

# KOKKELEXPERIMENTEN IN POLDER WASSENAAR

EINDRAPPORTAGE

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## AANBEVELINGEN VOOR VERVOLGSTAPPEN

1. Voor het bepalen van de mogelijke netto-opbrengst van binnendijkse kokkelteelt dienen de dichtheden van de kokkels en grootte van de kokkelakkers te worden opgevoerd, verschillende oogstmethoden te worden vergeleken en alle kosten van inzetten, onderhoud en oogsten in kaart worden gebracht;
2. Dit onderzoek zou in principe in Polder Wassenaar kunnen worden uitgevoerd, mits de toevoer van zeewater wordt opgehoogd. Tijdens het experiment bedroeg de toevoer ongeveer 1400 m<sup>3</sup> per tij, bij een volledige bedekking van de bodems van alle stroomgoten (16 x 150 m<sup>2</sup> = 2400 m<sup>2</sup>) met een dichtheid van 500 volwassen kokkels per m<sup>2</sup> (met een filtratie capaciteit van 1.3 l uur<sup>-1</sup> per kokkel en een droogvalduur van 6 uur per tij) zal ongeveer 10.000 m<sup>3</sup> per tij zijn. Zonder droogvalduur is dit dan 20.000 m<sup>3</sup> per tij;
3. Voor een jaarlijks bevoorrading van de kokkelakkers zijn er minstens 1000 jonge kokkels m<sup>-2</sup> nodig. Bij een oppervlak van 10 hectare (een minimaal oppervlak voor een commercieel aantrekkelijke kokkelkweek) moet de aanvoer van jonge kokkels dan minimaal 100.000.000 exemplaren per jaar zijn. De Nederlandse regelgeving staat op dit moment een maximum oogst van 10 kg jonge kokkels (vers gewicht inclusief schelp) per persoon per dag toe, wat overeenkomt met ongeveer 3500 individuen. Per jaar zouden er dan ruim 28.000 emmers nodig zijn om die kokkelakkers (10 ha) te vullen. Het uitvoeren van (grootschalig vervolgonderzoek en opzetten van een commerciële kokkelkwekerij vraagt dus om een structurele oplossing van het verkrijgen van voldoende uitgangsmateriaal, waaronder de mogelijkheid om hiervoor de regels aan te passen;
4. De hoge natuurlijke sterfte van jonge kokkels bij hoge dichtheden biedt mogelijk ruimte om die hoeveelheid jonge kokkels te oogsten die anders verloren zou gaan als gevolg van natuurlijke sterfte anders dan die predatie door vogels. Het kan overwogen worden om in het veldonderzoek uit te voeren naar de effecten van uitdunning op groei en overleving van de overblijvende kokkels, en de gevolgen hiervan voor de predatie door schelpdier-etende vogels;
5. Het is in theorie denkbaar dat een kokkelkweek zelfvoorzienend wordt als de mogelijkheid bestaat om jonge larven in te vangen die met het buitenwater meekomen of de larven binnen te houden die door de volwassen kokkels in het systeem worden geproduceerd. Het kan overwogen worden om experimenten uit te voeren naar de omstandigheden die het invangen dan wel het binnenhouden van de kokkelarven;
6. Voor de mogelijke uitrol van testfase naar commerciële kweek kan overwogen worden om criteria op te stellen over randvoorwaarden (zoals waterkwaliteit, afstand binnendijkse gebieden tot de zee, getijdeamplitude, hoogteligging binnendijkse gebieden, maximaal oppervlak kokkelakkers) en vervolgens gekeken worden welke binnendijkse gebieden langs de randen van het wad aan deze criteria voldoen (of zouden kunnen voldoen met enige aanpassingen);
7. Op basis van de (goede) ervaringen tijdens het Polder Wassenaar project wordt aanbevolen om de vervolgstappen zoveel mogelijk uit te blijven voeren in samengestelde teams van (potentiele) schelpdierkwekers, onderzoekers en terreinbeheerders (particuliere organisaties of overheid). Voor de ondernemers is het wel belangrijk dat dan op korte termijn een serieus commercieel perspectief wordt geboden, zoals een bestuurlijk besluit over het uitgangsmateriaal, om aangehaakt te blijven.

# ECOLOGICAL ENGINEERING FOR OPTIMALISATION OF LAND-BASED MARICULTURE OF COASTAL SHELLFISH

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## SAMENVATTING

1. Terwijl de vraag naar gezond en duurzaam geproduceerd voedsel uit zee toeneemt, staan de opbrengsten van de zeevisserij onder toenemende druk als gevolg van overbevissing en klimaatverandering. Binnendijkse aquacultuur van mariene schelpdieren kan een alternatieve en duurzame praktijk zijn om aan de vraag naar zeevruchten te voldoen;
2. In Noordwest-Europa zou de eetbare kokkel *Cerastoderma edule* een goede kandidaat kunnen zijn voor duurzame kweek aan de kust bij bevordering van groei, overleving en opbrengsten met behulp van een kosteneffectieve en op de natuur gebaseerde technologie. Dit schelpdier leeft ondiep ingegraven in de zeebodem en vergaart daar voedsel (pelagische en benthische microalgen) door het water op het grensvlak van water een zeebodem te filteren;
3. Experimenten in een testlocatie aan de Nederlandse waddenkust bevestigden dat de groeiomstandigheden kunnen worden verbeterd door een adequaat ontwerp en slimme operationele procedures. Het verlengen van de verblijftijd van het zeewater resulteerde in een verhoging van de waterkwaliteit voor de kokkels door vermindering van de sedimentbelastingen en het stimuleren van extra groei van de pelagische microalgen (fytoplankton), vooral tijdens de voorjaarsbloei. Deze verbeteringen in de groeiomstandigheden resulteerden in een verbeterde schelpgroei en vleesgehalte van de testkokkels die in het systeem waren uitgezet;
4. Hoewel kortere verblijftijden van het water resulteerden in een toename van de benthische microalgen (microfytobenthos), maar een gelijktijdige toename van het slibgehalte van het sediment leek een effectieve opname van deze algen te belemmeren waardoor de kokkels uiteindelijk daar niet goed groeiden;
5. Uit de resultaten van de experimenten concluderen we dat de groeiomstandigheden (met betrekking tot water- en sedimentkwaliteit) voor het kweken van kokkels aan land inderdaad kunnen worden verbeterd door middel van ecologische waterbeheer. De groei en overleving van kokkels kan verder worden verbeterd door de kokkels onder extreme weersomstandigheden (ijs winters, hittegolven) onder water te houden en door een continue natuurlijke toevoer van hoogwaardige rekruten binnen de kokkelkwekerij te creëren;
6. Onze bevindingen suggereren dat de aquacultuur van mariene schelpdieren in kustgebieden een duurzame vorm van voedsel en inkomen voor kustgemeenschappen kan opleveren, met name wanneer de schelpdierkwekerijen zich bevinden in laaggelegen verzilte kustgebieden waar traditionele landbouwpraktijk niet langer winstgevend is.

## ABSTRACT

1. Whilst the demand for nutritious and sustainable seafood is increasing, yields of marine fisheries are under increasing pressure resulting from overfishing and climate change. Inshore aquaculture of marine molluscs might be an alternative and sustainable practice to fulfil the demand for seafood.
2. In north-western Europe, the edible cockle *Cerastoderma edule* may be a good candidate for sustainable inshore cultivation if cost-effective and nature-based technology enhances cockle growth and survival and subsequently the yield of cockle farms. This suspension-feeding bivalve uses pelagic and benthic microalgae as food from the water-sediment interface.
3. Experiments in an inshore testing site confirmed that growth conditions can be improved by an adequate design and operational procedures. Increasing the residence time of the seawater resulted in an increase of the water quality for the cockles by reducing the sediment loads and stimulating extra growth of the pelagic microalgae (phytoplankton) in particular during the spring bloom. These improvements in the growing conditions resulted in an enhanced shell growth and meat content of the cockles transplanted into the system.
4. Although shorter residence times resulted in an increase of the benthic microalgae (microphytobenthos), the concurrent increase in the silt content of the sediment appeared to hamper effective filtration and subsequently overruled the benefits of the higher supply of the additional food source.
5. From the results of the experiments, we conclude that growth conditions (with respect to water and sediment quality) for inshore cultivation of cockles can indeed be improved by means of ecological engineering. Cockle growth and survival can be further enhanced by keeping the cockles submerged during extreme weather conditions (ice winters, heat waves), and by creating a continuous natural supply of high-quality recruits within the cockle farm.
6. Our findings suggest that inshore aquaculture of marine shellfish can provide sustainable food and income for coastal communities, in particular when the shellfish farms are located in low-lying salinized coastal areas where common agriculture practices are no longer profitable.

## KEYWORDS

*Cerastoderma edule*, survival, growth, phytoplankton, microphytobenthos, median grain size, silt content, flushing rates

## INTRODUCTION

Providing healthy and nutritious food, in particular animal proteins, for a growing human population living in coastal areas is one of the main challenges the world is facing (FAO 2018). Sustained sea-level rise in a warming world has led to serious salinization in coastal lowlands, however, that is jeopardizing common agriculture practices (Rozema & Flowers 2008). In addition, marine fishing yields have declined due to overfishing and impacts of climate change (Watson et al. 2013, Free et al. 2019). These developments call for innovative and sustainable ways of (seafood) production to secure stable long-term food supply with minimum ecological impacts (Waite et al., 2014, Gentry et al. 2019).

Inshore aquaculture of marine molluscs might be an alternative practice to sustainably exploit low-lying salinized coastal areas and fulfil the demand for seafood at the same time. In order to be environmentally safe, farming should be targeting a species of local origin to prevent the spreading of invasive species (Naylor et al. 2001). From a commercial point of view, the species should occur in high densities and be fast-growing, be low in maintenance efforts and easy to market (Kumar et al. 2018). Furthermore, the farm should make use of natural forces (such as a natural supply of food and tides to flush the system) to keep the ecological and carbon footprint as well as the exploration costs as low as possible (Guillen et al. 2019). Last but not least, the target species should be selected on the basis of their capacity to grow under warming conditions (Oyinlola et al. 2019).

In north-western Europe, the edible cockle *Cerastoderma edule*, an ecologically and economically important bivalve species found along the north-eastern Atlantic coast, may be a good candidate for sustainable inshore

culturing. Within European bays and estuaries, the cockle (*Cerastoderma edule*) is one of the most abundant mollusc species with recorded densities of more than 10,000 per m<sup>2</sup> (Tyler-Walters 2007). This species has a 1 to 2 years generation time (Malham et al., 2012) and can potentially grow to a commercial size (shell > 25 mm; Ricardo et al. 2015) within 1 year (own observations). Cockles (*Cerastoderma edule*) provide meat and shell by-products with a potential value of €11.3M per year (Carss et al. 2020). With Senegal in western Africa as the southern limit of its geographical distribution area (Hayward & Ryland 1995), this species is likely to be able to withstand the impacts of warming in more northern areas.

Cockles are suspension-feeding bivalves that obtain food particles from the water-sediment interface. During filtration of suspended particulate matter (SPM), cockles reject inorganic material and ingest organic material with living microalgae being the most important food source for cockles (Modéran et al. 2012). This selection mechanism allows for food intake to remain constant at increasing SPM concentrations, until the concentration becomes so high that selection is no longer effective (Navarro & Widdows 1997). Cockles also feed on microphytobenthos (Christianen et al. 2017, Jung et al. 2019), which occurs mainly on tidal flats and thrives in shallow and clear waters (Asmus et al. 1998). If living in the proximity of river outflows, cockles also take advantage of the local supply of freshwater algae (Jung et al. 2019).

Cost-effective and nature-based technology may improve the growth of cockles and subsequently the yield of cockle farms. Increasing the residence time of seawater before supplying it to the cockles, for example, can reduce the silt content of the coastal waters and consequently improve the food uptake efficiency of the cockles. Pelagic microalgal biomass can increase up to 67% per day under optimal light and nutrient conditions (Ly et al. 2014). Such an increase in food density could be artificially enhanced by increasing water residence time. A decrease in turbidity of the water can also stimulate the benthic food supply for the cockles when the growth of microphytobenthos is light-limited. This could result in faster shell growth (mm per day) and higher meat content ( $\mu\text{g}$  AFDW per mm<sup>3</sup> shell volume) of the cockles, and a higher production capacity (number of cockles per m<sup>2</sup> per year; m<sup>2</sup> of cockle farmland per m<sup>3</sup> of water) within the system.

Here, we present the results of experiments where we examined the impact of flushing rate on environmental (growth) conditions and on cockle performance indices at an inshore testing site. This site was built in a former polder on Texel, the westernmost island in the Wadden Sea, which is a coastal sea adjacent to the North Sea. Seawater was let into the site by means of siphoning and transported through a series of basins (raceways) by means of a system of lock gates and valves. The locks and valves were used to control the flow through the raceways. This study addressed the following questions: (i) Does flushing rates affect the amount of pelagic and benthic microalgae as a food source for cockles? (ii) Does flushing rates affect the net sedimentation of suspended silt fraction of the water? (iii) If flushing rates influence the growth conditions, do they also modify cockle survival, shell growth and meat content?

## MATERIAL AND METHODS

### STUDY SITE & EXPERIMENTAL SET-UP

The experiments were carried out in a hydrodynamic field laboratory, constructed in the “Polder Wassenaar” area on Texel (Fig. 1). The experiments were performed in 8 (numbers 1, 3, 5, 7, 9, 11, 13 & 15) out of the 16 raceways of the aquaculture system, each was 30 m long, 5 m wide and 0.48 m deep (below Dutch Ordinance Level). With incoming tide, seawater flowed into the so-called high-tide buffer (HTB) via an underground supply (siphon) through the sea dyke and subsequently into the raceways via small sluices. With outgoing tide, the water flowed back into the Wadden Sea from the raceways via small sluices into the low-tide buffer (LTB) and then via the siphon. Valves within the siphon ensured that water was always flowing in the same direction. The raceways that were not used for the experiment (numbers 2, 4, 6, 8, 10, 12, 14 & 16) were permanently closed at both ends.

The flushing rate (m<sup>3</sup> per tide) in each raceway was projected by means of the hydrodynamic SOBEM model (Deltares, version 3.7.13), which was originally developed for predicting and controlling irrigation systems, river and channel flows, water levels and surface water quality (Stelling & Duijnmeier 2003, Prinsen & Becker 2011). Based on the dimensions of the aquaculture system and tidal amplitudes in the adjacent Wadden Sea, this

model calculated the amount of inflowing water by adjusting the size of the inflow and outflow opening of the sluices at both ends of each raceway. Model predictions were validated with data on tidal heights within the system. Flushing rates were set on 600, 450, 250 and 50 m<sup>3</sup> water per tide, with two replicates (raceways) for each flushing rate and situated next to each other. The opening of the sluices in the system was set and fixed from 6 November 2018 until the end of the experiments.

Each raceway contained 9 circular enclosures for cockles with an inner diameter of 48.2 cm, consisting of 7 cm high PVC pipe with a 15 cm high gauze (6.3x6.3 mm mesh) attached to it to contain the cockles. These enclosures were pushed into the sediment to a depth of 7 cm, leaving 8 cm of gauze above the sediment surface. Two parallel experiments were run, where enclosures being sampled for one experiment were filled with fresh cockles for the other experiment. "Experiment 1" had a common seeding date (12 December 2018) and three sampling dates (16 January, 24 April and 3 July 2019), while "Experiment 2" had three seeding dates (6 February, 24 April and 3 July 2019) and one common sampling date (13 August 2019) (Fig. 2).

For both experiments, each enclosure was seeded with 40 juvenile cockles from two separate stocks, which were originally obtained from nearby tidal flats in November 2018 (used for transplants on 12 December 2018 and 6 February 2019) and in April 2019 (used for transplants on 24 April and 3 July 2019). For Experiment 1, cockles that were found dead within the enclosures were removed and replaced by living ones from the stock on 14 December 2018. During the experimental period, from 12 December 2018 to 13 August 2019, cockles and environmental conditions were regularly sampled within the enclosures and in the high tide buffer (Table 1, see Sampling methodology for details).

The siphon was cleaned regularly (once every 2 to 3 weeks), removing fouling to ensure a constant water flow throughout the experiments. The inflow of seawater was halted approximately once a week for different reasons, including maintenance of the siphon, maintenance of the raceways, during extreme high tide (to prevent the system from overflowing) and during very low temperatures (to prevent the cockles from freezing). The interruptions lasted from a few hours to a maximum of 2 days per event. The sluices were closed during dredging of the high tide buffer (to remove the surplus of sediment) which prevented turbid waters from entering the raceways.

## SAMPLING METHODOLOGY

To determine phytoplankton concentrations, water samples were taken from the high-tide buffer in front of each of the sluices connecting the HTB with the individual raceways at the time when water started to enter raceway 15 (Table 1). Water samples were kept cool and dark until filtration. A subsample of 500 ml from each sample was filtered over a pre-heated GF / F filter (with a mesh size of 0.7 µm). The material remaining on the filter was then dissolved in 20 ml of acetone (90%), after which the concentration of chlorophyll-a in this solution was measured with a Fluorescence Spectrophotometer F-2500 Hitachi. The measured concentration in the acetone was then converted back to the concentration in the water (mg CHLa m<sup>-3</sup>).

Microphytobenthos concentrations and sediment characteristics were determined from sediment samples taken in the enclosures in January, April and July 2019 (Table 1). The top 4 cm of the sediment was sampled three times with a sampling tube (17 mm diameter) in each enclosure and these samples were jointly stored in a glass jar. Upon arrival at the laboratory, the samples were immediately lyophilized and stored in the dark until further analyses.

Microphytobenthos concentrations in the sediment samples were determined by adding 20 ml of acetone (90%) to 2 grams of the lyophilized sediment. After centrifugation (separating solids and liquids), the liquid (acetone containing the dissolved chlorophyll-a) was diluted the following day, after which the concentration of chlorophyll-a was measured in 0.02 ml of this dilution, using a Fluorescence Spectrophotometer F-2500 Hitachi (in µg chlorophyll-a per ml liquid). These concentrations in the solution were then converted back to the concentrations in the sediment (µg CHLa g<sup>-1</sup>).

A weighed portion of homogenized lyophilized sediment was put through a 2 mm sieve and placed in 13 ml PP Auto-sampler tubes for determination of other sediment characteristics. Purified water (RO water, purified by reverse osmosis) was added and the sample was shaken vigorously on a vortex mixer for 30 seconds. The

median particle size ( $\mu\text{m}$ ) and proportion of silt (fraction of particles smaller than  $63 \mu\text{m}$ ) of sediments were determined using a Coulter LS 13 320 particle size analyser and autosampler. This device measures particle sizes in the range of  $0.04\text{-}2,000 \mu\text{m}$  in 126 size classes, using laser diffraction ( $780 \text{ nm}$ ) and PIDS ( $450 \text{ nm}$ ,  $600 \text{ nm}$  and  $900 \text{ nm}$ ) technology.

Cockle performance indices (total number, average shell length, average shell volume, average meat content and total biomass) were determined by collecting, counting, measuring and weighing all surviving individuals from 3 enclosures in each raceway (Table 1). All remnants of dead cockles were removed. The volume of the shell (SV;  $\text{mm}^3$ ) was calculated as  $\pi / 6$  (length x width x height) (Savari et al. 1991). The meat was removed of a subset of individuals from each enclosure, and its ash-free dry weight (mg AFDW) was determined as the difference between dry weight (mg DW; 48 hours at  $60^\circ \text{C}$ ) and ash weight (mg AW; 5 hours at  $560^\circ \text{C}$ ). Cockle meat content (mg AFDW  $\text{mm}^{-3}$ ) was calculated as the ash-free dry weight of the meat (mg AFDW) divided by the shell volume ( $\text{mm}^3$ ). The total biomass per enclosure (mg AFDW) was calculated as the total shell volume ( $\text{mm}^3$  per enclosure) times the average cockle condition (mg AFDW  $\text{mm}^{-3}$ ).

## STATISTICAL TREATMENT AND ANALYSIS

First, we tested if the observations on the various environmental conditions and cockle performance indices could be best explained by hypotheses stating that:

- There is one similar seasonal pattern for all raceways (H1);
- There is one seasonal pattern, with an additional effect of raceway (H2);
- There is one seasonal pattern, with an additional effect of flushing rate (H3);
- There is one seasonal pattern, with additional effects of flushing rate and relative distance to inlet (H4);
- There are different seasonal patterns per raceway (H5);
- There are different seasonal patterns per flushing rate (H6);
- There are different seasonal patterns per flushing rate, with an additional effect of relative distance to inlet (H7).

Testing of the latter three hypotheses (H5-H7), including an interaction term between smoothers, was possible only for large enough data sets as available for phytoplankton (Table 2). To enable a full comparison, the data were analysed with all effects both as a factor and as a smoother. The analyses of the other variables (raceway, flushing rate, distance) were restricted to effects as factors only.

First, we modelled the data by means of a Generalised Additive Mixed Effects Model (GAMM). To compare models, we followed an Information Theoretic approach (Anderson and Burnham 2002) and calculated differences  $\Delta i$  between the Akaike Information Criterion (AIC) of each model and the minimum AIC. Anderson and Burnham (2002) state that the level of empirical support for model  $i$  is:

- substantial if  $\Delta i$  is between 0 and 2 (these are models with similar AICs as the optimal model),
- considerably less if  $\Delta i$  is between 4 and 7, and
- essentially none if  $\Delta i$  is larger than 10.

We also calculated Akaike weights  $w_i$  (Anderson & Burnham 2002), which have the convenient ability that they can be interpreted as probabilities that a given model is judged the best model on repeated sampling. A weight for a particular model with value of 0.75, for example, implies that this model has a probability of 75% of being the best model within the series of models tested.

Secondly, we compared environmental conditions with the cockle performance indices by analysing the correlations between the values of the coefficients for distance to inlet as derived from the best GAMM model assuming that seasonal patterns and distance to inlet are additive factorial effects (H2). The outcomes of this correlation matrix are summarized by means of principal component analyses (PCA), where the 1<sup>st</sup> principal component (PC1) is the direction along which the samples show the largest variation and the 2<sup>nd</sup> principal component (PC2) is the direction uncorrelated to the first component along which the samples show the largest variation (Bro & Smilde 2014).

Statistical analysis was done using R 3.6.2 version (R Core Team 2013) in a R Studio (1.2.5033) environment (R Studio Team 2019), with using the package *mgcv* (version 1.8-31) for generalized additive mixed modelling (Wood 2019) and the package *MuMIn* (version 1.43.17) for calculating the Akaike weights (Barton 2020).

## RESULTS

### PHYTOPLANKTON

The average phytoplankton biomass in the high-tide buffer (HTB) increased from  $3.20 \pm 3.20$  mg CHLa  $m^{-3}$  in early February 2019 to  $13.77 \pm 4.23$  mg CHLa  $m^{-3}$  during the spring bloom at the end of April 2019 (Fig. 3a). Subsequently, the biomass of the pelagic microalgae decreased to  $4.17 \pm 2.13$  mg CHLa  $m^{-3}$  in mid-May, followed by an increase to  $8.47 \pm 2.28$  mg CHLa  $m^{-3}$  at the end of May 2019, and was variable from then onwards until  $6.96 \pm 1.87$  mg CHLa  $m^{-3}$  at the end of the sampling period in early August 2019.

Variation in phytoplankton biomass during the experiment along the HTB was best described by means of a model that took the interaction between seasonal dynamics and raceway number into account (Table 3). Apparently, the spring bloom was enhanced (occurred earlier in the year) in the high tide buffer following the direction of the main current (Fig. 4). Results also suggested that the blooms at the end of May and in early August occurred earlier (and were higher) in front of the higher numbered raceways (Fig. 4; Appendix 1). These findings imply that the cockles that were transplanted in the downstream raceways profited earlier from the pelagic food supply than the cockles in the upstream raceways.

### MICROPHYTOBENTOS

Biomass of microphytobenthos, averaged over all raceways, generally increased from  $1.75 \pm 0.84$  mg CHLa  $g^{-1}$  in mid-December 2018 to  $9.57 \pm 6.67$  mg CHLa  $g^{-1}$  by the end of April 2019 (Fig. 3b). The benthic microalgal biomass was similarly high ( $9.33 \pm 6.62$  mg CHLa  $g^{-1}$ ) in early July 2019, but had decreased to  $6.45 \pm 4.24$  mg CHLa  $g^{-1}$  by mid-August 2019.

The values of the Akaike weights  $w_i$  indicated that model H3 (microphytobenthic biomass variation was due to added effects of seasonality and flushing time) had the highest probability (65%) of being the best model tested within the experiment, followed by model H4 (there was an additional effect of the relative position of the raceway towards the inlet of seawater) (31%) (Table 4). The results suggest that biomass of microphytobenthos increased with increasing flushing rate and that the upstream raceways have a higher benthic microalgal biomass than downstream ones with a similar flushing rate (Fig. 5a; Appendix 1).

### SEDIMENT CHARACTERISTICS

Median grain size (MGS) of the sediment, averaged over all raceways, decreased from  $223 \pm 15$   $\mu m$  in mid-December 2018 to  $191 \pm 36$   $\mu m$  at the end of April 2019, followed by an increase from  $192 \pm 34$   $\mu m$  in early July 2019 to  $204 \pm 19$   $\mu m$  in mid-August 2019 (Fig. 3). Variation in MGS was best explained by model H3 (having a probability of 71% of being the best model) and model H4 (with a probability of 24%; Table 4). Raceways with a flushing rate of 600  $m^3$  per tide had relatively low values of MGS compared to the other flushing rates (Fig. 5b).

The average silt fraction (SF) of the sediment increased from  $6.7 \pm 2.7$  % in mid-December 2018 to  $21.3 \pm 13.4$  % at the end of April 2019, followed by a decrease from  $20.1 \pm 11.8$  % in early July 2019 to  $16.0 \pm 8.5$  % in mid-August 2019 (Fig. 3). Variation in SF was best explained by model H4 (having a probability of 70% of being the best model; Table 4). SF appeared to increase with increasing flushing rates, and higher for upstream raceways than for downstream raceways of similar flushing rates (Fig. 5c; Appendix 1).

## COCKLES

### Juvenile stock

The shell length (mm), shell volume (mm<sup>3</sup>) and meat content (mg AFDW mm<sup>-3</sup>) of the first juvenile cockle stock used for first transplants was lower (ca. 12 mm, ca. 600 mm<sup>3</sup> and ca. 0.014 mg AFDW per mm<sup>3</sup>) than that of the second stock used (ca. 20 mm, ca. 2750 mm<sup>3</sup> and ca. 0.033 mg AFDW per mm<sup>3</sup>) (Fig. 6).

### Experiment 1

The number of cockles in the enclosures decreased over time from 12 December 2018 onwards, with a survival of  $47 \pm 17$  % for cockles sampled in January 2019, of  $21 \pm 12$  % for cockles sampled in April and  $13 \pm 10$  % for cockles sampled in July 2019 (Fig. 7a). Variation in density (number in enclosures) was best explained by model H2 (having a probability of 39% of being the best model) (Table 5). Survival in the raceways with a flushing rate of 450 m<sup>3</sup> per tide was higher (> 4%) compared to raceways with different flushing rates (Fig. 8a; Appendix 2).

The shell length of the cockles generally increased over time from  $12.78 \pm 0.62$  mm on 16 January 2019 to  $24.38 \pm 1.18$  mm in 3 July 2019 (Fig. 7b). Variation in shell length was best explained by model H3 (having a probability of 59% of being the best model), model H4 (17%) and model H2 (24%) (Table 5). Shell length in the raceways with a flushing rate of 600 m<sup>3</sup> per tide was lower (almost 1 mm) compared to raceways with different flushing rates (Fig. 8b; Appendix 2).

On average, shell volume increased from  $717 \pm 109$  mm<sup>3</sup> on 16 January 2019 to  $4691 \pm 675$  mm<sup>3</sup> in 3 July 2019 (Fig. 7c). Variation in shell volume was best explained by model H3 (having a probability of 54% of being the best model), model H4 (17%) and model H2 (28%) (Table 5). Shell volume in the raceways with a flushing rate of 600 m<sup>3</sup> per tide was smaller (almost 800 mm<sup>3</sup>) compared to raceways with different flushing rates (Fig. 8c; Appendix 2).

On average, cockle meat content increased from  $0.020 \pm 0.002$  mg AFDW mm<sup>-3</sup> on 16 January 2019 to  $0.044 \pm 0.003$  mg AFDW mm<sup>-3</sup> in 3 July 2019 (Fig. 7d). Variation in condition was best explained by model H3 (having a probability of 56% of being the best model) (Table 5). Cockle conditions in the raceways with a flushing rate of 250 m<sup>3</sup> and 450 m<sup>3</sup> per tide were significantly smaller (more than 0.003 mg AFDW mm<sup>-3</sup>) compared those in the raceways with flushing rates of 50 m<sup>3</sup> and 600 m<sup>3</sup> per tide (Fig. 8d; Appendix 2).

Total biomass of the cockles in the enclosures increased from  $0.26 \pm 0.10$  mg AFDW per enclosure on 16 January 2019 to  $1.21 \pm 0.90$  mg AFDW in 3 July 2019 (Fig. 7e). Variation in condition was best explained by model H2 (having a probability of 31% of being the best model) (Table 5). Cockle conditions in the raceways 5, 7 and 13 were smaller (> 0.6 mg AFDW) compared to those in raceway 1 (Fig. 8e; Appendix 2).

### Experiment 2

The number of surviving cockles sampled in August varied for the different seeding dates, with a survival of  $25 \pm 19$  % for cockles transplanted in February, of  $49 \pm 28$  % for cockles transplanted in April and of  $39 \pm 22$  % for cockles transplanted in July 2019 (Fig. 9a). Variation in density (number in enclosures) was best explained by model H3 (having a probability of 79% of being the best model) and model H4 (22%) (Table 5). Survival in the raceways appeared to decrease with increasing flushing rates, with an average decline in the number of cockles of more than 12 individuals per enclosure for the flushing rate of 600 m<sup>3</sup> per tide compared to surviving numbers in the enclosures with a flushing rate of 50 m<sup>3</sup> per tide (Fig. 10a; Appendix 3).

The average shell length of the cockles sampled in August was larger for the cockles transplanted in April ( $28.33 \pm 1.17$  mm) than those seeded in February ( $24.27 \pm 4.19$  mm) and July ( $23.41 \pm 1.04$  mm) (Fig. 9b). Variation in shell length was best explained by model H4 (having a probability of 53% of being the best model) and model H3 (47%) (Table 5). Shell length appeared to decrease with increasing flushing rates, with an average decline of almost 2 mm for the flushing rate of 600 m<sup>3</sup> per tide compared to those at a flushing rate of 50 m<sup>3</sup> per tide (Fig. 10b; Appendix 3).

Mean shell volume of the cockles sampled in August was higher for the cockles transplanted in April ( $7317 \pm 874 \text{ mm}^3$ ) than for those seeded in February ( $5059 \pm 1638 \text{ mm}^3$ ) and July ( $4285 \pm 617 \text{ mm}^3$ ) (Fig. 9). Variation in shell volume was best explained by model H3 (having a probability of 63% of being the best model) and model H4 (37%) (Table 5). Shell volume appeared to decrease with increasing flushing rates, with an average decline of more than  $1300 \text{ mm}^3$  for the flushing rate of  $600 \text{ m}^3$  per tide compared to those at flushing rate of  $50 \text{ m}^3$  per tide (Fig. 10c; Appendix 3).

On average, the meat content of the cockles as sampled in August was higher for the cockles transplanted in February ( $0.048 \pm 0.007 \text{ mg AFDW mm}^{-3}$ ) than for those seeded in April ( $0.046 \pm 0.005 \text{ mg AFDW mm}^{-3}$ ) and July ( $0.044 \pm 0.005 \text{ mg AFDW mm}^{-3}$ ) (Fig. 9). Variation in cockle condition was best explained by model H2 (having a probability of 100% of being the best model) (Table 5). Cockle meat content was relatively high in raceway 15 ( $+ 0.011 \text{ mg AFDW mm}^{-3}$ ) and relatively low in raceway 9 ( $- 0.004 \text{ mg AFDW mm}^{-3}$ ) compared to raceway 1 (Fig. 10d; Appendix 3).

The total biomass of the cockles sampled in August was higher for the cockles transplanted in April ( $6.94 \pm 4.57 \text{ mg AFDW}$ ) than those seeded in February ( $3.05 \pm 2.72 \text{ mg AFDW}$ ) and July ( $3.01 \pm 1.44 \text{ mg AFDW}$ ) (Fig. 9). Variation in total biomass was best explained by model H3 (having a probability of 65% of being the best model) and model H4 (35%) (Table 5). Total biomass appeared to decrease with increasing flushing rates, with an average decline of almost  $5 \text{ mg AFDW}$  for the flushing rate of  $600 \text{ m}^3$  per tide compared to those at flushing rate of  $50 \text{ m}^3$  per tide (Fig. 10e; Appendix 3).

## CORRELATIONS

Microphytobenthos concentrations ( $\text{mg AFDW g}^{-1}$ ) were strongly and positively correlated with the silt fraction (%) of the sediment (Table 6). With respect to cockle performance indices, it was found that (i) total biomass was strongly and positively correlated with density, (ii) shell volume ( $\text{mm}^3$ ) was strongly and positively correlated with shell length (mm), and (iii) shell volume ( $\text{mm}^3$ ) was positively correlated with total biomass ( $\text{mg AFDW}$ ) for both Experiment 1 and Experiment 2 (Table 6). In Experiment 2, shell length (mm) and shell volume ( $\text{mm}^3$ ) as well as shell length (mm) and meat content ( $\text{mg AFDW mm}^{-3}$ ) were also positively correlated (Table 6).

Cockle meat content ( $\text{g AFDW mm}^{-3}$ ) in the raceways was positively correlated with phytoplankton concentrations ( $\text{mg CHLa m}^{-3}$ ) at the entrance of the raceways in both experiments (Table 6). For Experiment 1, cockle shell length (mm) was positively correlated with median grain size ( $\mu\text{m}$ ) and negatively with silt fraction (%) and microphytobenthos concentration ( $\text{mg CHLa g}^{-1}$ ) of the sediment (Table 6). Here, cockle shell volume ( $\text{mm}^3$ ) and median grain size ( $\mu\text{m}$ ) were also positively correlated (Table 6). For Experiment 2, positive correlations were found between cockle shell length (mm) and phytoplankton concentrations ( $\text{mg CHLa m}^{-3}$ ). In this experiment, negative correlations were found between cockle volume ( $\text{mm}^3$ ) and silt fraction (%), between cockle biomass ( $\text{g AFDW}$ ) and microphytobenthos concentration ( $\mu\text{g CHLa g}^{-1}$ ), and between cockle biomass ( $\text{g AFDW}$ ) and silt fraction (%) of the sediment (Table 6).

## PRINCIPAL COMPONENT ANALYSES

In Experiment 1, the 1<sup>st</sup> and the 2<sup>nd</sup> axes of the PCs explained 51.0% and 33.5% of the total variance within the data set (Fig. 11). The loadings of PC1 strongly suggest a more or less gradual change in environmental conditions and cockle performance indices from RW1 to RW8 (Fig. 12). This gradient is particularly related to the variance in cockle shell length (mm), cockle shell volume ( $\text{mm}^3$ ) and median grain size ( $\mu\text{m}$ ) being relatively large in raceways with lower flushing rates, and the variance in silt fraction (%) and microphytobenthos concentrations ( $\mu\text{g AFDW g}^{-1}$ ) of the sediment which were both relatively high in raceways with higher flushing rates (Appendix 4). The loadings of PC2 indicate a distinction between RW3 and RW4 (both with a flushing rate of  $450 \text{ m}^3$  per tide) and the other raceways (Fig. 12), in particular due to the high cockle density (number per enclosure) and total cockle biomass ( $\text{g AFDW per enclosure}$ ) and the relatively low values in cockle meat content ( $\text{mg AFDW mm}^{-3}$ ) and phytoplankton concentrations ( $\text{mg CHLa m}^{-3}$ ) in these two raceways (Appendix 4).

In Experiment 2, the 1<sup>st</sup> and the 2<sup>nd</sup> axes of the PCs explained 64.0% and 16.5% of the total variance within the data set (Fig. 11). As for Experiment 1, the loadings of PC1 of Experiment 2 also indicated a gradual change in environmental conditions and cockle performance indices from RW1 to RW8 (Fig. 12). Here, this gradient is

particularly related to the variance in cockle shell length (mm), cockle shell volume (mm<sup>3</sup>) and total cockle biomass (g AFDW per enclosure) being relatively large at raceways with lower flushing rates, and the variance in silt fraction (%) of the sediment which was relatively high at raceways with higher flushing rates (Appendix 4). The loadings of PC2 indicate a distinction between RW5 (with a flushing rate of 250 m<sup>3</sup> per tide) and RW1 and RW8, with flushing rates of 600 and 50 m<sup>3</sup> per tide respectively (Fig. 13). This difference was particularly due to a relatively large median grain size (µm) and a relatively low cockle meat content (mg AFDW mm<sup>-3</sup>) in this raceway compared to the two others (Appendix 4).

## DISCUSSION

### ECOLOGICAL ENGINEERING OF ENVIRONMENTAL CONDITIONS

Results on environmental conditions strongly suggest that increasing the residence time of seawater before supplying it to the cockles resulted in sedimentation of the silt content of the incoming seawater and in strengthening and enhancing the spring bloom of the phytoplankton from mid-April (> 10 mg CHLa m<sup>-3</sup> in RW5, 9, 10, 11 and 13) to as early as the end of January (> 10 mg CHLa m<sup>-3</sup> in RW11 and 13). Because growth of pelagic microalgae is light-limited in early spring (Loebl et al. 2009), the enhanced timing of the spring bloom was probably the result of a decrease in the turbidity. Such an advanced bloom could then also take advantage of the high concentrations of nutrients after the winter (Tillman et al. 2000), subsequently resulting in a high productivity and high biomass of phytoplankton. We cannot exclude, however, that the increased height of the blooms during the rest of the growing season were at least partly due to additional nutrient supply from the polder sediments.

If inflowing silt concentrations in the water were similar in all raceways, the raceways with the high flushing rates (600 m<sup>3</sup> per tide) would experience 12 times higher silt import rates than the ones with the low flushing rates (50 m<sup>3</sup> per tide). However, based upon the observations on phytoplankton as well as on the distance to inlet on silt fraction in the raceways, silt concentrations in the water entering the raceways was lower as the raceways were located further downstream. This implies that the differences in net import (silt per tide) was even larger between the raceways with high (600 m<sup>3</sup> per tide) compared to the ones with low (50 m<sup>3</sup> per tide) flushing rates.

Net sedimentation of the silt content of the incoming seawater is the difference between the import and export of this material (Lumborg & Pejrup 2005). If there were only import and no export of silt, then the silt fraction would have been accumulating during the full experimental period. However, after an initial period of silt accumulation from December 2018 to April 2019, the silt fraction in the raceways remained more or less constant until July 2019 and subsequently decreased. This implies that silt export also must have occurred. This seasonal pattern in sediment composition resembles the natural seasonal dynamics on tidal flats where silt accumulates during spring and summer and then washes away due to wave-generated resuspension (Chang et al. 2006).

The silt content of the sediment was strongly and positively correlated with the concentrations of microphytobenthos. On tidal flats, sediment type and bathymetry are considered as the main factors determining the spatial variation in MPB biomass (Varela & Penas 1985, Brotas et al. 1995, Ubertini et al. 2012). The observed correlation with sediment type is generally explained by microphytobenthos growth, promoted by low dynamic energy as being reflected by high silt contents of the sediment (Orvain et al. 2012, Daggert et al. 2020). However, highest silt and microphytobenthos concentrations in the aquaculture system were observed in the raceways which experienced the highest (but still low) flushing rates. Most likely, here a positive feedback occurred here between net silt accumulation and diatom growth, due to relatively high concentrations of nutrients in silt rich sediment compared to more sandy sediments as was also observed for tidal flats (Daggert et al. 2020).

In summary, the combination of increased distance to tidal inlet (+ 75 m) and reduced flushing rates (from 600 to 50 m<sup>3</sup> per tide) resulted in higher spring blooms of phytoplankton (+ 3 mg CHLa m<sup>-3</sup>) occurring much earlier in the season (weeks to months), in reduced silt fraction of the sediment (- 13 %) and in lower standing stocks of microphytobenthos (- 5.8 µg CHLa g<sup>-1</sup>).

## IMPACTS ON COCKLE PERFORMANCE INDICES

The maximum number of cockles in the experiment was  $9 \times 40 = 360$  individuals per raceway, implying a maximum density of 2.4 individuals per  $\text{m}^2$ . With a minimum flushing rate of  $50 \text{ m}^3$  per tide and an average depth of 0.3 m, the water renewal rate was  $1.1 \text{ m}^3$  per  $\text{m}^2$  per tide. Assuming a pumping rate of an adult cockle of  $1.3 \text{ l h}^{-1}$  (Foster Smith 1975), 2.4 cockles  $\text{m}^{-2}$  would pump  $0.02 \text{ m}^3$  per tide which is less than 2 % of the water renewal rate. This means that food supply was not likely to be limiting during the experiments, and that observed differences in cockle performance were more likely due to the quality of the food and/or growth-restricting environmental conditions.

The survival was relatively high in the raceways with intermediate flushing rates ( $250 \text{ m}^3$  per tide in Experiment 1,  $450 \text{ m}^3$  per tide in Experiment 2), and unrelated to the gradients in distance to tidal inlet (raceways 1 to 8). Apparently, mortality was determined by a local factor which was not included in the sampling scheme of the environmental conditions. On average, survival was highest (68%) for the cockles that were transplanted at the end of April and sampled in mid-August 2019, and lowest (12%) for cockles transplanted mid-December 2018 and sampled in July 2019. For Experiment 2, cockle survival was apparently related to the quality of the stock used for these transplants. This implies that a good stocking material is crucial for acquiring high survival rates during inshore culturing.

For both experiments, shell length (mm), shell volume ( $\text{mm}^3$ ) and meat content ( $\mu\text{g AFDW mm}^3$ ) appeared to be promoted by high phytoplankton and hampered by high microphytobenthos concentrations. The positive correlation between cockle growth and phytoplankton concentrations is in line with previous findings from field observations and (transplant) experiments (Jensen 1992, Kamermans 1993). The growth reduction of cockles in raceways which were relatively rich in microphytobenthos is, however, in contrast with many other findings (Sauriau & Kang 2000). Most probably, the surplus of food sources is overruled by the restricted feeding due to the high silt contents of the sediment as was also observed for cockles in waters with high ( $> 40 \text{ g DW m}^{-3}$ ) silt concentrations (Prins et al. 1991) and sediments of tidal flats with a high silt fractions (Kraan et al. 2010, Compton et al. 2013). This suggests that the supply of seawater with a high phytoplankton concentration and a low silt content favours cockle growth, and that cockles cannot profit from a high concentration of benthic microalgae when these grow in silty sediments.

## FUTURE REQUIREMENTS AND PERSPECTIVES

The results of the experiments showed that growth conditions (with respect to water and sediment quality) for inshore cultivation of cockles can be improved by means of ecological engineering. The design and maintenance of an inshore culturing facility to optimize growth conditions will depend on the temperature and composition of the seawater (with respect to nutrients, phytoplankton and silt content), the size and meat content of the seeding material, the intended yield (number of cockles per year of commercial size) and the surface area of the cockle fields. Assuming a commercially interesting yield of 500 marketable cockles per  $\text{m}^2$  and an individual cockle pumping rate of  $1.3 \text{ l h}^{-1}$  (Foster Smith 1975) during 6 hours per tide, the renewal rate should be around  $4 \text{ m}^3$  per  $\text{m}^2$  per tide (being  $40.000 \text{ m}^3$  per hectare).

Furthermore, controlling the inflow and outflow of the seawater can aid in protecting cockles against unfavourable conditions. Keeping the cockles submerged during extreme low and high temperatures will prevent mass mortality as has been observed in the field during severe winters and heat waves (Malhalm et al. 2012). Restricting the inflow when the seawater is brackish (after downpours and subsequent strong river run-off; Peteiro et al. 2018) or very turbid (following severe storms in shallow coastal waters; Chen et al. 2018) will limit the energy spent by the cockles to withstand such conditions (Peteiro et al. 2018) and subsequently result in better growth and survival. When bordering marine waters with variable water quality during the tidal cycle (e.g., the Wadden Sea water during outgoing tide is much more turbid than during incoming tide) may further improve the yield of the inshore cockle farm.

The growth and survival of the cockles within the aquaculture system appeared to be strongly related to the origin of the juvenile cockles that were seeded, including the timing when they were put into the raceways. This implies that the final yield of a cockle farm will not only depend on the conditions created within the raceways but also on the quality of the starting material. Assuming a mortality of 50% during farming, the starting density

should be 1000 juveniles m<sup>-2</sup> (being 10.000.000 juveniles per hectare). Juvenile cockles can be derived from fisheries or from hatcheries. While sourcing juvenile cockles from hatcheries is possible (Pronker et al. 2013), larval mortality is very high and this practice might be too costly to be sustainable. Because recruitment success of natural stocks varies strongly from year to year (Kristensen 1957, Dörjes et al. 1986, Ducrotoy et al. 1991), supply from fisheries might be too unpredictable for the required annual yield.

Recruitment failure is considered to be predominantly due to temperature-related high predation rates on post-larvae (Beukema & Dekker 2005, 2020) and /or high adult densities inhibiting post-larval settlement (André & Rosenberg 1991, Beukema & Dekker 2015). If so, nurseries could be set up with bare sediments and devoid of predatory shrimps and crabs, allowing successful settlement of larvae released from well-conditioned adult cockle stocks. Because a female cockle may produce up to 700.000 eggs (Honkoop & van der Meer 1997), it should be possible in theory to make cockle farms self-sufficient by rotating spawning stock areas, nurseries and harvesting grounds. Further increase of the yield could then be accomplished by selecting cockles for the parent stock which show fast growth and a high survival rate within the environmental conditions of the aquaculture system (Langdon et al. 2003). Getting there will then be part of maximizing yields and minimizing (environmental) costs by selection of the best location, developing the best design and optimize the procedures to operate inshore marine shellfish farms.

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## REFERENCES

- André, C., & Rosenberg, R. (1991) Adult-larval interactions in the suspension-feeding bivalves *Cerastoderma edule* and *Mya arenaria*. *Marine Ecology Progress Series*, 71, 227-234.
- Barton, K., 2020. Package 'MuMIn' (April 15, 2020). <https://cran.rstudio.com/web/packages/MuMIn/MuMIn.pdf>
- Beukema, J.J. & Dekker, R. (2005) Decline of recruitment success in cockles and other bivalves in the Wadden Sea: possible role of climate change, predation on postlarvae and fisheries. *Marine Ecology Progress Series*, 287, 149–167.
- Beukema, J.J. & Dekker, R. (2015) Density dependence of growth and production in a Wadden Sea population of the cockle *Cerastoderma edule*. *Marine Ecology Progress Series*, 538, 157-167.
- Beukema, J.J. & Dekker, R. (2020) Winters not too cold, summers not too warm: long-term effects of climate change on the dynamics of a dominant species in the Wadden Sea: the cockle *Cerastoderma edule* L.. *Marine Biology*, 167, 44 <https://doi.org/10.1007/s00227-020-3659-1>
- Carss, D.N., Brito, A.C., Chainho, P., Ciutat, A., de Montaudouin, X., Fernández Otero, R.M., Filgueira, M.I., Garbutt, A., Goedknecht, M.A., Lynch, S.A., Mahony, K.E., Maire, O., Malham, S.K., Orvain, F., van der Schatte Olivier, A., & Jones, L. (2020) Ecosystem services provided by a non-cultured shellfish species: The common cockle *Cerastoderma edule*. *Marine Environmental Research*, 158:104931. DOI: 10.1016/j.marenvres.2020.104931
- Chang, T.S., Bartholoma, A. & Flemming, B.W. (2006) Seasonal dynamics of fine-grained sediments in a back-barrier tidal basin of the German Wadden Sea (southern North Sea). *Journal of Coastal Research*, 22, 328-338.
- Chen, N., Krom, M.D., Wu, Y., Yu, D. & Hong H. (2018) Storm induced estuarine turbidity maxima and controls on nutrient fluxes across river-estuary-coast continuum. *Science of the Total Environment*, 628-629, 1108-1120.

- Compton, T.J., Holthuijsen, S., Koolhaas, A., Dekinga, A., ten Horn, J., Smith, J., Galama, Y., Brugge, M., van der Wal, D., van der Meer, J., van der Veer, H.W. & Piersma, T. (2013) Distinctly variable mudscapes: Distribution gradients of intertidal macrofauna across the Dutch Wadden Sea. *Journal of Sea Research*, 82, 103-116.
- Christianen, M.J.A., Middelburg, J.J., Holthuijsen, S.J., Jouta, J., Compton, T.J., van der Heide, T., Piersma, T., Damste, J.S.S., van der Veer, H.W., Schouten, S. & Olf, H. (2017) Benthic primary producers are key to sustain the Wadden Sea food web: stable carbon isotope analysis at landscape scale. *Ecology*, 98, 1498–1512.
- FAO, 2018 (<http://www.fao.org/state-of-fisheries-aquaculture>).
- Free, C.M., Thorson, J.T., Pinsky, M.L., Oken, K.L., Wiedenmann, J., Jensen, O.P. (2019) Impacts of historical warming on marine fisheries production. *Science*, 363, 979-983.
- Gentry, R.R., Ruff, E.O. & Lester, S.E. (2019) Temporal patterns of adoption of mariculture innovation globally. *Nature Sustainability*, 2, 949-956.
- Guillen, J., Natale, F., Carvalho, N., Casey, J., Hofherr, J., Druon, J.-N., Fiore, G., Gibin, M., Zanzi, A. & Martinsohn, J.Th. (2019). Global seafood consumption footprint. *Ambio*, 48, 111-122.  
<https://doi.org/10.1007/s13280-018-1060-9>
- Hayward, P.J. & Ryland, J.S. (1995) Handbook of the marine fauna of north-west Europe. Oxford University Press, Oxford.
- Honkoop, P.J.C. & van der Meer, J. (1998) Experimentally induced effects of water temperature and immersion time on reproductive output of bivalves in the Wadden Sea. *Journal of Experimental Marine Biology and Ecology*, 220, 227–246.
- Jensen, K.T. (1992) Dynamics and growth of the cockle, *Cerastoderma edule*, on an intertidal mud-flat in the Danish Wadden Sea: effects of submersion time and density. *Netherlands Journal of Sea Research*, 28, 335-345.
- Kamermans, P. (1993) Food limitation in cockles (*Cerastoderma edule* (L.)): Influences of location on tidal flat and of nearby presence of mussel beds. *Journal of Sea Research*, 31, 71-81
- Kumar, G., Engle, C. & Tucker, C. (2018) Factors driving aquaculture technology adoption. *Journal of the World Aquaculture Society*, 49, 447-476.
- Langdon, C., Evans, F., Jacobson, D. & Blouin, M. (2003) Yields of cultured Pacific oysters *Crassostrea gigas* Thunberg improved after one generation of selection. *Aquaculture*, 220, 227-244.
- Loebl, M., Colijn, F., van Beusekom, J.E.E., Baretta-Bekker, J.G., Lancelot, C., Philippart, C.J.M., et al. (2009). Recent patterns in potential phytoplankton limitation along the Northwest European continental coast. *Journal of Sea Research*, 61, 34-43.
- Lumborg, U. & Pejrup, M. (2005) Modelling of cohesive sediment transport in a tidal lagoon - An annual budget. *Marine Geology*, 218, 1-16.
- Ly, J., Philippart, C.J.M. & Kromkamp, J.C. (2014) Phosphorus limitation during a phytoplankton spring bloom in the western Dutch Wadden Sea. *Journal of Sea Research*, 88, 109-140.
- Mariani, S., Piccari, F. & De Matthaeis, E. (2002) Shell morphology in *Cerastoderma* spp. (Bivalvia: Cardiidae) and its significance for adaptation to tidal and non-tidal coastal habitats. *Journal of the Marine Biological Association of the United Kingdom*, 82, 483-490.
- Naylor, R.L., Williams, S.L. & Strong, D.R. (2001) Aquaculture - A Gateway for Exotic Species. *Science*, 294, 1655-1656.
- Modéran, J., David, V., Bouvais, P., Richard, P. & Fichet, D. (2012) Organic matter exploitation in a highly turbid environment: planktonic food web in the Charente estuary, France. *Estuarine, Coastal and Shelf Science*, 98, 126-137.
- Navarro, J. & Widdows, J. (1997). Feeding physiology of *Cerastoderma edule* in response to a wide range of seston concentrations. *Marine Ecology Progress Series*, 152, 175-186.
- Peteiro, L.G., Woodin, S.A., Wetthey, D.S., Costas-Costas, D., Martínez-Casal, A., Olabarria, C. & Vázquez, E. (2018) Responses to salinity stress in bivalves: Evidence of ontogenetic changes in energetic physiology on *Cerastoderma edule*. *Nature Scientific Reports*, 8, 8329.
- Prins, T.C., Smaal, A.C. & Pouwer, A.J. (1991) Selective ingestion of phytoplankton by the bivalves *Mytilus edulis* L. and *Cerastoderma edule* (L.). *Hydrobiological Bulletin*, 25, 93-100.
- Prinsen, G.F. & Becker, B.P.J. (2011) Application of SOBEK hydraulic surface water models in the Netherlands Hydrological Modelling Instrument. *Irrigation and Drainage*, 60, 35-41.

- Pronker, A.E., Peene, F., Donner, S., Wijnhoven, S., Geijssen, P., Bossier, P. & Nevejan, N.M. (2015) Hatchery cultivation of the common cockle (*Cerastoderma edule* L.): from conditioning to grow-out. *Aquaculture Research*, 46, 302–312.
- R Core Team (2013) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL <http://www.R-project.org/>.
- RStudio Team (2019) RStudio: Integrated Development for R. RStudio, PBC, Boston, MA URL <http://www.rstudio.com/>.
- Ricardo, F., Génio, L., Costa Leal, M., Albuquerque, R., Queiroga, H., Rosa, R. & Calado, R. (2015) Trace element fingerprinting of cockle (*Cerastoderma edule*) shells can reveal harvesting location in adjacent areas. *Nature Scientific Reports*, 5, 11932.
- Rozema, J. & Flowers, T. (2008) Crops for a salinized world. *Science*, 322, 1478-1480.
- Sauriau, P.-G. & Kang, C.-K. (2000) Stable isotope evidence of benthic microalgae-based growth and secondary production in the suspension feeder *Cerastoderma edule* (Mollusca, Bivalvia) in the Marennes-Oléron Bay. *Hydrobiologia*, 440, 317-329.
- Stelling, G.S. & Duijnmeijer, S.P.A. (2003) A staggered conservative scheme for every Froude number in rapidly varied shallow water flows. *International Journal Numerical Methods in Fluids*, 43, 1329-1354.
- Tillmann, U., Hesse, K.-J. & Colijn, F. (2000) Planktonic primary production in the German Wadden Sea. *Journal of Plankton Research*, 22, 1253-1276
- Waite, R., Beveridge, M., Brummett, R., Chaiyawannakarn, N., Kaushik, S., Mungkung, R., Nawapakpilai, S. & Phillips, M. (2014) Improving productivity and environmental performance of aquaculture. World Resource Institute.
- Wood, S.N., 2019. Package 'mgcv' (November 9, 2019) <https://cran.rstudio.com/web/packages/mgcv/mgcv.pdf>

**Table 1.** Number of samples taken during the experiment in or just before the raceways. For phytoplankton (PHYT), one sample per raceway was taken (just outside the entrance of the water flowing into the raceway). For microphytobenthos (MPB) and sediment characteristics (SED), three enclosures per raceway were sampled. At the start of the experiment ( $t_0$ ), samples for microphytobenthos and sediment characteristics were taken from all enclosures (9 per raceway). For Experiment 1, in total 772 living cockles were sampled from the enclosures, which is almost 27% of the maximum number if all cockles in this Experiment had survived during the experiment (being  $72 \times 40 = 2880$  individuals). For Experiment 2, 1091 living cockles were sampled from the enclosures, which is almost 38% of the maximum number if all cockles in this Experiment had survived during the experiment.

Date	Day nr	Environmental (growth) conditions			Cockle performance indices			
		PHYT	MPB	SED	Experiment 1		Experiment 2	
		sampled	sampled	sampled	seeded	sampled	seeded	sampled
2018-12-12	0		72	72	72			
2019-01-16	35		24	24		24		
2019-01-22	41	8						
2019-02-05	55	8						
2019-02-06	56						24	
2019-02-19	69	8						
2019-03-05	86	8						
2019-03-19	100	8						
2019-04-02	113	8						
2019-04-16	127	8						
2019-04-24	133		24	24		24	24	
2019-04-30	139	8						
2019-05-14	153	8						
2019-05-28	167	8						
2019-06-11	181	8						
2019-06-25	195	8						
2019-07-03	203		24	24		24	24	
2019-07-09	209	8						
2019-07-23	223	8						
2019-08-06	238	8						
2019-08-13	245		72	72				72
<b>SUM</b>		<b>120</b>	<b>216</b>	<b>216</b>	<b>72</b>	<b>72</b>	<b>72</b>	<b>72</b>

**Table 2.** Hypotheses and statistical models for the variation in environmental conditions and cockle performance indices during the experiment from 12 December 2018 to 1 August 2019. Seasonality (*i*) is based upon 15 (for phytoplankton) and 3 (for all other variables) sampling dates. The raceways (*j*) were numbered from 1 to 8, the flushing times (*k*) were 50, 250, 450 and 600 m<sup>3</sup> per tide. Models with smoothers could be applied for the data on phytoplankton concentrations only.

#	Hypothesis	Statistical model			
		Factors		Smoothers	
H1	There is one similar seasonal pattern for all raceways	H1f	$y_i = \beta_i + \text{SamplingDate}_i + E_i$	H1s	$y_i = \beta_i + s_i(\text{SamplingDate}) + E_i$
H2	There is one seasonal pattern, with an additional effect of raceway	H2f	$y_{ij} = \beta_i + \text{SamplingDate}_i + \text{RaceWay}_j + E_{ij}$	H2s	$y_i = \beta_i + s_i(\text{SamplingDate}) + \text{RaceWay}_j + E_{ij}$
H3	There is one seasonal pattern, with an additional effect of flushing time	H3f	$y_{ij} = \beta_i + \text{SamplingDate}_i + \text{FlushTime}_k + E_{ik}$	H3s	$y_i = \beta_i + s_i(\text{SamplingDate}) + \text{FlushTime}_k + E_{ik}$
H4	There is one seasonal pattern, with additional effects of flushing time and relative distance to inlet	H4f	$y_i = \beta_i + \text{SamplingDate}_i + \text{FlushTime}_k + \text{RelDist}_l + E_{ikl}$	H4s	$y_i = \beta_i + s_i(\text{SamplingDate}) + \text{FlushTime}_k + \text{RelDist}_l + E_{ikl}$
H5	There are different seasonal patterns per raceway			H5s	$y_i = \beta_i + s_{ij}(\text{SamplingDate}, \text{RaceWay}) + E_{ij}$
H6	There are different seasonal patterns per flushing time			H6s	$y_i = \beta_i + s_{ik}(\text{SamplingDate}, \text{FlushTime}) + E_{ik}$
H7	There are different seasonal patterns per flushing time with an additional effect of relative distance to inlet			H7s	$y_i = \beta_i + s_{ik}(\text{SamplingDate}, \text{FlushTime}) + \text{RelDist}_l + E_{ikl}$

**Table 3.** Values and differences of the Akaike Information Criterion (AIC), and the Akaike weight ( $w_i$ ) for statistical models 1–7 in Table 2 for phytoplankton biomass. Models for which the level of empirical support for model  $i$  is substantial (i.e. the value of  $\Delta_i$  is between 0 and 2) are underlined, the best fit (for which AIC has the lowest value) is printed in bold.

Model	Type	Microalgal biomass		
		Phytoplankton		
		AIC	$\Delta_i$	$w_i$
H1	Factors	643.74	1.11	0.000
	Smoothers	649.12	1.12	0.000
H2	Factors	623.41	1.07	0.000
	Smoothers	633.68	1.09	0.000
H3	Factors	628.88	1.08	0.000
	Smoothers	637.09	1.10	0.000
H4	Factors	627.57	1.08	0.000
	Smoothers	636.27	1.09	0.000
H5	Smoothers	<b>581.42</b>	1.00	<b>1.000</b>
H6	Smoothers	658.85	1.13	0.000
H7	Smoothers	658.10	1.13	0.000

**Table 4.** Values and differences of the Akaike Information Criterion (AIC), and the Akaike weight ( $w_i$ ) for statistical models 1–7 in Table 2 for sediment characteristics and microphytobenthic biomass. Models for which the level of empirical support for model  $i$  is substantial (i.e. the value of  $\Delta i$  is between 0 and 2) are underlined, the best fit (for which AIC has the lowest value) is printed in bold. Cells with best models have a blue colour.

Model	Sediment characteristics						Microalgal biomass		
	Median Grain Size			Silt Fraction			Microphytobenthos		
	Factors			Factors			Factors		
	AIC	$\Delta i$	$w_i$	AIC	$\Delta i$	$w_i$	AIC	$\Delta i$	$w_i$
H1	1962.79	35.29	0.000	1520.57	101.17	0.000	1220.46	89.79	0.000
H2	1931.82	4.32	0.051	1424.18	4.78	0.044	1135.13	4.46	0.044
H3	<b>1927.50</b>	<b>0.00</b>	<b>0.708</b>	1421.62	2.22	0.255	<b>1130.67</b>	<b>0.00</b>	<b>0.651</b>
H4	<u>1928.52</u>	<u>1.02</u>	<u>0.240</u>	<b>1419.40</b>	<b>0.00</b>	<b>0.701</b>	<u>1131.99</u>	<u>1.32</u>	<u>0.305</u>

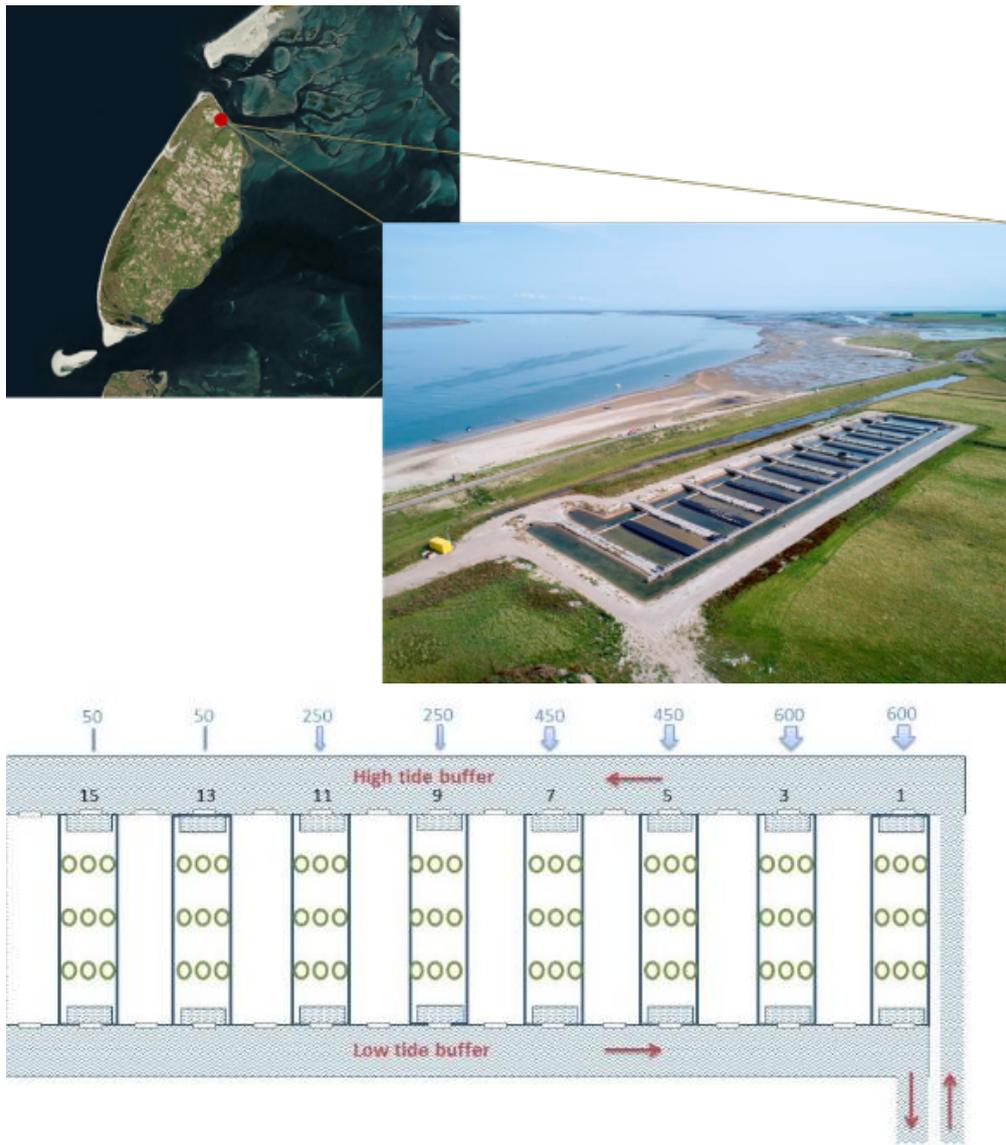
**Table 5.** Values and differences of the Akaike Information Criterion (AIC), and the Akaike weight ( $w_i$ ) for statistical models 1–7 in Table 2 for cockle performance indices. Models for which the level of empirical support for model  $i$  is substantial (i.e. the value of  $\Delta_i$  is between 0 and 2) are underlined, the best fit (for which AIC has the lowest value) is printed in bold. Cells with best models have a blue colour.

Index	Model	Experiment 1			Experiment 2		
		AIC	$\Delta_i$	$w_i$	AIC	$\Delta_i$	$w_i$
Density	H1	449.40	15.28	0.001	530.03	54.91	0.000
	H2	<b>434.12</b>	<b>0.00</b>	<b>0.391</b>	503.74	28.62	0.000
	H3	436.40	2.28	0.472	<b>475.12</b>	<b>0.00</b>	<b>0.785</b>
	H4	438.35	4.23	0.136	<u>477.12</u>	<u>2.00</u>	<u>0.215</u>
Shell length	H1	177.73	3.06	0.240	322.47	60.81	0.000
	H2	182.18	7.51	0.003	325.66	64.00	0.000
	H3	<b>174.67</b>	<b>0.00</b>	<b>0.591</b>	<u>262.54</u>	<u>0.88</u>	<u>0.470</u>
	H4	<u>176.64</u>	<u>1.97</u>	<u>0.165</u>	<b>261.66</b>	<b>0.00</b>	<b>0.530</b>
Shell volume	H1	1228.58	2.47	0.280	1266.48	108.61	0.000
	H2	1231.78	5.67	0.008	1267.59	109.72	0.000
	H3	<b>1226.11</b>	<b>0.00</b>	<b>0.541</b>	<b>1157.87</b>	<b>0.00</b>	<b>0.627</b>
	H4	<u>1227.90</u>	<u>1.79</u>	<u>0.170</u>	<u>1158.32</u>	<u>0.45</u>	<u>0.373</u>
Meat content	H1	-199.63	3.49	0.221	-174.11	43.85	0.000
	H2	<u>-202.80</u>	<u>0.32</u>	<u>0.070</u>	<b>-217.96</b>	<b>0.00</b>	<b>1.000</b>
	H3	<b>-203.12</b>	<b>0.00</b>	<b>0.564</b>	-179.86	38.10	0.000
	H4	<u>-201.18</u>	<u>1.94</u>	<u>0.145</u>	-193.06	24.90	0.000
Total biomass	H1	466.98	9.26	0.020	708.37	77.99	0.000
	H2	<b>457.72</b>	<b>0.00</b>	<b>0.313</b>	687.65	57.27	0.000
	H3	460.19	2.47	0.342	<b>630.38</b>	<b>0.00</b>	<b>0.651</b>
	H4	459.77	2.05	0.324	<u>631.03</u>	<u>0.65</u>	<u>0.349</u>

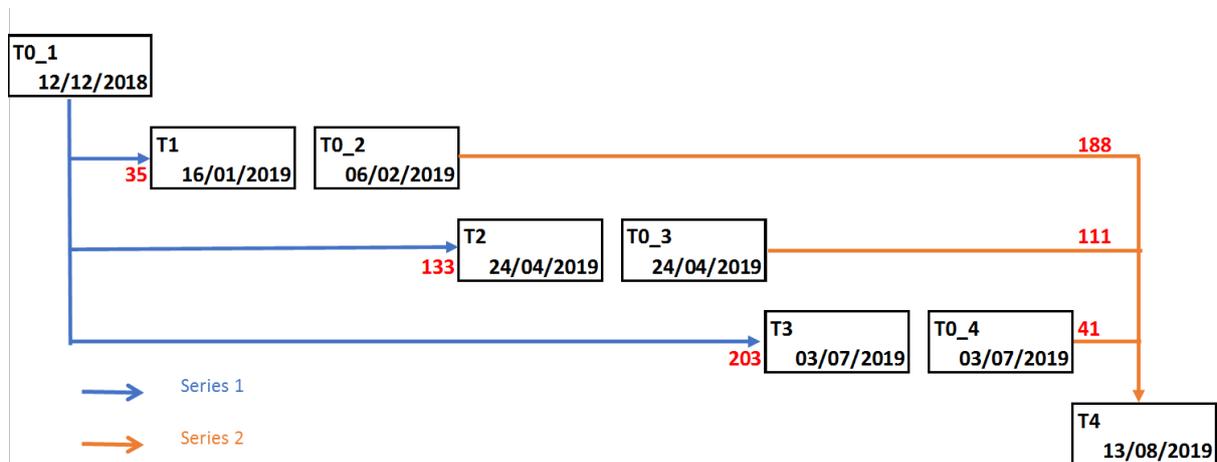
**Table 6.** Correlations and significance of correlations between environmental condition and cockle performance indices. Significant correlations ( $p < 0.05$ ) are printed in bold, correlations with  $p$ -values  $< 0.1$  are underlined. To aid comparison between Experiment, cells with positive correlations with  $p$ -values smaller than 0.1 are coloured blue and cells with negative correlations with  $p$ -values smaller than 0.1 are coloured orange. Significance values were not corrected for multiple comparisons.

Group	Variable 1	Variable 2	Experiment 1		Experiment 2	
			R	p	R	p
Environmental conditions	Phytoplankton	Microphytobenthos	-0.479	0.229	Similar to Experiment 1	
		Median grain size	0.239	0.569		
		Silt fraction	<u>-0.643</u>	<u>0.085</u>		
	Microphytobenthos	Median grain size	-0.604	0.113		
		Silt fraction	<b>0.960</b>	<b>0.000</b>		
Median grain size	Silt fraction	-0.600	0.116			
Cockle performance indices	Density	Shell length	0.312	0.452	0.544	0.164
		Shell volume	<b>0.714</b>	<b>0.047</b>	<u>0.675</u>	<u>0.066</u>
		Condition	-0.397	0.330	0.268	0.520
		Total biomass	<b>0.945</b>	<b>0.000</b>	<b>0.878</b>	<b>0.004</b>
	Shell length	Shell volume	<b>0.743</b>	<b>0.035</b>	<b>0.877</b>	<b>0.004</b>
		Condition	0.481	0.228	<b>0.813</b>	<b>0.014</b>
		Total biomass	0.382	0.350	<b>0.791</b>	<b>0.019</b>
	Shell volume	Condition	-0.132	0.755	0.521	0.186
		Total biomass	<b>0.747</b>	<b>0.033</b>	<b>0.760</b>	<b>0.029</b>
	Condition	Total biomass	-0.278	0.505	0.596	0.119
Cross correlations	Cockle density	Phytoplankton	-0.327	0.429	0.233	0.579
		Microphytobenthos	0.014	0.973	<u>-0.641</u>	<u>0.087</u>
		Median grain size	0.474	0.236	0.288	0.489
		Silt fraction	-0.046	0.913	<u>-0.691</u>	<u>0.058</u>
	Cockle length	Phytoplankton	<u>0.633</u>	<u>0.092</u>	<b>0.730</b>	<b>0.040</b>
		Microphytobenthos	<b>-0.714</b>	<b>0.047</b>	-0.493	0.215
		Median grain size	<b>0.810</b>	<b>0.015</b>	0.457	0.255
		Silt fraction	<b>-0.804</b>	<b>0.016</b>	<u>-0.696</u>	<u>0.055</u>
	Cockle volume	Phytoplankton	0.062	0.883	0.495	0.212
		Microphytobenthos	<u>-0.646</u>	<u>0.084</u>	-0.620	0.101
		Median grain size	<b>0.843</b>	<b>0.009</b>	<u>0.658</u>	<u>0.076</u>
		Silt fraction	<u>-0.648</u>	<u>0.082</u>	<b>-0.760</b>	<b>0.029</b>
	Cockle condition	Phytoplankton	<b>0.931</b>	<b>0.001</b>	<b>0.719</b>	<b>0.045</b>
		Microphytobenthos	-0.207	0.622	-0.172	0.685
		Median grain size	0.039	0.928	-0.023	0.957
		Silt fraction	-0.392	0.337	-0.418	0.303
	Cockle biomass	Phytoplankton	-0.277	0.506	<u>0.663</u>	<u>0.073</u>
		Microphytobenthos	-0.016	0.969	<b>-0.710</b>	<b>0.049</b>
		Median grain size	0.471	0.238	0.293	0.481
		Silt fraction	-0.071	0.868	<b>-0.834</b>	<b>0.010</b>

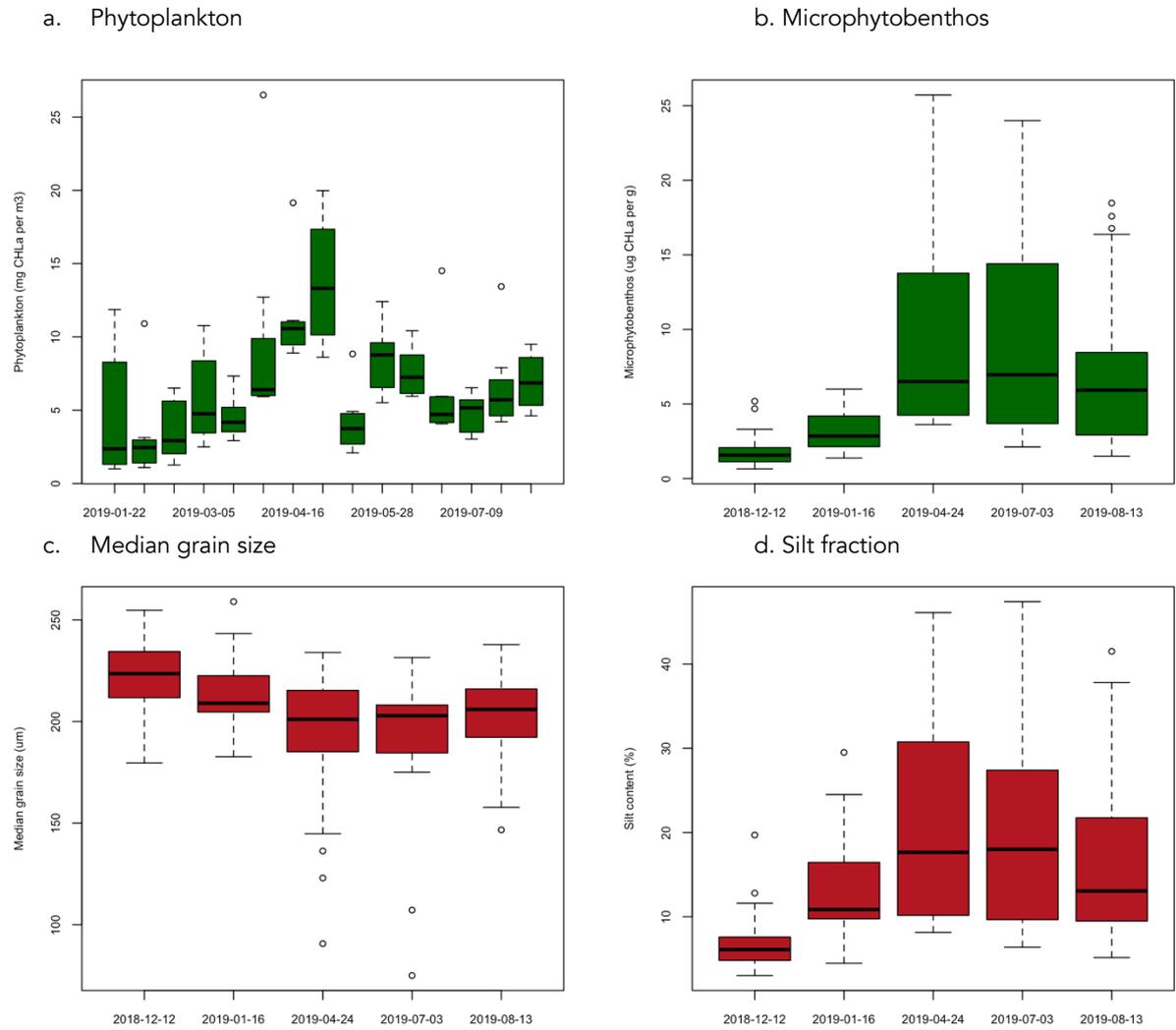
Figure 1. Location and dimensions of the experimental set-up, and location of sampling points.



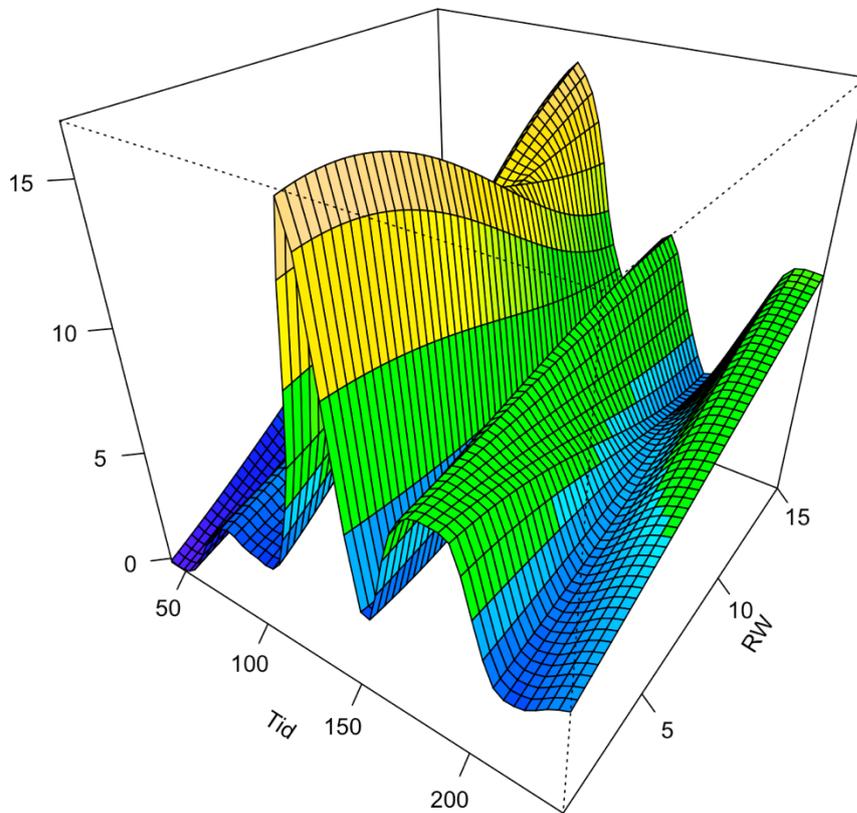
**Figure 2.** Sampling scheme for cockles divided into two experiments. For Experiment 1, the cockles were all transplanted into the enclosures on 12 December 2018 (in 9 enclosures per raceway, 72 enclosures in total), and sampled on 16 January, 24 April and 3 July 2019 (from 3 enclosures per raceway, being 24 enclosures per sampling period). For Experiment 2, the cockles were transplanted on 6 February, 24 April and 3 July 2019 (in 3 enclosures per raceway, being 24 enclosures per sampling period) and all sampled on 13 August 2019 (72 enclosures in total).



**Figure 3.** Environmental conditions at various sampling periods during the experiment from 12 December 2018 (t<sub>0</sub>) up to 13 August 2019. Phytoplankton concentrations (upper-left panel) were sampled in the high tide buffer directly in front of the sluices where the seawater entered the raceways. Microphytobenthos (upper-right) and sediment characteristics (bottom panels) were sampled in the enclosures in the raceways (see [Table 1](#) for the number of samples per sampling period).

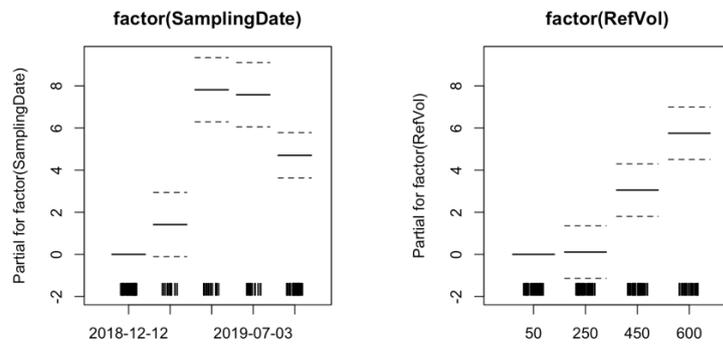


**Figure 4.** Estimated smoother for phytoplankton concentrations ( $\text{mg CHLa m}^{-3}$ ) at the entrances of the raceways between 22 January 2019 (day 41) and 6 August 2019 (day 238) obtained by Generalized Additive Mixed Modelling (GAMM) with a 2-dimensional smoother (see model H5 in Table 2). The vertical axis shows the fitted values ( $\text{mg CHLa m}^{-3}$ ) and the horizontal two axes the day number of the experiment (Tid; with values between 41 and 238) and the raceway numbers (from RW1 to RW8). Deviance explained is 74%.

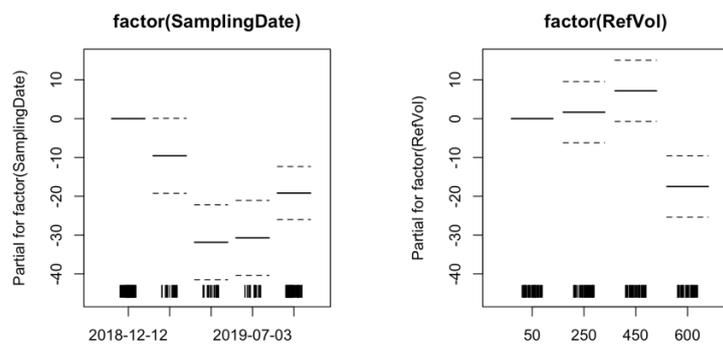


**Figure 5.** Coefficients of the best model fits on microphytobenthos concentrations ( $\mu\text{g CHLa g}^{-1}$ ), median grain size ( $\mu\text{m}$ ) and silt fraction (%) of the sediment as sampled in the enclosures within the raceways between 12 December 2018 and 13 August 2019, obtained by Generalized Additive Mixed Modelling (GAMM), for model H3 (microphytobenthos and median grain size) and model H4 (silt fraction) (see [Table 2](#) for description of the models).

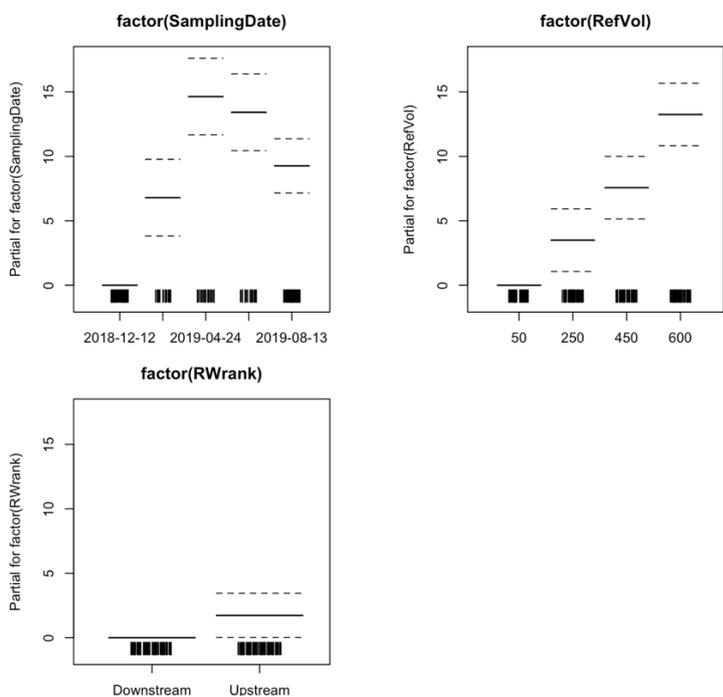
a. Microphytobenthos ( $\mu\text{g CHLa g}^{-1}$ ), intercept is  $-0.48 \mu\text{g CHLa g}^{-1}$ , deviance explained is 59.1%.



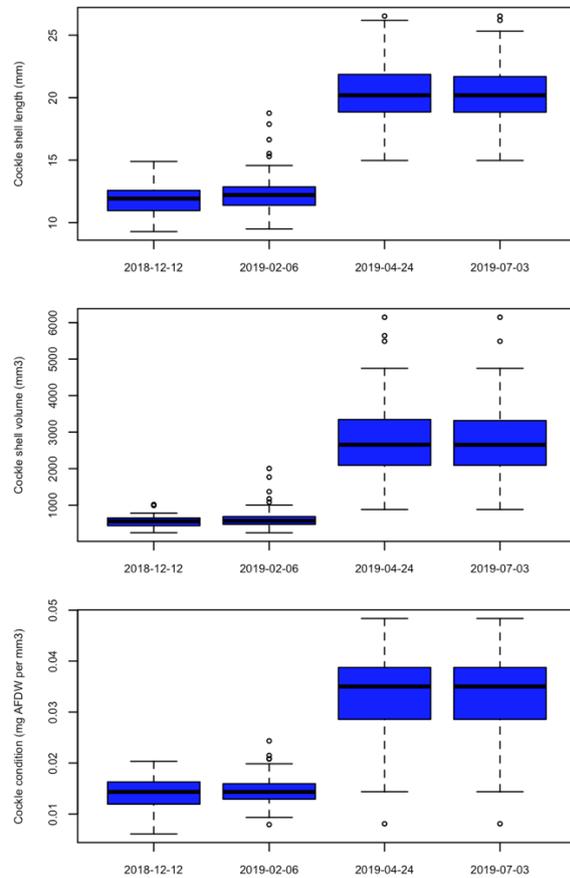
b. Median grain size ( $\mu\text{m}$ ), intercept is  $225 \mu\text{m}$ , deviance explained is 36%.



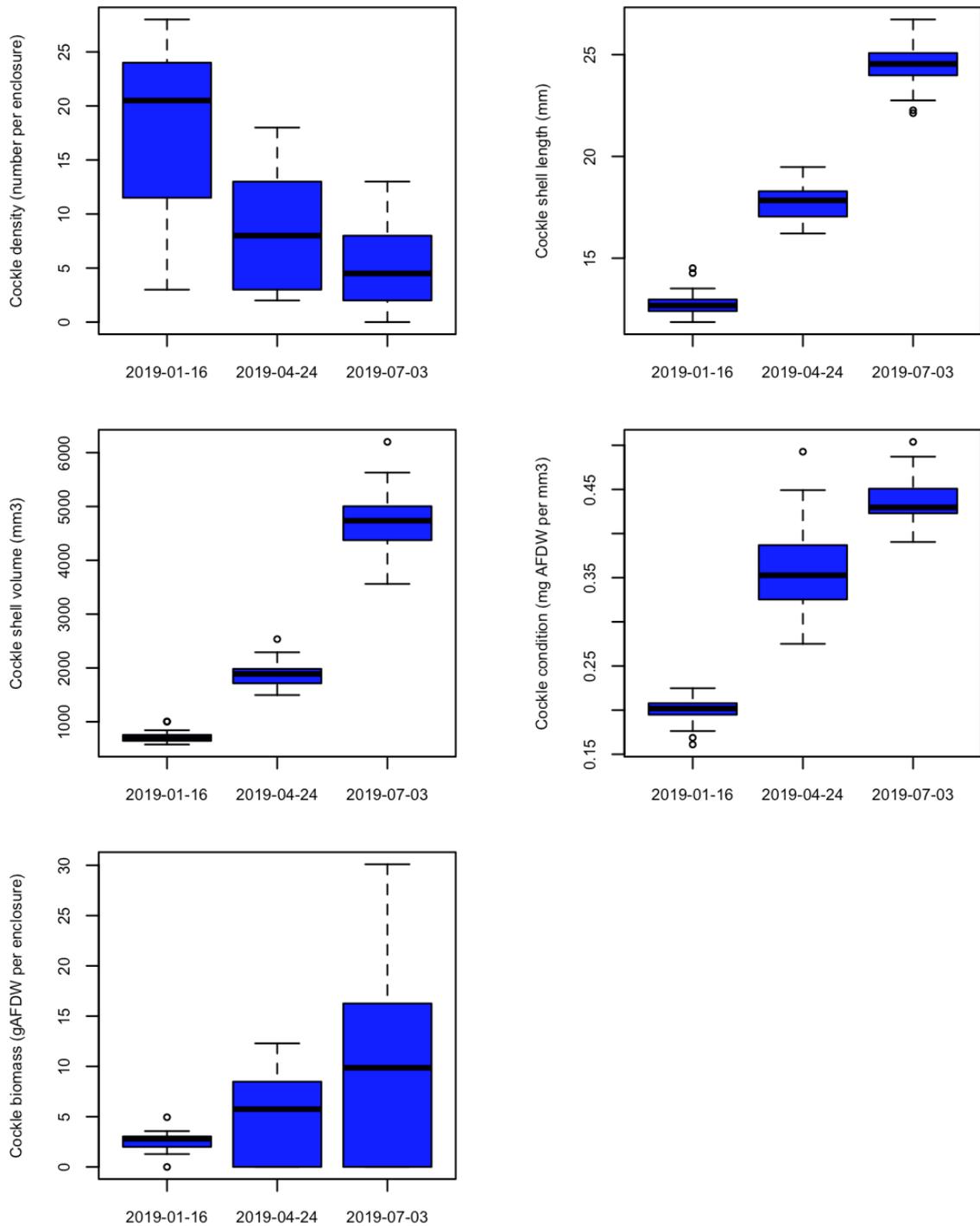
c. Silt fraction (%), intercept is  $-0.24\%$ , deviance explained is 58.7%.



**Figure 6.** Shell length (mm), shell volume (mm<sup>3</sup>) and condition (mg AFDW mm<sup>-3</sup>) of a subsample of cockles taken from the stock used for the transplants on 12 December 2018 (common t<sub>0</sub> for Experiment 1) and on 2 February 2019, 24 April 2019 and 3 July 2019 (respective t<sub>0</sub> for Experiment 2). The number of subsampled cockles for shell allometry ranged between 36 (December 2018) and 120 (February 2019) individuals, that for cockle condition between 27 and 111 individuals at the same dates.

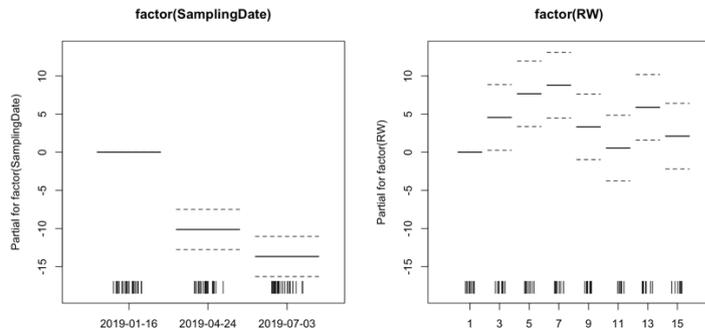


**Figure 7.** Cockle performance indices for Experiment 1, where all cockles were transplanted into the enclosures on 12 December 2018 (in 9 enclosures per raceway, 72 enclosures in total) and sampled on 16 January 2019, 24 April 2019 and 3 July 2019 (from 3 enclosures per raceway, being 24 enclosures per sampling period). Dates on the x-axis are, therefore, the dates when the cockles were sampled from the enclosures.

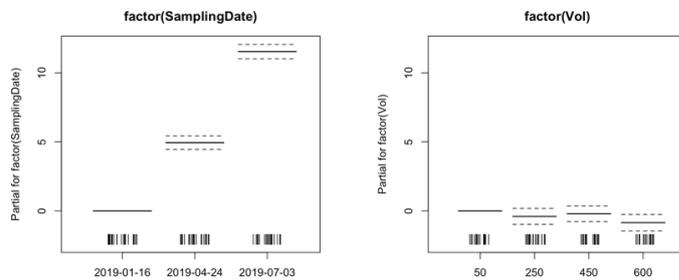


**Figure 8.** Coefficients of the best model fits on cockle performance indices as sampled in the enclosures within the raceways for Experiment 1 (with one common seeding date and three different sampling dates), obtained by Generalized Additive Mixed Modelling (GAMM), for model H2 (shell length and total biomass) and H3 (all other indices) (see Table 2 for description of the models).

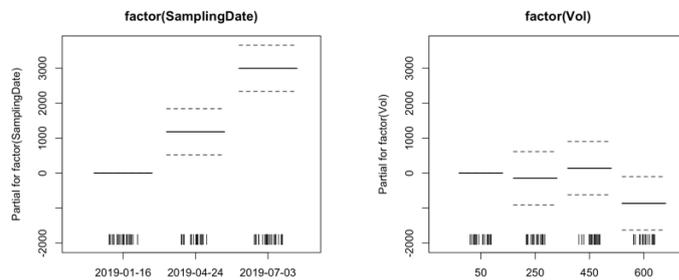
a. Density (number per enclosure), value intercept is  $14.6 \pm 1.4$  individuals, deviance explained is 70.4%



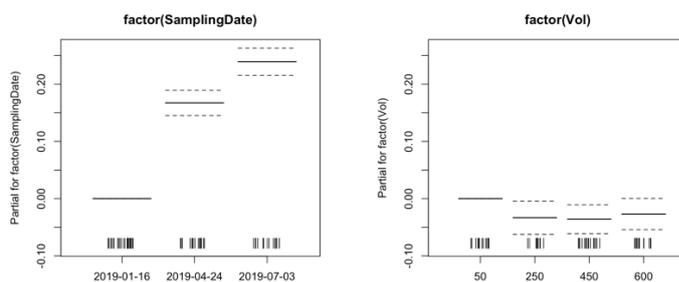
b. Shell length (mm), value of intercept is 13.2 mm, deviance explained is 97.1%.



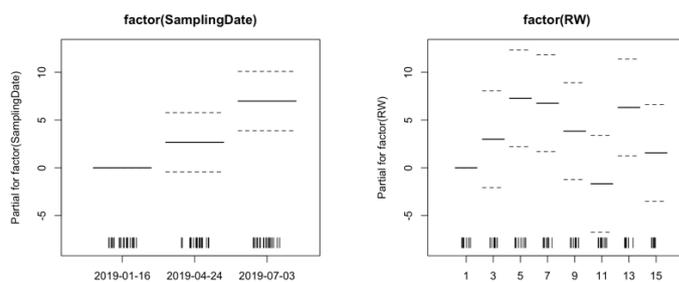
c. Shell volume ( $\text{mm}^3$ ), value of intercept is  $935 \text{ mm}^3$ , deviance explained is 58.2%.



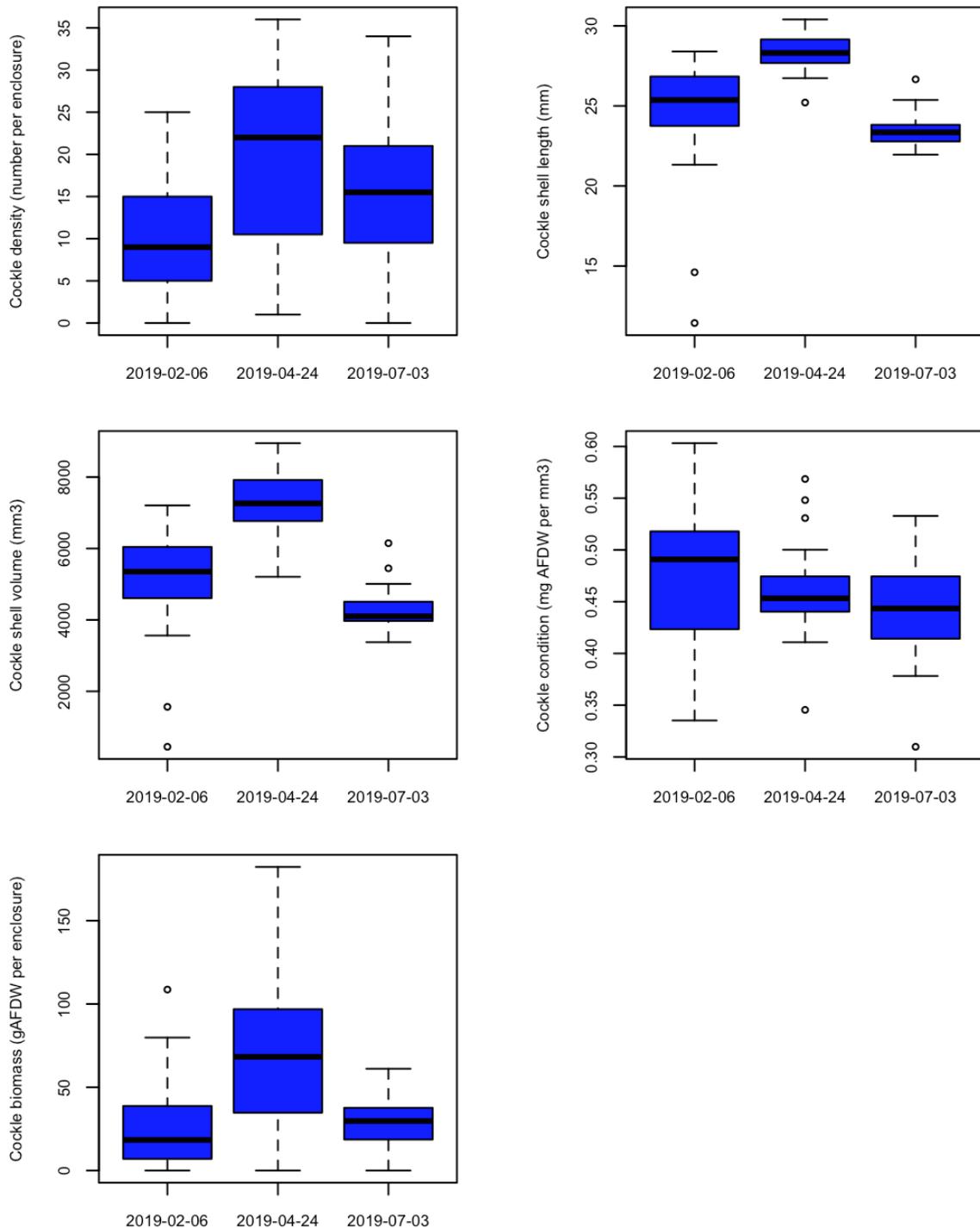
d. Meat content ( $\mu\text{g AFDW mm}^{-3}$ ), value of intercept is  $0.022 \mu\text{g AFDW mm}^{-3}$ , deviance explained is 91%.



e. Total biomass (g AFDW per enclosure), value of intercept is  $-0.080 \text{ g AFDW}$ , deviance explained is 41.7%.

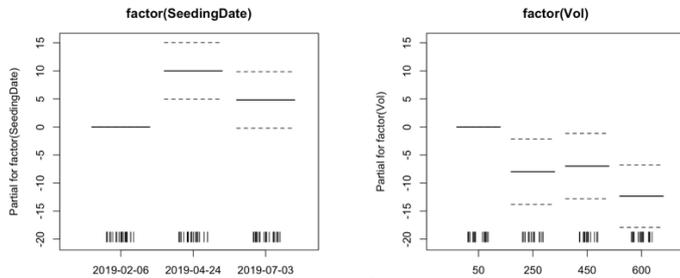


**Figure 9.** Cockle performance indices for Experiment 2, where the cockles were transplanted on 6 February 2019, 24 April 2019 and 3 July 2019 (in 3 enclosures per raceway, being 24 enclosures per sampling period) and all sampled on 13 August 2019 (72 enclosures in total). Dates on the x-axis are, therefore, the dates when the cockles were transplanted into the enclosures.

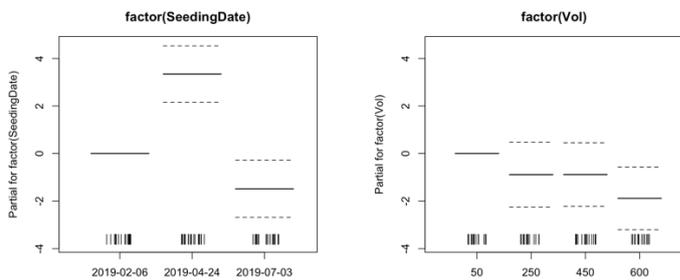


**Figure 10.** Coefficients of the best model fits on cockle performance indices as sampled in the enclosures within the raceways for Experiment 2 (with three different seeding dates and one common sampling date), obtained by Generalized Additive Mixed Modelling (GAMM), for model H2 (condition) and H3 (all other indices) (see [Table 2](#) for description of the models).

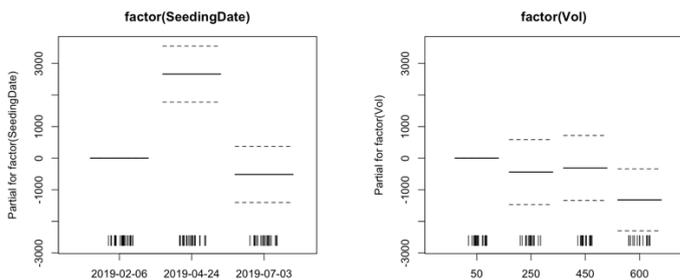
- a. Density (number per enclosure); intercept is  $17.2 \pm 2.4$  individuals, deviance explained is 37.5%



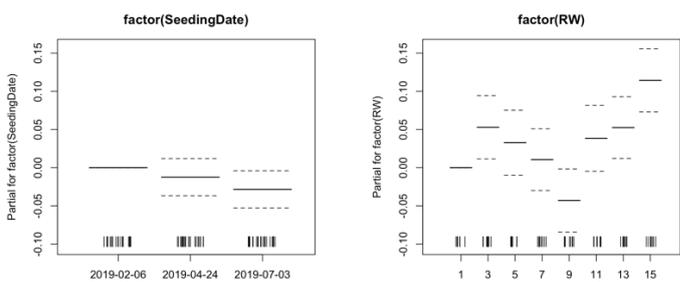
- b. Shell length (mm), value of intercept is 22.7 mm, deviance explained is 52.6%.



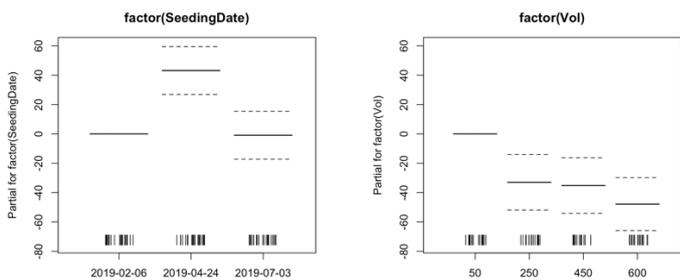
- c. Shell volume ( $\text{mm}^3$ ), value of intercept is  $3026 \text{ mm}^3$ , deviance explained is 56.3%.



- d. Meat content ( $\mu\text{g AFDW mm}^{-3}$ ), value of intercept is  $0.044 \mu\text{g AFDW mm}^{-3}$ , deviance explained is 62.7%.

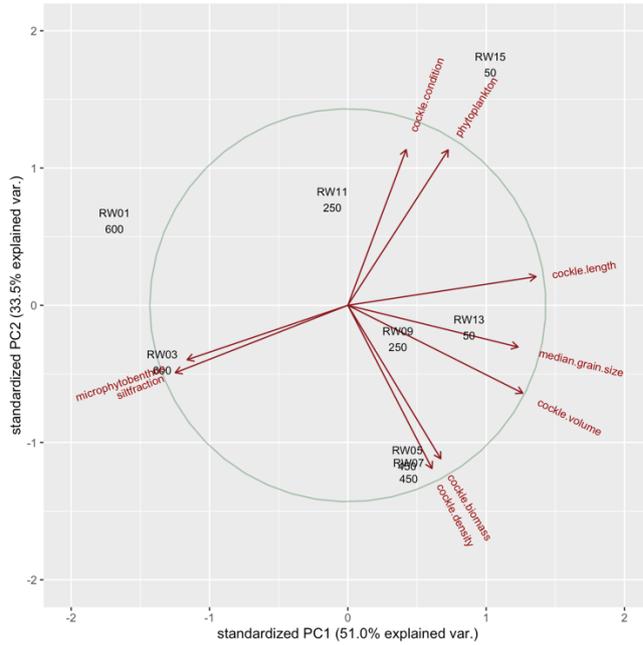


- e. Total biomass (g AFDW per enclosure), value of intercept is 5.76 g AFDW, deviance explained is 53.3%.

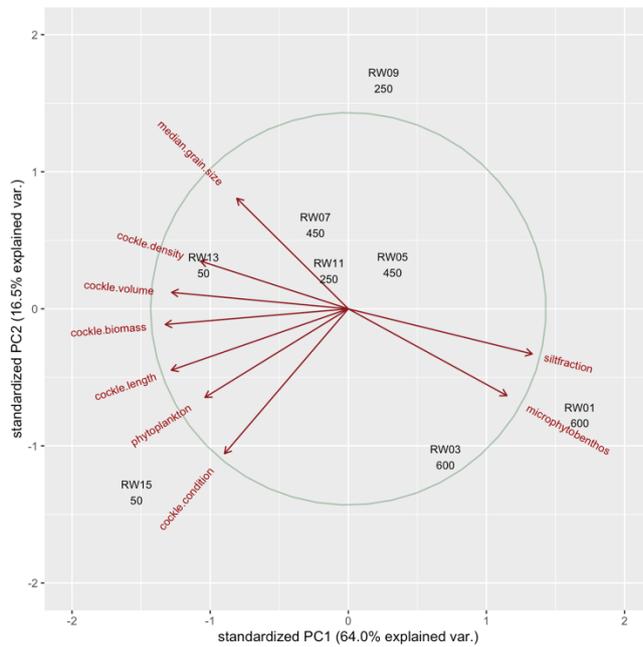


**Figure 11.** Biplots of principal component analyses of the raceway coefficients on environmental conditions and cockle performance indices as derived from Models 2 (see Table 2) on Experiment 1 (top panel) and Experiment 2 (bottom panel).

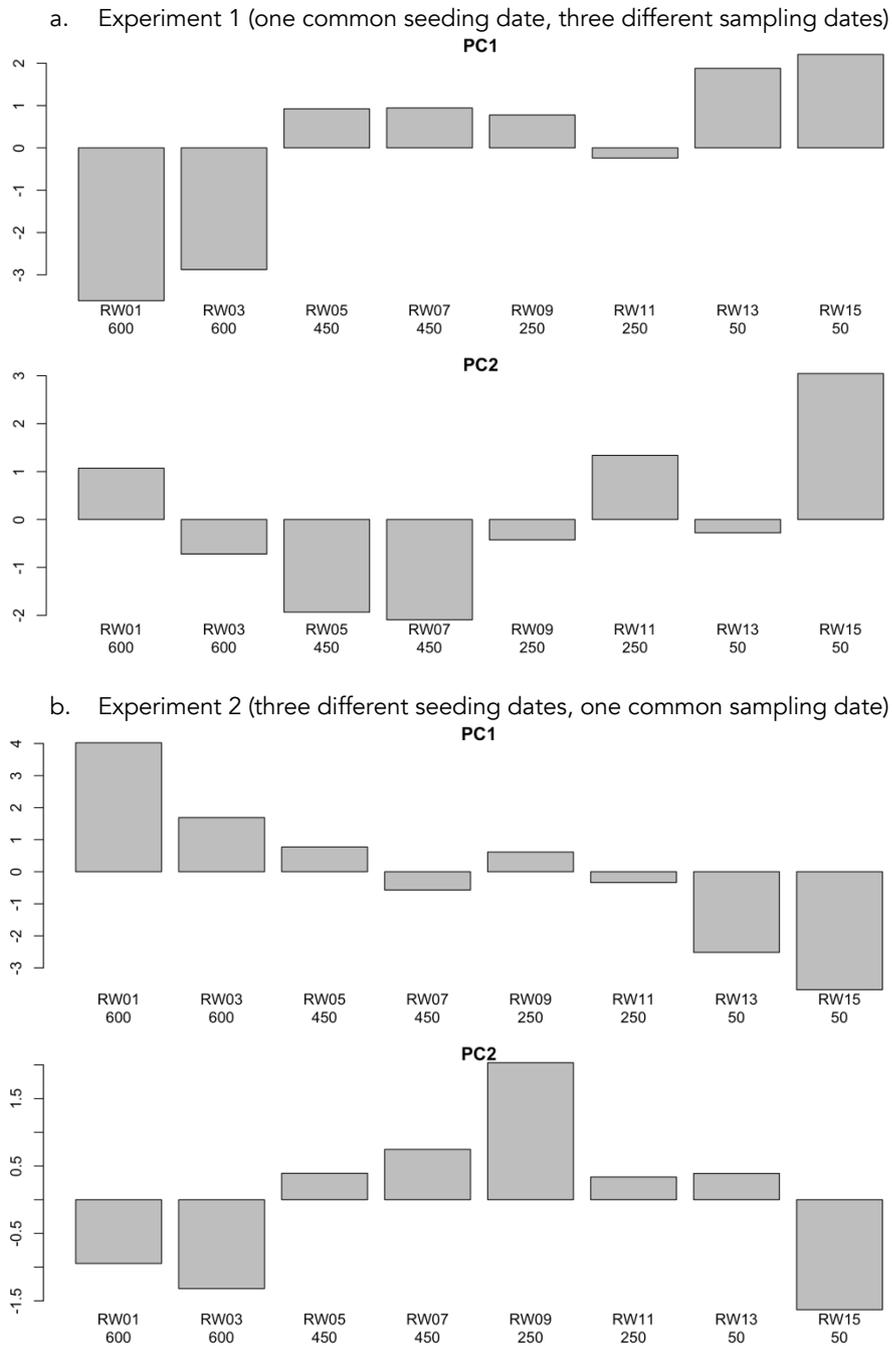
a. Experiment 1 (one common seeding date, three different sampling dates)



b. Experiment 2 (three different seeding dates, one common sampling date)



**Figure 12.** Loadings of 1<sup>st</sup> and 2<sup>nd</sup> axis of principal component analyses of the raceway coefficients on environmental conditions and cockle performance indices as derived from Models 2 (see Table 2) on Experiment 1 (top panel) and Experiment 2 (bottom panel).



**Appendix 1.** The estimates, estimated degrees of freedom and approximate significance for intercepts and smoothers of the best statistical model for environmental conditions (see [Table 2](#) for description of models).

### a. Phytoplankton

```

Formula:
CHLa ~ s(Tid, RW)

Parametric coefficients:
      Estimate Std. Error t value Pr(>|t|)
(Intercept)  6.729      0.224  30.04 <2e-16 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Approximate significance of smooth terms:
      edf Ref.df  F p-value
s(Tid,RW) 26.04  28.42 8.841 <2e-16 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.668 Deviance explained = 74%
GCV = 7.7724 Scale est. = 6.021 n = 120

```

### b. Microphytobenthos

```

Formula:
CHLa ~ factor(SamplingDate) + factor(RefVol)

Parametric coefficients:
      Estimate Std. Error t value Pr(>|t|)
(Intercept)  -0.4789      0.5400  -0.887  0.3761
factor(SamplingDate)2019-01-16  1.4155      0.7637   1.854  0.0652 .
factor(SamplingDate)2019-04-24  7.8177      0.7637  10.237 < 2e-16 ***
factor(SamplingDate)2019-07-03  7.5824      0.7637   9.929 < 2e-16 ***
factor(SamplingDate)2019-08-13  4.7042      0.5400   8.712  9.41e-16 ***
factor(RefVol)250      0.1069      0.6235   0.171  0.8640
factor(RefVol)450      3.0515      0.6235   4.894  1.98e-06 ***
factor(RefVol)600      5.7532      0.6235   9.227 < 2e-16 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.577 Deviance explained = 59.1%
GCV = 10.901 Scale est. = 10.497 n = 216

```

### c. Median grain size

```

Formula:
MedGS ~ factor(SamplingDate) + factor(RefVol)

Parametric coefficients:
      Estimate Std. Error t value Pr(>|t|)
(Intercept)  225.288      3.415  65.961 < 2e-16 ***
factor(SamplingDate)2019-01-16  -9.571      4.830  -1.981  0.0489 *
factor(SamplingDate)2019-04-24  -31.859      4.830  -6.596  3.44e-10 ***
factor(SamplingDate)2019-07-03  -30.717      4.830  -6.359  1.26e-09 ***
factor(SamplingDate)2019-08-13  -19.176      3.415  -5.615  6.26e-08 ***
factor(RefVol)250      1.665      3.944   0.422  0.6734
factor(RefVol)450      7.159      3.944   1.815  0.0709 .
factor(RefVol)600     -17.476      3.944  -4.431  1.51e-05 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.339 Deviance explained = 36%
GCV = 436.11 Scale est. = 419.96 n = 216

```

### d. Silt fraction (%)

```

Formula:
Vol63 ~ factor(SamplingDate) + factor(RefVol) + factor(RWrank)

Parametric coefficients:
      Estimate Std. Error t value Pr(>|t|)
(Intercept)  -0.2402      1.1354  -0.212  0.83268
factor(SamplingDate)2019-01-16  6.7972      1.4866   4.572  8.30e-06 ***
factor(SamplingDate)2019-04-24  14.6397     1.4866   9.848 < 2e-16 ***
factor(SamplingDate)2019-07-03  13.4206     1.4866   9.028 < 2e-16 ***
factor(SamplingDate)2019-08-13  9.2647      1.0512   8.813  4.98e-16 ***
factor(RefVol)250      3.5013      1.2138   2.885  0.00433 **
factor(RefVol)450      7.5726      1.2138   6.239  2.45e-09 ***
factor(RefVol)600     13.2517     1.2138  10.917 < 2e-16 ***
factor(RWrank)Upstream  1.7348      0.8583   2.021  0.04454 *
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.571 Deviance explained = 58.7%
GCV = 41.51 Scale est. = 39.781 n = 216

```

**Appendix 2.** The estimates, their standard error and the significance for intercepts and factors of the best statistical models for cockle performance indices of [Experiment 1](#) (see [Table 2](#) for descriptions of the models).

**a. Density**

Formula:  
Tn ~ factor(SamplingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	14.5972	1.7002	8.586	3.84e-12 ***
factor(SamplingDate)2019-04-24	-10.1250	1.3170	-7.688	1.38e-10 ***
factor(SamplingDate)2019-07-03	-13.6667	1.3170	-10.377	3.50e-15 ***
factor(RW)3	4.5556	2.1506	2.118	0.038169 *
factor(RW)5	7.6667	2.1506	3.565	0.000708 ***
factor(RW)7	8.7778	2.1506	4.082	0.000130 ***
factor(RW)9	3.3333	2.1506	1.550	0.126245
factor(RW)11	0.5556	2.1506	0.258	0.797013
factor(RW)13	5.8889	2.1506	2.738	0.008052 **
factor(RW)15	2.1111	2.1506	0.982	0.330097

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.66 Deviance explained = 70.4%  
GCV = 24.17 Scale est. = 20.813 n = 72

**b. Shell length**

Formula:  
Tn ~ factor(SamplingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	14.5972	1.7002	8.586	3.84e-12 ***
factor(SamplingDate)2019-04-24	-10.1250	1.3170	-7.688	1.38e-10 ***
factor(SamplingDate)2019-07-03	-13.6667	1.3170	-10.377	3.50e-15 ***
factor(RW)3	4.5556	2.1506	2.118	0.038169 *
factor(RW)5	7.6667	2.1506	3.565	0.000708 ***
factor(RW)7	8.7778	2.1506	4.082	0.000130 ***
factor(RW)9	3.3333	2.1506	1.550	0.126245
factor(RW)11	0.5556	2.1506	0.258	0.797013
factor(RW)13	5.8889	2.1506	2.738	0.008052 **
factor(RW)15	2.1111	2.1506	0.982	0.330097

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.66 Deviance explained = 70.4%  
GCV = 24.17 Scale est. = 20.813 n = 72

**c. Shell volume**

Formula:  
Tv ~ factor(SamplingDate) + factor(Vol)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	935.3	330.2	2.833	0.006111 **
factor(SamplingDate)2019-04-24	1180.7	330.2	3.576	0.000658 ***
factor(SamplingDate)2019-07-03	2996.0	330.2	9.075	3.21e-13 ***
factor(Vol)250	-146.9	381.2	-0.385	0.701172
factor(Vol)450	139.7	381.2	0.366	0.715292
factor(Vol)600	-864.6	381.2	-2.268	0.026613 *

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.55 Deviance explained = 58.2%  
GCV = 1.4269e+06 Scale est. = 1.308e+06 n = 72

**d. Meat content**

Formula:  
Tb ~ factor(SamplingDate) + factor(Vol)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	0.022354	0.001087	20.574	< 2e-16 ***
factor(SamplingDate)2019-04-24	0.016728	0.001106	15.119	< 2e-16 ***
factor(SamplingDate)2019-07-03	0.023909	0.001182	20.227	< 2e-16 ***
factor(Vol)250	-0.003328	0.001451	-2.294	0.02621 *
factor(Vol)450	-0.003589	0.001258	-2.853	0.00637 **
factor(Vol)600	-0.002693	0.001362	-1.977	0.05382 .

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.9 Deviance explained = 91%  
GCV = 1.3294e-05 Scale est. = 1.1817e-05 n = 54

**e. Total biomass**

Formula:  
Tw ~ factor(SamplingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	-0.07951	0.20029	-0.397	0.69276
factor(SamplingDate)2019-04-24	0.26705	0.15515	1.721	0.09019 .
factor(SamplingDate)2019-07-03	0.69844	0.15515	4.502	3.03e-05 ***
factor(RW)3	0.29886	0.25335	1.180	0.24266
factor(RW)5	0.72743	0.25335	2.871	0.00559 **
factor(RW)7	0.67567	0.25335	2.667	0.00975 **
factor(RW)9	0.38385	0.25335	1.515	0.13483
factor(RW)11	-0.16786	0.25335	-0.663	0.51007
factor(RW)13	0.63163	0.25335	2.493	0.01535 *
factor(RW)15	0.15551	0.25335	0.614	0.54158

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.332 Deviance explained = 41.7%  
GCV = 0.33544 Scale est. = 0.28885 n = 72

**Appendix 3.** The estimates, their standard error and the significance for intercepts and factors of the best statistical models for cockle performance indices of [Experiment 2](#) (see [Table 2](#) for descriptions of the models).

**a. Density**

Formula:  
Tn ~ factor(SeedingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	1.264	2.757	0.458	0.648265
factor(SeedingDate)2019-04-24	9.583	2.136	4.487	3.19e-05 ***
factor(SeedingDate)2019-07-03	5.625	2.136	2.634	0.010647 *
factor(RW)3	6.889	3.488	1.975	0.052692 .
factor(RW)5	4.889	3.488	1.402	0.165958
factor(RW)7	12.556	3.488	3.600	0.000633 ***
factor(RW)9	8.111	3.488	2.326	0.023319 *
factor(RW)11	6.556	3.488	1.880	0.064849 .
factor(RW)13	21.667	3.488	6.213	4.85e-08 ***
factor(RW)15	9.889	3.488	2.835	0.006171 **

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.449 Deviance explained = 51.9%  
GCV = 63.561 Scale est. = 54.733 n = 72

**b. Shell length**

Formula:  
TL ~ factor(SeedingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	22.6518	1.0775	21.022	< 2e-16 ***
factor(SeedingDate)2019-04-24	4.1252	0.7350	5.612	5.88e-07 ***
factor(SeedingDate)2019-07-03	-0.7950	0.7441	-1.068	0.28974
factor(RW)3	1.3271	1.2375	1.072	0.28799
factor(RW)5	1.4879	1.2375	1.202	0.23410
factor(RW)7	2.1539	1.2375	1.741	0.08707 .
factor(RW)9	0.5183	1.2375	0.419	0.67687
factor(RW)11	1.5489	1.2743	1.215	0.22912
factor(RW)13	2.0021	1.2375	1.618	0.11112
factor(RW)15	3.4139	1.2664	2.696	0.00918 **

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.452 Deviance explained = 52.6%  
GCV = 6.9992 Scale est. = 5.9699 n = 68

**c. Shell volume**

Formula:  
Tv ~ factor(SeedingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	3025.6	554.9	5.453	9.15e-07 ***
factor(SeedingDate)2019-04-24	2890.6	429.8	6.725	6.41e-09 ***
factor(SeedingDate)2019-07-03	-320.5	429.8	-0.746	0.45868
factor(RW)3	1321.7	701.9	1.883	0.06438 .
factor(RW)5	1469.7	701.9	2.094	0.04036 *
factor(RW)7	1855.0	701.9	2.643	0.01039 *
factor(RW)9	1363.3	701.9	1.942	0.05664 .
factor(RW)11	1237.5	701.9	1.763	0.08280 .
factor(RW)13	1746.0	701.9	2.488	0.01556 *
factor(RW)15	2216.6	701.9	3.158	0.00245 **

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.5 Deviance explained = 56.3%  
GCV = 2.5744e+06 Scale est. = 2.2168e+06 n = 72

**d. Meat content**

Formula:  
Tb ~ factor(SeedingDate) + factor(RW)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	0.043994	0.001795	24.507	< 2e-16 ***
factor(SeedingDate)2019-04-24	-0.001236	0.001216	-1.016	0.3141
factor(SeedingDate)2019-07-03	-0.002836	0.001217	-2.330	0.0237 *
factor(RW)3	0.005294	0.002073	2.554	0.0136 *
factor(RW)5	0.003275	0.002126	1.540	0.1296
factor(RW)7	0.001053	0.002021	0.521	0.6045
factor(RW)9	-0.004292	0.002065	-2.079	0.0426 *
factor(RW)11	0.003848	0.002157	1.783	0.0803 .
factor(RW)13	0.005252	0.002021	2.599	0.0122 *
factor(RW)15	0.011430	0.002065	5.536	1.03e-06 ***

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.562 Deviance explained = 62.7%  
GCV = 1.7356e-05 Scale est. = 1.4557e-05 n = 62

**e. Total biomass**

Formula:  
Tw ~ factor(SeedingDate) + factor(Vol)

Parametric coefficients:

	Estimate	Std. Error	t value	Pr(> t )
(Intercept)	5.76472	0.79307	7.269	8.70e-10 ***
factor(SeedingDate)2019-04-24	4.31670	0.81607	5.290	1.81e-06 ***
factor(SeedingDate)2019-07-03	-0.09126	0.81607	-0.112	0.911336
factor(Vol)250	-3.29836	0.94623	-3.486	0.000924 ***
factor(Vol)450	-3.51878	0.94623	-3.719	0.000443 ***
factor(Vol)600	-4.78567	0.90220	-5.304	1.72e-06 ***

---  
Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1

R-sq.(adj) = 0.495 Deviance explained = 53.3%  
GCV = 8.0582 Scale est. = 7.3256 n = 66

**Appendix 4.** Scores of the PCA of Experiment 1 and Experiment 2. The more the value of the variable deviates from 0, the stronger the correlation with that principal component.

a. Experiment 1 (one common seeding date, three different sampling dates)

	PC1	PC2
cockle.density	0.1984594	-0.47849586
cockle.length	0.4438226	0.08402952
cockle.volume	0.4124381	-0.25762515
cockle.condition	0.1375601	0.45623348
cockle.biomass	0.2191078	-0.44994878
phytoplankton	0.2366963	0.45542436
microphytobenthos	-0.3790822	-0.15973812
median.grain.size	0.4014566	-0.12232774
siltfraction	-0.4070667	-0.19777625

b. Experiment 2 (three different seeding dates, one common sampling date)

	PC1	PC2
cockle.density	-0.3115103	0.19967980
cockle.length	-0.3730005	-0.25665329
cockle.volume	-0.3722640	0.06883643
cockle.condition	-0.2608368	-0.60574895
cockle.biomass	-0.3857685	-0.06545885
phytoplankton	-0.3020928	-0.37053833
microphytobenthos	0.3343078	-0.36314764
median.grain.size	-0.2349643	0.46208897
siltfraction	0.3875155	-0.18868195