# FEEDING PREFERENCE OF THE JAPANESE OYSTER DRILL (*OCINEBRELLUS INORNATUS*) IN A NATURAL ENVIRONMENT

**Final Report**

**Delta Academy**
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# PREFACE

Conducting this research and writing this report has been a challenging and educational experience. I am very grateful to all my coworkers and supervisors, who helped me throughout the whole process. First, I would like to thank my supervisor, Eva, who guided my research and assisted me whenever I needed it. She always encouraged me to do my best, and made working at HZ University a great experience. I would like to thank my co-worker Niels as well, his technical skills helped facilitate the implementation of my various experiments; he was truly a pleasure to work with. Finally, I would like to thank all the other researchers who helped me out when I needed it, and helped me enjoy my time in the Netherlands.

# SUMMARY

The Japanese oyster drill, *Ocinebrellus inornatus*, is an invasive marine gastropod that predates upon benthic bivalves, including the cultivated Pacific oyster, *Magallana gigas*. *O. inornatus* was first discovered in the Oosterschelde estuary in the Netherlands in 2007 (Lützen et al., 2011), and has since established populations in cultivated oyster plots. *O. inornatus* has threatened the oyster farming industry in the area, causing up to 50% economic losses for local farmers (Smaal, et al. 2016).

To develop a novel method of reducing the impact of *O. inornatus* on oyster farms, research was conducted to gain insight into the feeding preference of *O. inornatus*. This novel method would involve farmers spreading the preferred prey in a border around their oyster plots to attract the *O. inornatus* away from the cultivated oysters.

An experiment was conducted in the Oosterschelde tidal zone where *O. inornatus* were placed inside enclosures with *M. gigas* from two different brood stocks, at a 1:1 predator to prey ratio. The first run of the experiment used *M. gigas* from Oosterschelde brood stock and French brood stock. The results from this experiment showed there was a significant feeding preference by *O. inornatus* on the French *M. gigas*. The second run used two different French *M. gigas* both raised in the Oosterschelde, one group being triploid raised in Yerseke and the other being diploid raised in Kattendijke. The results from this experiment showed a significant preference for the diploid *M. gigas* raised in Kattendijke.

Further research will have to be conducted to identify the specific cause of the feeding preference.

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# 1. INTRODUCTION

## 1.1. CONTEXT

The Pacific oyster, *Magallana gigas*, is a species of oyster that is cultivated around the world, with 4.4 million tonnes of oysters having been produced globally in 2003 ("*Crassostrea gigas* (Thunberg, 1793)", 2005). Oyster cultivation in the Netherlands has recently become threatened by the introduction of the invasive snail, the Japanese oyster drill, *Ocinebrellus inornatus*. *O. inornatu*s has caused high mortality within on-bottom cultivated oysters plots in the Oosterschelde estuary of the Netherlands. With little information being available on *O. inornatus*, combatting the snail has been difficult. HZ University has partnered with local oyster farmers to learn more about *O. inornatus* and develop and to compare counteractive measures to protect oyster plots.

## 1.2. AIM AND MAIN QUESTION

The experiment discussed in this report builds upon the food preference research previously conducted by Adryan Rademakers (2017), Dara Barbaran (2017), and Belma Colakovic (2018).

The main question being asked in this experiment is as follows:

"How is predation by *O. inornatus* affected when provided with Pacific oyster (*Magallana gigas*) from two different brood stock as possible food sources?"

### 1.2.1. SUB-QUESTIONS

1. How many successful boreholes were achieved, and how many occurred on each *M. gigas* per enclosure?
2. How were the *O. inornatus* spread at the end of the experiment throughout each enclosure?
3. How many total successful boreholes per origin of the *M. gigas* were achieved during the experiment?

## 1.3. BACKGROUND

### 1.3.1. BIOLOGY AND PHYSICAL DESCRIPTION OF *O. INORNATUS*

*O. inornatus*, commonly known as the Japanese Oyster Drill (Figure 1), is a predatory species of marine muricid native to the oceans around North-East Asia (Lützen et al., 2011). Typically, specimens will have 5-6 whorls (complete shell rotations) and approximately 8 axial ribs on the final whorl (van den Brink, 2010).

*O. inornatus* has been observed to have two breeding periods, spring and autumn (Fey et al., 2010). Because *O. inornatus* is gonochoristic, meaning each specimen has a single sex, mature individuals congregate to produce fertilized eggs (Fey et al., 2010). *O. inornatus* typically lay between 20 and 40 egg capsules during breeding periods (Lützen et al., 2011). Egg capsules are

bright yellow and contain between 10-15 individual embryos (Lützen et al., 2011). *O. inornatus* does not have a planktonic larvae phase, and as such the larvae hatch at 2 mm long and will keep the same form through to adulthood (Buhle et al., 2004).


**Figure 1: Typical *Ocinebrellus inornatus* shell appearance**

While *O. inornatus* prefer warmer water temperatures, they have been observed to withstand temperatures as low as 0 °C (Faasse et al., 2009), lower than what usually occurs in the Netherlands. This ability makes them particularly difficult to manage, as they can survive in a wide variety of habitats.

The diet of *O. inornatus* consists of mainly benthic bivalves such as oysters and mussels (Fey et al., 2010), though in some extreme cases cannibalism has been observed in the related American oyster drill, *Urosalpinx cinerea* (Carriker, 1955). To feed, *O. inornatus* follows effluent trails released by their prey and selects a suitable spot on the prey to feed (Carriker, 1981). *O. inornatus* feeds using its boring organ, called the radula, which it scrapes across the prey's shell while secreting sulphuric acid to assist the process (University of Rhode Island, n.d.). Depending on the shell thickness of the prey, this process can take anywhere from 1-14 days to complete before *O. inornatus* can feed on the prey (Fey et al., 2010).

### 1.3.2. INVASIVE HISTORY OF *OCINEBRELLUS INORNATUS*

*O. inornatus* was first discovered outside of its native range in 1924 in Puget Sound, United States (Lützen et al., 2011). *O. inornatus* was later discovered in 1995 to have established populations in Marennes-Oléron Bay, France where it then proceeded to travel north along the coast, to eventually establish a population in the Oosterschelde, Netherlands in 2007 (Lützen et al., 2011). The most likely cause of this spread is due to mass global transportation of *M. gigas* between oyster farms. Any attempts to regulate this transportation have been ineffective due to the large quantity of live oysters being transported between farms every year.

## 1.4. ECONOMIC AND ECOLOGICAL IMPACT

*O. inornatus* poses a significant threat to both the economic and ecological stability of affected areas, due to its rapid population growth rate and lack of a natural predator or parasite (Faasse et al., 2007). Predation by the introduced *O. inornatus* has inhibited the rehabilitation of the native European flat oyster (*Ostrea edulis*), whose population has been devastated historically by parasites including *Bonamia ostreae* (Ronza et al., 2018).

The economic losses by oyster farmers and related industries have been observed to be as high as 50% (Smaal, et al. 2016). In one year, the Netherlands produces approximately 5 million euro worth of *M. gigas* (Strietman, 2015) and as such, oyster farming is an important part of the economy. Oyster losses caused by *O. inornatus* could therefore have a significant impact on the lives of many Dutch farmers and businesses who rely on oyster cultivation for income.

## 1.5. RESEARCH GOAL

### 1.5.1. APPLICATION OF COLLECTED DATA

The aim of the research presented in this report, is to develop a novel method of reducing oyster mortality in cultivated oyster plots. Should a food preference be discovered, farmers will be advised to spread the preferred prey species around their oyster plots as a barrier, to prevent any *O. inornatus* from reaching the cultivated oysters.

### 1.5.2. PREVIOUS EXPERIMENTS

Research on *O.* inornatus by HZ University began with the experiments conducted by Adryan Rademakers in 2017, whose results directly inspired all future research. Her experiments provided the following conclusions: *O. inornatus* prefer feeding on smaller oysters, group feeding rates increase as prey species decrease, and *O. inornatus* prefer *M. gigas* over *O. edulis*. In previous experiments under controlled conditions (Barbaran, 2017), the *O. inornatus* were provided with 3 size classes of *M. gigas*, 1 - 3.6 cm, 3.61 - 6.6 cm, and 6.61 - 10 cm; the results from this experiment proved to be inconclusive. Another experiment was conducted (Colakovic, 2018) where small, half-grown *M. gigas* and *O. edulis* were provided to *O. inornatus* in natural, enclosed experiment settings. In this experiment, it was observed that during its 2-week duration, none of the preyed upon oysters had complete boreholes in them. Since the partial boreholes did not necessarily indicate that the *O. inornatus* intended to feed on the oyster, the data could not be used. This fragility meant that the shells of some of the preyed-upon spat had fallen apart, making it difficult to count exactly how many were preyed upon.

# 3. MATERIAL AND METHOD

## 3.1. LOCATION AND SPATIAL LAYOUT

The experimental site was located close to Yerseke Bank 119 in the Oosterschelde, near Yerseke, Netherlands. The site was in the intertidal zone which was only reachable during low tides. At this location there were 5 enclosures (Figure 2), each 1 m by 1 m, built out of 13 mm metal mesh. The enclosures were numbered 1-5, beginning at number 1 with the enclosure closest to the dyke.

**Figure 2: Photo of enclosure 1 in experiment 1, taken facing Yerseke**

# 3.2. EXPERIMENTAL PROCESS

## 3.2.1. Experimental **P**rocedure **O**verview

The experiment was conducted twice, both following the same procedure, but comparing *M. gigas* from different brood stocks in each run. In both experiments, 5 enclosures were used, and each enclosure was divided into quadrants. Two quadrants were randomly selected to each contain 50 evenly distributed *M. gigas* from different brood stocks. After the *M. gigas* were placed into the enclosures, 25 of the *O. inornatus* were selected and spread evenly in every quadrant, in total 100 painted *O. inornatus* were used per enclosure. Once all *O. inornatus* and *M. gigas* were in the enclosure, a photo (Figure 2) was taken and the enclosures were covered with 13 mm metal mesh. The experiments were left untouched for approximately 3 weeks. Experiment 1 was set in on May 21$^{rst}$, 2019 and withdrawn on June 7$^{th}$, 2019, totaling 17 days in the field. Experiment 2 was set in on June 21$^{rst}$, 2019 and withdrawn on July 12$^{th}$, 2019, totaling 21 days in the field. The following sections 3.2.2 – 3.2.5 provide further details on the experimental procedure.

## 3.2.2. ENCLOSURE DESIGN AND LAYOUT

The tops of the enclosures were covered with 13 mm metal fencing to prevent any *O. inornatus* from escaping. Each enclosure was divided into quadrants, lettered A - D beginning with the bottom left and proceeding clockwise (Figure 3). This lettering system was read with the dyke (shoreline) kept to the left side of the observer. Additionally, a water temperature logger was placed in enclosure 5.

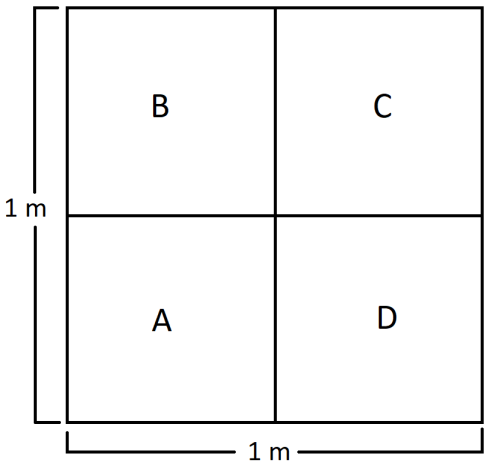


**Figure 3: Aerial view of enclosure setup with labelled quadrants**

### 3.2.3. *M. GIGAS* PREPARATION AND LAYOUT

Prior to the experiment, *M. gigas* were collected from farmers and stored at HZ University's SEA lab. The oysters were kept in tanks filled with filtered ground water and fed using *Tetraselmis sp.* (Figure 4) or *Rhodomonas sp.* algae three times each week. To ensure the health of the oysters, water in the tank was aerated and regularly cleaned. The first run of this experiment (experiment 1) compared the feeding preference of *O. inornatus* between *M. gigas* from brood stock in France and stock from the Oosterschelde. The second run (experiment 2) used *M. gigas* from two separate French brood stocks, both raised in the Oosterschelde. One group contained triploid *M.* gigas raised in Kattendijke and the other had diploid *M. gigas* raised in Yerseke. Refer to Table 1 and Table 2 on the following page for the oyster size data from experiments 1 and 2 respectively.

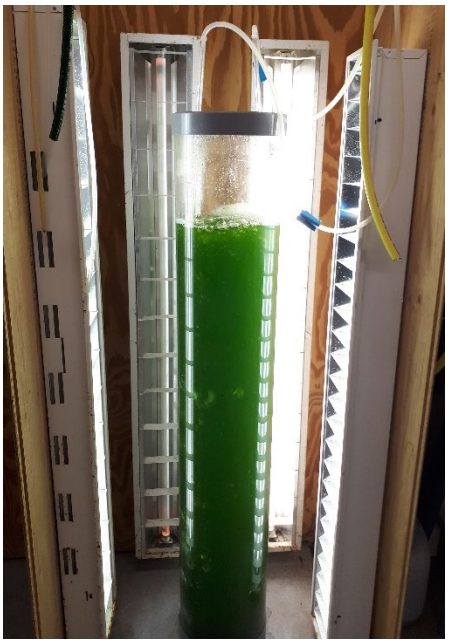

**Figure 4: Bubble column containing *Tetraselmis sp.***

**Table 1: Oyster Shell Measurements for Experiment 1**

| | French Stock (cm) | Oosterschelde Stock (cm) |
|---|---|---|
| **Average Length** | 65.35 ± 9.38 | 52.49 ± 6.9 |
| **Average Width** | 31.37 ± 3.74 | 30.02 ± 3.4 |
| **Average Height** | 17.47 ± 2.8 | 17.26 ± 1.94 |

**Table 2: Oyster Shell Measurements for Experiment 2**

| | Yerseke Stock (cm) | Kattendijke Stock (cm) |
|---|---|---|
| **Average Length** | 44.54 ± 6.92 | 47.81 ± 12.56 |
| **Average Width** | 28.9 ± 3.93 | 23.52 ± 4.27 |
| **Average Height** | 15.39 ± 2.76 | 13.08 ± 3.08 |

*M. gigas* were placed in two of the quadrants within each of the enclosures, chosen randomly using an Excel program. Each of these two quadrants contained *M. gigas* from different origins, with 50 being evenly spread in each of the two quadrants. The number of *M. gigas* used was determined such that there would be a 1:1 predator to prey ratio in each enclosure. Refer to Table 3 and Table 4 for the *M. gigas* distribution in experiments 1 and 2 respectively.

**Table 3: Distribution of French (Fr) and Oosterschelde (Os) *M. gigas* in Each Enclosure for Experiment 1**

| Quadrant | Enclosure 1 | Enclosure 2 | Enclosure 3 | Enclosure 4 | Enclosure 5 |
|---|---|---|---|---|---|
| **A** | x | **Os** | **Fr** | x | **Fr** |
| **B** | **Os** | x | x | **Fr** | **Os** |
| **C** | **Fr** | x | **Os** | x | x |
| **D** | x | **Fr** | x | Os | x |

**Table 4: Distribution of Yerseke (Ye) and Kattendijke (Ka) *M. gigas* in Each Enclosure for Experiment 2**

| Quadrant | Enclosure 1 | Enclosure 2* | Enclosure 3 | Enclosure 4 | Enclosure 5* |
|---|---|---|---|---|---|
| **A** | **Ka** | **Os** | x | **Ka** | x |
| **B** | x | **Fr** | **Ye** | x | **Fr** |
| **C** | x | x | **Ka** | **Ye** | x |
| **D** | **Ye** | x | x | x | **Os** |

* Indicates enclosures set up using Oosterschelde (Os) and French (Fr) *M. gigas* from Experiment 1

### 3.2.4. *O. INORNATUS* PREPARATION AND LAYOUT

The *O. inornatus* used in the experiment were stored in tanks at HZ University's SEA lab which were filled with filtered groundwater. The water was aerated and maintained at the same temperature as the current water in the Oosterschelde. To ensure the *O. inornatus* were actively seeking out food during the experiment, they were starved for 4 days prior to the start of the experiment. 500 *O. inornatus* were selected with shell length ranges of 40.5 - 45.9 mm for experiment 1 and 37 mm - 42.99 mm for experiment 2. These *O. inornatus* were then painted to make them more visible and to distinguish them from wild *O. inornatus*.

### 3.2.5. COLLECTION OF DATA

At the end of each experiment, prior to any disturbance of each enclosure, pictures were taken of the enclosure as well as each individual quadrant. Then, the *O. inornatus* and *M. gigas* were counted, and brought back to the lab for analysis. The number of partial and complete boreholes on each oyster were counted, and it was noted whether each oyster was dead or alive. Live, un-drilled oysters were kept for use in future experiments and all dead or partially drilled oysters were disposed of.

# 4. RESULTS

## 4.1. EXPERIMENT 1: FRENCH VS OOSTERSCHELDE *M. GIGAS*

An initial set of 5 enclosures were set up with the *M. gigas* and *O. inornatus* being collected 17 days later; data from enclosures 2 and 5 include the additional data that were collected during experiment 2. 176 out of the total 702 *M. gigas* recovered, had complete boreholes; 79 of these originated in the Oosterschelde and 97 originated in France. The percentage of drilled *M. gigas* recovered in each enclosure is displayed in Figure 5 below.

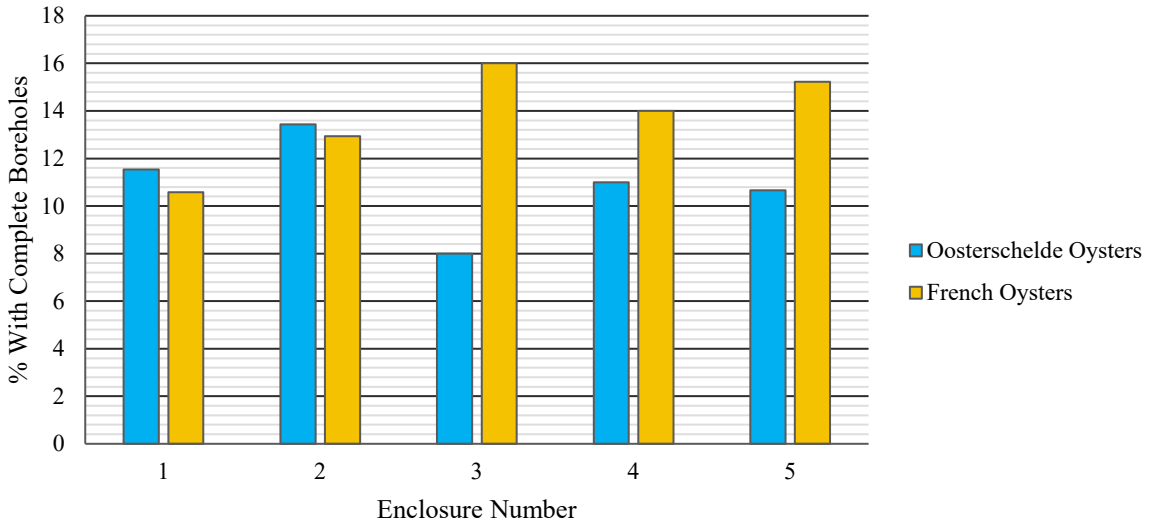


**Figure 5: Percentage of recovered *M. gigas* with complete boreholes in experiment 1**

A HOBO MX2202 water temperature logger was placed in enclosure 5 and programmed to take measurements every 5 minutes. The results, presented in Figure 6 on the following page, showed that water temperature did not significantly change through the duration of experiment 1. Peaks and troughs in the graph result from the logger being exposed to the atmosphere during hot days and cold nights respectively, at low tide. Excluding these peaks and troughs, the water temperature was about 17.75°C on average, with the high being approximately 19.5°C and the low being approximately 16°C.

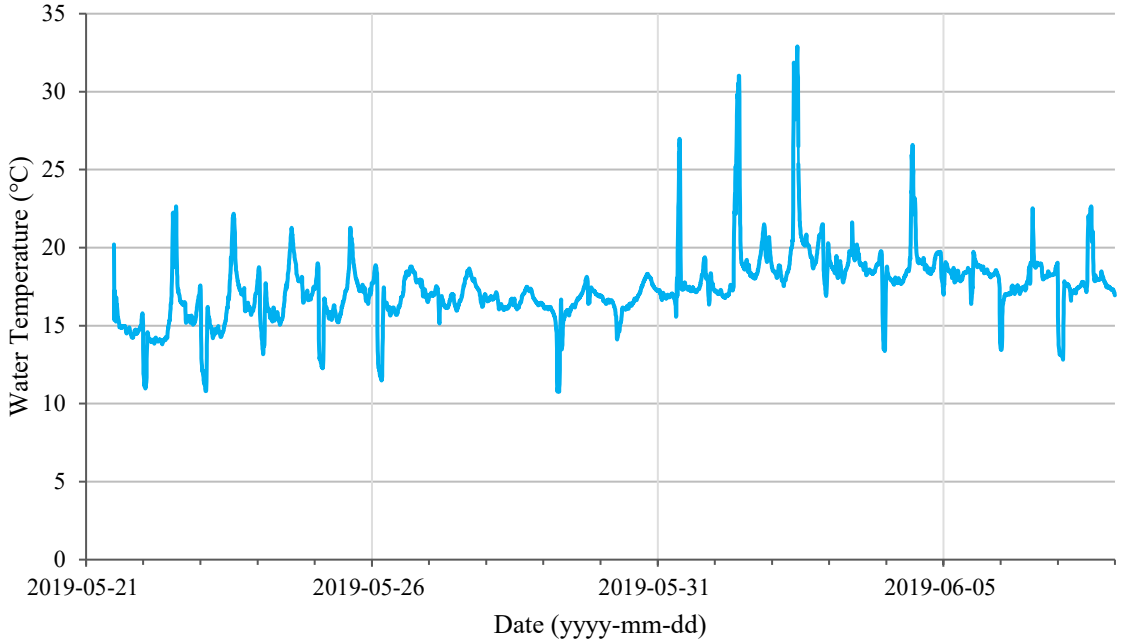


**Figure 6: Oosterschelde water temperatures for the duration of experiment 1**

### 4.1.1. RESULTS ANALYSIS

To determine if there was a significant difference in the predation on one brood stock of *M. gigas*, a hypothesis test was conducted. The test compared the average number of complete boreholes per brood stock, and all calculations were performed using the Excel Analysis ToolPak. Initially, an F-test was used to determine whether the variances between the two data sets were significantly different. It was concluded from this test that the variances were significantly different at the 5% level of significance ($p = 0.0247$). Then, a t-test was performed under the assumption of unequal variances to test the hypothesis that the French *M. gigas* were more preyed upon by *O. inornatus*. The test concluded that the average number of complete boreholes in *M. gigas* from French brood stock was significantly higher than the average found in *M. gigas* from Oosterschelde brood stock at the 5% level of significance ($p = 0.0285$).

## 4.2. EXPERIMENT 2: TWO DIFFERENT FRENCH STOCK, DIPLOID VS TRIPLOID

This experiment used two French brood stocks of *M. gigas* raised in different locations within the Oosterschelde. The stock raised in Kattendijke were diploid and the stock raised in Yerseke were triploid, meaning they are unable to reproduce. Results from experiment 2 were collected 21 days after the experiment was set in. Enclosures 2 and 5 were set up as a repetition of experiment 1, thus the following data exclude these enclosures. Of the 281 recovered *M. gigas*, a total of 181 had complete boreholes; 107 of these *M. gigas* were raised in Kattendijke, the remaining 74 were *M. gigas* raised in Yerseke. The percentage of drilled *M. gigas* recovered in each enclosure is displayed in Figure 7 on the following page.

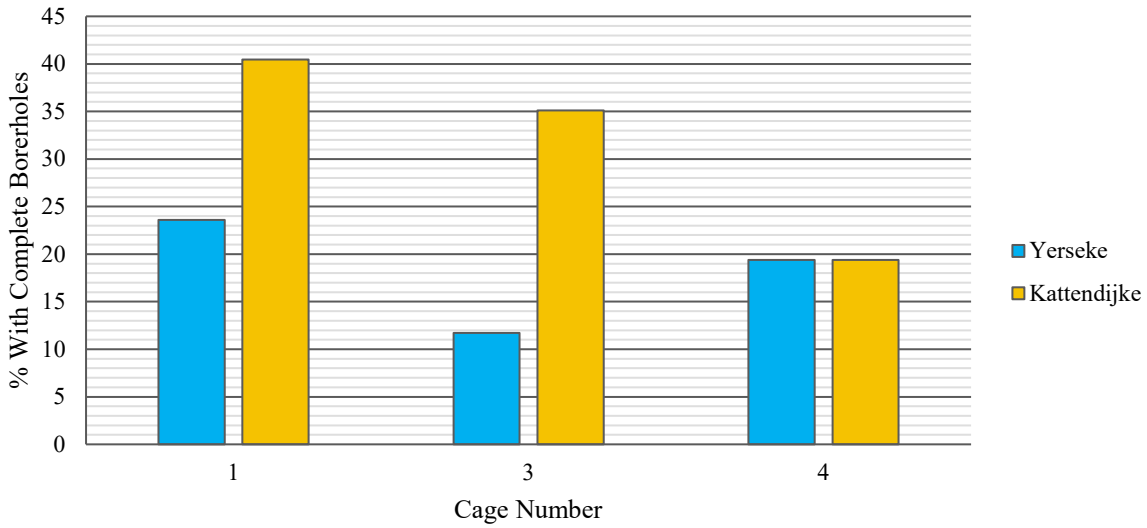


**Figure 7: Percentage of recovered *M. gigas* with complete boreholes in experiment 2**

The same HOBO MX2202 water temperature sensor used in experiment 1, was used to track water temperature during experiment 2. The data from the sensor for the duration of experiment 2, are displayed in Figure 8 below. These data show that the water temperature was more variable than it was in experiment 1, with warmer temperatures occurring around June 25$^{th}$ and June 30$^{th}$. Excluding peaks and troughs representing atmospheric temperature at low tide, the water temperature was about 21°C on average, with the high being approximately 23°C, and the low being approximately 19°C.

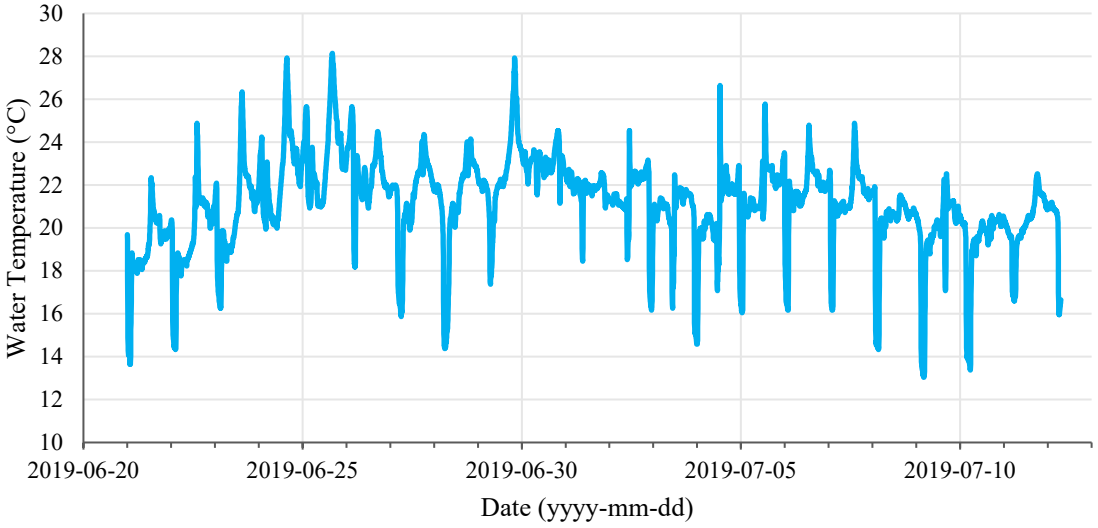


**Figure 8: Oosterschelde water temperatures for the duration of experiment 2**

## 4.2.1. RESULTS ANALYSIS

For experiment 2, the results were analysed in the same manner as experiment 1, using the number of complete boreholes on each brood stock of *M. gigas*. The following calculations were performed using the Excel Analysis ToolPak. Initially, an F-test was performed to determine whether the variances between the two sets of data were significantly different. The test showed that the variances were not significantly different at the 5% level of significance ($p = 0.2078$). Then a t-test was conducted under the assumption that the variances were equal

and concluded that the *M. gigas* raised in Kattendijke were more preyed upon than those raised in Yerseke at the 5% level of significance ($p = 2.6194x10^{-7}$).

# 5. DISCUSSION

## 5.1. EXPERIMENT 1: FRENCH VS OOSTERSCHELDE *M. GIGAS*

The results from this experiment showed that *O. inornatus* appeared to prefer preying upon the *M. gigas* originating from France, over those originating from the Oosterschelde ($p = 0.0285$). This preference may have been caused by a difference in shell thickness between the two *M. gigas* origins. It has been observed in past studies (Carriker, 1981; Lord et al., 2013) that the related *Urosalpinx cinerea*, commonly referred to as the American Oyster Drill, prefers drilling on thinner shells.

Another possible influencing factor would be the change in behaviour when feeding in groups, which has been observed in the related species *Stramonita haemastoma* (Brown et al., 1988), commonly referred to as the southern oyster drill. This study observed that predation rate increased when the drills fed in larger groups, thus if a higher density of *O. inornatus* occurred in the same location as the French *M. gigas*, higher than normal feeding may have occurred.

## 5.2. EXPERIMENT 2: TWO DIFFERENT FRENCH STOCK, DIPLOID VS TRIPLOID

The data collected from this experiment concludes that there was a significant predation preference of *O. inornatus* on the diploid *M. gigas* raised in Kattendijke, Netherlands ($p = 2.6194x10^{-7}$). This significant difference suggests that this result was not likely due to chance, and that some characteristic of the Kattendijke *M. gigas* makes them more attractive to *O. inornatus*. One notable observation was that the *M. gigas* from stock in Kattendijke had much more uniform and smooth shell surfaces (Figure 9). It is hypothesized that the smoother shells provide a more ideal drilling surface for *O. inornatus* by allowing it to obtain a stronger hold on the prey. The wave-like surface present on many of the Yerseke *M. gigas* may have made it difficult for *O. inornatus* to firmly attach itself.

**Figure 9: *M.* gigas raised in Yerseke (left) and Kattendijke (right)**

## 5.3. *O. INORNATUS* MANAGEMENT STRATEGIES

Due primarily to its high fecundity and currently established populations in the Netherlands, complete removal of *O. inornatus* from the natural environment would be unreasonable. Several management strategies have been tested, although none have yet been developed to match all the following criteria: cost-efficient, minimal extra labour, minimal environmental impact, time-efficient, and effective. The novel method investigated in this report aims to address all these criteria to provide an ideal solution for farmers.

The most commonly used management strategy is the manual removal of adult *O. inornatus* and egg capsules, although this strategy has minimal impact on already established populations of *O. inornatus* and thus is no longer enough. It has been suggested that the removal of adult *O. inornatus* would be significantly more effective than the removal of egg capsules (Buhle et al., 2004), however the authors point out that the labour cost involved in implementing both strategies must be considered before their use.

Another management strategy would be to use specialized fishing dredges on oyster plots to remove *O. inornatus*. This would involve the use of a dredge with a small enough mesh to catch the *O. inornatus*, while at the same time being large enough to let sand and other debris through. Only recently has an effective drill dredge been developed, designed by Oosterschelde oyster farmer Nico Boertjes. Upon processing an oyster plot 100 m by 75 m which had an *O. inornatus* density of 16.5 specimens per $m^2$, 100% of the live *O. inornatus* and 89% of the dead *O. inornatus* were found to have been removed from the plot (Hartog et al., 2017). However, this system is unable to remove the eggs of *O. inornatus*.

The use of tributyltin (TBT), a chemical component of certain anti-fouling marine paints popular in the mid 1900s, has also been observed to significantly reduce *U. cinerea* populations (Faasse et al., 2007). However, it was discovered that in areas with high rates of marine traffic while TBT was commonly used, many species of native marine snails were developing imposex and subsequently populations were drastically reducing (Santillo et al., 2002). The use of TBT has since been restricted, and its use to combat *O. inornatus* would pose too great of an environmental hazard.

Immersion of *M. gigas* in fresh water has been shown as an effective method of detaching *O. inornatus* from the *M. gigas*. A study showed that immersion time before detachment increased as the size of the *O. inornatus* increased, with an immersion time of 1.4 to 20.2 minutes expected for *O. inornatus* larger than 40 mm in length (Mueller et al., 1999). This is a very effective method of removal prior to transport of species between plots; however, it would be impractical to use this method while the oysters were in the ocean growing.

An established method of mitigating damage from *O. inornatus* is switching from bottom culture practices to off-bottom practices. In off-bottom oyster farms, oysters are raised in baskets or cages that are elevated from the sea floor away from the *O.* inornatus and are allowed to turn in the water. This method is very effective at keeping *O. inornatus* from feeding on cultivated oysters, but has a high initial and upkeep cost, thus it is not a viable option for many farmers.

## 5.4. THE NOVEL METHOD

Once further experiments have been conducted to confirm the results presented in this report, the novel mitigation method can be proposed to farmers. The farmers would be recommended to begin breeding the preferred prey of *O. inornatus* for use as bait to keep them out of the plots. The plots must be cleaned of all *O. inornatus*, possibly using a dredge, then the farmers could create a barrier of the preferred prey around their plots. This barrier would draw any *O. inornatus* present away from the cultivated oysters to feed on the bait. While cultivated oysters are growing, this barrier will need to be replenished.

# 6. CONCLUSIONS

The experiments described in this report compared whether the predation behaviour of *O. inornatus* was affected when presented with *M. gigas* of different origins. Upon analyzing the results of the two different runs of this experiment (experiment 1 and experiment 2), a significant predation preference was observed in both experiments. In experiment 1, comparing French and Oosterschelde brood stock, it was observed that the French *M. gigas* were more preyed upon, accounting for 97 of the 176 drilled *M. gigas*. In experiment 2, comparing French diploid stock raised in Kattendijke with French triploid stock raised in Yerseke, it was observed that the French diploid *M. gigas* raised in Kattendijke were more preyed upon, accounting for 107 of the 181 drilled *M. gigas*.

Although predation preferences were identified in both experiments, further experiments must be conducted before recommendations on mitigation strategies can be made to oyster farmers. Once the predation preference has been identified, then the proposed strategy of spreading desirable prey oysters around the cultivated oysters can be implemented.

# 7. RECOMMENDATIONS

It is recommended that future experiments into the feeding preference of *O. inornatus* focus on replicating the results observed during this experiment. In these future experiments, focus should be put into identifying why a preference was observed in this experiment. The main factors that should be monitored in the future are shell thickness of preyed upon oysters and the size class of preyed upon oysters.

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# APPENDICES

## APPENDIX A – HYPOTHESIS TEST RESULTS

**Table A - 1: F-Test Results from Experiment 1**

| | French Oysters | Oosterschelde Oysters |
|---|---|---|
| **Mean** | 0.29275362 | 0.226890756 |
| **Variance** | 0.23090664 | 0.187140025 |
| **Observations** | 345 | 357 |
| **df** | 344 | 356 |
| **F** | 1.23387095 | |
| **P(F<=f) one-tail** | 0.02475774 | |
| **F Critical one-tail** | 1.19238236 | |

**Table A - 2: t-Test Results from Experiment 1, assuming unequal variances**

| | French Oysters | Oosterschelde Oysters |
|---|---|---|
| **Mean** | 0.29275362 | 0.226890756 |
| **Variance** | 0.23090664 | 0.187140025 |
| **Observations** | 345 | 357 |
| **Hypothesized Mean Difference** | 0 | |
| **df** | 687 | |
| **t Stat** | 1.90647047 | |
| **P(T<=t) one-tail** | 0.02850307 | |
| **t Critical one-tail** | 1.64707464 | |
| **P(T<=t) two-tail** | 0.05700615 | |
| **t Critical two-tail** | 1.96342306 | |

**Table A - 3: F-Test Results from Experiment 2**

| | Kattendijke | Yerseke |
|---|---|---|
| **Mean** | 0.698529412 | 0.365517241 |
| **Variance** | 0.315849673 | 0.275191571 |
| **Observations** | 136 | 145 |
| **df** | 135 | 144 |
| **F** | 1.147744723 | |
| **P(F<=f) one-tail** | 0.207823583 | |
| **F Critical one-tail** | 1.321671433 | |

**Table A - 4: t-Test Results from Experiment 2, assuming equal variances**

| | Kattendijke | Yerseke Bank |
|---|---|---|
| **Mean** | 0.698529412 | 0.365517241 |
| **Variance** | 0.315849673 | 0.275191571 |
| **Observations** | 136 | 145 |
| **Pooled Variance** | 0.294864846 | |
| **Hypothesized Mean Difference** | 0 | |
| **df** | 279 | |
| **t Stat** | 5.1374653 | |
| **P(T<=t) one-tail** | 2.61938E-07 | |
| **t Critical one-tail** | 1.650333455 | |
| **P(T<=t) two-tail** | 5.23876E-07 | |

# APPENDIX B – EXPERIMENT 1 DATA

**Table B - 1: Data collected in the field for experiment 1, "drilled" includes partial boreholes**

| Cage and Zone | Number of OS Oysters | Number of Fr Oysters | Total Oysters in Cage | Number of OS Oysters Drilled | Number of Fr Oysters Drilled | Number of Drills |
|---|---|---|---|---|---|---|
| **1A** | 0 | 0 | | 0 | 0 | 16 |
| **1B** | 50 | 0 | | 14 | 0 | 33 |
| **1C** | 0 | 46 | | 0 | 14 | 35 |
| **1D** | 0 | 8 | 104 | 0 | 5 | 26 |
| **2A** | 88 | 0 | | 33 | 0 | 78 |
| **2B** | 1 | 44 | | 1 | 16 | 36 |
| **2C** | 2 | 9 | | 1 | 0 | 18 |
| **2D** | 5 | 52 | 201 | 1 | 18 | 66 |
| **3A** | 0 | 43 | | 0 | 17 | 35 |
| **3B** | 3 | 5 | | 3 | 2 | 16 |
| **3C** | 48 | 0 | | 8 | 0 | 22 |
| **3D** | 1 | 0 | 100 | 0 | 0 | 26 |
| **4A** | 5 | 0 | | 1 | 0 | 9 |
| **4B** | 0 | 49 | | 0 | 10 | 13 |
| **4C** | 6 | 1 | | 0 | 4 | 21 |
| **4D** | 39 | 0 | 100 | 13 | 0 | 54 |
| **5A** | 2 | 43 | | 1 | 18 | 51 |
| **5B** | 51 | 49 | | 15 | 12 | 58 |
| **5C** | 2 | 3 | | 2 | 2 | 25 |
| **5D** | 42 | 5 | 197 | 8 | 1 | 40 |

**OS: Oosterschelde brood stock**
**Fr: French brood stock**

**Table B - 2: Oyster (*M. gigas*) size class measurement data for experiment 1**

| | French Brood Stock | | | Oosterschelde Brood Stock | | |
|---|---|---|---|---|---|---|
| | **Length** | **Width** | **Height** | **Length** | **Width** | **Height** |
| **1** | 58.57 | 23.7 | 18.93 | 54.36 | 31.86 | 16.56 |
| **2** | 52.49 | 30.11 | 14.47 | 72.55 | 36.04 | 19.23 |
| **3** | 60.13 | 29.91 | 16.92 | 57.08 | 32.03 | 17.9 |
| **4** | 73.11 | 35.44 | 21.79 | 54.93 | 31.55 | 15.51 |
| **5** | 72.58 | 30.77 | 16.77 | 54.99 | 38.47 | 18.01 |
| **6** | 56.51 | 30.32 | 15.78 | 39.03 | 27.62 | 19.33 |
| **7** | 70.15 | 27.13 | 16.27 | 50.29 | 39.39 | 17.42 |
| **8** | 67.87 | 33.54 | 13.76 | 51.81 | 30.17 | 17.09 |
| **9** | 72.74 | 37.87 | 18.59 | 54.66 | 35.62 | 17.12 |
| **10** | 63.06 | 26.24 | 16.37 | 43.26 | 27.67 | 15.5 |
| **11** | 68.88 | 33.92 | 18.7 | 44.58 | 28.65 | 16.26 |
| **12** | 67.19 | 29.37 | 16.57 | 55.21 | 30.61 | 17.93 |
| **13** | 47.49 | 28.78 | 14.43 | 49.57 | 26.16 | 17.04 |
| **14** | 63.97 | 27.17 | 15.62 | 51.37 | 31.44 | 19.7 |

| | | | | | | |
|---|---|---|---|---|---|---|
| **15** | 70.79 | 32.03 | 20.04 | 51.26 | 32.08 | 16.7 |
| **16** | 60.8 | 32.25 | 13.47 | 47.16 | 30.52 | 18.98 |
| **17** | 66.81 | 32.92 | 24.8 | 49.96 | 32.43 | 19 |
| **18** | 58.13 | 32.45 | 14.56 | 61.54 | 30.72 | 16.22 |
| **19** | 77.83 | 34.15 | 23.25 | 52.55 | 29.8 | 17.59 |
| **20** | 56.94 | 28.66 | 14.05 | 48.48 | 33.59 | 20.23 |
| **21** | 69.74 | 29.36 | 17.14 | 56.79 | 26.06 | 20.21 |
| **22** | 71.97 | 32.11 | 15.55 | 52.89 | 27.57 | 15.23 |
| **23** | 65.62 | 31.2 | 15.93 | 49.69 | 24.9 | 15.2 |
| **24** | 71.46 | 34.21 | 21.68 | 49.37 | 29.65 | 17.41 |
| **25** | 67.75 | 32.93 | 17.85 | 59.17 | 27.1 | 15.79 |
| **26** | 76.98 | 27.74 | 16.02 | 35.28 | 25.58 | 13.35 |
| **27** | 48.78 | 30.54 | 14.96 | 67.31 | 26.06 | 15.97 |
| **28** | 55.4 | 28.88 | 13.69 | 53.24 | 30.96 | 19.05 |
| **29** | 53.48 | 30.42 | 15.84 | 60.96 | 25.17 | 14.67 |
| **30** | 75.81 | 33.83 | 20.76 | 48.17 | 32.19 | 19.04 |
| **31** | 78.89 | 39.83 | 16.56 | 50.68 | 29.35 | 16.86 |
| **32** | 81.49 | 30.01 | 18.28 | 41.01 | 29.36 | 17.58 |
| **33** | 68.59 | 32.06 | 22.38 | 46.35 | 31.04 | 19.07 |
| **34** | 75.16 | 33.99 | 22.97 | 47.31 | 27.79 | 15.21 |
| **35** | 71.03 | 28.02 | 16.43 | 59.22 | 28.59 | 12.99 |
| **36** | 54.23 | 29.69 | 18.04 | 47.06 | 31.13 | 19.35 |
| **37** | 68.7 | 32.91 | 17.49 | 56.09 | 28.09 | 14.51 |
| **38** | 68.31 | 33.92 | 15.98 | 45.77 | 26.35 | 18.67 |
| **39** | 76.63 | 38.33 | 17.57 | 56.93 | 32.01 | 17.64 |
| **40** | 85.44 | 33.56 | 14.94 | 47.35 | 35.78 | 20.51 |
| **41** | 68.2 | 32.85 | 22.23 | 64.26 | 31.36 | 18.72 |
| **42** | 58.9 | 30.68 | 17.64 | 52.33 | 25.26 | 21.36 |
| **43** | 58.04 | 25.97 | 17.6 | 46.61 | 32.18 | 17.95 |
| **44** | 68.88 | 42.85 | 16.03 | 54.03 | 32.35 | 13.91 |
| **45** | 64.68 | 32.86 | 13.16 | 49.4 | 25.62 | 16.27 |
| **46** | 53.49 | 29.93 | 19.83 | 52.64 | 26.5 | 17.53 |
| **47** | 55.11 | 22.81 | 17.14 | 57.49 | 32.25 | 18.93 |
| **48** | 44.04 | 30.52 | 19.67 | 57.28 | 29.07 | 15.01 |
| **49** | 53.05 | 27.24 | 15.42 | 62.73 | 29.73 | 15.78 |
| **50** | 71.65 | 32.38 | 19.55 | 52.23 | 25.6 | 15.67 |
| **Average** | 65.3508 | 31.3672 | 17.4694 | 52.4856 | 30.0214 | 17.2552 |
| **STD** | 9.38 | 3.74 | 2.80 | 6.90 | 3.40 | 1.94 |

# APPENDIX C – EXPERIMENT 2 DATA

**Table C - 1: Data collected in the field for experiment 2, "drilled" includes partial boreholes. Yellow rows represent cages set up as a repeat of experiment 1**

| Cage and Zone | Number of Kat/OS Oysters | Number of YB/Fr Oysters | Total Oysters in Cage | Number of Kat/OS Oysters Drilled | Number of YB/Fr Oysters Drilled | Number of Drills |
|---|---|---|---|---|---|---|
| **1A** | 25 | 0 | | 23 | 0 | 24 |
| **1B** | 13 | 4 | | 11 | 1 | 14 |
| **1C** | 0 | 12 | | 0 | 11 | 14 |
| **1D** | 7 | 28 | 89 | 6 | 18 | 31 |
| **2A** | 40 | 0 | | 12 | 0 | 45 |
| **2B** | 0 | 44 | | 0 | 16 | 26 |
| **2C** | 2 | 9 | | 1 | 0 | 7 |
| **2D** | 5 | 2 | 102 | 1 | 0 | 18 |
| **3A** | 2 | 3 | | 1 | 0 | 12 |
| **3B** | 5 | 35 | | 5 | 11 | 34 |
| **3C** | 40 | 5 | | 32 | 1 | 19 |
| **3D** | 1 | 3 | 94 | 1 | 0 | 18 |
| **4A** | 24 | 2 | | 11 | 1 | 20 |
| **4B** | 9 | 13 | | 6 | 11 | 13 |
| **4C** | 7 | 34 | | 4 | 14 | 28 |
| **4D** | 3 | 6 | 98 | 2 | 3 | 22 |
| **5A** | 2 | 0 | | 1 | 0 | 15 |
| **5B** | 0 | 49 | | 0 | 12 | 29 |
| **5C** | 2 | 3 | | 2 | 2 | 5 |
| **5D** | 42 | 0 | 98 | 8 | 0 | 27 |

**Kat: *M. gigas raised in Kattendijke***
**YB: *M. gigas* raised in Yerseke**
**OS: Experiment 1 *M. gigas* from brood stock in the Oosterschelde**
**Fr: Experiment 1 *M. gigas* from brood stock in France**

**Table C - 2: Oyster (*M. gigas*) size class measurement data for experiment 2**

| | Yerseke | | | Kattendijke | | |
|---|---|---|---|---|---|---|
| | **Length** | **Width** | **Height** | **Length** | **Width** | **Height** |
| **1** | 57.81 | 23.64 | 15.54 | 72.89 | 30.34 | 18.6 |
| **2** | 41.82 | 29.34 | 15.09 | 53.16 | 24.14 | 13.83 |
| **3** | 50.84 | 39.3 | 19.51 | 57.66 | 29.7 | 14.44 |
| **4** | 51.34 | 24.5 | 14.86 | 66.78 | 26.16 | 14.79 |
| **5** | 51.95 | 29.92 | 18.94 | 70.25 | 32.09 | 18.01 |
| **6** | 44.68 | 34.77 | 14.51 | 46.96 | 24.87 | 11.22 |
| **7** | 40.7 | 25.43 | 13.75 | 41.29 | 17.92 | 10.31 |
| **8** | 54.74 | 26.73 | 17.39 | 34.32 | 19.33 | 12.37 |
| **9** | 43.21 | 30.92 | 14.05 | 40.25 | 23.73 | 11.42 |
| **10** | 38.47 | 29.77 | 15.58 | 43.43 | 22.99 | 11.07 |
| **11** | 55.11 | 29.41 | 15.87 | 38.58 | 18.4 | 14.68 |

| | | | | | | |
|---|---|---|---|---|---|---|
| **12** | 53.33 | 29.93 | 21.43 | 51.67 | 23.49 | 13.52 |
| **13** | 53.29 | 33.74 | 18.06 | 37.58 | 18.32 | 11.75 |
| **14** | 45.85 | 31.21 | 15.34 | 39.64 | 22.52 | 9.73 |
| **15** | 38.64 | 25.65 | 13.26 | 33.68 | 23.12 | 11.2 |
| **16** | 49.12 | 30.49 | 18.49 | 35.23 | 18.75 | 8.6 |
| **17** | 45.94 | 27.67 | 14.15 | 27.59 | 19.32 | 10.01 |
| **18** | 41.66 | 24.34 | 14.5 | 33.5 | 20.59 | 10.76 |
| **19** | 48.15 | 30.77 | 14.21 | 30.32 | 16.6 | 9.96 |
| **20** | 49.1 | 39.01 | 16.23 | 38.68 | 22.12 | 14.5 |
| **21** | 44.83 | 35.14 | 15.7 | 45.48 | 20.56 | 11.98 |
| **22** | 45.99 | 32.29 | 18.16 | 46.86 | 21.97 | 11.83 |
| **23** | 37.12 | 26.6 | 12.5 | 42.76 | 24.11 | 12.3 |
| **24** | 34.7 | 22.72 | 14.03 | 45.93 | 23.17 | 11.74 |
| **25** | 38.91 | 29.94 | 11.46 | 45.5 | 21.96 | 11.95 |
| **26** | 38.86 | 25.71 | 11.54 | 32.55 | 19.58 | 10.1 |
| **27** | 53.39 | 26.84 | 18.48 | 71.22 | 27.45 | 18.27 |
| **28** | 36.8 | 30.48 | 18.47 | 44.93 | 21.16 | 10.2 |
| **29** | 41.94 | 24.99 | 14.23 | 43.75 | 28.31 | 13.36 |
| **30** | 40.71 | 26.28 | 14.91 | 58.1 | 26.8 | 18.29 |
| **31** | 39.24 | 30.74 | 15.64 | 37.79 | 18.59 | 14.83 |
| **32** | 48.65 | 26.04 | 20.94 | 53.74 | 28.81 | 17.09 |
| **33** | 39.06 | 32.05 | 13.52 | 61.18 | 25.98 | 15.16 |
| **34** | 35.65 | 22.19 | 11.27 | 71.27 | 34.63 | 18.09 |
| **35** | 41.4 | 25.05 | 14.94 | 63.91 | 26.98 | 16.73 |
| **36** | 53.19 | 31.37 | 19.95 | 52.87 | 22.81 | 10.74 |
| **37** | 46.84 | 32.92 | 17.82 | 53.46 | 21.52 | 11.89 |
| **38** | 37.6 | 29.32 | 10.84 | 65.46 | 28.86 | 17.69 |
| **39** | 31.28 | 20.49 | 9.93 | 57.52 | 27.93 | 13.29 |
| **40** | 36.74 | 23.92 | 13.56 | 50.2 | 22.8 | 11.12 |
| **41** | 58.13 | 29.11 | 19.01 | 52.66 | 27.8 | 14.21 |
| **42** | 36.29 | 26.82 | 12.27 | 74.72 | 30.32 | 22.51 |
| **43** | 32.42 | 28.19 | 12.3 | 48.23 | 17.74 | 8.41 |
| **44** | 38.31 | 26.19 | 12.64 | 52.73 | 21.12 | 11.63 |
| **45** | 44.44 | 30.25 | 16.63 | 49.15 | 28.43 | 14.58 |
| **46** | 40.47 | 29.79 | 13.11 | 39.11 | 18.84 | 8.92 |
| **47** | 50.39 | 29.47 | 16.03 | 37.28 | 21.75 | 9.68 |
| **48** | 51.87 | 31.34 | 15.49 | 31.69 | 22.63 | 12.85 |
| **49** | 44.82 | 33.43 | 13.59 | 35.29 | 21.27 | 11.85 |
| **50** | 51.31 | 29.03 | 19.83 | 31.8 | 17.75 | 11.94 |
| **Average** | 44.542 | 28.9048 | 15.391 | 47.812 | 23.5226 | 13.08 |
| **STD** | 6.92 | 3.93 | 2.76 | 12.56 | 4.27 | 3.08 |