

Oyster Shell Reclamation: Evaluating the Oyster Shell Curing Process and Potential Bacterial Pathogens from Restaurant to Reef Construction

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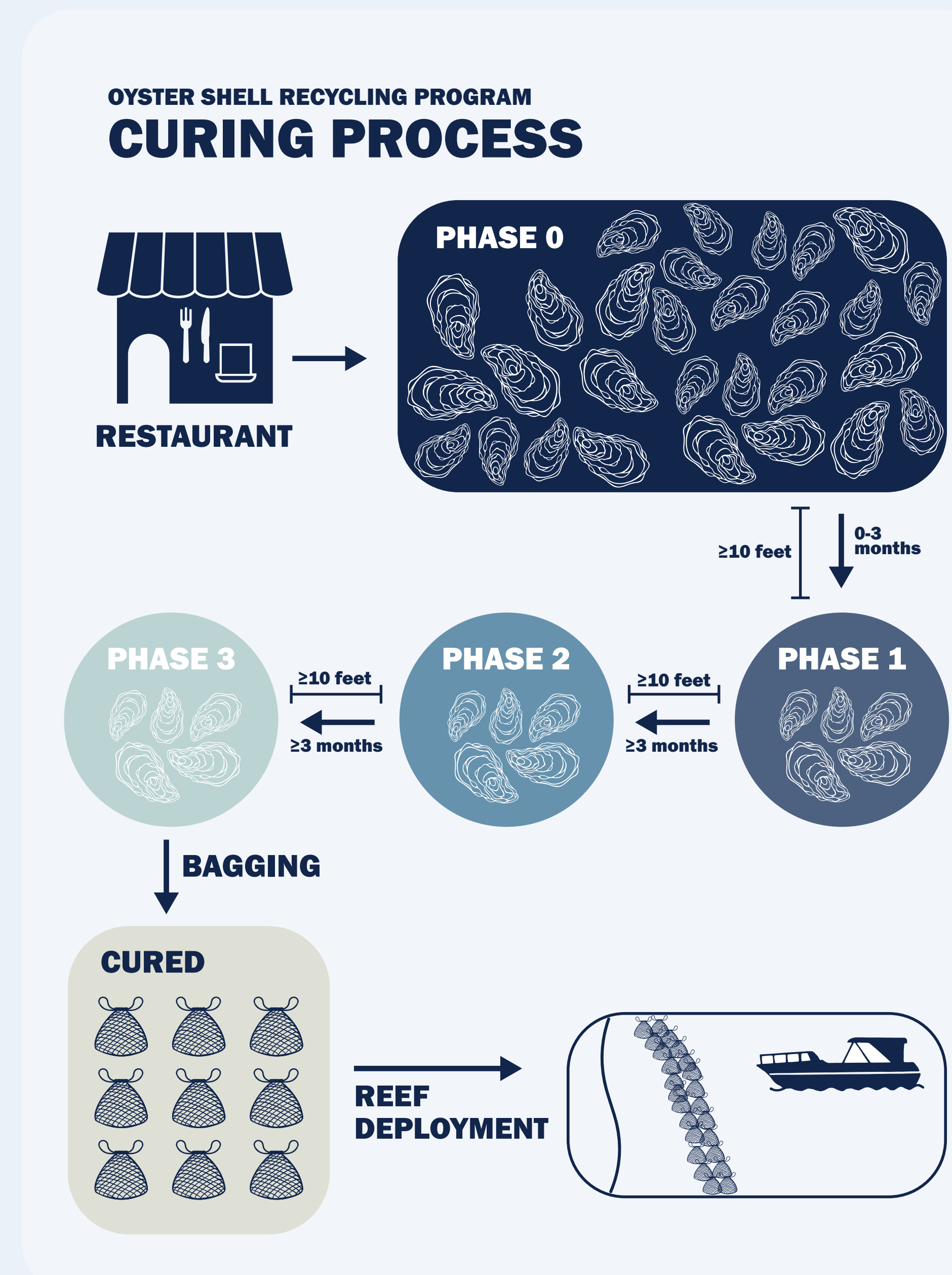
In Coordination with Coalition to Restore Coastal Louisiana

ABSTRACT

Understanding microbial communities in conservation and restoration programs that use food waste or human byproducts is critical to creating efficient and effective management strategies. The practice of oyster shell recycling, where spent oyster shells are taken from restaurants and diverted to artificial reefs, is a practice that is highly susceptible to pathogens. However, practices such as this one are valuable as citizen science programs that actively engage local communities in solutions to regional challenges. The introduction of bacterial pathogens has historically warranted a six month curing process. However, the process of curing the shells to remove pathogens is largely unstudied and thus has resulted in ambiguous curing timelines which could be too short and introduce pathogens to coastal water or too long and act as a drain on resources. In this study, we examined microbial communities throughout the oyster shell curing process to determine bacterial community shifts and identify potential pathogens being introduced into the waters where the artificial reefs are built.

BACKGROUND

- Oysters are not only a cultural food staple in coastal communities but also provide significant economic value in regions with established fisheries.
- Historically, oyster shells, which make up about 75% of an oyster's biomass, were considered waste and sent to landfills.²
- While there has been some use in commercial applications, such as cement production, livestock feed, and limestone substitution in fertilizer^{3,4}, some coastal community members are seeking to maximize oyster shells in locally impactful environmental management efforts.
- Numerous organizations across the country have been established over the past 20 years to take spent oyster shells from restaurants and deploy them back to the coast to create living shoreline barriers that mitigate wave action and create self-sustaining, biodiverse reefs.¹
- These reefs create a foundation for oyster larvae to attach⁶ and can provide a habitat for over 170 species such as fish, shrimp, and crabs.⁷
- These reefs address two major environmental problems in Louisiana: coastal land loss and the sustainability of fisheries.
- Coalition to Restore Coastal Louisiana (CRCL) has their own Oyster Shell Recycling Program.
- Shells are collected from restaurants in the Greater New Orleans Region and then transported to the CRCL Oyster Shell Recycling Site in Violet, LA where they undergo a minimum 6 month curing process to ensure that potential pathogens from the restaurants do not enter natural waterways during reef construction
- On a larger-scale, debris is dried and rinsed through natural weather patterns. On a microbial scale, ultraviolet light from the sun denatures DNA thus killing potentially pathogens.
- In the Gulf, *Vibrio splendidus* and other bacterial pathogens in this genus have been known to be pathogenic to oysters⁵ but transmission route and potential pathogens being introduced from restaurants has not been studied in depth.



The 6 month curing process for the oyster shells is separated into four phases delineated by time and space. Oyster shells are placed in Phase 0 for up to 3 months before they begin the 6 month curing process.

OBJECTIVES

- Identify pathogenic bacteria through the curing process
- Identify spatial or temporal relationships between bacteria through each phase of the curing process

Implication: create a recommended curing process that is efficient and safe.



Pointe-au-Chien Oyster Shell Reef on day of deployment in Montegut, LA (Photo courtesy of Coalition to Restore Coastal Louisiana)

METHODS

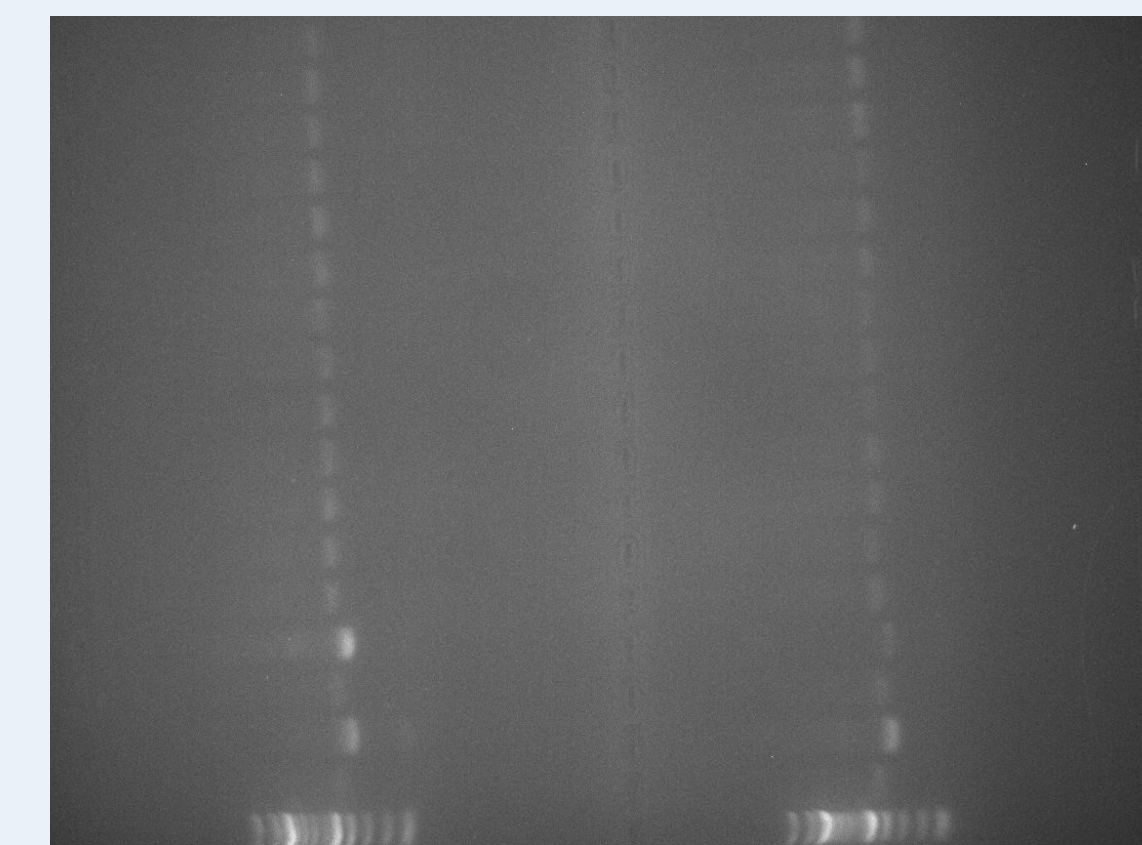
- 16 DNA samples were taken using sterile swabs and DNA shield for transport and downstream application
- Samples were taken from each phase in addition to the cured pile
- 8 of the 16 samples were taken from the outermost exposed part of the shell pile. The additional 8 of the 16 samples were taken 12 inches below the surface of the pile.
- DNA was isolated and is undergoing 16S Next Generation Sequencing to determine the diversity and relative quantities of bacteria at each phase
- Samples will be analyzed for temporal relation (through each phase of the curing process) and for spatial relation (from outermost exposed portion to 12 inches below) at each phase



Curing piles with flags notating where samples were to be taken



Sample being taken from oyster shell



Gel showing successful PCR amplification and barcoding

HYPOTHESIS & MOVING FORWARD

- Our hypothesis is that the current time-frame and method is more conservative than necessary thus providing an opportunity to increase efficiency in the curing process
- We expect to see a decrease in microbial diversity through the curing process as well as at each phase from the the outermost to the inner layer
- Contact with sun/air may be more important in lowering microbial density on oyster shells
- Knowing that there are potential protozoan and viral pathogens, future studies could analyze these microbes to gain a more comprehensive view of the curing process

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