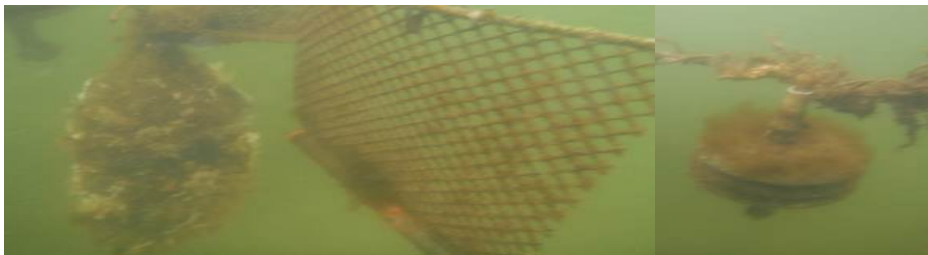


OYSTERECOVER: Testing the efficiency of different Spat Collectors

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Summary

This study identifies a way to develop more efficient spat collecting techniques that may compensate for the lower survival rate of the oysters caused by the *B. ostreae* parasite. It is part of the EU project Oysterecover aimed at investigating the causes and possible solutions to the decline in the European flat oyster (*Ostrea edulis*) due to the *Bonamia ostreae* parasite. Spat can be harvested directly from the field, or obtained by introducing substrate, also called collectors, in the water during the period that free-swimming larvae are present. Results of experimental testing of three selected spat collector types for flat oysters in three countries are presented. In an earlier stage of the Oysterecover project a review of the efficiency of different spat collectors was conducted with the intention of recommending three collector types to be tested in the field. From the review three spat collectors were selected for further testing in the field, these were mussel stockings, calcified Chinese hats and Vexar mesh. These collectors were tested in two different years. In 2011 all collectors were tested in the Netherlands, France and Denmark. In 2012 the three collectors were again tested in France while only the mussel stockings were tested in Denmark. For the second testing year in The Netherlands an earlier study of 2003 was used which included plastic tubes as a collector instead of Vexar mesh. The collectors were placed in the water in different weeks and stayed there for two months. After retrieval of the water, the amount of oyster spat per collector were analysed. Besides, in both studies in the Netherlands the oyster larvae present in the water over the experimental period was also monitored.

Results show that both the mussel stockings and calcified Chinese hats were successful in collecting oyster spat. The tubes tested in 2003 in the Netherlands were also successful. The Vexar mesh proved to be unsuccessful and was quickly disregarded as a possible option for spat collection. For the analysis, both the number of spat per collector and per cm² was calculated. There was a difference in results when the success of the spat collectors was analysed. When considering the number of spat collected per collector the mussel stockings were the most successful in all three countries, whereas the Chinese hats tended to be more successful when considering the number of spat collected per cm². This difference is due to the differing exposed surface areas onto which larvae can settle. In the mussel stockings the surface area of all the mussel shells are included in the calculations, however the larvae are unlikely to settle on the shells in the centre of the stocking as they are less exposed to the water, while because the entire surface area of the Chinese hat is equally exposed to the water, and the surface area is comparatively smaller than in the mussel stockings, more spat are likely to settle per cm².

The amount of spat collected varied between years. In general, in both Denmark and France fewer spat was collected in 2012 than in 2011. This shows that even with efficient collectors, other factors influence the availability of settling oyster spat. The results of the water samples showed that a peak in larval abundance was followed by a peak in oyster spat a week later. Fouling on the collectors was heavy in all three countries. The fouling was relatively easy to remove from the mussel stockings simply by removing the shells from the stocking. The calcium coating on the Chinese hats and tubes allowed easy removal of the fouling. Fouling on the Vexar mesh was particularly difficult to remove. To minimise the effects of fouling, timing the deployment of the collectors according to an increasing oyster larvae presence in the water is recommended. Aside from the number of spat collected on each collector type, other factors such as location, the available larvae in the water, financial cost and the following handling methods influence the actual efficiency of the collector type. Mussel shells appear to be the most successful collector in the Netherlands due to the method of direct re-dispersal of the young oysters into oyster culture plots. In France, Chinese hats are considered the most efficient spat collector because the efficiency of the handling methods rely on the removal of the young oysters from the collector and packing them into oyster bags with as little effort and bulk as possible. The oyster fishery in Denmark relies on wild oyster beds, so at present handling methods do not apply. However, considering only yield, mussel stockings appear to be the most successful spat collector type.

1. Introduction

The Oysterecover project is an EU project aimed at investigating the causes and possible solutions to the decline in the European flat oyster (*Ostrea edulis*) due to the *Bonamia ostreae* parasite (oysterecover.eu). Different aspects of transmission of the disease and demographic and genetic characterisation of tolerance to be used in restoration of natural beds and selective breeding programmes were investigated in the project. The Oysterecover study presented in this report concerns an alternative way to compensate for the lower survival rate of the oysters caused by the *B. ostreae* parasite. This is to enhance the amount of starting material by developing more efficient spat collecting techniques. Flat oyster (*Ostrea edulis*) larvae settle out of the plankton and onto hard surfaces where they develop into oyster spat. In order to settle, the larvae need appropriate hard surfaces and enough surface area. Spat collectors are designed to provide artificial surfaces for spat to settle on so that they can be collected, farmed and eventually sold by oyster growers.

In an earlier stage of the Oysterecover project a review of the efficiency (including economical profitability and user-friendliness) of different spat collectors was conducted with the intention of recommending three collector types to be tested in the field (Van den Brink, 2012). From that review three spat collectors were selected for further testing in the field. These were: Chinese hats, Vexar mesh and mussel stockings. Chinese hats are a standard oyster spat collector. They are used commercially in many countries, particularly in France. Chinese hats are textured plastic cones which can be assembled into towers. Research has shown that coating the hats in a calcium-based coating not only helps attract the settling oyster larvae, but makes it much easier to remove them once they have grown. Vexar mesh is a strong and sturdy plastic mesh available at any shellfish fishery hardware store. It is strong, but can be bent and it provides a large surface area for larvae to settle onto. This idea came from Bataller et al. (2006) who conducted a study in Canada where various collectors were tested and this mesh appeared the most efficient because not only did it collect a good amount of spat, but it was cheap, easy to store, easy to use and by bending it, it was easy to remove the oyster spat. Mussel shells are very successful in collecting larvae. Once the oysters grow, they will outgrow the shell they are attached to as the shell will disintegrate and break away, allowing the oyster to be almost unattached when it is full grown. However, if too many oysters settle close together, or they settle on a curved or creviced part of the shell, they can grow in a warped shape and not be appealing to consumers. In the Netherlands, oyster growers scatter mussel shells on the sea floor (600 shells per m²) and later gather them up when harvesting. However, with this method, efficiency is dependent on being able to gather as many shells up as possible after the spat fall in order to redistribute them in growing areas. To solve this problem oyster growers may keep all the mussel shells together in a plastic mesh stocking to allow easy retrieval.

These three collector types were then tested in the field in two different years in the Netherlands, Denmark and France. In 2011 all collectors were tested in all three countries. In France, all three collectors were tested again in 2012 while in Denmark the mussel stockings were only tested in 2012 again. A study from 2003 was used as a second year comparison for the Netherlands (Kamermans et al., 2004). The spat collectors were examined for their efficiency in providing a substrate on which larvae can settle, thereby concentrating the number of oyster spat.

2. Materials and Methods

Preparation of spat collectors

Chinese hats

Three replicate Chinese hats were assembled into towers on a centre tube with which it can be attached. The towers were then dipped into a calcium solution. In France and the Netherlands the solution was made from 50% calcium (calcium dihydroxyde and magnesium dihydroxyde) and 50% of water. In Denmark the solution consisted of a mixture of 40% lime, 60% sand and water added until you have a slurry/suspension of thin yoghurt. Once dipped in the calcium solution, the hats were left to dry in the sun for about three days until the calcium was completely hardened (*Figure 1 and 4*).



Figure 1. Prepared tower of three replicate Chinese hats used in the Netherlands.

Vexar Mesh

Replicate 30 x 30 cm squares of 14mm Vexar mesh were prepared by attaching a steel rod to one side to prevent it from floating (*Figure 2 and 4*).



*Figure 2. Prepared 30 x 30 cm square of Vexar mesh with attached steel rod used in the Netherlands.
One square was one replicate in the experiment.*

Mussel Stocking

In the current study small mesh sacks were filled with 50-200 mussel shells (depending on country and year) and sealed. One sack was used as one replicate (*Figure 3 and 4*).



Figure 3. Prepared replicate mussel stocking used in the Netherlands.

Replicates similar to those used in the Netherlands were also used in Denmark (*Figure 4a*). In France replicates were deployed as a whole set which could be easily retrieved (*Figure 4b*) .



Figure 4a. Mussel stockings, Chinese hats and Vexar mesh used in Denmark.



Figure 4b. The set of replicate Chinese hats, mussel stockings and Vexar mesh as used in France.

Deployment and retrieval of spat collectors

In the Netherlands, a series of poles joined by cable was set up in Lake Grevelingen (near Stampersplaat) in an open grid formation. Between 13 June and 26 August 2011 a series of three replicates of each collector type was attached to the cable (*Figure 5*). In Denmark (Limfjord), a similar installation was used onto which the test collectors were attached. In France (Bay of Brest), the collectors were deployed onto existing installations already being used for spat collection using Chinese hats. In all counties, a new series of collectors was deployed every week for 10 weeks so that the optimal time for larval settlement could be identified.

Once all the collectors had been deployed they were left until retrieval in November to allow the spat to grow to an easily visible size. The collectors were then retrieved from the water and brought to the lab

where they were meticulously analysed and all oyster spat was removed, the species were identified and the number of individuals per species counted.

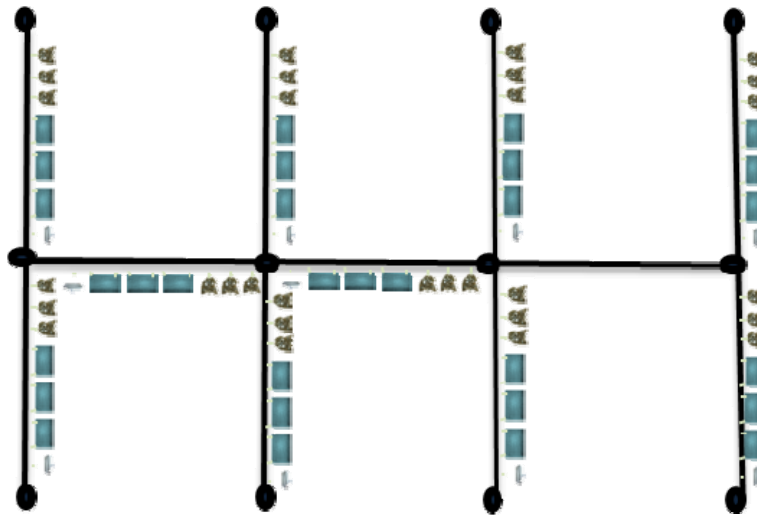


Figure 5. Schematic of the organisation of the pole/cable installation and how the collectors were arranged (one series deployed each week for 10 weeks) in Lake Grevelingen in the Netherlands.

Larvae collection

In the Netherlands, a sample of water from the same area was analysed each week for the presence of oyster larvae. This was done for the same ten weeks the deploying of collectors was conducted. A 100 μ m plankton net was used to filter 100 L of water. The samples were then fixed in 4% buffered formalin (diluted with sea water) for later microscopic examination in the lab. The numbers of flat oyster larvae, Pacific oyster larvae and larvae of other shellfish were counted.

2003 study in the Netherlands

Because available resources did not allow for a repeat of the experiment the following year in the Netherlands, results of the similar experiment conducted in 2003 by Kamermans *et al.* (2004) were used.

In the summer of 2003 several spat collectors were used to measure the recruitment of both the flat oyster, *Ostrea edulis* and the Pacific oyster, *Crassostrea gigas* in the Grevelingenmeer, The Netherlands on behalf of the local oyster fisheries. This report considers the data collected at Veermansplaat; the test location closest to the Stampersplaat installation used in 2011. Towers of 21 Chinese hats were used along with 5L onion sacks of mussel shells. 'Tubes' were also used in this experiment. These were long rough plastic cylinders about 1.2 m long and with a diameter of 2.2 cm in groups of 35. In this study, a 'collector' therefore refers to either a tower of 2 hats, a group of 5 tubes or 1 five litre sack of mussel shells (Figure 6). All collectors were suspended in the water column attached to a rope tied between two poles. New collectors were deployed every week for seven weeks, after which they were collected and analysed for the number of spat present.

Chinese hats	
Tubes	
Sacks of mussel shells	

Figure 6. Chinese hats (top), tubes (centre) and sacks of mussel shells (bottom) in week 3 of the spat collector experiment (from Kamermans et al. 2004).

Calculations of yield

The number of flat oyster spat was calculated both for each collector (whole replicate) as well as per cm² because the collectors were of different shapes and surface areas.

To determine the number of spat per cm² the following calculations were made:

$$Scm^2 = Sc / SA$$

Where Scm² is the number of spat per cm², Sc is the number of spat per collector and SA is the surface area of one collector replicate.

The surface area of each Chinese hat was taken from Kamermans et al. (2004) where it was calculated as 377 cm².

The surface area of the Vexar mesh was calculated to be ¼ of the surface area of the mesh in total due to the holes in the mesh. This was calculated to be 225 cm².

The surface area of the mussel stockings was determined by estimating the surface area of a single mussel shell and multiplying it by the number of mussel shells in the sack.

The surface area of a mussel shell was calculated with the equation from Reimer and Tedengren (1996):

$$SA = l \times (h^2 + w^2) \times e^{0.5} \times (\pi / 2)$$

Where l = length, h = height and w = width (all in cm). For consistency the measurements for an average sized shell was used so that the equation was:

$$4 \times (0.5^2 + 1.75^2) \times e^{0.5} \times (\pi / 2) = 34.3 \text{ cm}^2$$

In the Netherlands, an average of 71 mussel shells were used in each mussel stocking, ranging from 56-96 shells per stocking. In Denmark in 2011 46-58 shells were used per collector, and in 2012 50-183 shells were used per collector. In France about 200 mussel shells were used per collector.

Statistical analysis

Differences between collectors in number of spat were tested with Analysis of Variance (ANOVA). The difference in number of spat between two deployment weeks in Denmark was tested with a t-test.

3. Results

The Netherlands

2011

During the retrieval of the collectors it was apparent that there was a great amount of organisms attached to the collectors (fouling) on almost all collectors. However, to some degree the type of fouling species attached to the collector depended on which week the collector was deployed. Collectors from the first week of deploying were heavily fouled with mussels (*Mytilus edulis*), in the second week mussel fouling also appeared, but much less than in the first week. During the other weeks, the spat collectors were generally fouled with algae, bryozoans and ascidians to varying degrees. All three collector types were fouled on retrieval, but the fouling could be removed from the Chinese hats due to the calcium coating, and from the mussel stockings when the mussel shells were removed from the mesh. Fouling was difficult to remove from the Vexar mesh, and searching for oyster spat involved manually searching through the fouling (*Figure 7*).

Unfortunately in the second, third and fourth week during the experiment, three towers of Chinese hats were lost, and as all replicates were attached to the same central tube, no data was recovered for Chinese hats in these weeks.



Figure 7. Examples of collectors after retrieval from the first week of deployment (left) and the ninth week of deployment (right). Top: Chinese hats, Middle: Vexar mesh, Bottom: Mussel stockings.

Once the flat oyster spat was removed from the collectors and counted, the number of flat oyster spat per collector and per cm² was calculated and compared with the larval abundances for each week.

When considering the numbers of spat per collector, the mussel stockings appeared the most successful of the three collector types. Mussel stockings collected significantly more flat oyster spat per collector every week of the experiment (ANOVA, $F_{(1,41)}=0.003$, $p<0.01$ –data excluded from calculation for weeks where replicates went missing) (Figure 8). On the Chinese hats the most flat oyster spat was found on replicates deployed in week 29 (the fifth week of the experiment) with an average of 264 spat followed

by week 27 (the third week of the experiment) with an average of 223 spat. The number of spat found per Vexar mesh collector was negligible in comparison with the other collectors.

In the Netherlands, water samples were examined as well. Flat oyster larvae were always more numerous in the water column than the larvae of other shellfish species (*Figure 8*). There was an obvious peak in oyster larval abundance in the water column in both week 28 (the fourth week of the experiment) with 472 oyster larvae per 100 L and in week 33 (the ninth week of the experiment) with 450 oyster larvae per 100 L. The first peak in larval abundance in week 28 was followed by a peak in oyster spat collected on the mussel stocking a week later in week 29.

When considering the number of flat oyster spat per cm² the Chinese hats collected significantly more spat than the mussel stockings (ANOVA, $F_{(1,41)}=71303.74$, $p<0.01$ –data excluded from calculation for weeks where replicates went missing) (*Figure 9*). The most spat per cm² was found on Chinese hats in week 30 (the sixth week of the experiment) with 0.05 spat per cm² after which there was a gradual decrease with 0.034 spat per cm² in week 31 and 0.27 spat per cm² in week 32.

The peak in oyster spat per cm² in week 30 followed two weeks after the first peak in larval abundance in the water column in week 28.

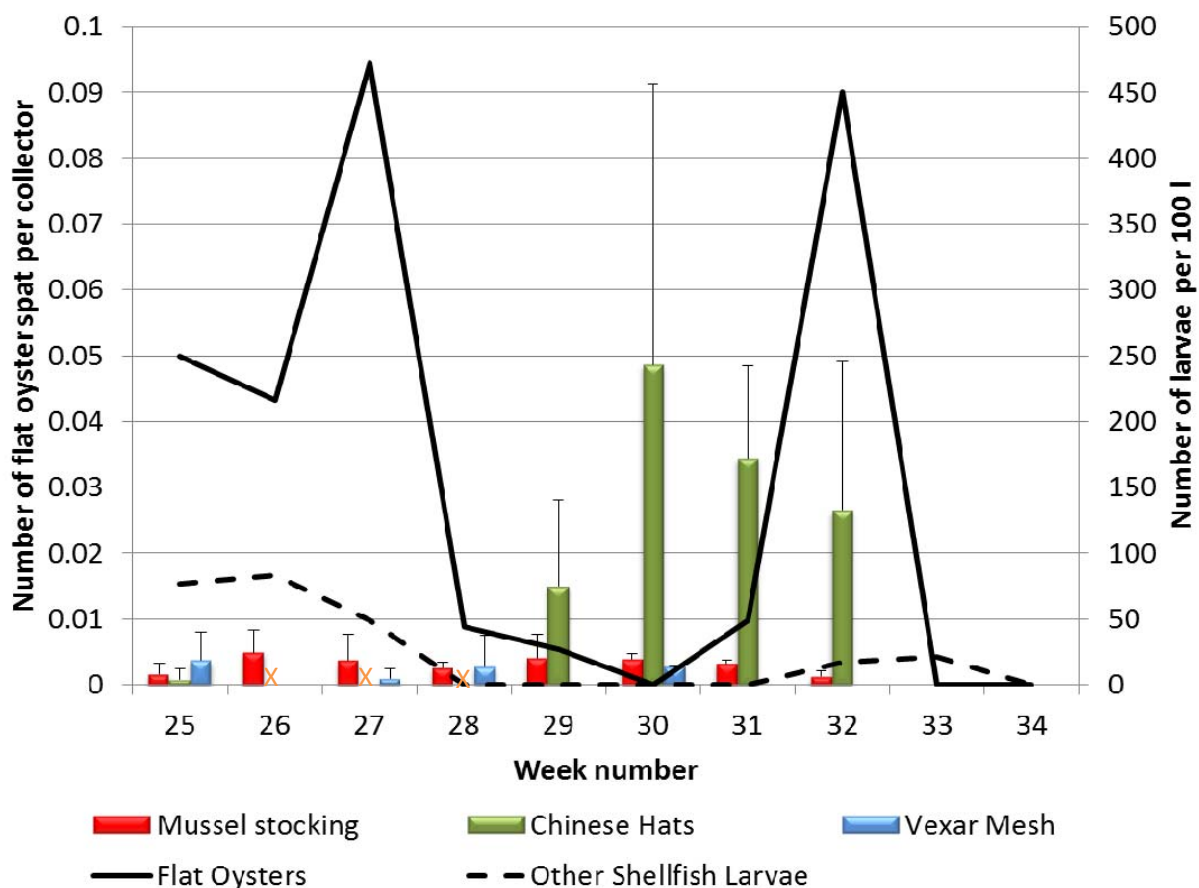


Figure 8. Bars represent the number of flat oyster spat found per collector (order as shown below graph) in 2011 over ten weeks of deploying a series of collectors each week (+ 1 S.D.) (left y-axis) and lines represent the number of flat oyster and other shellfish larvae per 100 L present in the water column at the same time in the Netherlands (right y-axis). X indicates Chinese hat collectors that went missing.

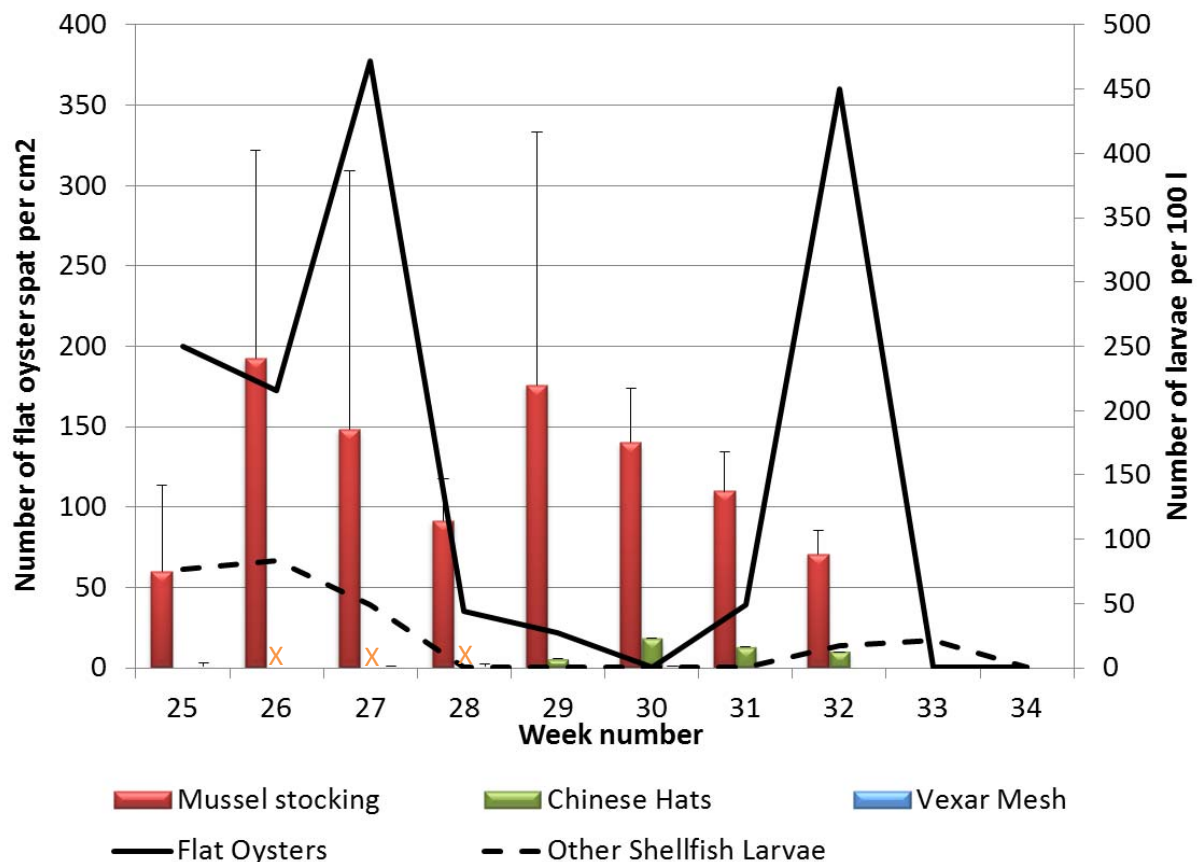


Figure 9. Bars represent the number of flat oyster spat found per cm² in 2011 over ten weeks of deploying a series of collectors (order as shown below graph) each week (+ 1 S.D.) (left y-axis) and lines represent the number of flat oyster and other shellfish larvae per 100 L present in the water column at the same time in the Netherlands (right y-axis). X indicates a Chinese hat collectors that went missing.

2003

Fouling was generally quite heavy on all collectors, but worst on the mussel sacks. The mussel sacks were most successful at collecting spat when considering both spat per collector and spat per cm² (Figure 10). The tubes were also successful at collecting spat in weeks 27-29, particularly in week 28 where more spat was collected on the tubes (672 spat) than on any other collector during the whole experiment. While spat was collected on the Chinese hats, there were much fewer compared with the other two collectors. As the peak in larvae numbers in the water appeared near the end of the deploying period, no trend relating larval *O. edulis* and spat collected was observed in the data.

There was a high abundance of Pacific oyster (*Crassostrea gigas*) larvae in the water in 2003 (Figure 10) compared with 2011 (Figure 128 and Figure 139) and in 2003 *C. gigas* dominated the collectors (Kamermans et al. 2004). Regardless of this competition for space, more flat oyster spat was collected per cm² in 2003 compared with 2011.

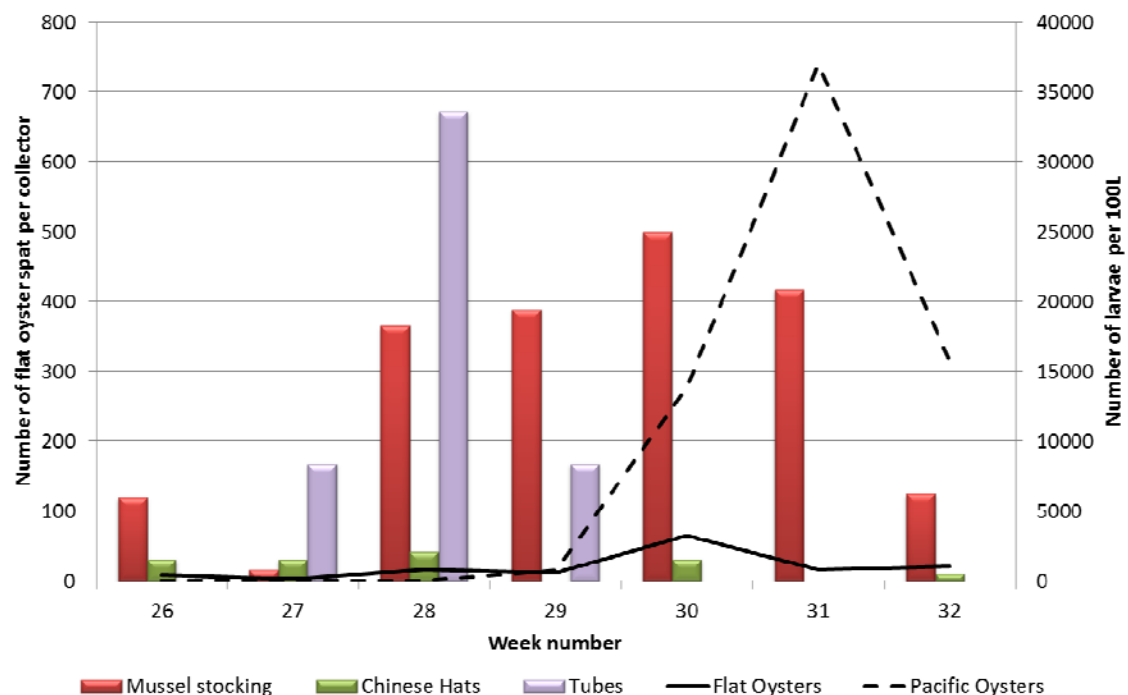


Figure 10. Bars represent the number of flat oyster spat found per collector (order as shown below graph) in 2003 over seven weeks of deploying a series of collectors each week (left y-axis) and lines represent the number of flat oyster and Pacific oyster larvae per 100 L present in the water column at the same time in the Netherlands (right y-axis, data for standard deviation not available).

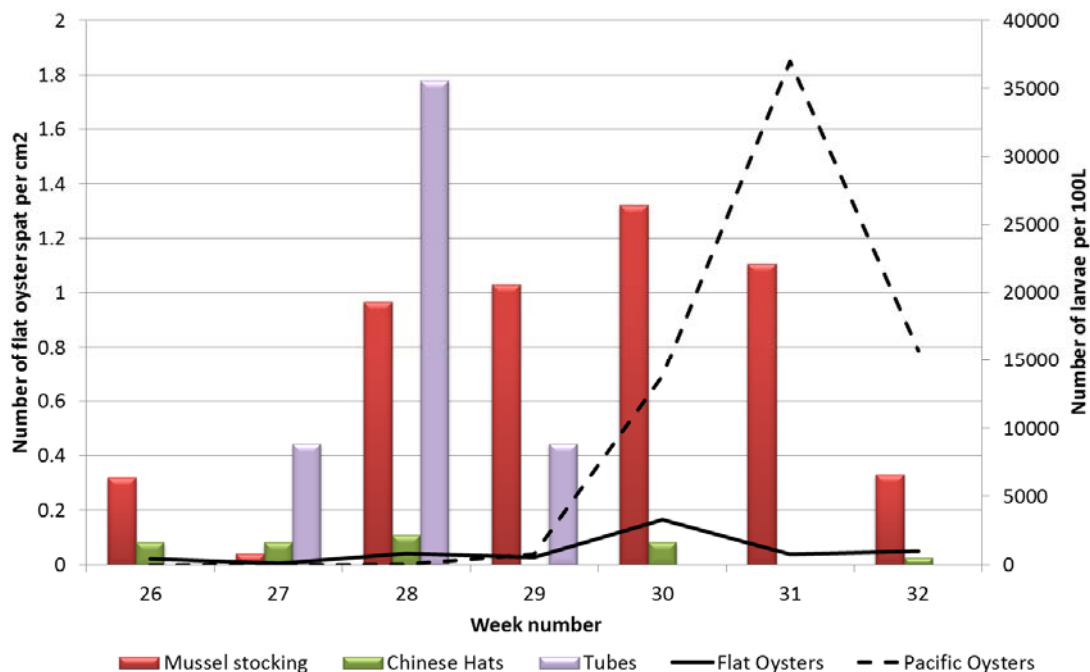


Figure 11. Bars represent the number of flat oyster spat found per cm² in 2003 over seven weeks of deploying a series of collectors (order as shown below graph) each week (left y-axis) and lines represent the number of flat oyster and Pacific oyster larvae per 100 L present in the water column at the same time in the Netherlands (right y-axis, data for standard deviation not available).

Denmark

In Denmark heavy fouling, consisting mostly of ascidians and barnacles, was observed. The most fouling was present on collectors deployed in the first four weeks of the experiment. As in the Netherlands, a lot of mussels were found on the collectors deployed in the first weeks. In some cases, particularly on collectors deployed in the first week of the experiment, the fouling by ascidians was so heavy there was little room available for oyster spat to settle and/or grow.

The largest spat was found on the Chinese hats (an average of 7 mm), and these were generally easy to remove. Spat on the mussel shells were noticeably smaller (an average of 3 mm) and more fragile and broke easily with handling. In several cases the Vexar mesh had come loose from the installation and had to be retrieved from the bottom. These replicates had no attached spat.

2011

Significantly more flat oyster spat was found on the mussel stockings than the Chinese hats in Denmark per collector (Figure 12) (ANOVA, $F_{(1,40)} = 822276$, $p < 0.01$), while there was no significant difference in number of spat collected between the Chinese hats and mussel stockings per cm^2 ($F_{(1,40)} = 1.16$, $p > 0.01$). No spat were found on the Vexar mesh from any week (some were lost or retrieved from the bottom).

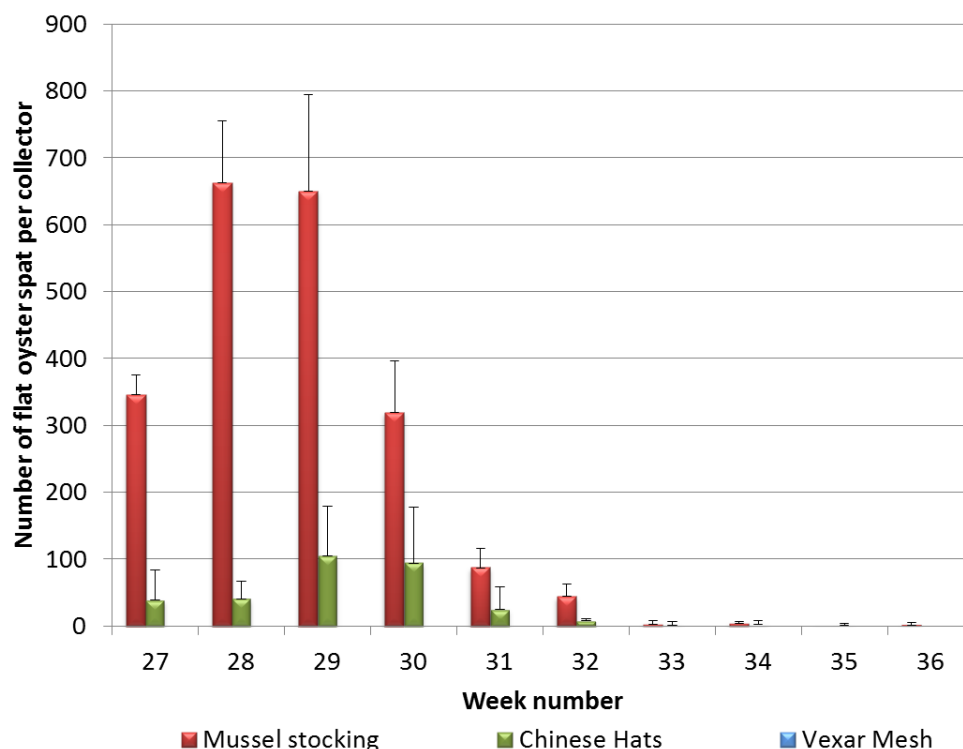


Figure 12. The number of flat oyster spat found per collector (order as shown below graph) in 2011 over ten weeks of deployment a series of collectors each week in Denmark (+ 1 S.D.).

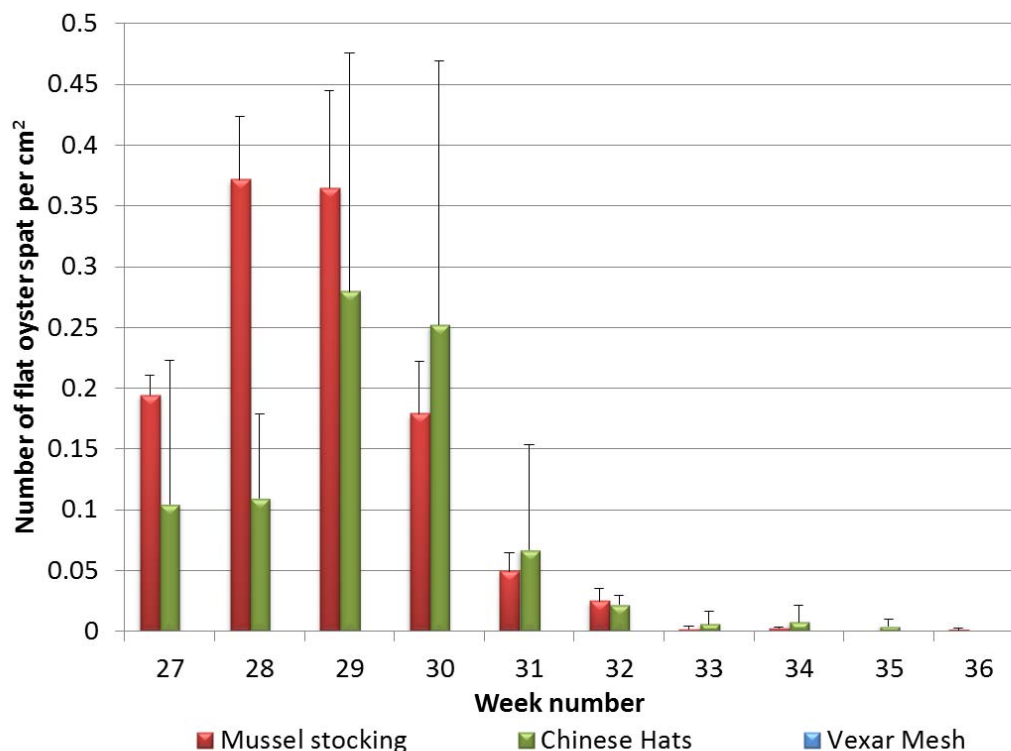


Figure 13. The number of flat oyster spat found per cm² in 2011 over ten weeks of deployment a series of collectors (order as shown below graph) each week in Denmark (± 1 S.D.).

2012

In 2012 only the mussel stockings were tested in Denmark to indicate the differences in spat collection between years. Nine replicate mussels stockings were deployed in the water on 11-7-2012 (week 28) and nine more were deployed a week later on 18-7-2012 (week 29). These weeks gave the highest yield in 2011. The average of the replicates deployed on each date was compared for whole mussel stockings (Figure 14) and cm² (Figure 15). Although replicates deployed on 18-7-2012 collected on average more spat (23 spat per collector and 0.0048 spat per cm²) than those deployed on 11-7-2012 (17 spat per collector and 0.0044 spat per cm²), there was no significant difference between the two weeks (t-test $p > 0.1$). This is caused by a high degree of variation between the replicates. Compared to 2011 (Figure 12 and Figure 13) there were much fewer spat collected on the mussel stockings in 2012.

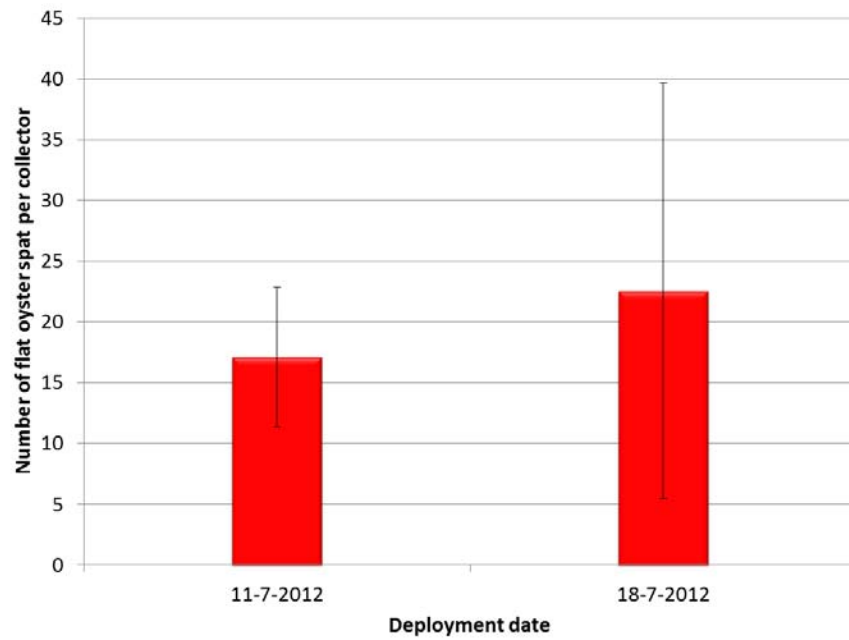


Figure 14. The number of flat oyster spat found per mussel stocking in 2012 over two weeks of deployment a series of nine replicates each week in Denmark (± 1 S.D.).

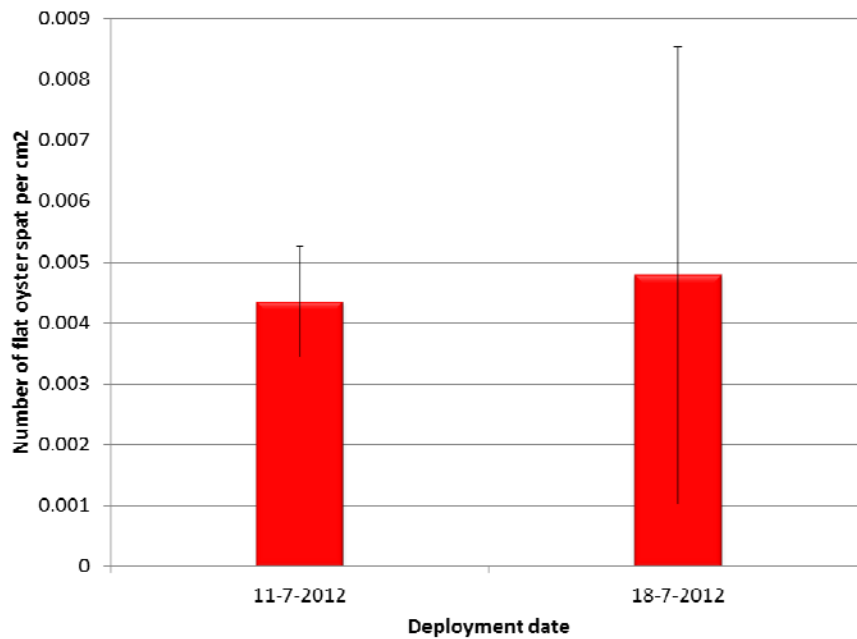


Figure 15. The number of flat oyster spat found per cm² of mussel stocking in 2012 over two weeks of deployment a series of nine replicates each week in Denmark (± 1 S.D.).

France

2011

In France, collectors were fouled mostly with mussels. All collectors were lost in weeks 30 and 33, while in week 27 the mussel stockings were lost and could not be retrieved. Mussel shells collected the most spat in France when considering the number of spat collected per collector and Chinese hats collected the most spat per cm² (Figure 16 and Figure 17). When considering the number of spat per collector, the most spat was collected in week 28 on the mussel stockings (548 spat) while when considering the number of spat collected per cm² the most spat was collected in week 27 on the Chinese hats (0.24 spat). Vexar mesh collected by far the smallest number of spat. The most spat was collected in the first weeks of the experiment while in the last week no spat was found on any of the collectors.

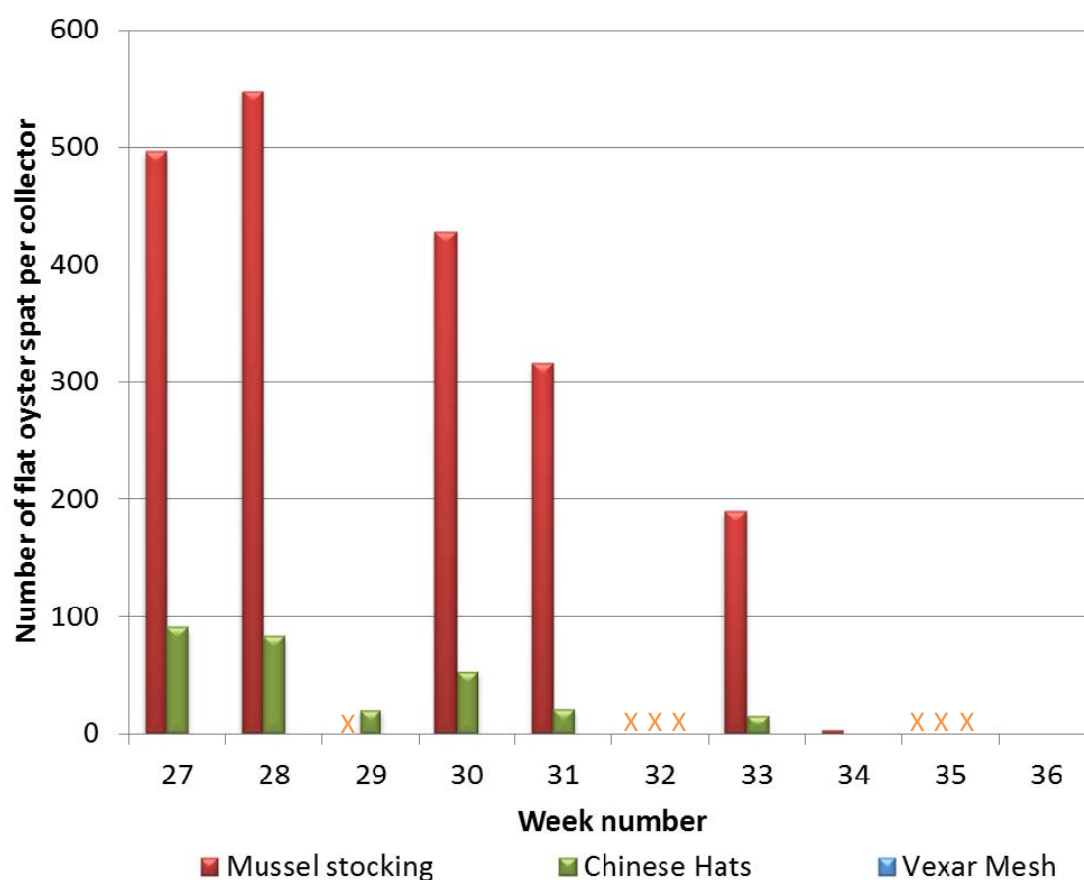


Figure 16. The number of flat oyster spat found per collector (order as shown below graph) in 2011 over ten weeks of deployment a series of collectors each week in France (data for standard deviation not available). X indicates a collector that went missing.

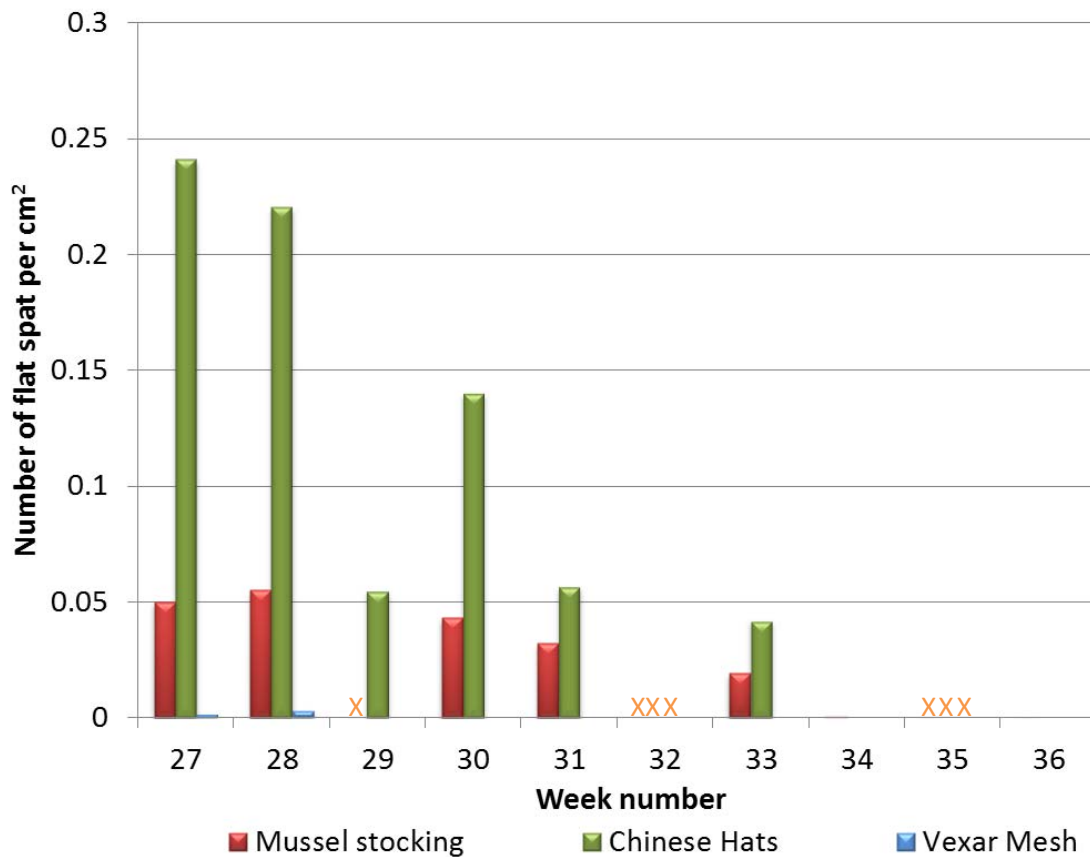


Figure 17. The number of flat oyster spat found per cm² in 2011 over ten weeks of deployment a series of collectors (order as shown below graph) each week in France (data for standard deviation not available). X indicates a collectors that went missing.

2012

In 2012 a similar trend was observed. Again fouling of the collectors took place. More spat was collected on the mussel stocking when considering the number per collector (Figure 18), while more spat per cm² was collected on the Chinese hats (Figure 19). In 2012 the most spat was collected in week 33 with a maximum of 4130 spat per collector (mussel stockings) and 0.1 spat per cm² (Chinese hats). After a high yield in week 33 no spat were collected in week 35. Collectors from week 34 and 36 were missing. The Vexar mesh only collected spat in weeks 29 and 30, but considerably fewer than the other collectors when considering per collector, but more than the mussel stockings when considering per cm². Compared to 2011 (Figure 15 and Figure 16) there were fewer spat collected in 2012, with the exception of week 33 where there was far more spat than in 2011.

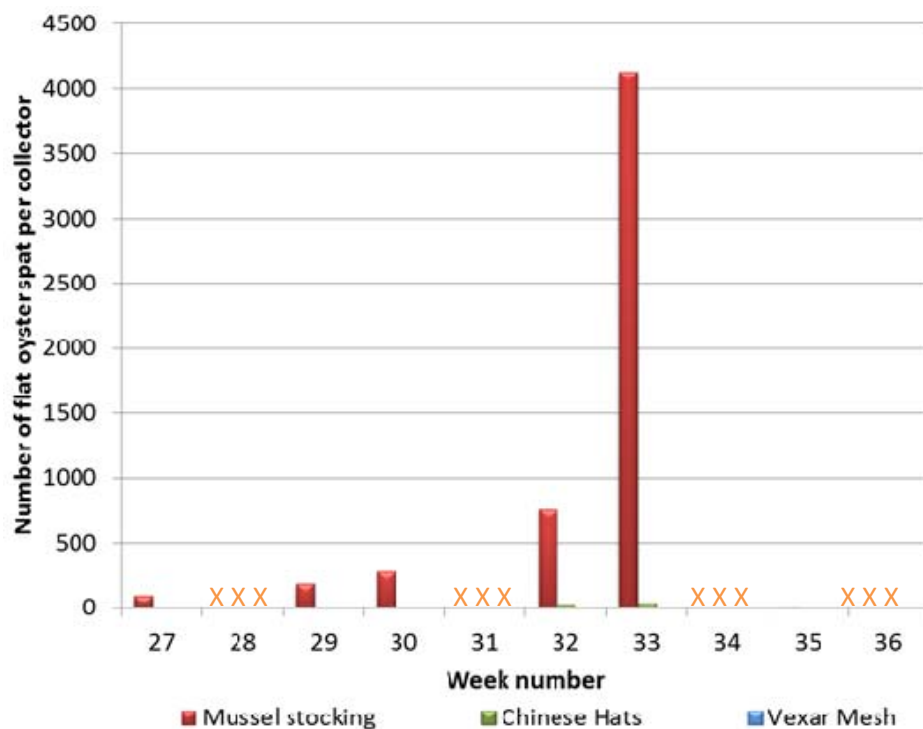


Figure 18. The number of flat oyster spat found per collector over ten weeks of deployment a series of collectors (order as shown below graph) each week in France (data for standard deviation not available). X indicates a collectors that went missing.

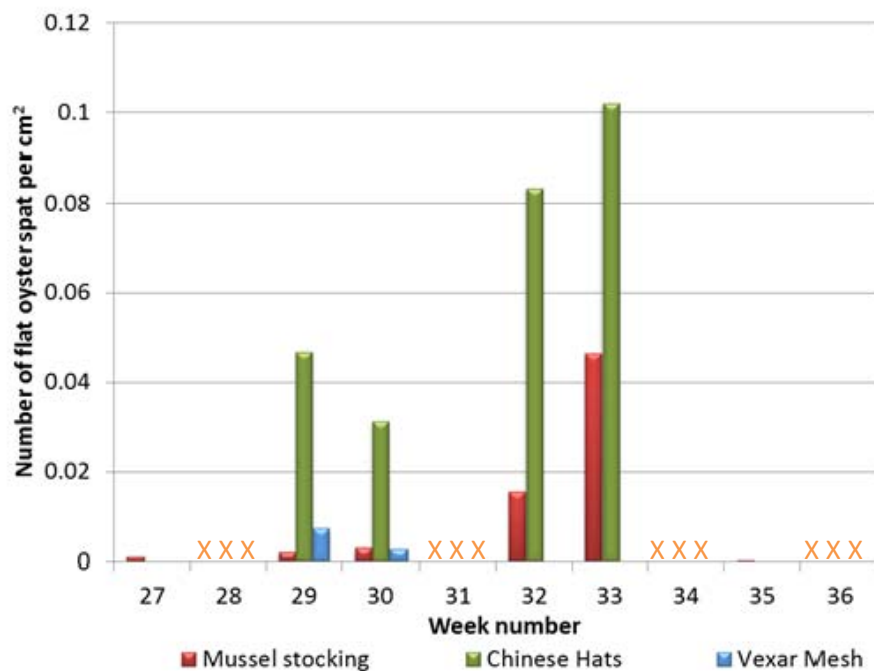


Figure 19. The number of flat oyster spat found per cm² in 2012 over ten weeks of deployment a series of collectors (order as shown below graph) each week in France (data for standard deviation not available). X indicates a collectors that went missing.

4. Discussion

The aim of this study was to enhance the amount of starting material by developing more efficient spat collecting techniques as an alternative way to compensate for the lower survival rate of the oysters caused by the *B. ostreae* parasite. The mussel stockings and calcified Chinese hats tested provided the type of surface on which oyster spat can successfully settle and grow. The tubes tested in 2003 in the Netherlands were also successful. The Vexar mesh proved to be particularly unsuccessful and was quickly disregarded as a possible option for spat collection.

There was a difference in results depending on the calculations used to compare the success of the spat collectors (whether number of spat per whole collector or per cm²). When considering the number of spat collected per collector the mussel stockings were the most successful in all three countries. Whereas when considering the number of spat collected per cm², the Chinese hats were generally more successful (except in Denmark, 2011 where the numbers of spat on the mussel stockings and Chinese hats were almost comparable). This is most likely due to the available surface area provided by the two collectors. Mussel shells provide favourable surface area on both sides and in the crevices of the shell. Depending on how many mussel shells comprised each mussel stocking, the surface area can be quite large. However, if the volume of a mussel stocking is too large, the usable surface area decreases because the mussels in the centre of the stocking are no longer accessible to the larvae who tend to only settle on the outside shells directly exposed to the water. This was seen in the Dutch and French mussel stockings which were essentially a sack filled with many (around 71 in the Netherlands and 200 in France) shells. The shape of these stockings resulted in many shells filling the inside of the sack with only those on the outside effectively exposed to the water. The results therefore show a large amount of spat collected per whole collector (on the outside shells only), and due to the large number of shells, very few spat collected per cm². In contrast, the stockings used in Denmark had fewer shells (around 50), and therefore had more exposed surface area, and accordingly a high number of spat collected both per whole collector and cm². In comparison, the Chinese hats provide less total surface area than the mussel stockings, but all of the surface area is exposed equally to the water. This allowed a more reasonable and calculable difference between the number of spat collected per whole collector and per cm². The tubes used in the Netherlands in 2003 showed a similar situation where more surface area is exposed equally to the water, and therefore they also collected reasonably high numbers of spat both per collector and per cm². Therefore, the difference in exposed surface area should be taken into account when interpreting these results. Considering these differences, it seems that if the surface area of the mussel shells could be optimally utilised, the mussel shells would be the most successful spat collector.

The amount of spat collected varies between years. One highly successful season of spat collection is no guarantee that the following season will be equally successful. This can be seen in the difference in results of the mussel stocking in Denmark in 2011 and 2012. Much fewer spat were collected in 2012 compared with 2011. Furthermore, there was much variation between individual replicates, indicating that general success in spat collection is very difficult to predict. In France there were also generally fewer spat collected in 2012 than in 2011, with the exception of week 33 where there was far more spat than in 2011.

The number of larvae in the water and the number of surviving spat vary between years and the onset of spawning seasons can vary. The availability of larvae in the water can be influenced by temperature, available food, competition, and the general health of the spawning adults. Therefore, regardless of how efficient a spat collector is, the yield is difficult to predict and initially depends on the number of surviving larvae in the water.

Fouling was a problem in all three countries and on all spat collectors. Algae, bryozoans and ascidians all accumulated and grew on the collectors, smothering the settled oyster spat or preventing larvae from settling in the first place. Furthermore, mussel (*Mytilus edulis*) and Pacific oyster (*Crassostrea gigas*) spat also settled on the collectors, competing with *O. edulis* spat for space. Due to the calcium coating, fouling was possible to remove from the Chinese hats and tubes with some effort, while on the Vexar mesh the fouling was thick and particularly difficult to remove. The fouling on the mussel stockings was easier to remove as the sack, onto which most of the fouling was attached, could simply be opened and the relatively clean shells removed for inspection. The timing of which the collectors are deployed may help to decrease the problems caused by fouling. In the Netherlands there appeared to be a spike in spat collected about one to three weeks after a spike in the number of larvae observed in the water. In order to minimise the fouling on the collectors preventing the oyster spat from settling, the collectors should be deployed as close to the settling of the larvae as possible. It would therefore be useful to regularly sample the larvae in the water, and deploy the collectors only when a high number of larvae are detected. However, because oyster breeding success varies from year to year, it is therefore difficult to predict when a peak in larvae numbers occurs, or what a 'high number' of larvae for that year will be. Therefore deploying collectors when larvae numbers are detected to be on the increase will be a feasible option.

All of the tested collector types require preparation, financial cost and can be labour intensive. The effort involved in the preparation of each collector; filling the stockings with mussel shells, building towers of Chinese hats and calcifying Chinese hats and tubes must be considered when comparing the efficiency of the collectors, but it is difficult to quantify in order to compare. According to Van den Brink (2012), the cost of the equipment can be compared per collector as Chinese hats: €10.50, Mussel stockings (24 x 5L sacks + shells) €6.00 and tubes €35.00. This suggests that mussel shells are the cheapest option except that Chinese hats and tubes can be re-used (if properly cleaned) whereas the mussel shells cannot. However, the re-use of the Chinese hats or tubes would require more labour to properly clean them, and may still not be of sufficient condition to re-use.

Considering the information gathered during these experiments, it appears that the most efficient means of spat collection are mussel shells well exposed to the water. Oyster growers in the Netherlands utilise a method of dispersing mussel shells on the bottom and gathering them up after the oyster larvae have settled and grown into spat of manageable size. This method involves human and mechanical labour during collection (but little in preparation). However there is the possibility of bottom sediment smothering the spat if it covers the shells, and some loss as not all mussel shells will be retrieved and predation on the oyster spat. Nevertheless, there is also minimal fouling as the mussel shells lie on the bottom as opposed to being suspended in the water column, and there is maximum mussel shell surface area exposed to the water. If certain adjustments could be made, mussel stockings may be even more successful. One can think of suspending long, narrow mussel stockings in the water with maximum exposed shell surface area at a commercial scale. Or in some other way elevating the mussel shells slightly so that they are not in direct contact with the sediment. For example, making collection more efficient by perhaps dispersing the mussel shells on an open net that can be easily gathered up. Workers at a local fishery supplies Warehouse in Yerseke, the Netherlands, Machine Fabriek W. Bakker B.V., have developed a machine which can quickly fill long, narrow mussel stockings for use by commercial oyster fishers. If cost and labour could be kept to a minimum, this may be an efficient development in oyster spat collection.

The handling of the spat following collection also plays a role in the efficiency of collectors. In the Netherlands oyster growers collect spat and re-disperse them almost immediately on the bottom in oyster culture plots so they can grow into adults. This method favours the use of mussel shells as a collector because the mussel shells initially provide a portable hard surface for the spat and young

oysters, which will disintegrate as the oyster grows, thus resulting in an unattached adult oyster that can be collected again for sale.

In France, however, spat are generally collected on Chinese hats. This is due to the lack of easily available mussel shells and the opinion of the fishers that mussel stocking is too labour intensive to prepare and to collect and that Chinese hats are more successful in collecting spat. Furthermore, in France the young oysters are mechanically removed from the Chinese hats and packaged in large oyster bags for transport and acclimation on shore or in deep water to remove fouling. The oysters are later brought to growing areas and left to grow inside oyster bags (Mathieu Hussenot, CRC Bretagne Nord pers. comm.). With this method mussel shells would provide too much bulk inside the bags and therefore increase the cost of the handling.

In Denmark the oyster fishery is based on wild oysters so is not relevant in this case. If oyster farming would start in a new location, the choice of collector should depend on the on-growing method: mussel shells when on-growing takes place on culture plots and Chinese hats for on-growing in oyster bags.

Judging the efficiency of spat collectors involves various considerations including the measurements used, the location, financial cost, the available larvae in the water and the following handling methods. All of these factors must be taken into account and will most likely result in different spat collectors being considered the most efficient in different situations.

5. Conclusions

Both the mussel stockings and calcified Chinese hats were successful in collecting oyster spat. The Vexar mesh proved to be unsuccessful. Fouling on the collectors was heavy in all three countries. The fouling was relatively easy to remove from the mussel stockings simply by removing the shells from the stocking. The calcium coating on the Chinese hats and tubes allowed removal of the fouling. Fouling on the Vexar mesh was particularly difficult to remove. Timing the deployment of the collectors according to an increasing oyster larvae presence in the water is recommended to minimise the effects of fouling. Mussel shells appear to be the most successful collector in the Netherlands due to the method of direct re-dispersal of the young oysters into oyster culture plots. In France, Chinese hats are considered the most efficient spat collector because the efficiency of the handling methods rely on the removal of the young oysters from the collector and packing them into oyster bags with as little effort and bulk as possible. The oyster fishery in Denmark relies on wild oyster beds, so at present handling methods do not apply. However, considering only yield, mussel stockings appear to be the most successful spat collector type.

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7. Quality Assurance

IMARES utilises an ISO 9001:2008 certified quality management system (certificate number: 57846-2009-AQ-NLD-RvA). This certificate is valid until 15 December 2015. The organisation has been certified since 27 February 2001. The certification was issued by DNV Certification B.V. Furthermore, the chemical laboratory of the Environmental Division has NEN-AND-ISO/IEC 17025:2005 accreditation for test laboratories with number L097. This accreditation is valid until 1 April 2013 and was first issued on 27 March 1997. Accreditation was granted by the Council for Accreditation.

8. References

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