





## THESIS APPROVAL SHEET

Title of Thesis: Advancing Technology for Restoration: An Investigation of the Solar Oysters Production System

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Advisor: Tamra Mendelson

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### **Housing and Residence Life, George Mason University**

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Acker-Carter, D., Giron, A., Hampton, A., Rowell, K., "Future Oceans – Bridging Sustainability priorities for Early Career Professionals" Sustainability Research Innovation 2021

## ABSTRACT

Title of Document:                   ADVANCING TECHNOLOGY FOR  
RESTORATION: AN INVESTIGATION OF  
THE SOLAR OYSTERS PRODUCTION  
SYSTEM

Darryl Acker-Carter, Degree, 2023

Directed By:                         Professor, Tamra Mendelson, Biological  
Sciences

The Solar Oyster Production System (SOPS) was developed to expand growing capacity while decreasing the area required to grow oysters and reducing labor demands. SOPS is the first system in the world to grow oysters on continuously rotating ladders, however, little is known about the effect of consistent rotation on oyster growth. The goal of this study was to understand (1) the impact of rotation on growth of the *Crassostrea virginica* and (2) how the performance of oysters on SOPS compares to performance on traditional gardening methods. Oysters were deployed on rotational and static ladders on SOPS as well as oyster gardening cages provided by the Chesapeake Bay Foundation (CBF). The findings from this study reveal how consistent rotation affects shell shape and condition, and they provide baseline metrics for the further development of the technology.

ADVANCING TECHNOLOGY FOR RESTORATION: AN INVESTIGATION  
OF THE SOLAR OYSTERS PRODUCTION SYSTEM

By

Darryl Acker-Carter

Thesis submitted to the Faculty of the Graduate School of the  
University of Maryland, Baltimore County, in partial fulfillment  
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Estuarine and Environmental  
Science  
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## Chapter 1: Oyster Biology and Farming in the Chesapeake Bay

### Overview

Oysters have been providing critical ecosystem services for the Chesapeake Bay and a sustainable food resource for Chesapeake Bay residents for well over a millennium, but human activity has devastated this natural resource. Solar Oysters LLC has invented a new oyster growing technology, the Solar Oyster Production System (SOPS), that could revolutionize the way oysters are grown for restoration and aquaculture. This technology could also open the door for a new generation of diverse farmers and conservationists.

My master's thesis research investigates the efficacy of the Solar Oysters technology for aquaculture and restoration. Here, I outline the history of oysters in the Chesapeake Bay, oyster biology in relation to feeding and growth, different types of oyster farming practices, the effect of tumbling on oysters, and different biofouling management practices in the oyster aquaculture industry. Understanding the biology of oyster growth is crucial, as the one of the main foci of this study is the evaluation of oyster growth on SOPS. Tumbling is a key feature of SOPS, as tumbling aids in the development of shell shape and can be a tool for biofouling management on the system.

### The Oyster in the Chesapeake Bay

The eastern oyster, *Crassostrea virginica* (Gmelin 1791), is a water filtering bivalve native to estuarine ecosystems from the Gulf of St. Lawrence in Canada throughout

east coast of the United States, the Gulf of Mexico, and even in parts of the West Indies (Sellers & Stanley, 1984). Wild oyster populations globally have been declining over the past century primarily due to overfishing, pollution, and disease (Wilberg et al., 2011). As a result of the decline of the wild oyster population, the filtering capacity of oyster populations has been reduced by 80 % (Ermgassen et al., 2013). In the Chesapeake Bay region, the eastern oyster was known for its abundance and its versatility as a natural resource, but today, the oyster population in the Bay is less than 1% of its original population prior to the colonization of the Americas (Wilberg et al., 2011).

As a result of the decline in eastern oysters, the Chesapeake Bay is losing vital ecosystem services that the mollusk provides, such as: improving water quality through water filtration and bio deposition, providing habitat for important recreational and commercial fish species, increasing landscape diversity, and carbon sequestration (Coen et al., 2007). Additionally, oysters increase their feeding activity during the spring in relation to the rising temperatures, when they would feed on the annual phytoplankton bloom. The larger phytoplankton blooms in the past years have become a major issue due to the decline of the eastern oyster's abundance (Newell, 1988). Oysters are essential to the Chesapeake Bay ecosystem, and their decline has presented a challenge to people and wildlife who call it home. Thus, efforts to restore their population have been taking place for several decades.

In 2014, the states making up the Chesapeake Bay watershed signed the Chesapeake Bay Watershed agreement, committing to reducing nutrient pollutants, fostering stewardship, improving water quality, and creating and maintaining sustainable fisheries. This led to the goal of restoring oyster populations within ten tributaries of the Bay by 2025 (Agreement, 2014). In order to meet this goal, a variety of restoration actions took place, such as planting seed from hatcheries and habitat enhancement by adding additional substrate. A variety of different organizations, including government agencies such as the Maryland Department of Natural Resources, the Virginia Marine Resource Commission, and non-profit organizations like the Chesapeake Bay Foundation and the Oyster Recovery Partnership, use a combination of these restoration actions to aid in oyster restoration efforts (Kennedy et al., 2011). One of the more popular options is using spat-on-shell oysters to add reef substrate and oyster spat at the same time. Although the 2014 Chesapeake Bay Watershed agreement only addresses 10 specific tributaries, thousands of oyster bars have received restoration plantings or actions.

Even with all the restoration efforts taking place, oyster populations in the bay are far from being truly recovered. One of the shortcomings of restoration efforts lies with the monitoring efforts of restored reefs, where restoration practitioners lack substantive quantitative information on the impact of restoration efforts across the bay (Kennedy et al., 2011). Oyster restoration in the bay faces several additional challenges: the impact of disease, habitat degradation, and unpredictable recruitment. Recruitment in particular is one of the leading factors that is limiting the success of



oyster restoration, as there are many factors influencing recruitment, such as water quality and substrate availability. Restoration practitioners can enhance recruitment in the short term, but establishing long term recruitment proves to be more challenging, especially with the increase in dead zones and declining water quality in the bay (Mann & Powell, 2007).

A study by Kennedy et al (Kennedy et al., 2011) reviewed oyster restoration efforts in the bay up to 2006 to answer questions about restoration success or failure and data collected from monitoring activity. They concluded that there are not enough data available to determine if current efforts were successful. Yet, we are seeing short term success in restoration efforts in terms of planting oysters in the designated tributaries in the watershed agreement, with 7 of the 10 tributaries having completed their restoration efforts (Maryland and Virginia Oyster Restoration Interagency Workgroups, 2022). One popular oyster restoration practice is oyster gardening, a practice where oysters are grown near the water's surface off piers and docks to expose them to higher concentrations of phytoplankton, their food source (Goldsborough & Meritt, 2001). This practice allows people and communities with access to waterfront properties to participate in oyster restoration. Although a lot of restoration has occurred, work remains to be done examining the impacts of restoration and examining recruitment dynamics on the restored reefs.

## Oyster Biology

### Feeding

*Crassostrea virginica* is a filter-feeding bivalve. Their gills are primarily used for respiration, but they also play an essential role in feeding. Oysters engage in suspension filter feeding as their gills use different ciliary tracts to take particles in from the pumped water to the mantle cavity where the particles go through the digestion process (Gossling, 2003). There is a positive allometric relationship between filtration rate and tissue dry weight, meaning that the larger the oyster, the greater the filtering capacity (Gossling, 2003).

One of the oyster's key physiological features that provides an ecosystem service is the excretion process. Oysters will absorb organic matter through the digestion process, leaving the inorganics to be excreted as a fecal ribbon or as pseudofeces, which are carried away from the oyster by water flow (Galstoff, 1963). In their pseudofeces, there is matter that the oyster did not select for consumption, such as excess nutrients and other inorganic matter. Their pseudofeces become a part of the sediment where it undergoes denitrification, a process whereby microbes transform into harmless dinitrogen gas, unable to be used by algae that may drive the eutrophication process (Newell et al., 2002). Denitrification is a naturally occurring process that can be amplified by the filtration and bio deposition of oysters. Being able to do this at a high volume and efficiently, oysters tend to have higher absorption efficiencies than other bivalves, which can lead to faster growth (Galimany et al, 2017).

## Growth

Many factors can influence the growth of an oyster. After spawning, oysters see the greatest and fastest shell growth within its first three months of life. Environmental factors such as temperature, salinity, food availability, turbidity, and exposure out of water are the main factors influencing growth (Sellers & Stanley, 1984). How these factors fluctuate can also play a role in how oysters grow. Additionally, growth is directly related to phytoplankton density and abundance as it is the main food source for oysters (Sellers & Stanley, 1984).

The three dimensions of bivalves are the shell height (the distance between the umbo or hinge of the oyster and the bill), length (the distance between the anterior and posterior margin parallel and the hinge), and the width (the greatest distance between the top and bottom shell of a closed oyster) (Galtsoff & U.S. Fish and Wildlife Service, 1964). The shape of an oyster's shell is variable and largely dependent on its surrounding environment. An oyster's shell will grow where there is space. As a result of their need for space, an oyster may grow a shell that is long and slender or a shell that is round. Oysters have the capacity to grow both shell and their bodies (meat) simultaneously although not necessarily at equal rates. For many calcifying species including the eastern oyster, tissue growth is more energetically costly than shell growth (Watson et al., 2017).

## Current oyster aquaculture gear and farming practices

### Extensive Aquaculture

Extensive aquaculture is a type of farming in which the natural ecosystem of the raised species provides support for production. Cageless on-bottom oyster aquaculture, a type of extensive aquaculture, was one of the first forms of oyster aquaculture. One of the more traditional practices uses already existing oyster reefs, and adding additional oyster cultch (oyster shell, limestone, alternative substrate) to the structure. Farmers will cultivate them over the season and harvest in 2-3 years after oyster have grown to market size (minimum 76 mm).

Extensive culture allows for high levels of production; however, cageless on-bottom culture, which is common in the Chesapeake, can experience high levels of predation, irregular shell shape, and even poor recruitment year to year (Walton & Swann, 2021). Oysters from cageless on-bottom oyster culture are primarily targeted for the shucked market rather than the half-shell market. On-bottom culture can be a good investment depending on where the farmer's leases are, as consistent maintenance of the oysters and gear is not always needed, and it can save cost on fuel and labor. On the other hand, a farmer cannot see if an oyster has reached market size until harvest which can result in harvesting inefficiencies (Webster, 2007).

### Intensive Aquaculture

Intensive aquaculture refers to culture that controls the aspects that affect growth and production. For oyster aquaculture this is caged culture. With the challenges that arise

from extensive, intensive was created to improve the control a farmer has over the product. Intensive uses floating cages, rafts, or bottom cages to grow oysters, and there are several advantages to this style of production compared to on-bottom culture, such as: promoting faster growth by controlling stocking densities, decreasing predation, biofouling control, improving shell shape, and increasing the consistency of the produced product (Walton et al., 2012). A variety of different off-bottom systems have been developed to meet the needs of farmers.

Floating systems are the most common form of off-bottom culture and can be in the form of floating rafts or cages. Most floating systems in Maryland are near shore, and floating systems are a common practice for commercial purposes due to the higher survival of oysters in the floating system and ability to maintain and influence the condition of the oyster (Webster, 2007). As it is a newer system in the bay, particularly in Maryland, the permitting process to obtain leases for floating systems can be challenging, as there are several state departments involved in the process, and the department's involvement can result in a lengthy permitting period. One of the biggest concerns with floating systems is their proximity to near shore environments, as elevated levels of bacteria create a health concern for the oysters if they were to reach the market and were consumed. Despite these challenges, floating culture in Maryland has existed and operated within the state for the past three decades and continues to grow (Webster, 2007).

Adjustable long-line systems are a common off-bottom practice that use a tensioned line strung between pilings within a riser post that has interval height adjustment levels within the water column. This gives the farmer the ability to raise and sink their oysters in and out of the water as they see fit, creating a system where farmers can desiccate their oyster cages and control biofouling (Walton et al., 2020). This system is not currently used in Maryland.

A FlipFarm system is another off-bottom system that was developed in New Zealand. FlipFarm systems allow for a streamlined process of managing oysters for biofouling using the flip bag method. In this system, floating cages are connected by a singular line that runs through all the cages, allowing the gear to stay together in the open water. When a farmer decides they need to flip their cages for conditioning, biofouling management, or pest control, they will use a helix flipper or a FELIX. The FELIX is a modified rack that is attached to the side of a motorized boat that can flip the baskets in a FlipFarm system. The line of oyster baskets is fed through the FELIX, and as the boat moves along the line, the baskets are flipped (Moore et al., 2021). Flip farming reduces the labor need for controlling fouling, allows for efficient harvesting, and improves the economic viability of an oyster farm (FlipFarm New Zealand, 2023). As flip farming is a newer system, studies are still needed to evaluate the effect of the system on shell shape and condition, but it does have a promising outlook for the future of automated oyster aquaculture. This system is not currently used in Maryland.

## Tumbling Oysters

Tumbling oysters is a practice whereby oysters are rolled on top of each other, such that the extremities of the oyster shell are trimmed or broken. Tumbling is used as a farming practice since it can make an oyster more cupped and improve its depth and width in relation to its height (Cheney, 2010). Several studies have been conducted to identify the ideal shell shape, aiming for a 3:2:1 ratio in relation to shell height, length, and width respectively, and the tumbling process can promote the oyster to grow its shell to this ideal ratio (Thomas et al., 2019, Mizuta & Wikfors, 2019). Additionally, when repairing its shell, oysters produce higher levels of glycogen, and this is thought to improve the flavor of the oyster (Cheney, 2010). Tumbling is usually conducted simultaneously with shell washing which can be used to remove some of the biofouling that may occur on the oyster (Marshall & Dunham, 2013). Between improving the shape and flavor of the oyster, as well as decreasing biofouling, tumbling also allows for creating a uniform shape of the oyster, decreasing the variation in shell shape and size across a farm. Ultimately, tumbling can therefore lead to the development of an oyster that is more marketable to consumers.

Tumbling oysters can be a very useful tool to any oyster farmer to create a marketable oyster; however, there are some downsides to tumbling oysters. Oysters that are tumbled can experience lower growth rates in height, length, and width, which is primarily seen in oysters that are 18 months or less in age (Robert et al., 1993). However, one study suggests that it is unclear whether the increased glycogen

reserves in an oyster from the tumbling process is concurrently reducing the growth rate (Poirier & Quijón, 2022). Another thing to consider with tumbling oysters is the labor cost that comes with removing oysters from the water, tumbling them, and putting them back into the water. Tumbling oysters have both advantages and disadvantages but the use of the practice will ultimately be up to the farmer and what works best for them on their farm.

### Biofouling Management

One of the key challenges of any aquatic operation is finding an appropriate way to manage biofouling. In bivalves like the eastern oyster, fouling on the shell will occur, because the oyster has no mechanism to remove or prevent it. In terms of building a healthy ecosystem, biofouling may not be a problem. However, in aquaculture, biofouling can lead to competition between individuals, reduction of flow within cages, and even mortality events, which is why proper biofouling management is essential (Watson et al., 2009). In bivalve aquaculture, biofouling has been estimated to account for 20-30% of all production cost; however, it is key to note that type and amount biofouling will vary from the cultivated species and the geographic area that the aquaculture practice is taking place (Lacoste & Gaertner-Mazouni, 2015).

To control biofouling, farmers can use methods such as mechanical scraping/brushing, power washing, flipping floating bags, UV exposure, high salinity dip, freshwater rinse or even gear rotation (Adams et al., 2011). The method of control used differs between farms, and so will the financial cost and efficiency. The



cost of biofouling can be increased labor, damage to gear, or even fuel or energy costs. Finding a cost-effective control method is essential to every oyster farm (Adams et al., 2011). Certain biofouling methods can have different effects on the production of oysters on a farm. In long line systems, for example, when a farmer raises the oyster cages out of the water to desiccate, those oysters have less time in the water feeding, which may influence their overall condition. However, if a proper desiccation pattern is in place, overall growth of the oyster may not be as negatively affected (Mallet et al., 2009). Regardless, even though removal practices can reduce the overall growth and health of an oyster, these strategies are essential to implement to ensure that biofouling is controlled, and the stock makes it to market (Sievers et al., 2017).

### *The Solar Oyster Production System*

A new technology is currently in development that seeks to combine the benefits of off-bottom culture and tumbling into one aquaculture system, called the Solar Oyster Production System, or SOPS (SolarOysters, LLC). SOPS is an automated oyster aquaculture system that uses rotating ladders to slowly tumble oysters throughout the day, in order to influence shell shape and improve the meat quality of the oyster ( Figure 1.1). The prototype platform is 40 by 25 feet long, with 5 rotational ladders powered by 12 solar panels. Each ladder can hold 115 SEAPA (an Australian aquaculture gear supplier) baskets, and each of the baskets has an optimal stocking density of 450 one-inch oysters or 90 three-inch oysters. SOPS was designed to address the challenges of labor cost from tumbling and biofouling control through

air drying and UV exposure. Additionally, using the solar panels, the system aims to reduce the energy costs needed to tumble oysters and control biofouling.

The SOPS technology has the potential to revolutionize new ways to grow oysters for restoration, commercial aquaculture, and attract a new generation of oyster farmers.

However, to get to that point, the technology must be evaluated to quantify how well oysters grow on the platform, and if the SOPS rotational function is successful in improving the shape and condition of the oyster.

The aim of this study is to evaluate the performance of oysters on SOPS and assess how growth, condition, and biofouling differ between SOPS's rotational ladders, static ladder, and most importantly how growth on SOPS compares to traditional oyster gardening methods. This is because SOPS shows potential for use in the restoration sector as well as the commercial aquaculture space. Thus, this study evaluates the performance of both spat-on-shell and seed oysters on the platform.

## Chapter 2: Evaluation of oyster growth on the Solar Oysters Production System (SOPS)

### Introduction

The Solar Oysters Production System (SOPS) is the only oyster aquaculture system that uses an automated continuous rotating system to grow oysters (Fig 2.1). SOPS has five ladders that constantly rotate throughout the day, tumbling the oysters on the platform (Figure 2.2). The rotational function of the ladders is designed to influence the shape of the oyster shell to make uniform shell shape and to promote the development of a deeper cup in the oyster.

In traditional aquaculture, the practice of tumbling involves placing oysters into a rotating metal cylinder. This rotation trims off the extremities, thickens the shell, promotes the development of a deeper cup in the oyster, and even increases flavor through the promotion of glycogen for repairing the chipped shell (Robert et al., 1993, Cheney, 2010). The process of tumbling oysters is costly in both time and money as it requires a farmer to haul oysters out of the water, put them through a tumbler, and then put them back into the water. In oyster restoration practices, tumbling oysters does not occur often if at all. One of the main functions of mechanical tumbling in commercial aquaculture settings is to be used as a grader sorting the large oysters from the smaller ones. The grading process is not often used in restoration practices as the varying sizes of the oysters is not a concern, as the primary focus is to get as many oysters as possible regardless of the irregular sizes of the oysters. One of the beneficial aspects of tumbling is the promotion of the deeper

cup to produce an oyster with higher meat content. This could be beneficial for restoration as oysters with more meat could potentially increase their survival when they are transplanted to a reef.

SOPS includes the tumbling process within the growing system by rotating continuously and producing the same tumbling action within the grow out cages themselves. This minimizes the need for additional resources, as the system is being powered through photovoltaics.

Being the first the first system of its kind, it is not known how oysters will perform or if they will even grow on this system. The aim of this study is therefore to evaluate (1) if SOPS can grow oysters for restoration and commercial aquaculture and (2) if the continuous rotational function of SOPS is able to increase the sizes of oysters of oysters on the system. Therefore, for this study we seek to answer two questions: “Do oysters grow to similar sizes and experiences similar survival rates on SOPS as in conventional restoration gardening baskets?” and “does rotating oysters in a continuous system affect shell height, condition, and survival?”. The study examined the performance of the system by measuring oyster shell height, survival, meat condition index, fan ratio, and cup ratio in different growing treatments. Condition metrics (meat condition index, fan ratio, cup ratio) are often used in the commercial aquaculture industry to identify qualities of highly marketable oysters (Brake et al., 2003). Additionally, meat condition can be an indicator of oyster growth, as oysters under stressful conditions will put more energy into developing their shell rather than

their bodies or meat (Cheney, 2010). Therefore, by quantifying various metrics of oyster growth, this study will evaluate the effectiveness of SOPS for oyster aquaculture and restoration.

### Site Description

SOPS was located at Maritime Applied Physics Corporation (MAPC) in Baltimore, Maryland along the mouth of the Patapsco River (Figure 2.3). It operates off the company's floating dock. This area of Baltimore is industrial and historically, oysters did not grow here. This is primarily due to the lower salinity due to the freshwater coming from the Patapsco River.

The 40 x 25-foot platform is outfitted with five ladders, each with baskets that can rotate on a continuous loop. The ladders are lengths of chain separated by steel bars every 12 inches, from which can hang, at maximum capacity, 578, 29"x6"x11" SEAPA (SEAPA Oyster Basket Systems | Single Seed Farming Technology, n.d.) baskets made from a recyclable polypropylene plastic (Figure 2.2).

### Experimental Design

In October 2021, SolarOysters LLC received spat-on-shell oysters from the Chesapeake Bay Foundation's (CBF's) Oyster Restoration Center in Shady Side, Maryland. Spat-on-shell oysters were produced using eyed larvae from the University of Maryland Center for Environmental Science's Horn Point Oyster Hatchery, located in Cambridge, Maryland. Oyster larvae were introduced to remote setting tanks with

ambient water and allowed 3 days to set prior to moving to flow-through conditions. Oyster spat were held in the tanks a further 7 days prior to being moved into an in situ nursery until they were transferred to SOPS. At the same time, SolarOysters LLC received 1,200 (8mm) seed oysters from Hoopers Island Oyster Company. All spat-on-shell and seed oysters were placed on SOPS in October 2021.

SOPS has five ladders (Figure 2.2). Three out of the five ladders were operational for the bulk of this study. For this study, Ladder 1 will be referred to as "Rotational Ladder 1" and Ladder 4 will be referred to as "Rotational Ladder 2". Rotational Ladder 1 and Rotational Ladder 2 are continuously rotating ladders, with each rung moving one position every 12 minutes, completing a full cycle every four and a half hours. Each basket on the rotating ladders had a total of one hour exposed from the water each day. Ladder 3 was static (referred to as "Static Ladder") and only experienced rotation for cleaning and when oysters were sampled. All baskets on each ladder were cleaned to reduce fouling only when sampled (every three weeks).

From December 2021 to February 2022 all spat-on-shell oysters were sorted into SEAPA baskets of 120 mother shells each. The stocking density of 120 mother shells per basket was selected as that number of shells filled the baskets to be 40% full and would allow room for growth without needed to split the baskets ladder on in the study. Shells without live oysters were removed from the platform to ensure that all the shells on SOPS had live oyster spat. On average, each mother shell had  $2.03 \pm 1.08$  spat per shell. Originally, 134 spat-on-shell oyster baskets were distributed on

SOPS, approximately evenly distributed across the rungs of each ladder (27 to 30 baskets per ladder). On May 5th, critical mechanical failure on 2 of the rotating ladders and the baskets from those 2 ladders were redistributed to the remaining 2 ladders so that the new number of baskets per rotation ladder was approximately 30 baskets.

In addition to the spat-on-shell baskets, three baskets of seed oysters (10mm) were on Rotational Ladder 2, and two seed baskets were on the Static Ladder, each having 240 seed oysters. Spat on shell oysters is the typical product used in restoration where seed oysters are typically grown in cages for commercial aquaculture. On Rotational Ladder 2, seed baskets were distributed evenly across the vertical dimension (top, middle, bottom). On the Static Ladder, prior to August 3rd, one seed basket was placed near the surface and one near the bottom rung. On August 3rd, one of the seed baskets on the Static Ladder was split into two baskets due to the oysters' large size. Each of these new baskets contained 120 seed oysters. When the baskets were split, the third basket was placed in the middle of the ladder.

Every basket on SOPS was assigned a number, and each number was entered in a random number generator to ensure statistical randomness in sampling. Colored zip ties were used to indicate the row of each basket on the ladder. This allowed for easy identification of the baskets as well as providing a way to know the starting position and depth of each of the sampled baskets. Eleven total baskets were lost across the platform due to a failure in the SEAPA basket clips on May 5th. Although this

reduced the total number of baskets on the platform, it did not affect the number of baskets sampled nor did it delay any sampling. MAPC replaced all clips on all baskets on SOPS with a new design that prevented baskets from falling off with rotation and wave action. This delayed sampling in May by an extra week.

In addition to oysters in SEAPA baskets on SOPS, five cages from the Chesapeake Bay Foundation's (CBF) oyster gardening program were attached to cleats on the side of SOPS opposite the dock, to act as a control representing traditional oyster gardening methods. In each of those cages were 120 mother shells with live spat-on-shell oysters, replicating the number of shells in the SEAPA baskets. The CBF cages were cleaned manually with a brush to control biofouling once every three weeks and baskets on SOPS were cleaned the same week as the CBF cages. Thus, oysters were distributed across three different growing regimes: two rotating SOPS ladders, one static SOPS ladder, and five CBF gardening cages.

Oyster survival, growth, condition index, and water quality

Sampling occurred every three weeks, starting the week of April 20th, 2022, through the week of October 24, 2022. During each sampling event, five spat-on-shell baskets and all seed baskets were sampled from each of the three SOPS ladders (N=21) and the CBF cages (N=5). From each spat-on-shell basket or cage, ten shells were haphazardly selected, and the number of live and dead oysters were recorded. Percent live oysters was estimated each sampling period, as the proportion of live oysters divided by number of live plus dead oysters per ladder ( $\% \text{ live} =$



$\frac{\text{Number of oysters alive}}{\text{total number of live and dead oysters}}$ ). Oyster height (length in millimeters from umbo to bill) was measured for every oyster (live or dead) in the sample.

For seed oysters, all baskets were sampled at each sampling period. Ten seed oysters per basket were haphazardly selected and measured for three shell metrics (length, height, and width). All measurements were taken to the nearest 10<sup>th</sup> of a millimeter with an Accusize Industrial Tools (Accusize Industrial Tools, Inc.) digital caliper.

For condition index, on October 28th, 6 mother shells were taken haphazardly from each of 5 baskets on Rotational ladder 1, Rotational ladder 2, the Static Ladder, and the CBF cages, totaling 120 mother shells. In addition, 6-7 seed oysters were selected haphazardly from each of the three baskets on the Static Ladder and Rotational Ladder 2, totaling 40 seed oysters. All oysters were taken to the lab for condition indexing. All oysters, including all spat and seed oysters, were measured for shell height, length, and width. These metrics were used to calculate the fan ratio

( $\text{fan ratio} = \frac{\text{Shell length}}{\text{Shell Height}}$ ) and the cup ratio ( $\text{cup ratio} = \frac{\text{Shell width}}{\text{Shell Height}}$ ) (Mercado-

Silvia, 2005). Oysters were then shucked and dried in a Fisherbrand™ Isotemp™ drying oven (22.2 x 20.9 x 28.3 in) to a constant standardized weight at 90 C for at least 48 hours. The condition index was estimated using the following equation:

$\text{meat condition index} =$

$\frac{(\text{Dry meat weight } g \times 100)}{\text{internal cavity volume } g}$  (Mercado-Silvia, 2005), where internal cavity volume is

determined by the total wet mass of the oyster, minus the shell wet mass.

Water quality data were collected once per week, from April through October, in two ways. First, using a YSI ProDSS (Yellow Springs Instruments Inc/ Xylem, Inc) Multiparameter Digital Water Quality Meter, estimates of dissolved oxygen (DO), salinity, and temperature were taken next to the front (Bay side), middle (Static Ladder), and rear (shore side) of SOPS at three different depths: surface, 3 meters (middle of the ladders), and 6 meters (which is the bottom of the harbor just past the lowest rung on SOPS). Second, I placed a YSI EXO1M (Yellow Springs Instruments Inc/ Xylem, Inc), a continuous water quality sonde, in an empty SEAPA basket on the Rotational Ladder 2. The YSI EXO1M experienced the same conditions of a basket full of oysters on one of the rotating ladders, moving one stage on the ladder every 12 minutes. The YSI EXO1M collected estimates of DO, salinity, and temperature.

### Statistical Analysis

The following statistical analyses were conducted to address the two overarching questions. For the first question, asking whether SOPS can be used to grow oysters for restoration and aquaculture, I compared metrics between oysters grown in two treatments: SOPS baskets and CBF cages. A two factor ANOVA was conducted for shell height and percent live using treatment type and time as fixed effects.

For the second question, asking whether the continuous rotational function of SOPS can improve the growth of oysters, I compared metrics between oysters grown in four treatments: the two SOPS rotating baskets, SOPS static baskets, and CBF cages. The

rationale for splitting the two static regimes (SOPS static and CBF cages) into two separate treatments is that SOPS uses SEAPA baskets smaller in height to the CBF cages and are designed to optimize oyster growth, while CBFs gardening program uses cages made from vinyl-coated wire mesh, similarly used in the construction of crab pots. The type of growing system used could influence growth and survival, and it is important to consider that potential effect (Thomas et al., 2019). The two rotational ladders were separated into separate treatments to control for potential differences that may occur spatially across SOPS, with Rotational ladder 1 being furthest from shore and Rotational Ladder 2 being closest to shore. Rotational Ladder 1 will be exposed to more wave action than Rotational Ladder 2 that is surrounded by the other ladders on the platform.

The first statistical test conducted for the second question, with four treatments, was a two-factor ANOVA testing for the effect of treatment and time on shell height. An additional two factor ANOVA was conducted to test for the effect of treatment and time on the percent of oysters alive in the sample. Additional one-way ANOVAs were conducted to test the effect of treatment on meat condition index, fan ratio, and cup ratio for both spat-on-shell and seed oysters. Analyses were performed in R-4.2.2 for Windows, with an alpha level of 5% or a p-value  $\leq 0.05$  considered significant. If significant effects were detected, a Tukey's post-hoc comparison was performed to compare treatment means.

## Results

### Water Quality

Average dissolved oxygen (DO) for the study ranged between 0.375 mg/L to 5.77mg/L, decreasing over time until the sixth sampling period in August, when it began to increase again (Figure 2.4), resulting in anoxic growing conditions for the study. Temperature increased as the study went into the summer months but began to decrease as the study reached October. Salinity increased over time, reaching a high of 15 ppt by October (Figure 2.5). DO and temperature (C°) decreased with depth (Figure 2.4, 2.6). DO, temperature and salinity did not noticeably differ around various sites on SOPS. However, salinity did increase with depth, dissolved oxygen decreased with depth, and temperature did not drastically change with depth (Figure 2.6).

The YSI EXO1M sonde collected data throughout the course of the day showing the water quality conditions that oysters on a rotating ladder experienced. As the sonde increased in depth, salinity increased, reaching salinities of 10 ppt or greater and as the sonde reached the surface, salinity decreased (Figure 2.7). Temperature did not vary much with depth or throughout the day (Figure 2.8). Dissolved oxygen decreased with depth and increased as the sonde reached the surface (Figure 2.9). A diurnal pattern between water quality parameters was not present.

Question 1: Do oysters grow as well on SOPS as in conventional restoration gardening baskets?

Spat-on-shell in the CBF cages had a significantly higher average shell height (mean shell height = 49.48 mm) compared to the oysters on SOPS (mean shell height = 39.488) (Figure 2.10, Table 1,  $p < 0.001$ ). A significant effect of system type on proportion of oysters alive was also detected. The oysters in the CBF cages had a significantly low number of oysters alive than the baskets on SOPS (Figure 2.11, Table 2,  $p < 0.03$ ).

Question 2: Does rotating oysters in a continuous system affect shell height, condition, and survival?

For spat-on-shell oysters, a significant effect of treatment group was detected for average height (Table 3,  $p < 0.001$ ). The Tukey's post-hoc test revealed that the significant effects of treatment on shell height were between: Rotational Ladder 1 and the Static Ladder (Table 4,  $p < 0.001$ ), Rotational Ladder 2 and the Static Ladder (Table 4,  $p < 0.001$ ), Rotational Ladder 1 and the CBF cages (Table 4,  $p < 0.001$ ), and Rotational Ladder 2 and the CBF cages (Table 4,  $p < 0.001$ ). In other words, no differences were detected between the two rotational treatments and the two static treatments. Height was greatest for the spat-on-shell oysters in the CBF cages, followed by the oysters on the Static Ladder, Rotational Ladder 2, and Rotational Ladder 1 (Figure 2.12).

There was a significant effect of treatment on seed oyster shell height (Table 5,  $p < 0.001$ ) with those on the static ladder being higher than seed on the rotating ladder.

Sampling period was also a significant factor, with shell height across both treatments (Figure 2.13, Table 5,  $p < 0.001$ ).

#### Percent live: Four treatments

In the spat-on-shell oysters, a significant difference in the percent of oysters alive was detected across treatments (Table 6,  $p < 0.005$ ). A Tukey's post hoc comparison revealed a significant difference between the CBF cages and all other treatment groups (Rotational 1, Rotational 2, and Static Ladder) (Table 7,  $p < 0.05$ ). The CBF cages had a lower proportion of live oysters at each sampling event (Figure 2.14). In the seed oysters, no significant difference was detected between treatments in the proportion of live oysters (Table 8,  $p = 0.598$ ). The seed oysters on the Rotational and the Static ladders had a similar proportion of oysters alive (Figure 2.15).

#### Condition index

In the spat-on-shell oysters, a significant effect of treatment was detected for meat condition index (Table 10,  $p < 0.001$ ). Significant differences in condition index were detected between Rotational Ladder 1 and the Static Ladder (Table 11,  $p < 0.05$ ), Rotational Ladder 2 and the Static Ladder (Table 11,  $p < 0.05$ ), and the CBF cages compared to all treatment groups (Table 11,  $p < 0.001$ ). On average, the CBF cages had a significantly higher condition index than all other treatments, followed by Rotational Ladder 2, Rotational Ladder 1, and the Static Ladder (Figure 2.12). On SOPS, the spat-on-shell oysters on the rotational ladders (Rotational Ladder 1 and 2) had significantly higher condition index than the oysters on the Static Ladder (Table 11,  $p < 0.001$ ).

For the seed oysters, a significant difference in meat condition index was detected between the Rotational Ladder and the Static Ladder (Table 12,  $p < 0.001$ ), with the oysters on Rotational Ladder 2 having a significantly higher condition (Figure 2.16).

#### Fan Ratio

For the spat-on-shell oysters, no significant difference was detected for fan ratio across treatment groups (Table 13,  $p > 0.078$ ), with the spat-on-shell oysters having a similar fan ratio (Figure 2.17). In the seed oysters, no significant difference was detected between the Rotational Ladder and the Static Ladder (Table 14,  $p > 0.442$ ), with seed oysters on the Rotational Ladder having a similar fan ratio than those on the Static Ladder (Figure 2.17).

#### Cup Ratio

For the spat-on-shell oysters, no significant difference was detected for cup ratio across treatment groups (Table 15,  $p > 0.168$ ), with spat-on-shell oysters having a similar cup ratio (Figure 2.18). In the seed oysters, a significant difference was detected (Table 16,  $p < 0.001$ ) between the Rotational Ladder and the Static Ladder (Table 16,  $p < 0.001$ ), with seed oysters on the Rotational Ladder having a higher average cup ratio or a deeper cup than those on the Static Ladder (Figure 2.18).

#### Spat Per Shell Analysis

At the start of the experiment there was an average of 2.05 spat per shell across all baskets and treatments. By the end of the study, the average spat per shell were statistically different between treatments ( $p < 0.001$ , Table 2.17) with the on the Static

Ladder having the most spat per shell followed by Rotational Ladder 1, Rotational Ladder 2, and the CBF cages having the lowest spat per shell average with 1 spat per shell (Figure 2.19) There was a significant difference average spat per shell between the CBF cages, the Static Ladder and Rotational Ladder 1 and there was a significant difference in spat per shell between the Static Ladder and Rotational ladder 2 (Table 2.18 ).

### Discussion

The objectives of this research were to evaluate the growth of oysters on SOPS and the Chesapeake Bay Foundation's oyster gardening cages. An additional goal was to investigate the impact that rotation has on oyster growth, survival, and condition index. The results of the study indicated that the effect of growing regime on the performance of oysters is complex. Oysters on SOPS experienced higher survival but lower meat condition and shell height compared to traditional gardening cages. However, rotation on SOPS was able to improve meat condition, fan ratio, and cup ratio for seed oysters, but not for spat-on-shell oysters, compared to oysters on the Static Ladder.

### *Growth & Condition*

Baltimore is not the most ideal location to grow oysters. Oysters require salinities of at least 8ppt and DO of at least 3.2 mg/L to grow optimally (Oesterling & Petrone, 2012). Salinities and DO concentrations thus reached levels below the optimal conditions for oyster gardening (Figures 2.4 & 2.5). For spat-on-shell oysters, oysters



on the Static Ladder and the CBF cages had a higher average shell height than the spat-on-shell on the rotational ladders throughout the study. In other words, oysters that were not tumbled achieved a greater shell height. The purpose of tumbling oysters is to increase the strength of the shell, make the shell more cupped, and smooth the extremities of the shell (Cheney, 2010). The design of SOPS's rotational function is to replicate the same effect that tumbling has on shell shape and growth. By design of the platform and the nature of tumbling, we would expect to reduce height in the oysters on rotating ladders as we are intentionally chipping at the shell. Here, this seems to have generated a substantial difference in average height, with spat-on-shell oysters on the Static Ladder and in CBF cages having a much higher shell height compared to those on the rotational ladders. Spat-on-shell oysters on Rotational Ladder 1 had the lowest average shell height in the study. This result is consistent with the observation that when an oyster shell experiences breakage the growth rate of the shell is slowed (Mizuta & Wikfors, 2019).

Additionally, wave action plays a role in the shaping of oyster shells, similarly breaking the edge of the shell as seen in the practice of tumbling oysters (Brake et al., 2003). Wave action therefore could be another potential factor explaining some of the variation in shell height, as some oysters are experiencing tumbling in addition to wave action. Rotational Ladder 1 is the outermost ladder on SOPS. It faces the Patapsco River and is most exposed to natural wave actions and the wake of ships passing by, which may contribute to the lower shell height seen in Rotational Ladder 1. In a study by Bishop and Peterson, oyster growth was lower in the floating racks

than in the fixed suspended racks, suggesting that wave action does inhibit growth of oysters (Bishop & Peterson, 2005). Taking into consideration the effect of wave actions and tumbling on oyster growth, it is fair to suggest that the current frequency of the rotating ladders may be leading to the negative effect on spat-on-shell oyster growth we observed. A negative effect of tumbling and wave action on shell height is further supported by growth observed in the seed oysters. The seed oysters on the Static Ladder were significantly larger than the rotating seed oysters, at the final sample period.

The condition index of spat-on-shell and seed oysters further demonstrates the effect of rotation on oysters. The spat-on-shell oysters in the CBF cages had the highest average condition index followed by Rotational Ladder 1, Rotational Ladder 2, and the Static Ladder having the lowest average condition index. It was surprising to see the CBF cages with the highest condition, as I predicted that the oysters on rotating ladders to have a higher condition. It is key to note as the Static ladder also had the same opportunity as the CBF cages to be submerged under water and the same opportunity to feed continuously yet, potentially due to the location of the Static Ladder being surrounded by two additional ladders on each side, the amount of wave action it receives is significantly reduced. The two rotational ladders had extremely similar condition indexes. This strongly suggests that the rotational ladders did play a role in improving the condition index of the oysters, as they had a higher condition index compared to the oysters on the static ladder. However, it is important to keep in mind that rotation also may be playing a role in the limited shell growth seen in these

oysters on those ladders. The ladders are moving one stage every 12 minutes and that frequency in movement may not be the most optimal for oysters on this system as the time they have to feed is reduced.

There was not a significant difference detected in the fan and cup ratio of the spat-on-shell oysters across growing regimes (Table 4). This observed effect was expected, as spat-on-shell oysters' fan and cup are driven by the space they have available, which is dependent on the size of the mother shell the oyster sits on and other oysters that sit on the same mother shell.

In the seed oysters, condition, cup ratio, and fan ratio are more important, and the oysters on the rotating ladder had a significantly better meat condition and cup ratio than the seed on the Static Ladder. The purpose of tumbling oysters is to improve these metrics and the data here support that the platform was successful in mimicking the same effect of a traditional oyster tumbler to improve condition.

### *Survival*

For the spat-on-shell oysters, there was a significant difference between the CBF cages and all the other spat-on-shell oysters on the SOPS platform in terms of the proportion of oysters alive in the sample. The Static Ladder, rotational ladder 1 and rotational ladder 2 on the platform had a similar performance in the proportion of oysters found alive in in each sample with some variation between each treatment but differences were not significant. The CBF cages had a significantly reduced

proportion of oysters alive at each sampling event compared to the other growing regimes.

A significant difference was detected between survival and time for the seed oysters (Table 8,  $p < 0.001$ ). The analysis detected a difference between time but not treatments. This is most likely due to a drop in survival for both rotating and static treatments at the third sampling period in early July.

Around sampling periods 3 and 4 there was a drop in survival in all treatments but most dramatically in the CBF cages. This mortality may be a result of the drop in salinity around those sampling points due to a freshet caused by heavy rain. Since the CBF cages were only at the water's surface, which saw the lowest salinity (Figure 2.5), the oysters in those baskets did not have an opportunity to be at lower depths and higher salinity concentrations and may have died due to the freshet. Another explanation may be the shape of the baskets. All the baskets on SOPS are SEAPA cages, and when the baskets are submerged the oysters are distributed evenly across the basket giving them more space to filter and grow. The cages provided by the Chesapeake Bay Foundation are rectangular cages. When they are submerged, the oysters tend to cluster in one spot rather than distribute themselves across the baskets. This clumping could have led to more competition between oysters and resulted in some of the mortality observed. Additionally, those cages had a higher abundance of other wildlife from mud crabs, blue crabs, and even blister worms that might have preyed upon the young oysters (personal observation).

The hypothesis that mortality in the CBF cages might be an effect of gear rather than location is further supported when looking at the seed oysters. The seed on the rotational and Static Ladder performed very similarly and at times the same when it came to the number of oysters alive in the basket.

It is also important to note that during the first 3 sampling events there was a large spawning event of *Victorella pavid*a that covered the oysters, primarily affecting the CBF cages and the Static Ladder on SOPS, though it was found on oysters in every treatment. The additional stress this event could have had on the oysters might explain some of the mortality seen in the study, especially in the CBF cages as they had the highest abundance of the *V. pavid*a on the cages and the oysters (personal observation). A limitation of the sampling design is that the data that we collected on mortality in the oysters is from a sample of the random basket selected and in turn the basket was also a sample of the total baskets on the ladder.

The spat per shell numbers were different between treatment groups at the end of the study and this may have resulted from the lower rates of survival seen in the CBF cages. The oysters in the CBF cages had significantly less spat per shell than the shells on SOPS. The CBF cages experienced a significant die-off earlier in the study, and this likely led to the lower spat per shell number in the CBF cages at the last sampling period. However, since there were less oysters competing for space and food within the cages that may be a potential factor contributing to higher condition

and shell height in the cages. It is important to note that an increased proportion of oysters alive at the last few sampling periods in CBF cages does not mean there are more oysters alive in the baskets. By the end of the study two cages did not have any oysters alive left in them so only 28 oysters were identified in the CBF cages after following the sampling procedures at the last sampling event.

### Conclusion

Growing both seed and spat-on-shell oysters on the platform provided a unique perspective on how SOPS can be used for oyster restoration and commercial aquaculture. For restoration, oysters that are raised to a size where they are less susceptible to poor water quality and threats from predators have higher survival and can improve restoration success (Mann & Powell, 2007). However, limitations on space typically preclude holding spat-on-shell oysters in tanks or other protected grow-out conditions for more than 10-14 days. Oyster gardening provides a means to grow out a small number of spat-on-shells to larger size to supplement larger plantings of 10–14-day oysters that are deployed. It also provides an opportunity for community members to participate directly in restoration through husbandry of the oyster cages.

The data from this study suggest that CBF's current gardening cages for oyster restoration are not the most adept at maintaining high survival during the growing season. Rotation on SOPS can lead to lower shell heights for both spat-on-shell and seed oysters but oysters grown on the system experience higher survival. In terms of

condition metrics (meat condition index, fan ratio, cup ratio), seed oysters were more influenced by rotation than the spat-on-shell oysters. The difference between spat-on-shell and seed oysters was expected as oysters set on a mother shell grow differently than free growing oysters. SOPS was designed to raise seed oysters for commercial aquaculture industry and high survival in the samples and condition metrics observed suggest SOPS will achieve its objectives. However, the significantly smaller size in the rotating seed oysters was not expected. Ultimately there are tradeoffs to rotating oysters on SOPS, trading size for better meat condition and cup ratio. Markets exist for different sizes and shapes of oysters and support the idea that bigger is not always better. It will be up to a farmer to decide if the tradeoff is worth it for their needs.

Oysters were on SOPS for a full year, and none of the oysters reached the 3-inch market size. Although it takes on average 18 months for oysters to reach market size, SOPS (both static and rotational ladders), is not reducing the time needed to get to market size which is not a negative against the system since the study conducted was a few months shorter than the time needed to grow oysters to market size.

Additionally, these oysters were growing in relatively poor conditions with respect to dissolved oxygen and salinity. However, the market characteristics of high condition index and the deeper cups seen in the seed oysters on the rotational ladder demonstrates the system's potential to produce an oyster with highly marketable traits of a deeper cup and more meat in relation to its shell.

SOPS is recommended for restoration use as this study demonstrates SOPS' potential to raise oysters on the system with higher survival rates. Oysters are smaller on rotation ladders, but SOPS does not have to deploy the 12-minute rotational pattern and can be made to do what is best for the restoration practitioner. The oysters on the static ladder were larger than those on the rotating ladders but the oysters on the rotating ladders did still reach a size to where they would be tolerable to changes in their environment and predators on a reef as they are beyond the juvenile stage and size (Galtsoff & U.S. Fish and Wildlife Service., 1964).

Due to the lower dissolved oxygen and salinity levels in Baltimore, the location where SOPS operated is not conducive for optimal oyster growth. Salinity is one of the key factors for optimal survival, growth in shell height and condition. A study conducted from 2016-2018 at the University of Maryland Center for Environmental Science Horn Point Laboratory saw high mortality rates due to the abnormally low salinity concentration in 2018, even though the laboratory is in waters optimal for oyster growth (Hood et al., 2020). The waters where SOPS operated also had below optimal salinity concentrations but the oysters on SOPS did not experience the same rates of mortality seen in the Hood et al study. The oysters in the CBF cages did experience low survival rates (below 50%) but in some of the treatments in the Hood et al study there are survival rates of 36% or lower (Hood et al., 2020). The difference in survival rates supports the argument that SOPS can increase the survival rates of oysters.



In the same study, researchers at the Horn Point lab looked at oyster growth between several different gear types including SEAPA baskets (on a longline system) that was used in the study conducted for this thesis. In the growing season from 2016-2018 the Hood et al. study, the oysters in the SEAPA baskets grew to be around 43.8 mm from starting around 19.8 mm which is similar to the growth seen on the SEAPA baskets on SOPS (Figure 2.10) (Hood et al., 2020).

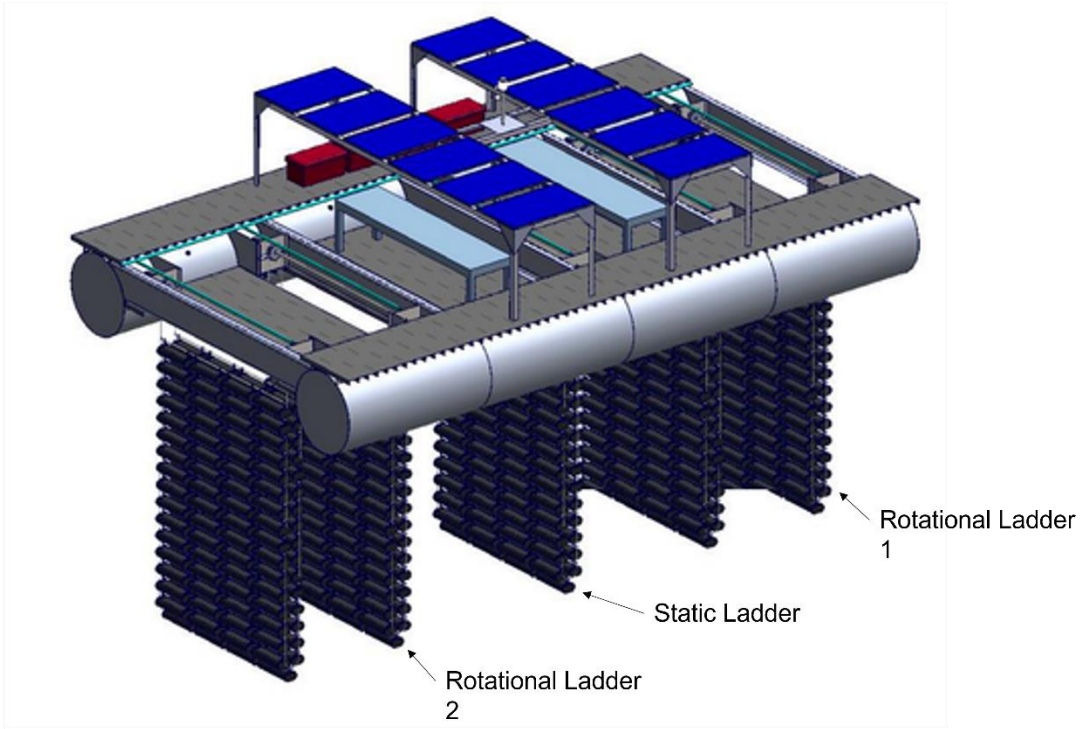
Looking at the condition index of oysters, with the CBF cages having the highest condition index, I hypothesize that this is most likely related to the oysters being near the surface where phytoplankton is most abundant. A study by Thomas et al found that oysters raised near the surface during the grow out period have higher condition indexes, which is similar to what was observed in the CBF cages(Thomas et.al, 2019). This goes back to the benefits of oyster gardening and raising oysters near the surface. The condition and cup ratio were better on the rotating ladder, they did produce smaller oysters, which is not inherently a negative as the size and shape of the oyster can be sold and profitable depending on how it is marketed when it is sold (Mizuta & Wikfors, 2019).

It is recommended that studies conducted for the further development of SOPS focus on finding an optimal rotation pattern. Rotating oysters every 12 minutes may have led to the smaller shell heights observed in the study. Overall, the study showed that SOPS can successfully grow oysters and influence shell shape while supporting the technology's potential as a tool for aquaculture and restoration.

## Figures



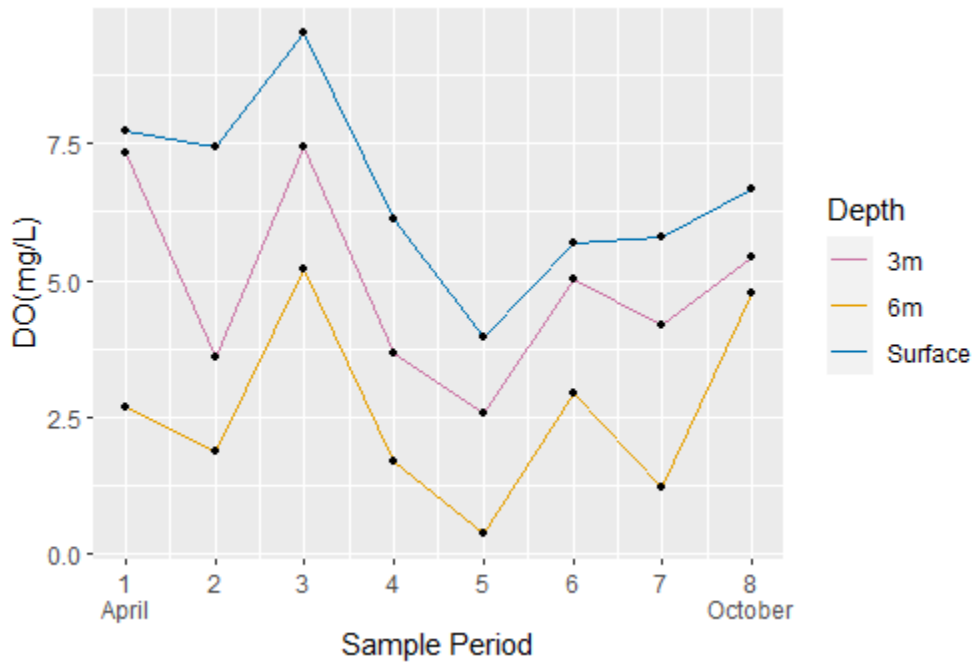
**Figure 2.1 Solar Oyster Production System (SOPS) on floating dock at the Maritime Applied Physics Corporation**



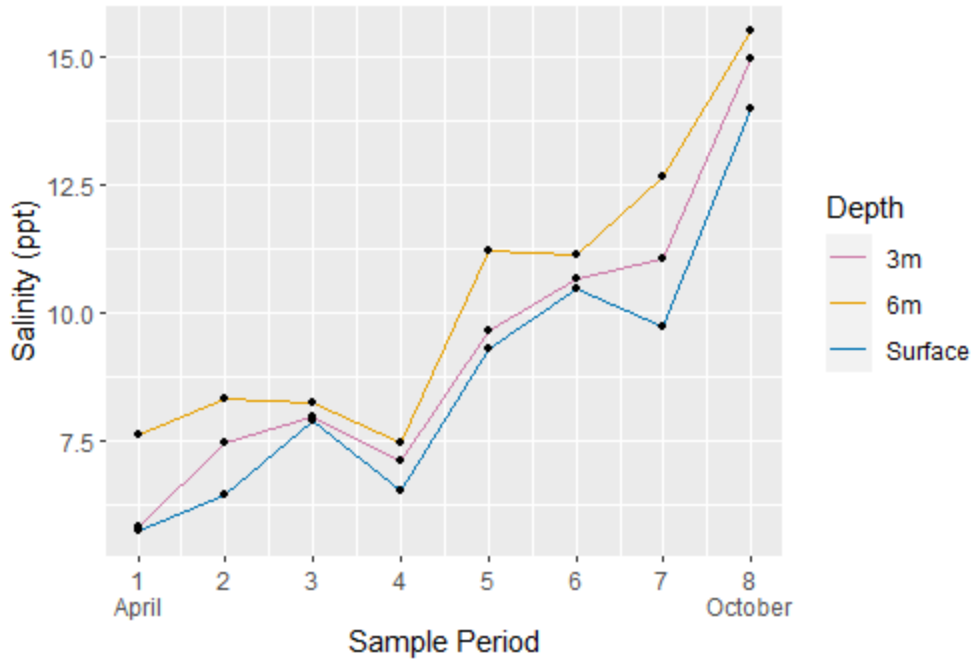
**Figure 1.2 3D Model of SOPS with the 3 operational ladders on the system**



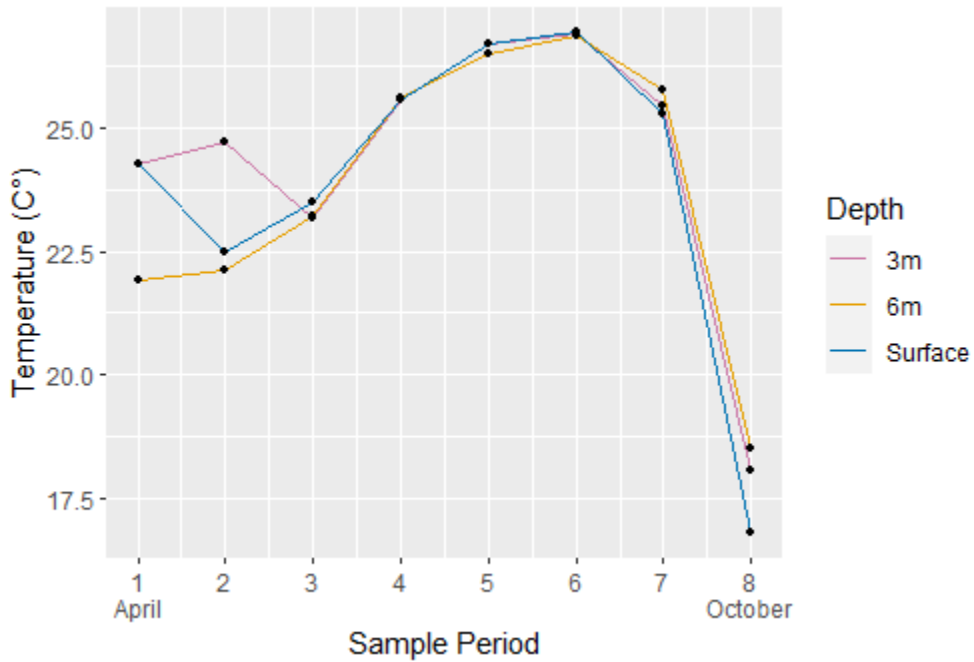
**Figure 2.3 Location of the SOPS at MAPC in Baltimore, MD**



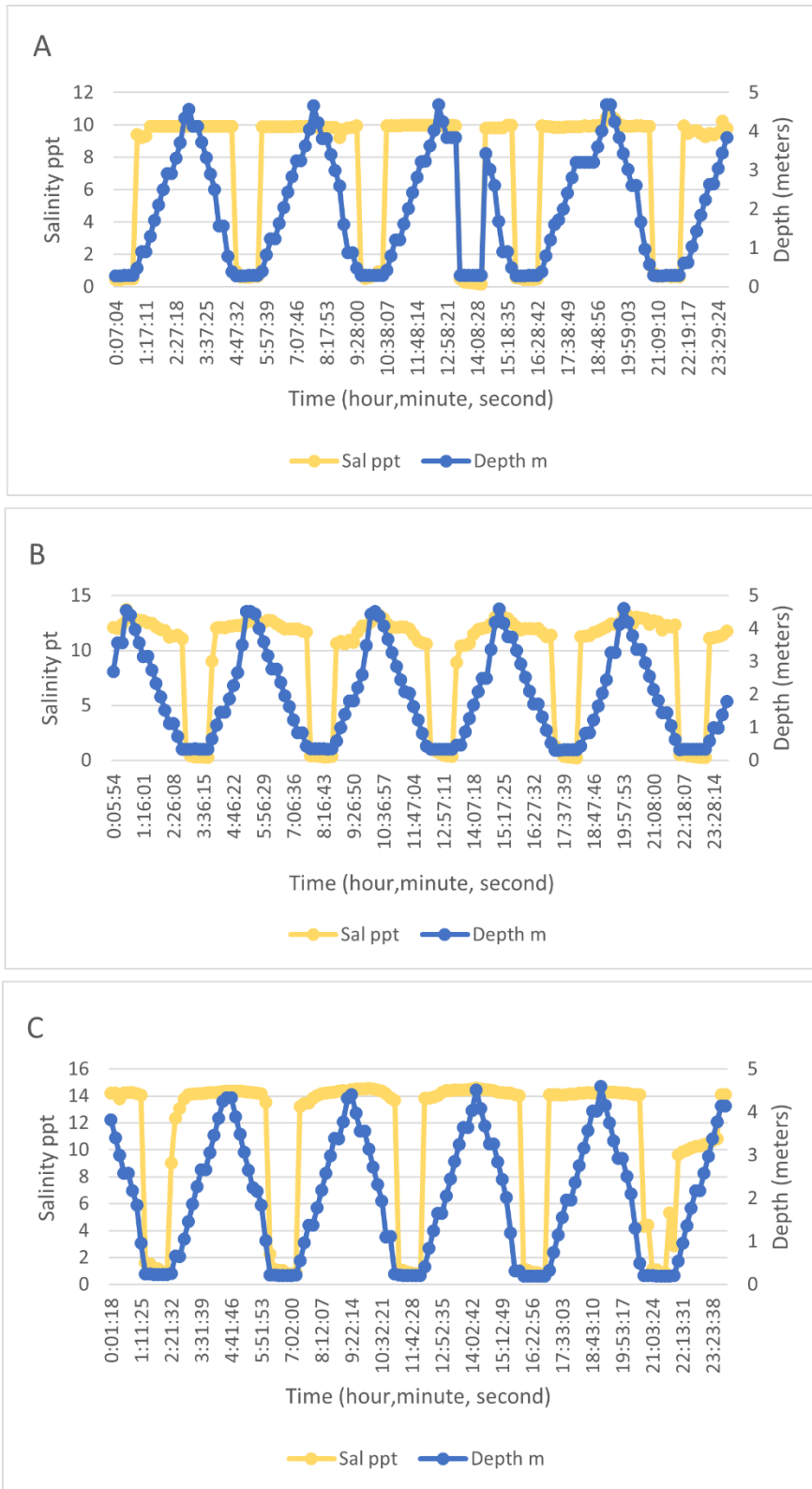
**Figure 2.4 Dissolved oxygen (mg/L) at three depths at SOPS from April 2022 through October 2022 using the YSI ProDSS Multiparameter Digital Water Quality Meter**



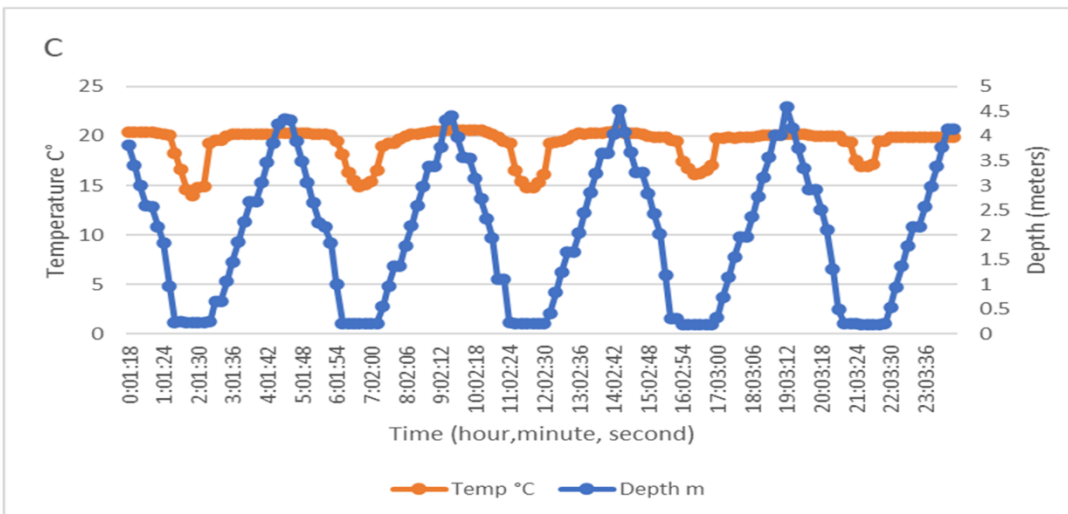
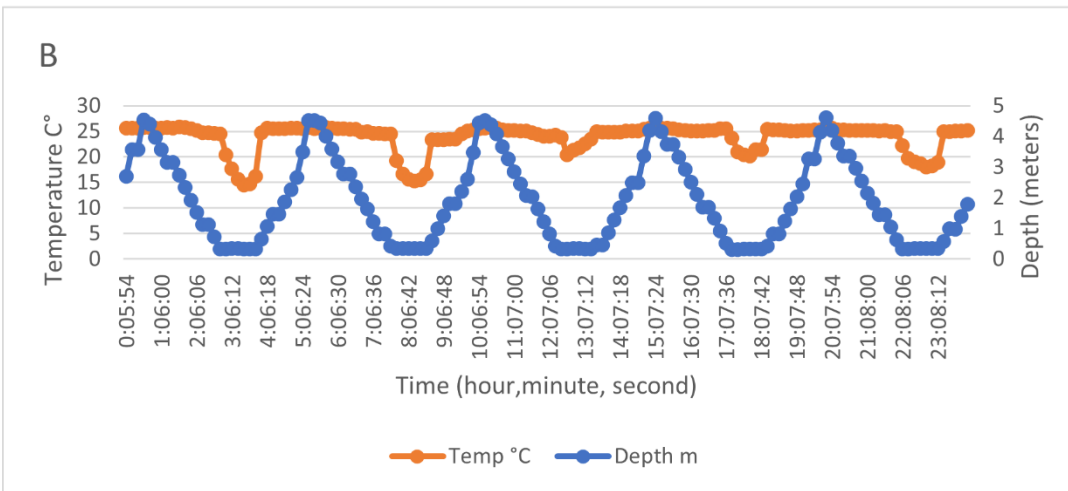
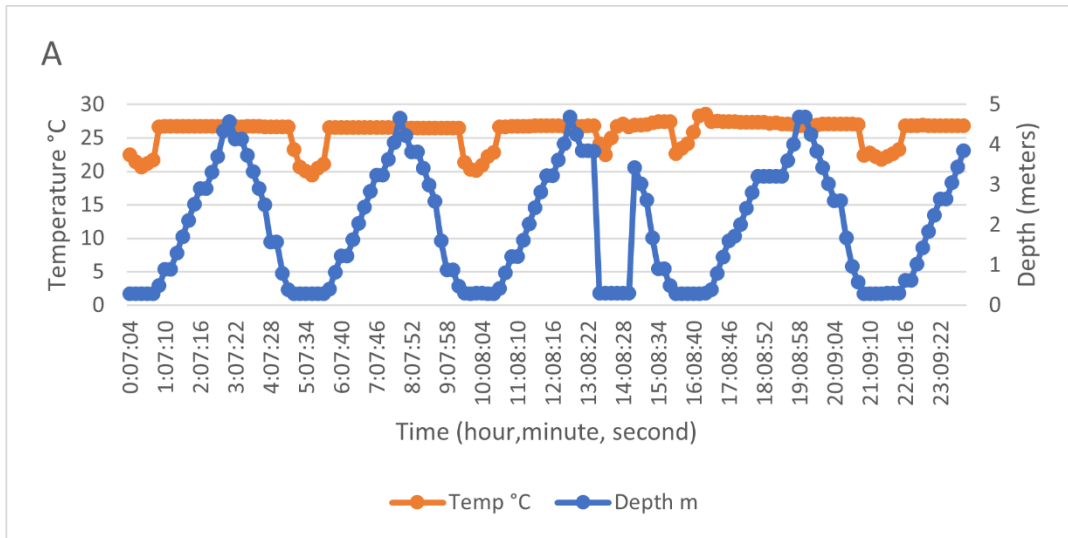
**Figure 2.5 Salinity (ppt) at three depths at SOPS from April 2022 through October 2022 using a YSI ProDSS Multiparameter Digital Water Quality Meter**



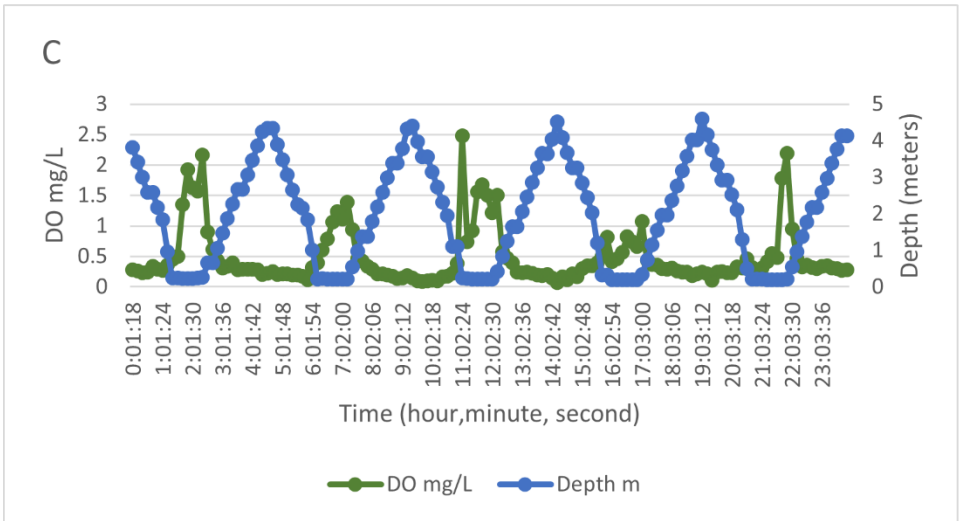
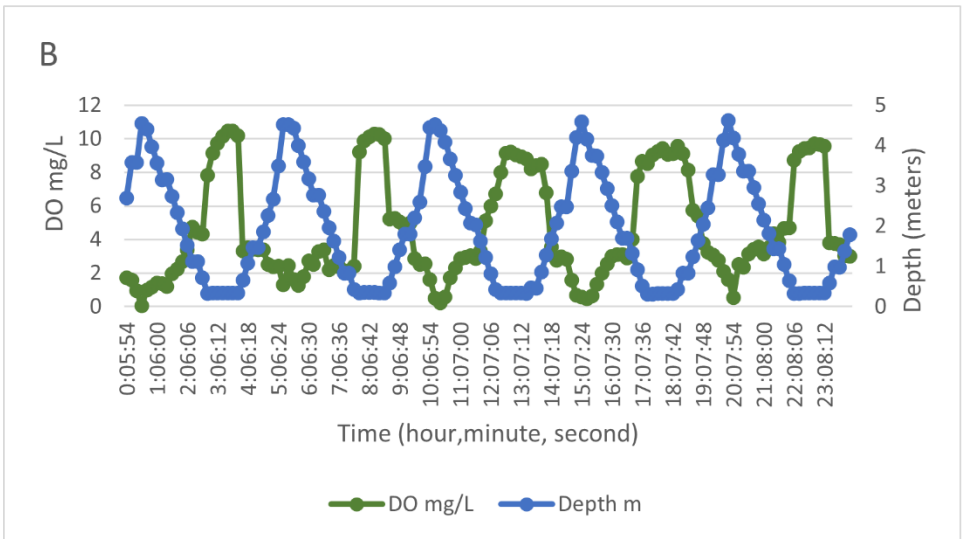
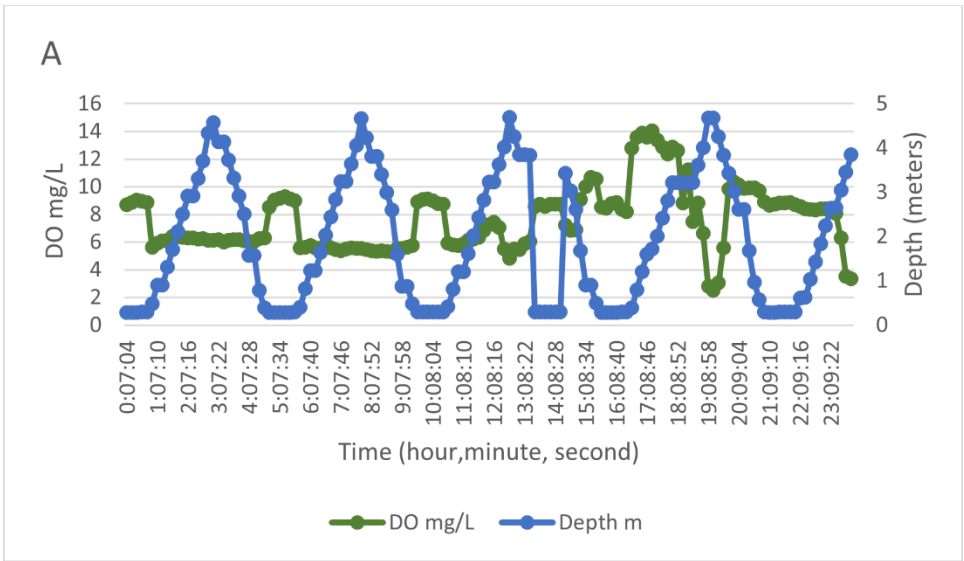
**Figure 2.6 Temperature (C°) at three depths at SOPS from April 2022 through October 2022 a YSI ProDSS Multiparameter Digital Water Quality Meter**



**Figure 2.7 Salinity concentrations collected on a rotating ladder throughout a day using a YSI EXO1M at three different months (A) Augusts 15<sup>th</sup> , (B) September 16<sup>th</sup> , (C) October 1<sup>st</sup>**

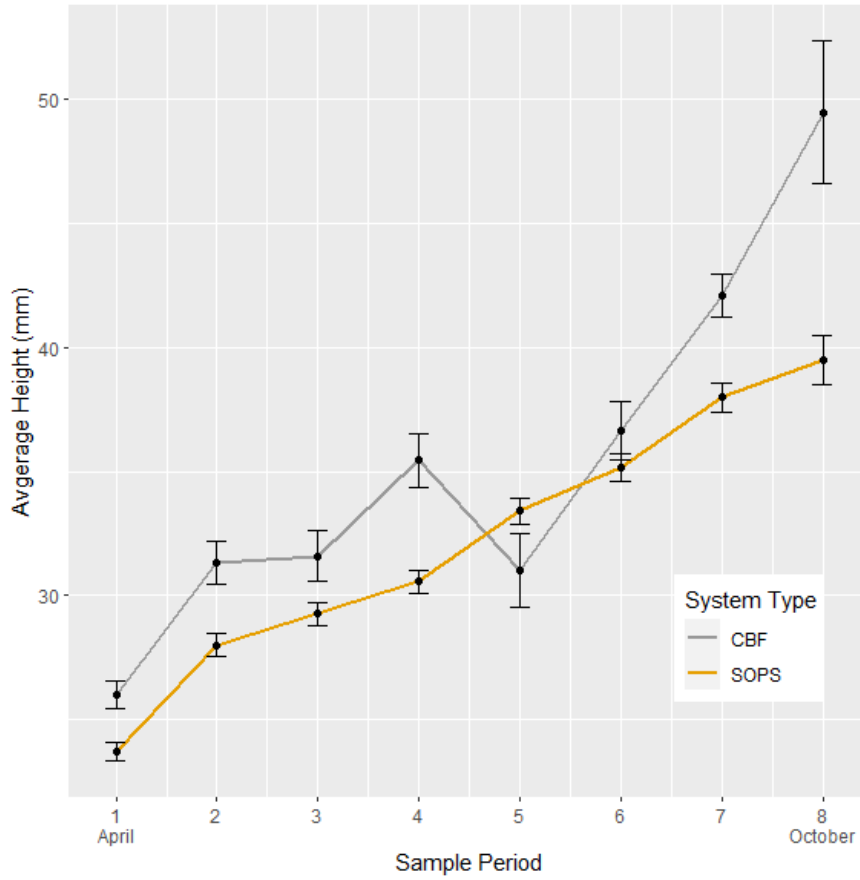


**Figure 2.8** Temperature C° collected on a rotating ladder throughout a day using an YSI EXO1M at three different months (A) Augusts 15<sup>th</sup> , (B) September 16<sup>th</sup> , (C) October 1<sup>st</sup>

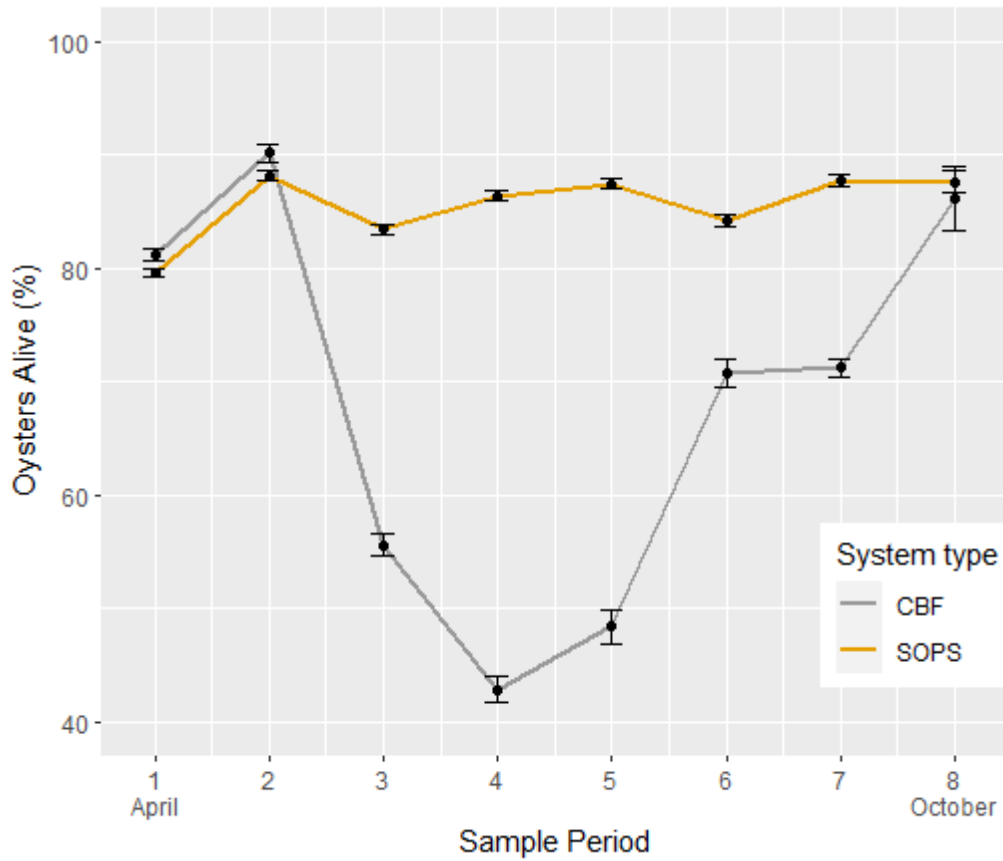




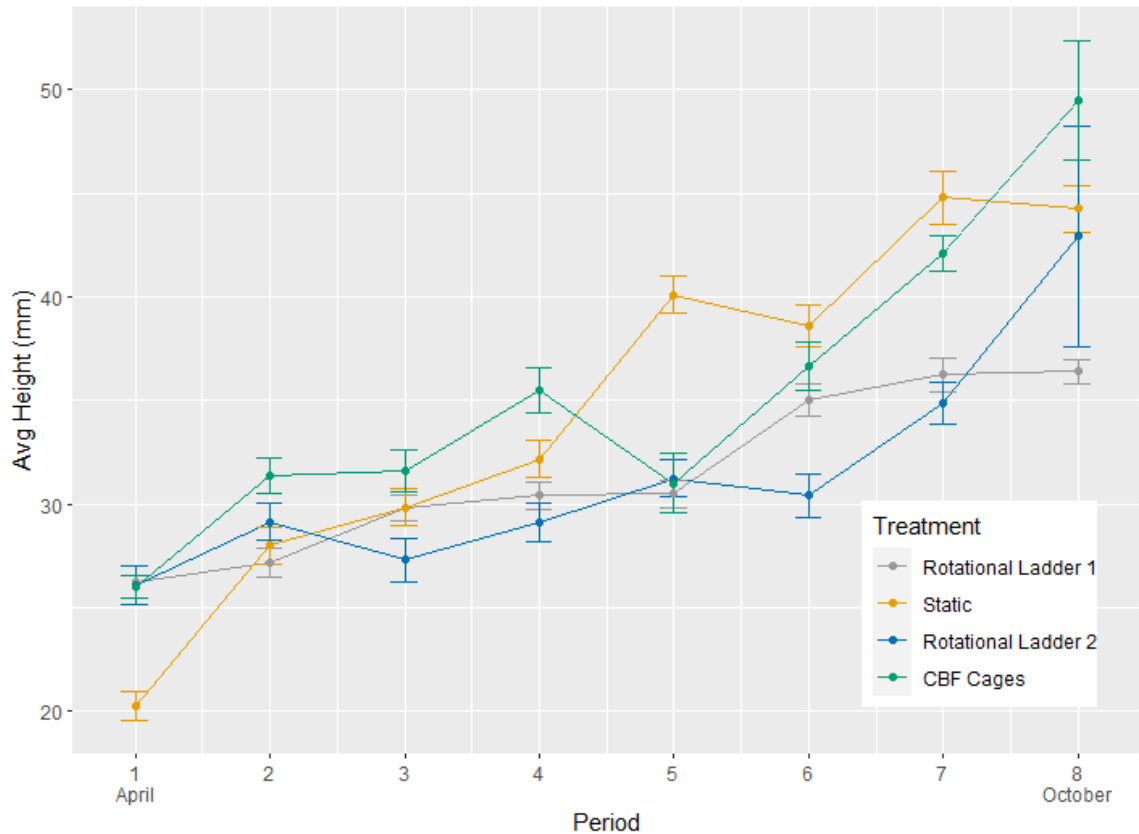
**Figure 2.9 DO concentrations collected on a rotating ladder throughout a day using an YSI EXO1M at three different months (A) Augusts 15<sup>th</sup> , (B) September 16<sup>th</sup> , (C) October 1<sup>st</sup>**



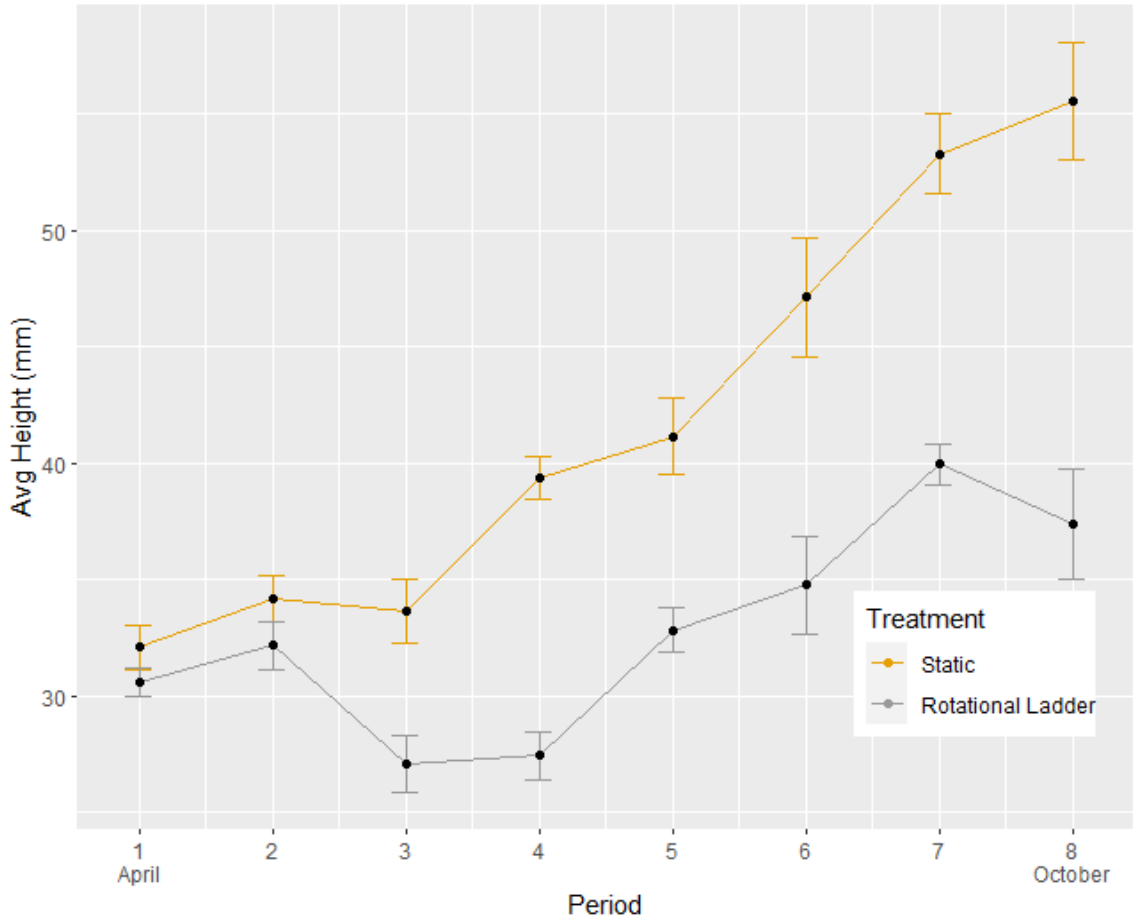
**Figure 2.10 Mean shell height +/- standard deviation in millimeters for spat-on-shell oysters in SOPS baskets (orange) vs CBF cages (black) across 8 sampling periods from April through October 2022. Significant differences were detected across treatments ( $p < 0.001$ ).**



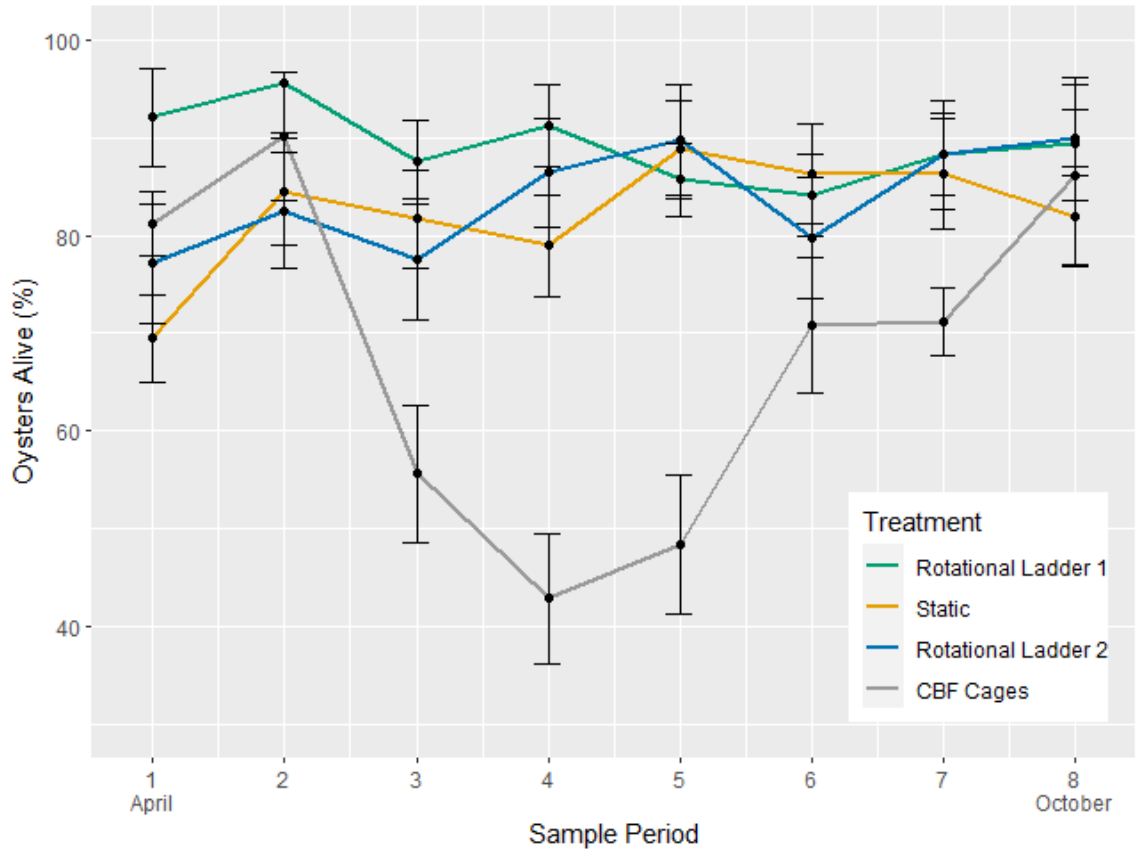
**Figure 2.11 Proportion of spat-on-shell oysters alive +/- standard deviation in SOPS baskets (orange) vs CBF cages (black) across 8 sampling periods from April through October 2022. Significant differences were detected across treatments at all sampling periods ( $p < 0.001$ )**



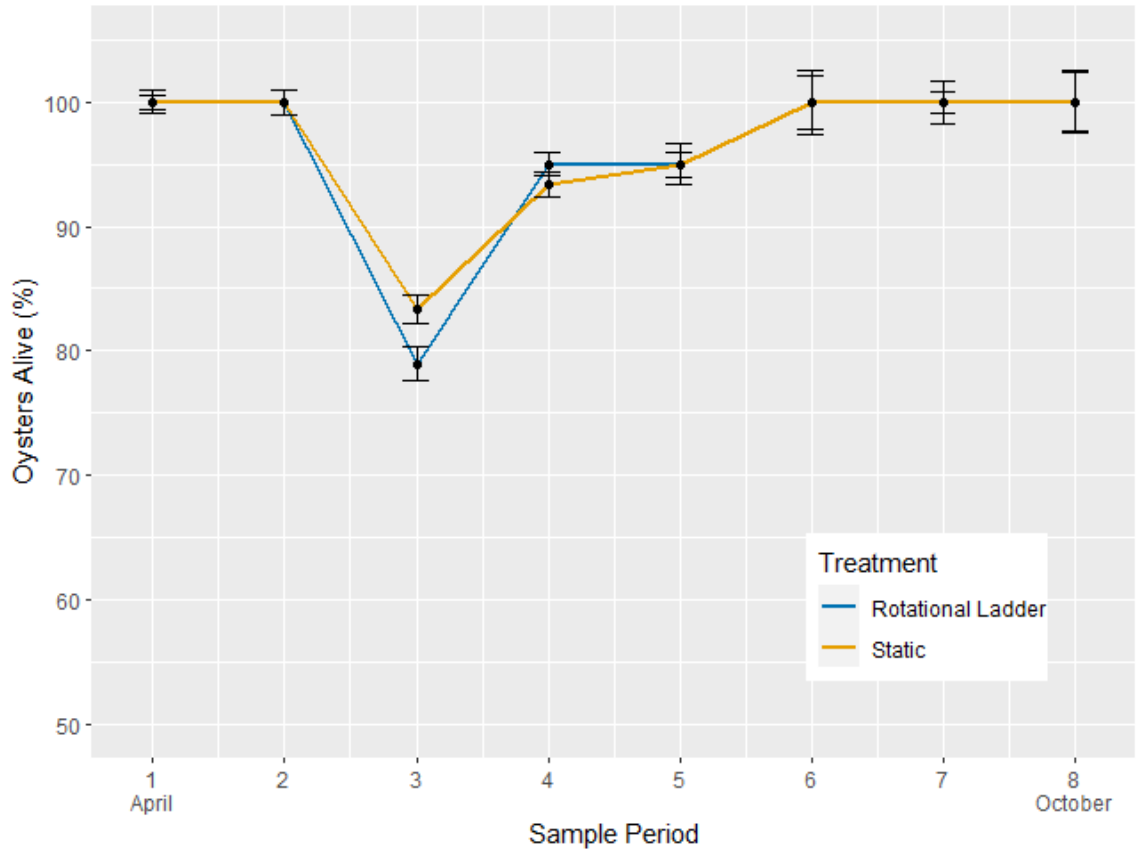
**Figure 2.12 Mean shell height +/- standard deviation in millimeters for Spat-on-Shell in SOPS rotational and static ladders vs CBF cages across 8 sampling periods from April through October 2022. Significant differences between average shell height were detected across treatments at all sampling periods ( $p < 0.001$ )**



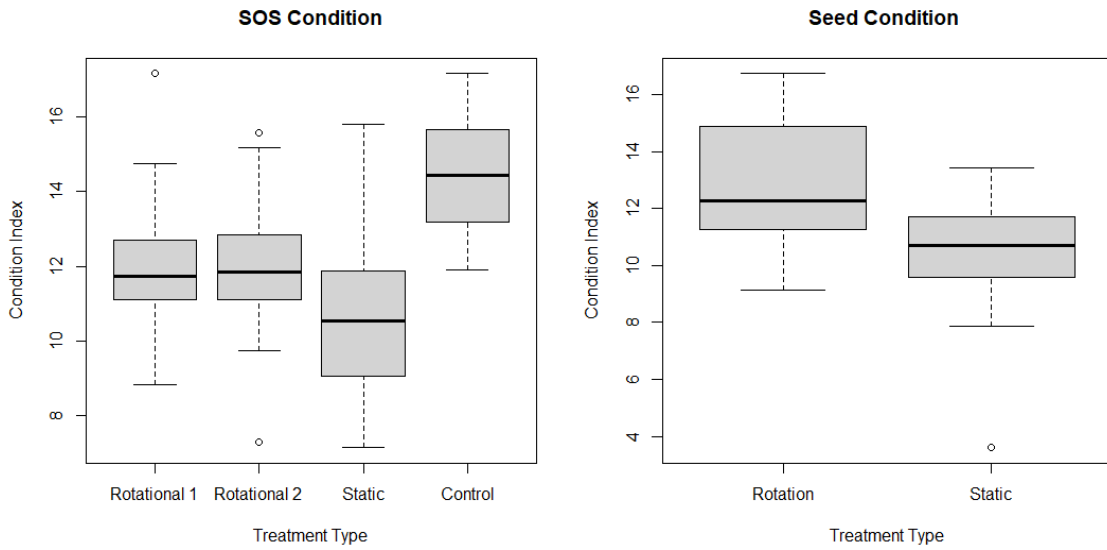
**Figure 2.13 Mean shell height +/- standard deviation in millimeters for seed oysters on SOPS's rotational and static ladders across 8 sampling periods from April through October 2022. Significant differences in height between treatments were detected across treatments at all sampling periods ( $p < 0.001$ )**



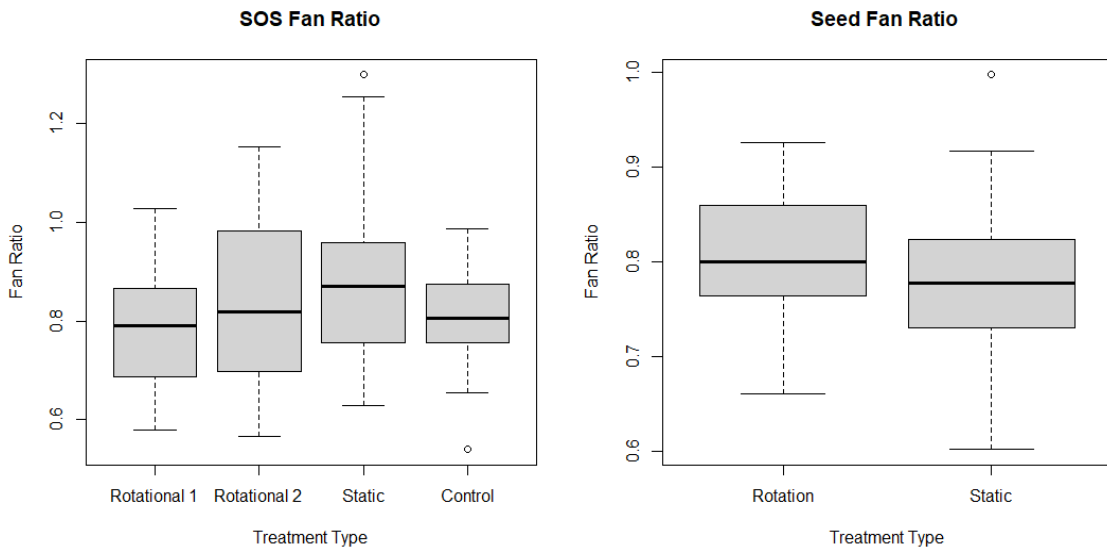
**Figure 2.14 Proportion of spat-on-shell oysters alive +/- standard deviation across treatments over 8 sampling periods from April through October 2022. Significant differences were detected between the rotational ladders, static treatment of SOPS static ladder and CBF cages ( $p < 0.001$ )**



**Figure 2.15 Proportion of seed oysters alive +/- standard deviation between treatments over 8 sampling periods from April through October 2022. Significant was detected between the percentage of oysters alive and sampling period ( $p < 0.001$ )**

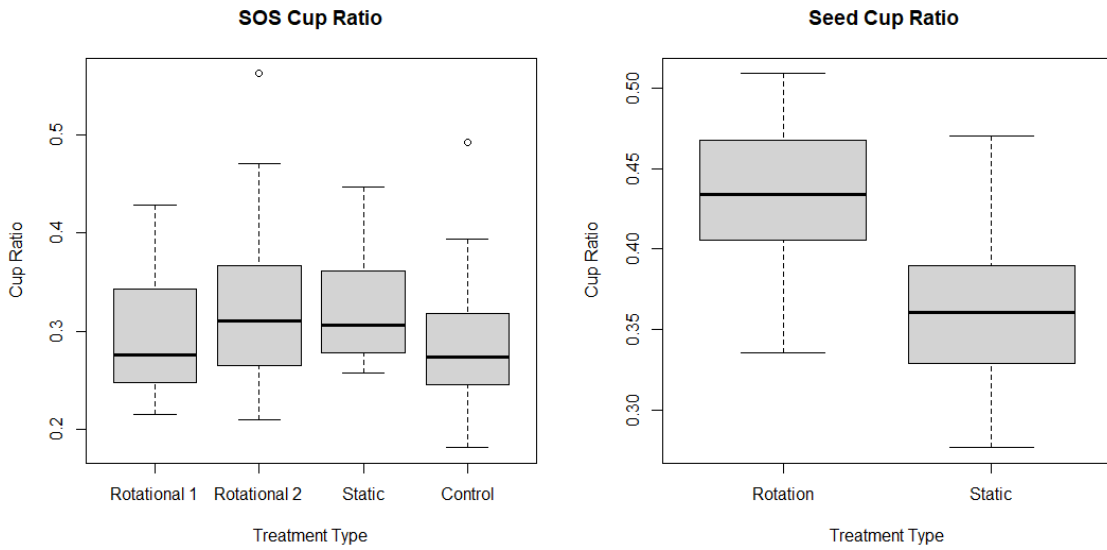


**Figure 2.16** Spat-on-shell & Seed condition index between growing regimes ( $p < 0.001$ ).

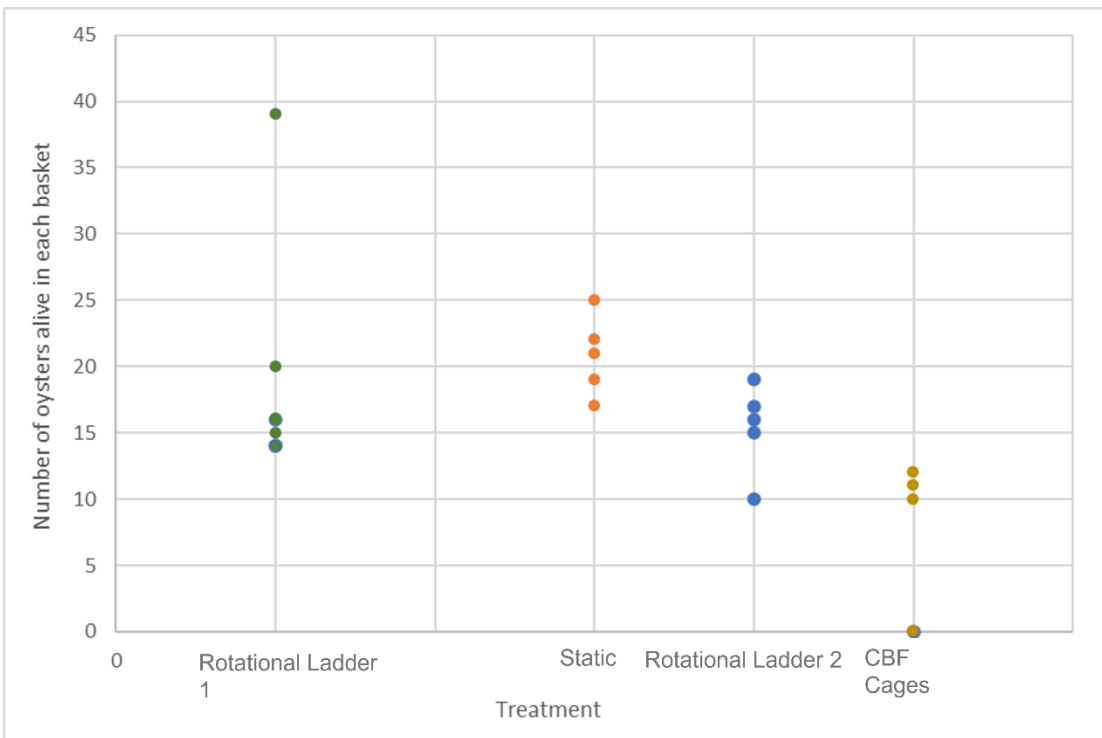


**Figure 2.17** Spat-on-shell & Seed fan ratio between growing regimes.





**Figure 2.18** Spat-on-shell & Seed cup ratio ( $p < 0.001$ ) between growing regimes.



**Figure 2.19** Total Oysters sampled baskets at the last sampling period between treatment groups.

Tables

**Table 1. Spat-on-shell Shell Height on SOPS & CBF Cages - Two-Way ANOVA between system type and time**

Variable	Df	Sum Sq	Pr(>F)
System	1	5850	< 0.0001
Sample Period	7	130873	< 0.0001
System: Sample Period	7	1621.3	0.0004

**Table 2. Spat-on-shell Survival on SOPS & CBF Cages - Two-Way ANOVA between system and time**

Variable	Df	Sum Sq	Pr(>F)
<b>System</b>	<b>1</b>	<b>1194.7</b>	<b>0.03</b>
Sample Period	7	0.9772	0.51174

**Table 3. Spat-on-shell Shell Height - Two-Way ANOVA between treatment and time**

Variable	Df	Sum Sq	Pr(>F)
Treatment	3	13247	<0.001
Time	7	124526	<0.001
Treatment: Time	21	24001	<0.001

**Table 4. Average Spat-on-shell Shell Height - Tukey's HSD between treatments**

Contrast	Difference	Pr(>F)
<b>RL 1- Static</b>	<b>-3.2669</b>	<b>&lt;0.0001</b>
RL 1- RL 2	0.0954	0.9998
<b>RL 1- CBF Cage</b>	<b>-3.9810</b>	<b>&lt; 0.0001</b>
<b>Static- RL 2</b>	<b>3.3623</b>	<b>&lt; 0.0001</b>
Static- CBF Cages	-0.7141	0.6710
<b>RL 2- CBF</b>	<b>-4.0764</b>	<b>&lt; 0.0001</b>

**Table 5. Seed Shell Height - Two-Way ANOVA between ladder type and time**

Variable	Df	Sum Sq	Pr(>F)
Ladder	1	0.46	0.5969
Sample Period	7	9683.8	< 0.0001
Ladder: Sample Period	7	1621.3	< 0.0001

**Table 6. Spat-on-Shell Proportion of oysters alive - Two-Way ANOVA between treatment and time**

Variable	Df	Sum Sq	Pr(>F)
<b>Treatment</b>	<b>3</b>	<b>1921.89</b>	<b>0.002</b>
Time	7	677.22	0.446

**Table 7. Proportion of Spat-On-Shell oysters - Tukey's HSD between treatments**

Contrast	Difference	Pr(>F)
RL 1- Static	7.02	0.501
RL 1- RL 2	0.0954	0.702
<b>RL 1- CBF Cage</b>	<b>21.00</b>	<b>0.002</b>
Static- RL 2	-1.65	0.987
<b>Static- CBF Cages</b>	<b>13.97</b>	<b>0.046</b>
<b>RL 2- CBF</b>	<b>15.62</b>	<b>0.023</b>

**Table 8. Seed Survival - Two-Way ANOVA between treatment and time**

Variable	Df	Sum Sq	Pr(>F)
Treatment	1	0.46	0.598
Time	7	609.01	<0.001

**Table 9. Spat-on-shell Condition - One-Way ANOVA between treatment**

Variable	Df	Sum Sq	Pr(>F)
Treatment	3	209.37	<0.001

**Table 10. Average Spat-on-shell Meat Condition - Tukey's HSD between treatments**

Contrast	Difference	Pr(>F)
<b>RL 1- Static</b>	<b>-3.2669</b>	<b>0.0093</b>
RL 1- RL 2	0.0954	0.9977
<b>RL 1- CBF Cage</b>	<b>-3.9810</b>	<b>&lt; 0.0001</b>
<b>Static- RL 2</b>	<b>3.3623</b>	<b>0.0161</b>
<b>Static- CBF Cages</b>	<b>-0.7141</b>	<b>&lt; 0.0001</b>
<b>RL 2- CBF</b>	<b>-4.0764</b>	<b>&lt; 0.0001</b>

**Table 11. Seed Meat Condition - One-Way ANOVA between treatments**

Variable	Df	Sum Sq	Pr(>F)
Treatment	1	80.635	<0.001

**Table 12. Spat-on-Shell Fan Ratio - One-Way ANOVA between ladders**

Variable	Df	Sum Sq	Pr(>F)
Treatment	1	80.635	0.079

**Table 13. Seed Fan Ratio - One-Way ANOVA between ladders**

Variable	Df	Sum Sq	Pr(>F)
Treatment	1	0.00344	0.442

**Table 14. Spat-on-shell Cup Ratio - One-Way ANOVA between treatments**

Variable	Df	Sum Sq	Pr(>F)
Treatment	3	0.02401	0.1683

**Table 15. Seed Cup Ratio- One-Way ANOVA between ladders**

Variable	Df	Sum Sq	Pr(>F)
<b>Treatment</b>	<b>1</b>	<b>80.635</b>	<b>&lt; 0.0001</b>

**Table 16. Spat on shell - One-Way ANOVA between treatment**

Variable	Df	Sum Sq	Pr(>F)
Treatment	3	47.07	<0.001

**Table 17. Average Spat per shell - Tukey's HSD between treatments**

Contrast	Difference	Pr(>F)
RL 1- Static	-0.453	0.161
RL 1- RL 2	0.460	0.162
<b>RL 1- CBF Cage</b>	<b>1.000</b>	<b>&lt; 0.002</b>
<b>Static- RL 2</b>	<b>0.913</b>	<b>0.002</b>
<b>Static- CBF Cages</b>	<b>1.453</b>	<b>&lt; 0.001</b>
RL 2- CBF	0.540	< 0.271

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