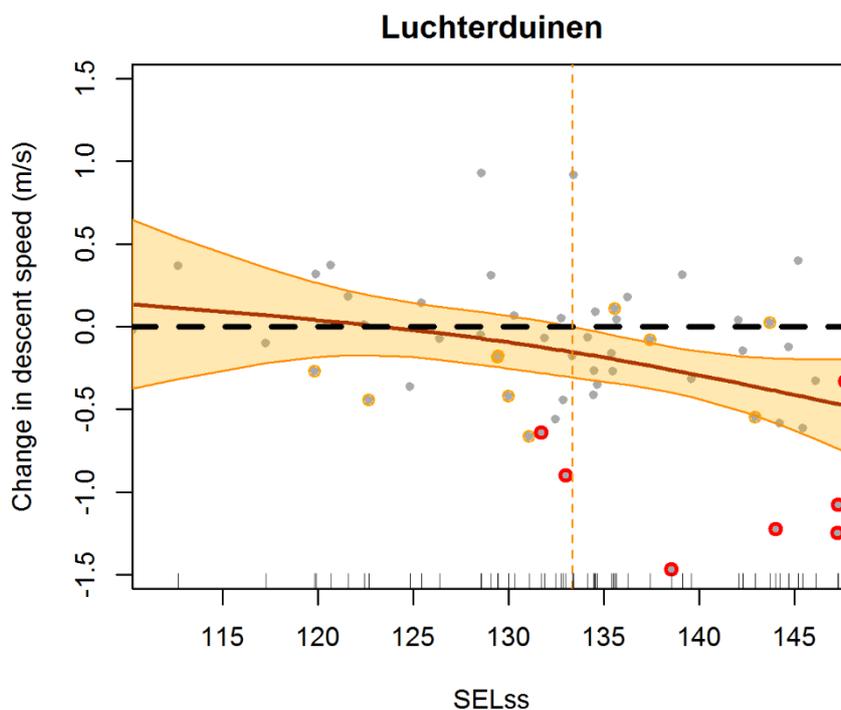


Figure 17. Unweighted Sound Exposure Level from a single strike (SEL_{ss} , in dB re 1 μPa^2) during pile-driving and change in descent speed. For more details, see Figure 15. SEL_{ss} estimates assumed no effect of wind (i.e. wind speed of 0 m/s). The solid red line represents the mean estimate, and the shaded orange area the 95% confidence interval (with 2.5% and 97.5% lower and upper limits, respectively). Thick red circles are exposures where the descent speed drops significantly during piling. The lighter orange circles are exposures where significant changes in other behavioural response variables were observed (i.e. average dive depth or change in (horizontal) movement). The orange vertical line (at 133 dB re 1 μPa^2) indicates 97.5% certainty of a significant decrease in descent speed. For SEL_{ss} exceeding ~ 137 dB re 1 μPa^2 , the majority of exposures (10 out of 18) showed a significant behavioural response in one of the dive or movement variables.

3.3.4 Spatial movement in the vicinity of pile-driving

There was large variability in the change in movement direction during pile-driving. Depending on the exposure, individuals moved either towards or away from the pile-driving events. For the Luchterduinen windfarm, avoidance was not apparent, but for Gemini, there was on average more movement away from pile-driving (Figure 18). For example, up to 40 km from Gemini, grey seals moved away from pile driving in 19 of the 25 exposures. The 2.5% lower confidence band was above zero up to 33 km distance, which indicated that seals on average showed a statistically significant avoidance up to this distance. For both Luchterduinen and Gemini, there were very few exposures at small distances (<20km), because seal density was low in the direct vicinity. This explains the larger confidence intervals in this region. Also, the number of GPS locations (every 15 minutes, but often less

frequent) that could be used in the analysis was much smaller than for the dive analysis, and hence the statistical power for this analysis was much smaller. A model was also fitted to simulated false pile-driving scenarios, where the pile-driving period was shifted backward or forward randomly between 0 and 24 hours. These analysis did not show a dependency with distance to the 'false' pile-driving times (p-value=0.94). This shows that tendency to move away correlated with pile-driving and were not the consequence of the analytical procedure used.

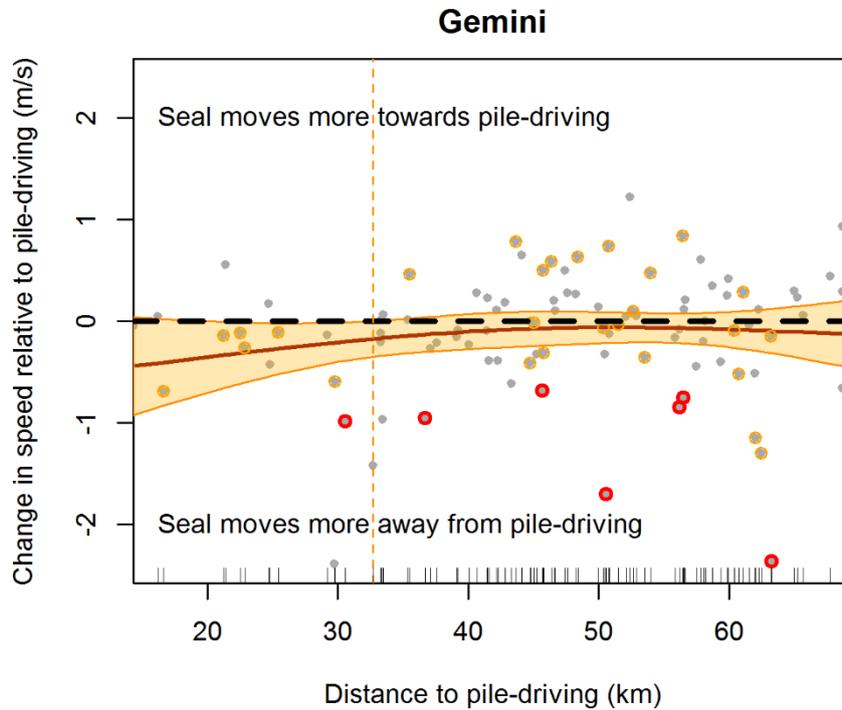


Figure 18. Change in movement speed before and after pile driving started relative to the Gemini pile driving site. Negative values indicate that the movement of seals was directed more away from the site. The shaded area represents the 95% confidence interval of that mean estimate. For Gemini, grey seals < 33km from pile-driving tended to move away .

4 Discussion

Grey seals are abundant, top-level predators in the North Sea. Their numbers, distribution and density in this area have been increasing since the 1980s, possibly in response to their relatively recent protection from human hunting (Lambert 2002, Brasseur *et al.* 2015). The North Sea is however also a region of increasingly dense human activity, with heavily utilised shipping lanes, oil and gas installations, seismic surveys, dredging, fishing, explosive clearances and windfarm constructions (Degnbol & Wilson 2008, Ducrotoy & Elliott 2008). All these activities can produce sounds underwater that can be detected by marine mammals and may influence their behaviour (Southall *et al.* 2007). There has been minimal effort to understand how grey seals might be impacted by anthropogenic underwater sounds, particularly compared to efforts to measure such impacts on the harbour porpoises (Carstensen *et al.* 2006, Tougaard *et al.* 2009, Dähne *et al.* 2013, Kastelein *et al.* 2013b, Hastie *et al.* 2015). Recently, however, there have been several studies of acoustic abilities of grey seals (Götz & Janik 2011, Hastie *et al.* 2014, Stansbury *et al.* 2014), and given their protected status, governments, for example in the Netherlands, UK and Denmark, have included grey seals as species of interest in impact assessments of industrial activities (e.g. Skeate *et al.* 2012).

In this study, we investigated potential responses of grey seals to pile-driving for offshore windfarms on the Dutch continental shelf. The approach was to attach tracking devices that transmitted location and diving data, to grey seals that could move within the vicinity of the windfarms. Seals were captured at sandbars as close to the respective windfarms as possible. As grey seals can move over great distances (McConnell *et al.* 1999, Matthiopoulos *et al.* 2004), there was no certainty that the tracked seals would remain in the area. Moreover, there was a time gap between when seals could be caught (by Dutch law, haul out sites are closed between May 15th and September 1st) and the designated pile-driving periods (again by Dutch law, between July and December). A positive result for the study was however that 20 tracked seals (in water depths >10 m near or within the Wadden Sea or Delta) were within 70 km of pile-driving events, resulting in 261 exposures. For several exposures however, there were no GPS locations near the start of the pile-driving, and 175 exposures could be used for the analysis.

An important caveat in the results is that the interpretations of responses were derived from remotely recorded broad-scale, movement compared to the expected reaction of the seals and dive data that were limited to depth only: in average, only 2-3 locations were recorded per hour. Fine scale responses, such as altered sinuosity in travel or changed heart rate, that could have been indicative of an impact on the individual, could not be measured. Therefore, we probably overlooked events where seals might have reacted to the pile-driving.

4.1 Seal dive behaviour and pile-driving

An important finding of this study was that most often grey seals reduced descent speed (presumably diving more diagonally) and reduced their bottom time during pile-driving events, certainly on average when within 36 km from the pile-driving and occasionally at distances well beyond this. Grey seals are predominantly benthic feeders (Thompson *et al.* 1991), so a reduction in bottom time has a direct effect on their food intake and means the individuals would need to work harder to gain required resources during times when there was no pile-driving, or forage elsewhere.

Changes in behaviour that corresponded in time to pile-driving would provide an indication that the grey seals were detecting pile-driving. Such changes were particularly evident in the seals' dive profiles. There was, however, considerable variation in the response. On occasions a change in dive pattern was evident coinciding with, particularly, the commencement of pile-driving, while on other occasions a reaction to the pile-driving was either not evident, or not exactly synchronised with the pile-driving.

4.2 Directional movement during pile-driving

Similar to the behavioural changes observed in the diving profiles, there was large variability in the change in movement, with grey seals moving both towards and away from the pile-driving site. A significant effect was observed for seals near the Gemini wind park, but not for Luchterduinen, where less data were available. *On average* seals near Gemini would move away from the pile-driving site, and we can state with 97.5% certainty that this was significant at least up to 33 km. Up to 30 km the vast majority of seals moved away from pile-driving (19 out of 25). This pattern was not observed for the randomly created pile-driving events.

It is not known if at these large distances the seals were able to accurately determine the direction from which the pile-driving sound was coming, as due to bottom and surface reflection and resonance, the sound may be received as a rumbling noise rather than the impact heard at closer range. This could explain the unexpected seal movements observed. On one occasion a seal was swimming in a large circle at a high speed (~ 1.8 m/s) for nearly an hour (**Figure 11**). During another exposure, a seal suddenly changed its course, but the new course was not directed away from pile-driving (i.e. **Figure 12**). Also it is unknown if the seals could comprehend that the sound source (pile-driving) was not moving. For example, a ramp-up in pile-driving energy might be perceived by the seal as something approaching, which could make some seals increase speed or head towards perceived safe areas (shallow or deeper water), and others might attempt to determine the direction of the approach (for example by zig-zagging). Prior experience with the pile-driving in the area might influence the directional response of a seal during an exposure, because they could have a better idea of the direction to the pile-driving, its potential duration, and what behaviours or movements could minimise any potential disturbance from it, while maximising prey acquisition.

On several occasions, particularly in relation to Luchterduinen windfarm, seals did move into shallower water during pile-driving (e.g. see Appendix), but again, this response was not consistent. Occasionally, a seal would move toward deeper water during pile-driving. The potential that water depth could influence movement patterns of grey seals during pile-driving events requires further investigation.

4.3 Sound exposure levels

This study indicates that a behavioural response by grey seals to pile-driving occurred in response to SEL_{ss} of 133 dB re 1 $\mu\text{Pa}^2\text{s}$ (1 m above the bottom, unweighted, assuming no wind-effect). This was below the threshold level of SEL_{ss} = 145 dB re 1 $\mu\text{Pa}^2\text{s}$ at which a review conducted prior to installation of Luchterduinen windfarm had predicted that seals would take evasive action (Heinis 2013): that level was based on the hearing ability and response of captive harbour seals (Kastelein *et al.* 2013b). One explanation for the discrepancy between assumed and actual levels at which grey seals might respond might be that grey seals are more responsive than harbour seals to pile-driving sound. However, a preliminary inspection of the dive-profile plots for harbour seals also showed occasional responses at larger distances (e.g. ~ 40 km, See Supplementary **Figure 19**).

Another explanation might be that there is individual variability in the response of grey seals, both in the wild and in captivity. The two harbour seals exposed in captivity (Kastelein *et al.* 2013b) might be less sensitive, only responding at higher SEL_{ss}. Or the context (i.e. being in captivity) might influence the probability and strength of the response.

Finally, an explanation might be that the current sound model propagation estimates for SEL_{ss} are inaccurate. Indeed, Binnerts and de Jong (2016) stress that the SEL_{ss} estimates for distances beyond 15 km are unreliable, whilst by far most of our measurements are beyond 15 km. Moreover, for higher wind speeds, mostly due to unknown effects of wind-related surface attenuation, and more inshore locations, where most seals are found, the estimates are likely less reliable (Binnerts & de Jong 2016). The SEL_{ss} measurements for Luchterduinen at 46.6 and 47.1 km were 120 and 118 dB re 1 $\mu\text{Pa}^2\text{s}$, respectively. The estimates by the Aquarius sound propagation model were 132 and 119 dB re 1 $\mu\text{Pa}^2\text{s}$, respectively. These results suggest that the SEL_{ss} estimates used in this study are more likely overestimates, rather than underestimates. This would imply that grey seals might respond to even lower received SEL_{ss} (i.e. < 133 dB re 1 $\mu\text{Pa}^2\text{s}$).

4.4 Individual variability in behavioural response

Although specific responses, notably a reduction in bottom time, could be correlated with the pile driving, even at large distances, there was large variability in measured response between and within individuals. On different occasions, grey seals were observed to: stop resting at the surface, increase their time at the surface, decrease the dive time spent near the maximum depth, increase their time at depth, or show no apparent response (see Figure 7 to Figure 14). One reason for the observed individual variability in the response might be individual differences in hearing abilities or sensitivity to sound. Harbour seals tested in captivity and in the field, have demonstrated significant differences between individual and within individual variability in hearing sensitivity (Kastelein *et al.* 2013a, Lucke *et al.* 2016). Old age, for example, can generally reduce hearing sensitivity, as was demonstrated in harbour seals (Lucke *et al.* 2016).

In many cases, no response was observed. When seals are exposed to pile-driving, we expected that they would often switch from a foraging state (straight descent and long bottom time), towards a transit state (more diagonal movement and less time near the bottom). A more diagonal movement (and lower vertical descent speed) would indeed be a more efficient route to maximize movement in the horizontal plane. Seals, however, also need to take current speed into account, and current speed varies by depth (Wagenaar & Eecen 2010). Depending on the direction of travel and the direction of the current, it may be more efficient to travel as close to the bottom – where current speed is often lowest – as possible. In that case, it can be challenging to differentiate between foraging and fleeing.

Seals may also respond differently because pile-driving sounds would be masked to different degrees over time. For example, strong wind or other anthropogenic sound sources, such as nearby shipping, dredging, or fishing activity, but also presence of underwater dunes or gullies might attenuate the propagation of sound and could reduce the seals chance of detecting the pile-driving sounds.

Finally, individual qualities of each seal could influence how it responds to a stressor, such as anthropogenic sound. For example, the startle response of animals can be influenced by their nutritional status, sex or hormonal condition (Plappert *et al.* 2005). Thus, there are numerous exogenous and endogenous reasons for there being a broad range of responses to pile-driving. In this study, there was large variability in the observed responses, but we did not find significant evidence that this variability was the result of between-individual. However it should be noted that the number of individuals exposed at small distances (tens of kilometres), was probably too small for such an analysis.

4.4.1 Potential prior experience with pile-driving

An important consideration for the interpretation of these data is that information is not available on the prior experience of individual seals to pile-driving, nor what exposure they had to other potentially disturbing activities at the site. During the months prior to pile-driving at Luchterduinen, there was increased shipping and sonar activity over the area, the area was searched for unexploded ordnance, dredging was required to lay cables to each turbine, and layers of stones were dropped at each monopile site to provide scour protection. Therefore some individual seals with prior experience of windfarm construction may have already moved to other areas prior to the commencement of pile-driving. Hence, in this study we could only measure behavioural response of those individuals that remained in the vicinity of the construction sites.

In recent years, there has been considerable pile-driving activity in the North Sea including in close proximity to the Dutch coastal zone. Twenty kilometres north of Luchterduinen, 36 towers were installed at OWEZ windfarm in 2006 and 60 were installed at PAWP in 2007. Also since 2008, 181 towers have been installed at three Belgian windfarms, 60-70 km south west of our seal capture sites in the Delta, and 415 have been installed at three UK windfarms, within 170 km of the Dutch Delta region. Potentially, all the grey seals we tracked had heard at least one pile-driving event previously. They could have been aware that a pile-driving event would last a particular duration (e.g. 2 hours) and then stop for a duration (e.g. 12 to 24 hours). The varying degrees of prior experience of the tracked seals with pile-driving (potential habituation for some individuals) could have mitigated reactions to later pile-driving events.

In addition, while pile-driving at windfarms would have been one source for sound on the Dutch coastal zone in 2014 and 2015, other sources included shipping activity, dredging, fishing and occasional retrieved ordnance explosions (e.g. see von Benda-Beckmann *et al.* 2015). Responses to sounds from these sources could mask potential responses to the pile-driving at windfarms, and complicate interpretations.

Hence, there are multiple reasons why detection of actual changes in seal behaviour coinciding with pile-driving could be difficult, especially because all data come from back-mounted tracking devices. That the data, particularly the dive-profiles and speed data, frequently indicate changes in behaviour that overlap in time with pile-driving, is of interest and requires further investigation. This represents the first study to record overlap between movement and behaviour of individually tracked grey seals and pile-driving, and the results have relevance to future pile-driving activities in the Netherlands and elsewhere. This study suggests that for grey seals the SEL response threshold is lower, and impact distance is higher than previously thought (based on two captive harbour seal study Kastelein *et al.* 2013b), and therefore that more grey seals might be exposed and impacted during pile driving activities.

4.4.2 Potential reasons to tolerate pile-driving

Some individual grey seals that were exposed to pile-driving, continued to return to the vicinity of the windfarms on subsequent trips and, accordingly, received multiple exposures. Potentially, the motivation to frequently visit the area was that it contained prey resources which the seals perceived to be more available to it, than prey resources elsewhere. The seals might choose to accept the risk of pile-driving rather than take the risk of leaving a known foraging area to seek prey elsewhere.

Grey seals have a high fidelity to breeding sites (Pomeroy *et al.* 1994, Twiss *et al.* 1994, Pomeroy *et al.* 2000), and probably also to moulting and resting sites (Karlsson *et al.* 2005). At sea, there are also indications of local preferences and site-fidelities (Oksanen *et al.* 2014), although long term data are scarce. Seals operate in an open environment through which they continually balance the metabolic costs and gains of remaining in an area with the metabolic costs and gains of moving elsewhere. The motivation to shift is typically less than the motivation to remain, because shifting exposes the seal to unpredictable variables (such as prey, predators, disturbance and conspecific competition). This is a known phenomenon that complicates simplified optimal foraging theories (Kamil *et al.* 1993), because it appears as if the animal is not foraging optimally. For the three grey seals that frequently moved into the area inshore from Luchterduinen before and during pile-driving, the perceived benefits of the prey resources there, did apparently exceed the perceived costs of visiting the area and being exposed to pile-driving, and did not induce them to forage elsewhere.

Seals exposed to a disturbance in an area, could continue to visit the area in that season, because of the anticipated costs of moving, but avoid the area in subsequent years, due to the memory of the disturbance. Accordingly, the knowledge that pile-driving occurred in the vicinity of Luchterduinen might influence foraging area selection by a seal in following years. However, detecting such a reaction could require tracking of the same individual in later years, and knowledge of inter-annual variability in foraging ranges of individuals. Multi-year data from the same individual do currently not exist.

4.5 Potential population-level consequences

This study shows an evasion reaction and change in dive behaviour for at least some grey seals exposed to pile-driving at a distance of several tens of kilometres away from the pile-driving site. Not all individuals appear to respond, but this could be (partly) because of our inability to detect the changes in behaviour, given the large individual variability. For those individuals that do evidently respond, a switch from a foraging state to a more transit state seems apparent. How much reduction precisely is caused by the exposure to pile-driving is difficult to estimate. Either they switch completely to a non-foraging behavioural state, or continue feeding, but do so less efficiently. Given the potential large effect range, a large section of the population could be influenced.

Whether this ultimately impacts the population as a whole (e.g. reduced survival or reproduction), is difficult to determine, and depends on many factors, e.g. seasonal variation in food requirement and energy reserves. An individual on the verge of dying of starvation, might be pushed over the limit and die as a consequence of human disturbance. While at other times of the year or other individuals, the temporary inability to feed might be compensated for and therefore have no measurable consequences.

Another process that will play a role is whether there is density dependent competition for food resources between seals. Reduced local foraging abilities (e.g. due to disturbance), may potentially lead to less depletion and hence a higher remaining prey-availability for future feeding trips or other individuals of the population. Although challenging, these questions are fundamental to answer when attempting to estimate the population-level effects of disturbance.

4.6 Cumulative effects of underwater sound

One of the striking results of this study is that grey seals can respond to pile driving at large distances, significant changes in behaviour were seen even beyond 48 km. Pile driving is a relative loud sound source, but underwater explosions are even louder (albeit infrequent and only single impulsive sound). Also, seismic surveys are loud and produce low frequency sounds. In addition, there are sound sources which have lower source levels (e.g. shipping, dredging or operating windfarms), but are more abundant and continuous. Hence, seals (and other marine mammals) will be exposed to anthropogenic sound on a regular basis, and may be influenced more than previously assumed. Since the hearing abilities of harbour and grey seals is centred more around the lower frequencies (compared to e.g. harbour porpoises), and most anthropogenic sound is at similar lower frequencies, they are likely to be particularly sensitive to anthropogenic sound. Moreover low frequency sounds are known to carry further than high frequency.

So far, most research attention has focussed on temporal and permanent hearing loss, this is potentially more relevant for harbour porpoises as they rely on echo-location to navigate and detect prey and would potentially suffer directly from a loss in hearing. However, the ability to acoustically detect prey might also be important for harbour and grey seals (Stansbury *et al.* 2014). When it comes to assessing the impact of underwater sound on their behaviour, a multitude of factors play a role. Perhaps the best starting point is to assess whether the sound is audible (given a specific background noise), and subsequently how marine mammals respond to this and what the (population-level) consequences are.

4.7 Future research

This study was focussed on testing for a behavioural response in relation to pile-driving, and determining what type of response is invoked, and at what distance (or SEL). Much more extensive analysis can be carried out on the existing dive and location data. One candidate analysis is to carry out a more individual- and context-specific analysis: Is the animal exposed multiple times, and what was the sound-exposure level? At what section of its feeding trip was it exposed? And what was its behaviour prior to pile-driving (i.e. feeding, transiting, resting)?

Although there is room for improvement with the existing seal tracking data, there are also substantial shortcomings that might prevent a more in depth analysis. The GPS location data (only available when at the surface), is often temporarily sparse, and hence cannot be used to measure fine-scale sinuosity in movement. Dive data only provides information on movement in the vertical direction. For example, the existing dive data cannot differentiate between a slow vertical descent or ascent, and a faster more diagonal ascent and descent. Also changes in the proportion of time near maximum depth here assumed to be indicative for foraging, might also reflect other types of behaviour (e.g. transiting). A small proportion of time at bottom might be indicative of successful foraging, or of foraging on pelagic fish in mid-water. To classify the behaviour of seals more precisely, more fine-scaled movement data is required that can measure the 3-D movement underwater, but also detect prey-capture events (i.e. apparent in rapid acceleration of the neck). This could be attained by using accelerometers. The

challenge however is to classify the vast amount of detailed data which is then collected into meaningful behavioural states.

Another 'limitation' of this study is that both Luchterduinen and Gemini were located at relative large distances from the nearest haul-out site (>50km). Therefore, only those seals that venture into the vicinity of de wind parks were exposed, leading to relative few exposures at close range (<15km). The density of seals is substantially higher near the haul-out sites. Some of the future construction sites, e.g. Borssele near the Belgium-Dutch border are also much closer to the haul-out sites. Consequently, the construction of these parks and simultaneous tagging effort should lead to a much larger sample size of exposures at smaller distances. This would allow us to more clearly define how seals respond in response to pile-driving, and subsequently test whether similar behavioural changes also occur at larger distances.

5 Concluding Remarks

When considering the influence of sound on marine mammals, several types of impacts can be discerned, e.g. temporary or permanent hearing loss; masking – which can reduce the ability to communicate, navigate or find prey; or disturbance – which can reduce the efficiency of the ongoing behaviour (e.g. feeding), or cause a flight response. This study has focused on disturbance by investigating changes in dive-patterns and movement direction during temporal and spatial overlap with pile-driving for the installation of offshore wind-turbine towers. The study shows that observed behavioural changes occur at tens of kilometres, which, compared with current understandings and controls on underwater sound production, are relatively large distances. Hence, even low sound exposure levels may lead to a large number of marine mammals (in this case grey seals) being influenced, particularly if such activities are located in regions of high marine mammal densities, e.g. in the vicinity of haul-out, breeding and/or feeding sites, or migration routes.

Pile-driving is one of numerous anthropogenic sound sources that are detectable by marine mammals in the North Sea. Other sound sources, e.g. shipping, seismic surveys and underwater explosions, are present and, hence, disturbances by anthropogenic sounds are likely to occur on a regular basis. Similar to pile-driving, the effects of other anthropogenic sound sources are still poorly understood. Part of the reason for this has been our inability to follow an individual at large distance from these sound sources. However, the rise of smaller and effective animal-borne tracking devices combined with advanced statistical analysis techniques (e.g. Michelot *et al.* 2016, DeRuiter *et al.* 2017) provides an exciting opportunity for more in-depth understandings on how individual animals cope with all sound sources present in their environment.

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7 Quality Assurance

Wageningen Marine Research utilises an ISO 9001:2008 certified quality management system (certificate number: 187378-2015-AQ-NLD-RvA). This certificate is valid until 15 September 2018. The organisation has been certified since 27 February 2001. The certification was issued by DNV Certification B.V.

Justification

Report C006/18

Project Number: 4312100043

The scientific quality of this report has been peer reviewed by a colleague scientist and a member of the Management Team of Wageningen Marine Research

Approved: Dr. Tobias van Kooten
Senior Scientist

Signature:



Date: 5 December 2018

Approved: Jakob Asjes
Manager integration

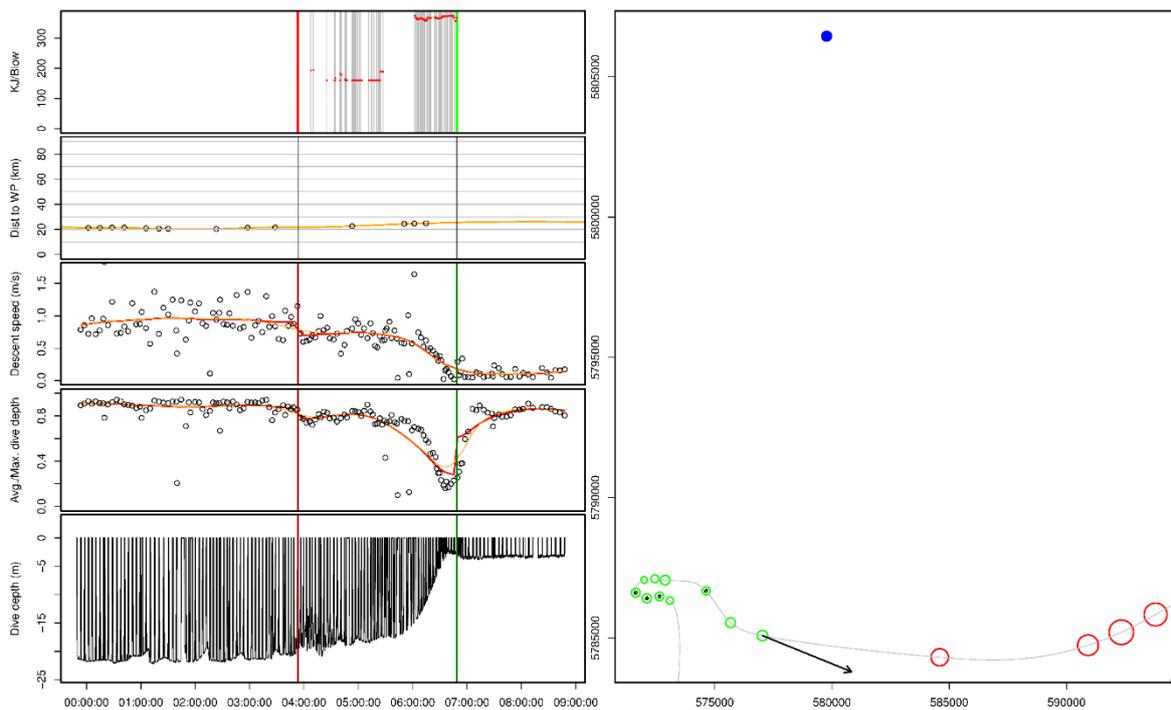
Signature:



Date: 5 December 2018

Appendix 1 Example of movement inshore

An example of movement by a grey seal (hg46L-Z037) into shallow water during a pile-driving event, on 3 August 2014, the first Luchterduinen pile-driving event encountered by this seal. The map provides locations of the seal (connected by interpolated lines), the yellow oval approximates the seal's position during pile-driving. Note that the seals' movement inshore pre-empted the commencement of pile-driving. The graph indicates dive depth against time of day. Black lines show diving behaviour of the seal, typically from the surface to the bottom (which shallows over time from 22 m to <5 m after 6:30). The pink shading distinguishes stages during pile-driving, from a soft-start to continues hammering at high power. The numbering and colour scale on the 0 m (surface) line represent horizontal speed of the seal (in metres per second), which increases during the pile-driving.



Supplementary figure

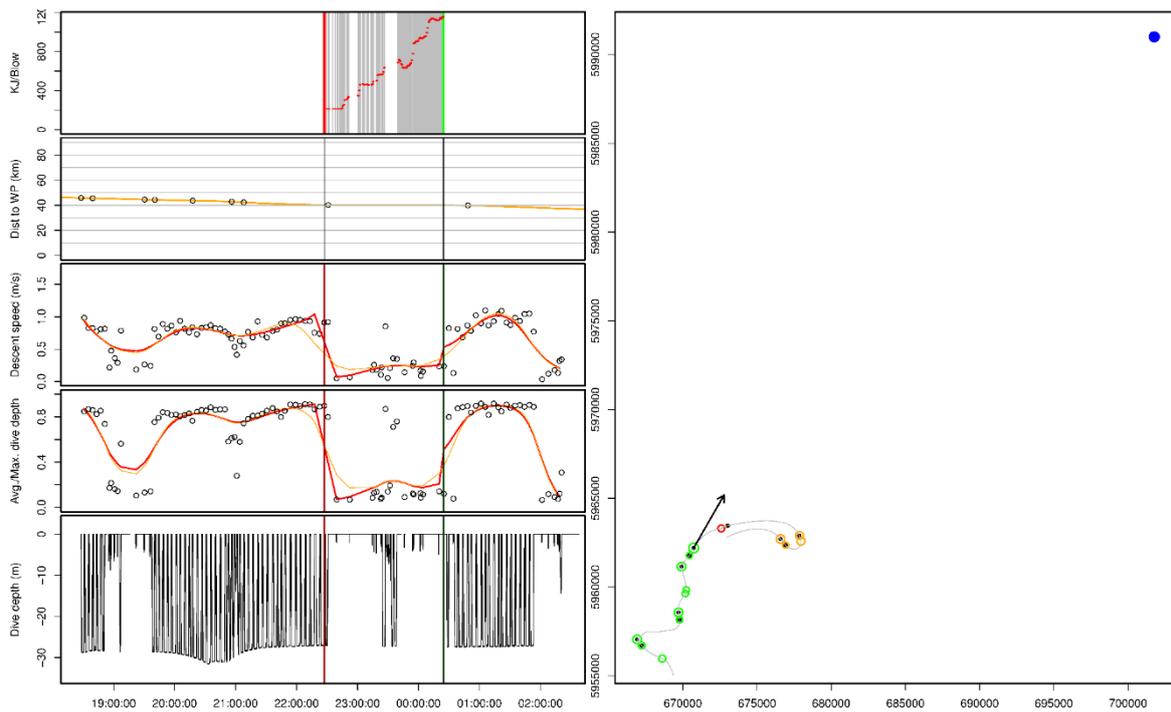


Figure 19. Example of a response to pile driving by a harbour seal (seal pv61-115-15, 40 km away on 24 September 2015 (monopole V4). Coinciding with commencement of pile-driving (top figure), the seals' diving behaviour became irregular (bottom figure left), descent speeds declined, and average dive depth (as fraction of the water depth) decreased, implying that seals stayed more time near the surface or mid-water, and less time near the bottom. After pile-driving ceased, more routine dive patterns resumed. The right figure indicates the movement of the seal in relation to pile-driving site (blue dot). The colours represent locations prior (green), during (red) and after (orange) pile-driving. The small black arrow is the expected seal location (based on previous direction and speed) after commencement of piling.

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Wageningen Marine Research is the Netherlands research institute established to provide the scientific support that is essential for developing policies and innovation in respect of the marine environment, fishery activities, aquaculture and the maritime sector.

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is specialised in the domain of healthy food and living environment.

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'To explore the potential of marine nature to improve the quality of life'

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