Identifying and Prioritizing Research Required to Evaluate Ecological Risks and Benefits of Introducing Diploid *Crassostrea ariakensis* to Restore Oysters to Chesapeake Bay

Report of the STAC Workshop
December 2-3, 2003
Annapolis, Maryland

STAC Publication 04-002

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Publication Date:
February 2004

Publication Number:
04-002

Cover photo of *C. virginica* provided by the Virginia Institute of Marine Science.

STAC Administrative Support Provided by:
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EXECUTIVE SUMMARY

Heavy fishing pressure, habitat degradation and high disease mortality have driven native oyster (*Crassostrea virginica*) populations to historic low levels in Chesapeake Bay. In response, the states of Maryland and Virginia are considering introducing the Asian Suminoe oyster (*C. ariakensis*) with the goal of establishing a naturalized, self-sustaining population.

Neither the potential risks nor the potential benefits of such an introduction are adequately known at this time. The scientific community agrees that an introduction of diploid *C. ariakensis* is likely to be irreversible (NRC 2004), and that the spread of *C. ariakensis* beyond the borders of Chesapeake Bay is inevitable if a self-sustaining population is established. Further, the potential for novel interactions between oyster pathogens— those resident in the Bay and others that may emerge—and *C. ariakensis* is uncertain and impacts may be unpredictable both for this oyster and for other species over time. Given the long-term implications of an introduction, sound scientific information must form the basis of the environmental impact statement (EIS) that will assess the proposed introduction as well as other alternatives.

The Scientific and Technical Advisory Committee (STAC) of the Chesapeake Bay Program convened a workshop of research scientists in Annapolis on December 2-3, 2003 to discuss and prioritize research needed to fill critical gaps in our ability to predict risks and benefits that might result from an introduction of diploid *C. ariakensis* to Chesapeake Bay. *The outcome of this effort represents a disciplined approach to prioritize research needs—only those that were considered to be most important are reported here.* The specific research recommendations found in this report address issues of the genetics, biology and ecology of *C. ariakensis* that should be clarified prior to a final decision on the introduction of diploid individuals to Chesapeake Bay.

Meeting organizers and attendees were specifically focused on research to inform the primary proposal put forward by Maryland and Virginia - i.e., the introduction of 'Oregon' C. *ariakensis*. – because of the urgency of this issue. Previous workshops have provided guidance on research needs for native oyster restoration and oyster disease—issues of critical importance to Chesapeake Bay. Because of the scarcity and limited genetic diversity of the 'Oregon strain', recommendations below that specifically target this organism should be considered to be more generally applicable to whichever stocks of *C. ariakensis* are considered for introduction. In addition, wild-strain and disease-tolerant *C. virginica* will clearly need to be used as the benchmarks against which to assess the risks and benefits of *C. ariakensis*.

Research is needed to address the following four critical questions that must be answered to assess both the risk and the potential for success of an introduction of the 'Oregon strain' of *C. ariakensis*:

1) Can self-sustaining populations of *C. ariakensis* be established in Chesapeake Bay, and is there a greater likelihood of successful restoration using 'Oregon' or other strains of *C. ariakensis* than using wild- or disease tolerant strains of *C. virginica*?
2) What risks does *C. ariakensis* pose to *C. virginica* and other bivalve species, within Chesapeake Bay and in regions outside the Chesapeake?
3) What ecosystem services (*e.g.,* water quality improvement through filtration, provision of vital habitat) might be provided by *C. ariakensis* relative to those already demonstrated for *C. virginica*?
4) Will *C. ariakensis* accumulate human pathogens to a greater degree than *C. virginica*, thereby impacting the economic viability of the fishery?

There are global challenges that must be met to insure timely completion of research needed to answer these questions. Sufficient investments to facilitate multi-institutional multidisciplinary, integrated approaches, and support necessary infrastructure are essential. In addition, a commitment to independent peer review of proposals for funding, coordination of research efforts and ongoing independent technical review of outcomes must be integral to the EIS process. There was a consensus that most of the priority data gaps can be filled within a 5-year time frame if these challenges are met, and if sufficient funding is available to allow projects to start during Years 1 and 2

Overarching Recommendations of the Workshop:
In addition to specific research recommendations, several overarching recommendations should guide the design and scope of research:

- Research on *C. ariakensis* should use *C. virginica* as the baseline comparison against which to weigh benefits and risks.
- Responses of *C. ariakensis* should be compared with those of wild-stock and selectively bred, disease-tolerant 'strains' of *C. virginica*.
- Continued development of a well-integrated, science-based program for native oyster restoration is required, including adequate scale of effort, research on optimal restoration methods, and research on sources of disease resistance.
- The potential for important phenotypic and genotypic variation in *C. ariakensis* and *C. virginica* needs to be incorporated in the design and scope of research to ensure the generality of results and to determine if the 'Oregon strain' of *C. ariakensis* is the best choice for a *C. ariakensis* introduction.
- The ability to predict responses of diploid *C. ariakensis* from experiments using triploid animals must be evaluated since some experiments, including field deployments, can only be done with triploids of the non-native species.
- Models of various types are needed to integrate the results of completed and future research, to provide a tool for prediction, and to inform economic and risk analyses. Modeling and experimental work should progress hand in hand through an iterative process.

High and Essential Priority Research Recommendations:
Specific research recommendations rated as *essential, high or moderate* priority are listed in Table 1 and described in the body of the report. Research considered essential and of the highest priority to address the four critical questions noted above includes:

- *Oyster Disease: Bonamia* susceptibility; Herpes virus vertical transmission; the potential for increased disease transmission between species;
- *Reproduction and Genetics:* Genetic variation among *C. ariakensis* strains, and phenotypic variation within strains, that would affect which strain is best suited for introduction; reproductive rates and processes of adults, reproductive interference between *C. ariakensis* and *C. virginica;.*
- *Physiology:* Growth, environmental tolerances and other vital responses of adults and larvae; behavioral and settlement responses of larvae; postsettlement mortality
- *Ecological interactions among oyster species*: Competition between *C. ariakensis* and *C. virginica*; how the timing of reproduction is likely to affect the outcome of competition.
- *Environmental Services:* Growth form and reef building potential of *C. ariakensis* under a variety of environmental conditions; consequences of those growth forms to other organisms.
- *Modeling and Prediction:* Models to predict larval dispersal, the potential for population growth, and habitat effects on these processes; incorporation of results into oyster population models.
- *Human Pathogens:* Identification of research needs related to human pathogens by those with expertise in shellfish sanitation, potentially including monitoring of pathogen loading and retention.

This document represents the judgment of scientific experts from both within and outside the Chesapeake Bay region. The credibility of decisions surrounding the proposed introduction of *C. ariakensis* depends upon the quality of the science that underpins the decision-making process— a process that ultimately, will require management and the scientific communities to work in concert to achieve an outcome in the best interest of the long-term health of Chesapeake Bay.

WORKSHOP REPORT

BACKGROUND

Native oyster (*Crassostrea virginica*) populations in the Chesapeake Bay are at historic low levels as a result of heavy fishing pressure and habitat degradation during the 19th and 20th centuries and recent high mortalities due to the diseases Dermo and MSX. In response, the states of Maryland and Virginia are considering introducing the Asian Suminoe oyster (*C. ariakensis*) into the tidal waters of the Bay with the goal of establishing a naturalized, reproducing, and self-sustaining population of this non-indiginous species of oyster.

Neither the potential risks nor the potential benefits of introducing *C. ariakensis* to the Chesapeake Bay are adequately known. The current understanding of the biology and ecology of *C. ariakensis* is insufficient to predict whether an introduction will provide desired benefits or have a substantial adverse impact within the Bay or other Atlantic Coast estuaries over short or long time scales. The National Academy of Sciences (NAS) has concluded that an introduction of diploid *C. ariakensis*, either as a result of intentional pursuit of the diploid introduction strategy or as a consequence of extensive triploid aquaculture, likely would be irreversible (NRC 2004).

Given the long-term implications of an introduction, it is important that sound scientific information be available to inform the decision-making process. To help address this need, the Scientific and Technical Advisory Committee (STAC) of the Chesapeake Bay Program convened a workshop in Annapolis on December 2-3, 2003. The workshop was requested and funded by the Chesapeake Bay Program and NOAA. Researchers with expertise in oyster biology, invasive species, and estuarine ecology from within and outside the Chesapeake Bay region participated in plenary sessions and workgroups charged with identifying critical information gaps and providing advice on priority research and monitoring required to meet those information needs. The workshop did not address social or policy issues, but instead focused on identifying scientific information that could contribute to an understanding of the ecological consequences of policy alternatives and decisions. A small workgroup discussed economics issues and risk assessment. However, very few experts in these areas attended the meeting and the workgroup did not propose research needs except to meet the currently proposed EIS scope and timeline; economics research recommendations are not included in this report.

The workshop deliberations used the recent NAS report and other available research plans[1] as a starting point and considered a broad a range of issues. While recognizing that

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Aquaculture of Triploid Crassostrea ariakensis in Chesapeake Bay. A Symposium Report. E. Hallerman, M. Leffler, S. Mills. S. Allen 2001 Maryland and Virginia Sea Grant College Programs

The Suminoe oyster, Crassostrea ariakensis, in Chesapeake Bay: Current Status and Near-term Research Activities W.L. Rickards, P.Ticco 2002, Virginia Sea Grant College Program the final decision-making framework has yet to be identified, a draft strawman proposal of the states' (Maryland and Virginia) preferred action and alternatives to be considered in the programmatic environmental impact statement (EIS) was provided to participants by the Maryland Department of Natural Resources (MD-DNR) for context and guidance (Appendix I; MD-DNR December 2003 draft).

The central focus of the draft MD/VA strawman document was the proposal to introduce fertile, diploid, 'Oregon strain' C. ariakensis to Chesapeake Bay, and the need to evaluate the risks and benefits of this action. Accordingly, the primary focus of research discussions and recommendations at the workshop was the urgent need to evaluate risks and benefits associated with this proposed action.

The draft MD/VA strawman proposal also identified six alternatives to the introduction of 'Oregon strain' *C. ariakensis*, three of which received considerable attention during workshop deliberations:

- Continued restoration using wild-type *C. virginica* from Chesapeake Bay,
- Restoration utilizing *C. virginica* strains that had been bred for enhanced resistance to Dermo and MSX, and
- Restoration using strains of *C. ariakensis* other than the single proposed strain derived from stocks from Oregon.

These alternatives received attention because it was considered impossible to evaluate the risks and benefits of 'Oregon' *C. ariakensis* without comparing its performance to *C. virginica* and other *C. ariakensis.* The workshop scope and goals did not include development of specific recommendations on research required to determine how to best utilize native oysters (wild or disease-tolerant strains) for restoration, the likelihood of successful restoration using native oysters, or strategies and techniques to improve disease tolerance of native oysters.

The purpose of this workshop was to:

1) ***Identify specific information needed,***
2) ***Recommend research needed to collect that information in a timely manner, and***
3) ***Prioritize research required to predict the risks and benefits of introducing fertile diploid C. ariakensis to Chesapeake Bay relative to the risks and benefits***

of other alternatives proposed to meet the goal of restoring oysters in Chesapeake Bay.

The workshop did not:

1) Specifically evaluate impacts on an oyster fishery (with the single exception discussed under ‘critical issues’, below)
2) Evaluate the full suite of economic, social and management issues, or
3) Develop actual risk assessment and decision frameworks.

Subsequent to this workshop, a Notice of Intent for the U.S. Army Corps of Engineers to prepare a Programmatic Environmental Impact Statement was published in the Federal Register (January 5, 2004, Vol. 69, Number 2: 330-332). This Notice of Intent contains the same proposed action and list of alternatives as the draft DNR outline used to guide the discussions at this workshop.

Applying Science to Risk Analysis

Because risk-benefit analyses will form the foundation for the decision-making process regarding a planned introduction of *C. ariakensis*, workshop participants sought to identify existing uncertainty in critical biological, ecological, and economic areas and prioritize information that is needed to reduce that uncertainty.

Given this context, it was apparent to all participants that definitive data *can* be collected to address many important and specific issues. However, because of the complexity, duration, and cost of research to understand critical aspects of a number of high priority issues, defining risks and benefits will require a substantial commitment on the part of the research and management communities. ***Given the magnitude of the decision, and the importance of understanding unintended consequences or outcomes of an introduction, it is important that decision makers give full and careful consideration to the workshop conclusions and the prioritized research recommendations presented in this document.***

Workshop Structure

The workshop was divided into two parts: 1) plenary sessions with presentations from noted experts, and 2) facilitated workgroups that built upon the plenary talks to develop detailed research priorities. A complete list of speakers and workgroups is found in Appendix II.

Plenary Sessions:

Plenary presentations and discussions addressed the state of the knowledge *of* C. ariakensis***, the use of scientific data and the importance of identifying information that reduces uncertainty in a risk-based decision analysis process. Plenary speakers addressed the geographic spread of non-native species, the ecological impact of invasive species and the influence of ecosystem health on ‘invasibility’ within the***

context of disease ecology and community and population interactions. Economic considerations and the history of oyster introductions in Asia, North America and Europe were also discussed.

Themes and charges to workgroups:
Workgroups provided the opportunity to develop consensus on questions that must be answered to inform the decision-making process regarding a *C. ariakensis* introduction in Chesapeake Bay. Recognizing that expertise on some issues may have been concentrated in a few individuals, facilitators worked with the workgroup participants to develop recommendations for a phased, prioritized research program.

Particular emphasis was placed on:

- The importance of the scientific data for decision-making,
- The sequence of research efforts, and
- The required duration of research efforts.

What emerged from this effort is an identification of information gaps and a prioritization of what will be needed to fill those gaps in order to make an informed decision regarding introducing diploid *C. ariakensis*. All issues identified by the workgroups were considered important on some level, but some were given lower priority with respect to their relevance to the decision-making process. Low priority issues were not included in workgroup recommendations, and are not included in this report. Thus *the high-to-moderate ranking of all topics in this report should not be viewed as a non-prioritized 'laundry list', but instead reflects considerable restraint by participants in limiting research recommendations to topics that have the potential to make a substantial contribution to the ability to predict risks and benefits of introducing diploid C. ariakensis to Chesapeake Bay.*

WORKSHOP RECOMMENDATIONS AND CONCLUSIONS:

<u>Clarification regarding the 'Oregon strain' of *C. ariakensis*</u>

The MD/VA draft recommendations for the EIS that was available at the time of the workshop and the subsequent USACOE Notice of Intent specifically identify the 'Oregon strain' of *C. ariakensis* as the proposed source for a diploid introduction. The workshop participants were nearly unanimous in their belief that it was important to consider a range of both phenotypic and genotypic variation within this species in assessing the risks and benefits of an introduction. We were nevertheless conscious that diluting the research effort and limited resources across a wide variety of stocks or strains of *C. ariakensis* could delay acquiring the information needed to inform the management decision. Thus, it would seem to be necessary to apply some limits to the call for investigating an extremely wide range of *C. ariakensis* stocks.

The term 'Oregon strain' has been used to refer to descendents of a small number of *C. ariakensis* that were originally introduced by accident in shipments of *C. sikamea* from

Japan to Oregon during the 1970's. A small number of these animals were spawned in a hatchery and attempts were made to establish populations from northern California to Washington. There are no reports of any of these populations becoming successfully established, however (NRC 2004 and references therein). At the present time there are no confirmed *C. ariakensis* in Oregon, either in the wild or in hatcheries (C. Langdon, pers. comm.). One commercial aquaculture company in Washington has a limited supply of *C. ariakensis* in their hatchery (85 diploid animals), but no field populations are established in Washington State (Bill Dewey, Taylor Shellfish Farms, pers. comm.). The initial importation of this stock of oysters to the east coast was to Rutgers University in the early 1990's, where they were kept in quarantine systems and further inbred. This is one of several genetic bottlenecks where the breeding population may have been represented by as little as six or fewer adults.. Presently there are only a few dozen diploid *C. ariakensis* from this strain at Rutgers (Ximing Guo, Rutgers University, pers. comm..). Offspring from this Rutgers line have been spawned in the VIMS hatcheries at Gloucester Point and Wachapreague. The total numbers of the animals currently available totals just over a thousand animals. This is almost certainly too few animals to initiate an aggressive introduction effort. Moreover, given the very restricted nature of the original gene pool these stocks have very reduced genetic diversity relative to wild stocks. Because of successive inbreeding in the hatchery, these stocks are in great danger of suffering from inbreeding depression, which can severely limit their fitness. It would seem inevitable that an introduction would require that more brood stock be obtained from the native range of *C. ariakensis*.

The current state of our knowledge suggests that there are genetically distinct Northern (from north China, southern Japan, and probably Korea) and Southern (from south China and perhaps Vietnam) stocks. The 'Oregon strain' oysters represent a very restricted subset of the Northern stock. Thus, the proposed action of introducing the 'Oregon strain' will seemingly require obtaining additional Northern stocks of *C. ariakensis* from Asia, which themselves exhibit phenotypic and genotypic variations. The minimal approach, therefore, to clarifying risks and benefits associated with an introduction will require that the diversity associated with the Northern stocks be incorporated into research efforts.

Recommendations below that specifically target the 'Oregon strain' should be considered to be more generally applicable to whichever stock of C. ariakensis is considered for introduction.

Consensus from Plenary Sessions:

There was general consensus on a number of important points raised in the plenary presentations and these became the basis for discussions and research recommendations in subsequent workgroup sessions:

- There are <u>major</u> gaps in the understanding of the genetics, biology and ecology of *C. ariakensis* that should be addressed prior to a final decision on the introduction of diploid individuals to Chesapeake Bay.
- A risk-based decision framework that considers both the potential negative impacts and the benefits of *C. ariakensis* relative to other proposed alternatives should help prioritize research needs and guide the decision-making process. Research that reduces uncertainty in predicting risks and benefits is of highest priority.
- The effect of any single non-native species, including oysters, can range from economically beneficial to economically and ecologically harmful across the variety of locations in which it becomes established.
- The spread of *C. ariakensis* beyond the borders of Chesapeake Bay is inevitable if a self-sustaining, population of diploid *C. ariakensis* is established. Research will therefore need to consider the potential risks outside Chesapeake Bay.
- The potential for novel coupling between oyster pathogens (resident and non-resident) and an introduced species is uncertain and impacts may be unpredictable both for the host and for other species over time.
- Within the context of the U.S. oyster industry, the economic value of a restored Chesapeake Bay fishery is low because an adequate supply of oysters exists nationwide to meet current market demand. In a broader context, investment for ecological services may yield greater economic impacts, but there is still considerable scientific uncertainty as to whether restoration utilizing *C. ariakensis* can be implemented in a manner that will result in significant, positive improvements in ecosystem services provided by oysters in Chesapeake Bay.

<u>Risks to Chesapeake Bay and the Likelihood of Achieving Desired Benefits:</u>

Three critical issues that defined the major risks and likely benefits of introducing diploid *C. ariakensis* to Chesapeake Bay were identified in research recommendations from all three workgroups focusing on biological and ecological processes. Although the expertise of each workgroup varied, the fact that all ultimately reached similar conclusions with regard to the importance of these broad questions accentuates the need to address them as a foundation for the decision-making process.

The three critical issues identified by all workgroups were as follows:

Issue 1: Will C. ariakensis thrive in Chesapeake Bay and provide greater likelihood of success for oyster restoration efforts than would use of wild- or disease tolerant strains of *C. virginica*?

Issue 2: What risks does C. ariakensis pose to C. virginica and other bivalve species both within Chesapeake Bay and in regions outside the Chesapeake?

Issue 3: What are the risks and benefits to ecosystem services likely to be provided by *C. ariakensis* relative to *C. virginica* ?

In addition, there was agreement on the potential importance of a fourth issue proposed by one of the workgroups:

Issue 4: Although the workshop did not specifically address risks and benefits to fisheries, one fishery-related question was deemed sufficiently pressing to include in the research recommendations: ***Will C. ariakensis accumulate human pathogens or E. coli to a greater degree than C. virginica,*** thus increasing the incidence or extent of fishery closures?

The importance of these issues, and how much uncertainty is considered acceptable for each, is a policy question – not strictly a scientific one. For example, it is up to the decision-makers whether to consider risks to native oysters in the decision-making process. The guidance provided in this document is a listing and explanation of the research that scientific experts from within and outside the Chesapeake Bay region recommend as important to substantially reduce uncertainty in predicting risks and benefits of an introduction of diploid *C. ariakensis* to Chesapeake Bay. The workshop was not designed to determine whether those risks were socially, politically or economically acceptable, or whether the potential benefits were valuable to society.

Overarching Recommendations:

While each workgroup developed a detailed list of priorities, a number of common, overarching themes emerged across all the groups. These represent global challenges to decision-making and are critical issues that should guide the design of research and the scope of research funding:

Research on C. ariakensis should use C. virginica as the benchmark against which to weigh benefits and risks.

- The risk of introducing a non-native species will never be zero. Therefore, it is important to obtain sufficient information to predict whether the benefit of a *C. ariakensis* introduction is likely to substantially exceed benefits of restoration utilizing wild or selectively-bred strains of the native oyster species.
- Both the likelihood of success of restoration efforts, and the benefits expected to accrue from similar-sized populations of the various oysters should be considered.

Responses of C. ariakensis should be compared with those of wild-type and selectively bred, disease-tolerant 'strains'of C. virginica.

- Generation of comparative databases will provide a context regarding the ecological performance and characterization of the two species, and potentially which *C. virginica* or *C. ariakensis* 'strains' would most likely become self-sustaining populations in the Chesapeake Bay.
- This report does not make specific recommendation on the development and testing of disease tolerance in *C. virginica*. These topics are important and were the focus of a separate Sea Grant-sponsored workshop held during September 2003 (workshop report ref to be added to be added in final draft).

The ability to predict responses of diploid C. ariakensis and C. virginica from triploid animals should be determined.

- Many experiments, especially those requiring field deployments, will need to utilize triploid *C. ariakensis* because of the danger of accidental release of fertile animals or their gametes to the field. There are potential differences in growth, tolerance of environmental stress, and disease susceptibility between diploid and triploid animals because of the energy that diploids expend on reproduction. The magnitude and existence of differences between diploid and triploid oysters is important to determine in order to use results of research on triploids to predict risks and benefits of an introduction of diploid *C. ariakensis.*

Continued development of a well-integrated, science-based program for native oyster restoration is required, including adequate scale of effort, research on optimal restoration methods, and research on sources of disease resistance.

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- Although the design of future native oyster restoration efforts was beyond the scope of the current workshop, the need to continue efforts on native oyster restoration was a clear and emphatic priority of workshop participants.
- While previous oyster restoration efforts in the Bay have shown mixed results, recent research studies (e.g., using disease-tolerant native oyster 'strains') have shown promise. The reasons for past failures as well as development of new strategies for oyster restoration should be investigated. Continued restoration research efforts will contribute important information for both native oyster restoration and on protocols for introducing *C. ariakensis* into the Bay, should that decision be made.

The potential for important phenotypic and genotypic variation in C. ariakensis and C. virginica needs to be incorporated in the design and scope of research. We do not know if the 'Oregon strain' C. ariakensis is the type of C. ariakensis that would have the greatest potential to form self-sustaining populations in Chesapeake Bay, whether it would provide the greatest ecological benefits if it does become established, or whether it poses the lowest risks. The generality of results will be greater, and the degree of uncertainty in characterizing risks will be reduced by approaches that compare and contrast different oyster strains and consider the range of environmental conditions in Chesapeake Bay and other Atlantic and Gulf Coast estuaries.

- Across its geographic range, *C. virginica* varies in physiological tolerances, vital rates and susceptibility to various pathogens. At present, there is no reason to assume that this would not also be the case with *C. ariakensis*—necessitating research across a spectrum of environmental conditions.
- It is important to determine whether variation observed in the field in the native range of *C. ariakensis* results from genotypic variation or environmental cues. Genetic markers and phylogenetic characterization is essential for further studies.

Models of various types are needed to integrate results of a range of experiments and to provide a tool for prediction, as well as to inform economic and risk analyses.

Modeling and experimental work should progress hand in hand through an iterative process.

- A diversity of modeling approaches is available, and several different modeling approaches should be encouraged. These could range from simple mass-balance box models to more sophisticated spatially-explicit models that are linked with hydrodynamic-larval transport models and consider a range of local habitat conditions. Model development should be conducted in a phased/sequenced (iterative) approach between data collection and model development to insure that the models provide ecologically relevant outputs and that the proper types of data are being collected for input into the models.

The best way to generate information that will contribute to decision-making on this issue is through multi-institutional, multidisciplinary, integrated and interactive approaches.

- Coordinated research efforts among scientists, research/academic institutions and state/federal agencies is required at field sites within the Chesapeake Bay and in regions along the East and Gulf Coasts of the US. Care should be taken to standardize research protocols so that data generated can be statistically compared across regions in the Bay (and outside the Bay). Collaborative research simultaneously conducted at locations that span the range of salinities and other environmental variables to which oysters, pathogens, predators and competitors could be exposed will help ensure that evaluations of risks and benefits of a *C. ariakensis* introduction are not idiosyncratic as a result of conditions at a single location. Finally, given the relatively tight timeline to assess the effects of the potential *C. ariakensis* introduction, coordinated efforts will insure the most efficient and rapid means of data collection.

<u>Specific Research Recommendations:</u>

The following section of the report lists and explains the specific research questions and data needs recommended as high or moderate priority by workgroups to address the four major issues identified. Considerations listed under ‘Overarching Recommendations’, such as the need to investigate multiple strains and use *C. virginica* as a benchmark, provide strong guidance on the design of studies. Workshop participants were specifically instructed to include topics that were the focus of current studies unless those studies had already reached conclusive results. These recommendations are also summarized in Table 1.

The lack of specific research recommendations on native oyster restoration techniques and disease-tolerant *C. virginica* strains in this report is not a reflection of the importance of these two issues. Rather it reflects the attempt on the part of participating researchers to keep the focus on the primary proposal by MD and VA (i.e., the introduction of diploid *C. ariakensis*) and to avoid duplication of previous workshops that focused specifically on native oyster restoration and oyster disease.

Although 17 essential and high-priority research topics were identified, it is important to note that there are several cases where more than one question can be addressed within a single study. For example settlement substrate preferences of *C. ariakensis* and *C. virginica* can be studied individually and in combination to address issues on the potential for *C. ariakensis* to thrive or become a nuisance, and also whether it will increase or decrease settlement substrate available to native oysters. Similarly, studies to predict growth may be efficiently combined with studies of reproductive rates and timing. There are also cases where more than one study, or a complex, multi-institutional approach is required. For example, interactions with other species and the extent to which *C. ariakensis* performs the same ecosystem services as *C. virginica* may vary among different regions of the Bay.

ISSUE 1.Will *C. ariakensis* survive and reproduce in Chesapeake Bay and provide greater likelihood of success for oyster restoration efforts than would use of wild- or disease tolerant strains of *C. virginica*?

General Considerations

Evaluating the risks and the benefits of an introduction of *C. ariakensis* relative to other alternative actions will necessarily depend upon (1) predicting not only if, but where, populations of this oyster will become established and (2) predicting population growth potential in the Chesapeake Bay and other regions. Ultimately, most issues related to the potential ecological and economic impacts of introducing this species will be dependent upon the size and spatial distribution of populations that become established. Indeed, the current level of interest in this species is driven by early indications that it has potential to survive and grow in the region, rather than from information about the economic or ecological role of this species worldwide.

Estimating the potential for *C. ariakensis* populations to become established and grow requires information about disease susceptibility, physiological requirements, reproductive biology, habitat requirements and population growth parameters under a broad spectrum of environmental conditions. It is important to obtain information not only on individual stressors, but also on multiple stressor interactions. There was general recognition by the workshop participants that an appreciation of the range of phenotypic variation and underlying genotypic variation would be essential to understanding how *C. ariakensis* would perform in a range of habitats if introduced to the Chesapeake Bay.

Diseases, Harmful Algal Blooms, and Toxins

The susceptibility of *C. ariakensis* to pathogens, pests, harmful algal blooms and toxins could have very significant consequences for the establishment and growth of the species. The benefit of using this non-native species for restoration instead of the native oyster may be particularly dependent on any differences in the susceptibility of *C. ariakensis* relative to wild- or disease-tolerant strains of *C. virginica*. Results of previous and ongoing field trials using triploid *C. ariakensis* in Chesapeake Bay indicate that this species is resistant or has low susceptibility to *Haplosporidium nelsoni* (MSX) and *Perkinsus marinus* (Dermo). However, several other known disease-causing pathogens may represent significant risks if *C. ariakensis* is susceptible to them. Understanding the risk of disease mortality under conditions in Chesapeake Bay and areas where *C. ariakensis* may eventually spread is considered to be of great importance. In addition, there are numerous transport mechanisms, including ballast water, that may introduce pathogens from other areas that may impact *C. ariakensis*. There was a strong recommendation that the individual and interactive effects of host species, host density dependence, host spatial distribution and environmental factors (e.g., salinity be considered in studies of disease transmission.

Research recommendation: <u>Determine the susceptibility of *C. ariakensis* to known disease-causing parasites and pathogens.</u> Specific pathogens of concern are listed below in order of priority. The workgroups also identified a number of other issues that warranted consideration, including susceptibility to pest species (e.g, *Polydora* spp. and boring sponge), harmful algal blooms (e.g, *Prorocentrum*), and toxins, but none were considered high or medium priority research issues.

- *Bonamia* spp. – *B. ostrea* is present in *Ostrea edulis* oysters in Maine and in France and a *Bonamia*-like parasite has been detected in *C. ariakensis* in flow through systems in France. In addition, a species of *Bonamia* recently has been detected at high prevalence and is believed to have caused extensive oyster mortality in *C. ariakensis* being held in Bogue Sound, North Carolina. The impact of *Bonamia* on *C. ariakensis* under environmental conditions relevant to the mid-Atlantic region is a critical research need. **Timeframe: 2 years. Priority: Essential**

- Herpes virus – Herpes viruses have been found in *C. ariakensis* in its native range and are reported to cause larval mortality in hatcheries in other oyster species. Vertical transmission, from parent to offspring, is suspected, but needs to be verified through laboratory experiments. If vertical transmission occurs there are implications for brood stock and hatchery management. **Timeframe: 2-4 years. Priority: High**

- *Perkinsus* species other than *P. marinus* – Other species of *Perkinsus* exist in Chesapeake Bay and in other areas of the world. Of particular importance is *Perkinsus olseni*, which occurs in the eastern Atlantic Ocean. The susceptibility of *C. ariakensis* to infection to other *Perkensis* sp. and their pathogenicity should be investigated. **Timeframe: 2 – 4 years. Priority: Medium**

In addition, sufficient ***funds should be held in reserve*** to screen *C. ariakensis* used in any studies for pathogens if high morality rates or other indications of disease occur. As increasing numbers of *C. ariakensis* are exposed to a broad range of Chesapeake Bay and other Atlantic estuary environments, it is possible that pathogens not specifically identified above as high priority for specific studies may emerge as potentially important disease agents.

Genetics-related issues

Much of our information on the biology of *C. ariakensis* in the field has been obtained from research performed on reproductively sterile triploid oysters. Of major concern is that we do not know how useful this information is in predicting the potential performance of diploid oysters in the natural environment. Consequently, predictions based on the response of triploids held under field conditions in the Chesapeake Bay may not adequately predict the survival and potential distribution of diploids. In addition to growth differences between triploids and diploids there are questions concerning whether or not their stress tolerances to such factors as low dissolved oxygen, harmful algal blooms, and salinity fluctuations are similar. The responses of triploids and diploids need to be intercalibrated under uniform conditions so that data obtained from triploids can be appropriately corrected when being used to predict the response of diploids.

In addition to using triploid *C. ariakensis* as a tool for predicting the field performance of diploids, their use in aquaculture was proposed by the NAS report as at least a near-term alternative to a diploid introduction. It is now known that as these triploids age, some individuals gradually become capable of producing gametes. There is still incomplete knowledge of the reproductive biology of triploids, how reversion to diploidy varies across different genetic strains used for either scientific experiments or commercial production, and thus what risk triploids may pose of resulting in a diploid introduction.

A strain of *C. ariakensis* originally derived from oysters introduced to Oregon has been used in most of the triploid studies undertaken at VIMS and is currently being considered as the preferred candidate for introduction of diploids to Chesapeake Bay. Within the genus *Crassostrea,* individual species have often been observed to exhibit considerable phenotypic variation. Our current knowledge suggests that *C. ariakensis* may similarly exhibit significant phenotypic variation across its range. Clarifying the genetic versus environmental basis of this variation will be crucial to predicting how *C. ariakensis* will perform in an introduced environment.

Research recommendations:

- <u>Determine the efficacy, and quantify the uncertainty, associated with using triploids as surrogates for diploids.</u> Studies comparing the response of diploids and triploids may need to be conducted in the natal environment of *C. ariakensis* or areas where they have already been introduced, (e.g.,Oregon) in order to examine responses under field conditions, as well as under laboratory quarantine conditions. Studies should address not only growth differences, but responses to environmental stress and disease susceptibility. **Timeframe: 2 – 4 years. Priority: High**

- <u>Investigate the stability of triploids and their longer-term reproductive potential.</u> Reversion of some triploid oysters to a 'mosaic' state (i.e., individual oysters containing both triploid and diploid cells) is a progressive process and the frequency of mosaics in the population increases over time. Thus, evaluating the reproductive potential of these mosaics would require 3 – 5 years in a newly initiated project. There are currently mosaics in Virginia that are up to 3 years old that are being held in quarantine conditions. A project initiated with them could be accomplished in a **Timeframe of 1 – 2 years. Priority: Medium under the current proposal for the introduction of diploid *C. ariakensis.*** However, if a conclusion is reached that the risk posed by a diploid introduction is unacceptably high, a fuller evaluation of risks posed by triploids will be imperative.

- <u>Elucidate genetic variation in selected *C. ariakensis* stocks from Asia and determine if this variation is related to important physiological and ecological traits that will affect the risks and benefits associated with introducing it to the Chesapeake Bay.</u> Of particular importance is determining whether or not different strains exhibit varying reproductive traits, physiological tolerances (e.g., salinity, dissolved oxygen, sedimentation) and growth forms. **Timeframe: 2 – 3 years. Priority: High**

Potential for Population Growth and Sustainability

The potential for population growth and sustainability are the factors upon which all of the ecological and economic benefits of an introduction of diploid *C. ariakensis* will depend. Characterization of potential population performance by *C. ariakensis,* in conditions representative of the range of environmental variation within the Chesapeake Bay and in regions to which the species would be likely to spread, was therefore considered crucial by workshop participants. There was broad consensus that reliable estimates of vital rates (e.g., size-specific growth, fecundity and mortality) will be critical to predicting potential population growth. In addition, data on larval behavior, dispersal patterns and mortality rates, larval settling preferences and post-settlement mortality are required. Assessing oyster population performance over a range of environmental conditions found within the Bay requires particular focus on the effects of salinity, temperature, sediment type/load, oxygen, food quality/quantity, predation and competition. Multi-factorial studies using combinations of these variables are also required.

There was also consensus that it was important to evaluate a diversity of 'strains' of *C. ariakensis*. This will require additional work in Japan, China and potentially Korea to collect and genetically identify oyster populations possessing different growth forms and living in different environmental conditions (discussed above).

This approach will provide valuable insight into the diversity of population dynamics of *C. ariakensis* and which 'strain(s)' will be most successful in the Bay. One scenario could be that several different 'strains' (e.g., low and high salinity ones, low and high turbidity ones) may be required in order to insure the successful introduction of *C. ariakensis* into the Bay ecosystem.

Research recommendations:

- Determine the physiological responses (e.g., growth, feeding and survival) to a range of environmental factors (e.g., salinity, D.O., temperature and seston) of adults and larvae of 'Oregon strain' *C. ariakensis* relative to responses of *C. virginica* and other *C. ariakensis* stocks. The workshop participants stressed that these studies need to embrace some of the range of genotypic variation of *C. ariakensis* in an effort to identify the stocks most suited for growth and survival in the desired areas of the Chesapeake Bay, and to compare performance of *C. ariakensis* with that of *C. virginica*. **Timeframe: 2 – 4 years. Priority: High**

- Determine vital reproductive processes and rates (, e.g., gametogenesis, spawning, fecundity, sex change) in Chesapeake Bay and other U.S. East Coast environmental conditions. This information will be especially crucial for parameterizing demographics models necessary for predicting population growth rates. **Timeframe: 5 years. Priority: High**

- Obtain information on the physiological and behavioral characteristics of *C. ariakensis* and *C. virginica* larvae to parameterize a comparative larval dispersal model. It is important to determine the environmental cues that affect the vertical position of planktonic larvae in the water column and to clarify any differences in this behavior among *C. ariakensis* strains under consideration. **Timeframe: 2 years. Priority: High**

- Determine settlement cues and substrate preferences for *C. ariakensis.* Since adequate substrate for settlement is a limiting factor for *C. virginica* populations in many areas of Chesapeake Bay, understanding the settlement cues and substrate requirements for *C. ariakensis* will be important for insuring successful establishment of populations. Additionally, this information is needed to evaluate the potential of *C. ariakensis* to become a nuisance fouling organism. **Timeframe: 2 years. Priority: High**

- Evaluate post-settlement mortality rates for *C. ariakensis*. Early post-settlement mortality rates can be extremely high in oysters and can have dramatic effects on population dynamics. Thus, it is important to determine how relative mortality rates of juvenile *C. ariakensis* and *C. virginica* are affected by low dissolved oxygen concentrations, sedimentation and predation. The latter may be especially important because differences in shell thickness and morphology of the two species may influence size-specific vulnerability to predators. **Timeframe: 2 – 3 years. Priority: High**

- Predict the likelihood that *C. ariakensis* and/or *C. virginica* will develop self-sustaining populations in the Chesapeake Bay under current conditions. Specifically, it is important to address how long it will take to develop populations of sufficient size and density to provide the desired ecological benefits and to support fisheries production. The research required for making such an evaluation is described above and should include laboratory studies with diploids, field deployments with triploids and studies of *C. ariakensis* in its native environment, in order to collect data on disease susceptibility, reproductive rates, settlement requirements and juvenile mortality rates. These data should be used to parameterize a demographic model to predict potential population growth rates, given current environmental conditions (including substrate limitation and high sedimentation rates) and disease in the Bay. It is important to model not only wild strain *C. virginica*, but also strains selectively bred for increased disease tolerance in order to predict which alternative provides the greatest potential for oyster restoration. Modeling efforts will hinge on assumptions that efforts to rehabilititate additional settlement substrate will continue or be expanded. **Timeframe:** Much of the laboratory and field data (described in preceding sections) can be collected over a 3-5 year period. Model development (which will likely involve adaptations of one or more of several existing oyster demographic models) can proceed concurrently and will likely require **1 – 1.5 years. Priority: High**

ISSUE 2. How will *C. ariakensis* affect the native oyster, *C. virginica,* in the Chesapeake and in regions outside of the Chesapeake?

General Considerations

Non-native species can cause or contribute to the decline of native species if the non-native establishes large, self-sustaining populations or increases disease problems. Species most strongly affected tend to be those with similar habitat requirements and those that may be affected by the same pathogens and parasites. Thus, the species most likely to be at risk due to an introduction of *C. ariakensis* is *C. virginica*. There are also mechanisms by which establishment of a second oyster species could enhance *C. virginica* growth or survival by altering habitat or disease dynamics in ways that would create a more favorable environment for the native oyster species. An important question is therefore whether the introduction of *C. ariakensis* to Chesapeake Bay would cause further decline or even local extinctions of *C. virginica,* or would increase *C. virginica* abundance both within and outside Chesapeake Bay.

Habitat preferences and environmental tolerances of different strains of *C. ariakensis* may overlap with native *C. virginica* to greater or lesser degrees, and therefore have differing effects on *C. virginica*. For example, if a management goal is to minimize risk to native oysters, a decision could be made to introduce a *C. ariakensis* strain that is less tolerant of low salinities than *C. virginica*, if such a strain exists. Thus, information on potential effects of *C. ariakensis* on *C. virginica* could provide the basis for decisions on how an introduction is done, as well as whether or not to introduce *C. ariakensis*.

Some of the data needed to address this issue will be generated by the experiments and models recommended under Issue 1. These studies could provide data and predictions on environmental tolerances and predicted population dynamics of several strains of *C. ariakensis* as well as native *C. virginica*. In addition, experimental tests directly examining the potential effect of *C. ariakensis* on *C. virginica* through altering disease dynamics, reproductive potential, growth and survival are needed. Because these factors may differ within and outside Chesapeake Bay, coordinated complementary studies should be conducted in northern and southern Atlantic Coast estuaries.

Disease dynamics

A non-native species could potentially influence the prevalence and intensity of disease in native species by acting as a source and increasing transmission of pathogens, or by acting as a sink and decreasing pathogen supply and transmission. It is possible for a species to act as a source or sink if it becomes infected, whether or not that species suffers significant mortality from the pathogens, and it is possible for a resistant species to reduce disease transmission among susceptible individuals. Analogs in the human health realm would be carriers of disease who show no symptoms but infect others, and the protection provided to the unvaccinated population by the large number of individuals who have been vaccinated. The workgroup rated experiments to determine the potential for *C. ariakensis* to increase disease prevalence and intensity in *C. virginica* and other bivalves to be a higher priority than experiments to determine the potential for *C. ariakensis* to reduce disease prevalence and intensity. Laboratory, mesocosm, and triploid outplant experiments may provide useful information on the effect of *C. ariakensis* on disease dynamics in *C. virginica* and other bivalves.

Research recommendations :

- Determine whether *C. ariakensis*, even if not impacted itself, is likely to serve as a reservoir and facilitate transmission of pathogens to other bivalve molluscs. For example, if *C. ariakensis* is infected with *Bonamia*, will infected hosts facilitate transmission to other molluscs that may be impacted? Pathogens noted in the disease section under Issue 1 are of particular concern, but there may be others. It will be important to consider key variables of disease transmission including host density, spatial distributions, disease intensity and effects of the physical environment. **Timeframe: 2 year laboratory effort after susceptibility is determined. Priority: High**

- Determine whether *C. ariakensis* has the potential to function as a pathogen sink and reduce transmission rates of known pathogens to native oysters or other organisms. Laboratory experiments and triploid outplants can provide insight into the mechanisms underlying this process as well as provide broad estimates of rates. **Timeframe: 2-3 years. Priority: Moderate.**

- Develop and parameterize a model of disease dynamics including *C. ariakensis* and potential native hosts and a range of spatial scales. Data from small-scale experiments and field studies should help parameterize the model. **Timeframe: >5 years** needed to obtain meaningful results if data from field studies are included. **Priority: Moderate**.

Reproductive potential

The potential for either oyster species to maintain and increase population abundance when growing in close proximity may depend on whether congeners directly interfere with each other's reproductive success. Gametes from different oyster species spawning simultaneously have the potential to start the initial stages of fertilization even though such fertilization cannot be completed successfully, or to complete fertilization but produce embryos that do not survive. *C. ariakensis* sperm are capable of fertilizing *C. virginica* eggs but result in nonviable embryos; hence there may be a reproductive 'sink' and reduced larval abundances where the two species co-occur. This lost reproductive potential could reduce the reproductive success of both the native oysters and introduced diploid species, hence exacerbating the decline of oyster populations baywide.

Research recommendation:

- Estimate the potential for interspecific gamete competition between *C. ariakensis* and *C. virginica* to influence the ability of either species to thrive in Chesapeake Bay. There was some disagreement among participating researchers as to the best approaches - which aspects of the problem can be realistically examined in the lab, which would need to be modeled, and whether additional field data would be required. Such differences of opinion highlight the importance of peer review of proposals. **Timeframe: 1-2 years. Priority: High.**

Ecological interactions between *C. ariakensis* and *C. virginica*

Achieving the simultaneous goals of persistence of *C. virginica* in Chesapeake Bay and establishment of a self-sustaining population of *C. ariakensis* may be difficult if *C. ariakensis* consistently outcompetes *C. virginica*, but may be enhanced if *C. ariakensis* increases settlement or survival of *C. virginica*. Competition for space is of key concern since it is widely recognized that appropriate space for settlement and growth is a limiting resource in Chesapeake Bay. While there is less agreement regarding whether food is a limiting resource in the Bay, recent studies between the introduced oyster, *Crassostrea gigas*, and the native oyster, *Saccostrea commercialis*, in Australia indicate that feeding interactions may be important in determining competitive outcomes.

Research recommendations :

- <u>Determine the potential extent, magnitude and outcome of competition for space and food between *C. ariakensis* and *C. virginica*.</u> Studies should involve a combination of laboratory experiments (using diploid and triploid organisms) and limited field deployments (using triploid organisms) whenever possible. Studies are needed with mixed size classes of the two species (e.g, small *C. ariakensis* and large *C. virginica*) in order to obtain as complete a picture as possible regarding the nature and magnitude of inter-specific interactions between the two species. Both direct interactions (e.g, overgrowth, undercutting) and indirect interactions (e.g., feeding interactions, reproductive risks, reproductive timing, disease transmission) should be addressed. One important issue is whether *C. ariakensis* will increase or decrease settlement substrate for *C. virginica*. Interaction studies should be conducted for a diversity of environmental conditions (these could be incorporated with multi-factorial environmental parameter experiments outlined in the Population Process phase above). **Timeframe**: **3-5 years** (Timeline is influenced by the need to collect data on several life stages and size groups of oysters). **Priority : High.**

- <u>Assess the timing of reproduction of *C. ariakensis* and *C. virginica* under a range of environmental conditions.</u> For many benthic species in which space is a limiting resource for population growth, timing of reproductive (e.g, who gets there first) is critical in determining competitive interactions. This information is also critical to assess the potential for interspecific fertilization to depress reproductive success when the two species grow in close proximity. **Timeframe: 2-3 years. Priority: High .**

- Determine whether the presence of *C. ariakensis* in the Bay is likely to influence predation pressure on the native oyster. The presence of *C. ariakensis* on predation rates on *C. virginica* may be affected by factors such as whether one species is more abundant than the other, and whether *C. ariakensis* exerts a different influence on predator population dynamics or distributions than does *C. virginica*. The susceptibility of each species to predators is discussed under Issue 1. However, additional modeling or experiments may help elucidate how this mechanism may increase or reduce risk to *C. virginica*. **Timeframe: 3 years. Priority: Moderate.**

- Incorporate data generated in these experiments into the oyster population dynamic models (discussed under Issue 1) in order to begin examining Bay-wide predictions of the interactions between the two species. For example: Are there specific habitats/regions within the Bay which may be of greater or lesser concern for negative effects of *C. ariakensis* on *C. virginica*? Can we identify habitat/strain combinations that would yield population growth and coexistence of both species? **Timeframe:** Model development will begin during data collection and could be completed **1-2 years after data are available**. **Priority: High.**

ISSUE 3. How will *C. ariakensis* impact Chesapeake Bay ecosystem services and functions?

General Considerations

A major goal of oyster restoration in Chesapeake Bay, whether the ultimate decision is to implement restoration using wild-strain Chesapeake *C. virginica*, selectively-bred strains of *C. virginica,* or *C. ariakensis*, is to restore the ecosystem services historically provided by *C. virginica*. The potential 'benefits of a rehabilitated oyster resource include: improving water quality by filtering phytoplankton, suspended solids and organic particles from the water' and 'providing important habitat for oysters, finfish, crabs and a diversity of other species; rehabilitating an oyster population…' (Appendix I, MD-DNR 2003). Thus, it is important to compare the ability of *C. ariakensis* to fulfill those ecosystem services relative to the benefits that would be derived from *C. virginica*.

Preliminary laboratory experiments suggest that the filtration rate of the two oyster species is similar (Newell, unpublished), and that differences in filtration capacity would therefore be determined by the density, size and distribution of oysters of each species. However, qualitative observations (Luckenbach, unpublished) suggest the potential for differences between *C. ariakensis* and *C. virginica* that could influence the ecosystem services they provide. *C. ariakensis* has been observed to produce a variety of growth forms in its native habitat – ranging from individuals growing in mud, to a 'rock oyster' morphology, to consolidated reef structures similar to those formed by *C. virginica*. The shell of *C. ariakensis* may also be thinner than that of *C. virginica*. Differences in growth forms, habitat and shell morphology could result in differences between *C. virginica* and C. *ariakensis* in their suitability as habitat, provision of pseudofeces as a food base for consumers, susceptibility to predators, and provision of other ecosystem services.

The workgroup that discussed these issues recognized that virtually all of the ecological impacts of *C. ariakensis* on Chesapeake Bay would be related to the potential for population growth of this species in various environments. Consequently, they emphasize the critical importance of collecting appropriate data and modeling population growth potential as discussed under Issue 1, above. Additionally, they identified several specific research needs related to community- and ecosystem-level processes.

Research recommendations:

- <u>Examine the growth form and reef building potential of *C. ariakensis* under a variety of environmental conditions, and the consequences of those growth forms to other organisms</u>. Observed variation in the growth form of *C. ariakensis* may be related to genetic differences among populations in Asia or to variation in environmental conditions. Studies are needed that help predict how *C. ariakensis* will grow in Chesapeake Bay and whether the growth form of *C. ariakensis* will provide the habitat value formerly provided by *C. virginica*. For instance, do *C. ariakensis* shells provide adequate nesting sites for reef resident fishes and the structure to facilitate high rates of secondary production utilized by economically and ecologically important fish species? **Timeframe: 3 – 5 years. Priority: High.**

- <u>Evaluate the effect of *C. ariakensis* on the abundance of potentially important associated predators and interactions with competitors</u>. For instance, if *C. ariakensis* is likely to be a reef-forming oyster in the Chesapeake Bay, will it support similar numbers of blue crabs and xanthid crabs as a *C. virginica* reef? What are its interactions with other space competitors (e.g., barnacles and sea squirts)? **Timeframe: 3 – 5 years. Priority: Medium.**

- <u>Examine the potential impacts of *C. ariakensis* feeding on trophic structure and biogeochemical cycling.</u> Determine feeding rates, feeding preferences, and biodeposition by *C. ariakensis* to evaluate whether or not significant differences exist between it and the native oyster. If significant differences are found, modeling efforts should be initiated and combined with other predictive models under development, to estimate its impacts on the ecosystem. **Timeframe: 2 – 3 years. Priority: Medium.**

<u>Issue 4.</u> Will *C. ariakensis* accumulate human pathogens or *E. coli* to a greater degree than *C. virginica?*

The workshop did not include participants with specific expertise in human health or shellfish sanitation. However, the question above was raised and workshop participants considered it of **Essential Priority** to evaluate. If *C. ariakensis* accumulates higher burdens of pathogens, or indicators of fecal contamination, than similar-sized *C. virginica*, increased fishery closures could be triggered and negatively impact the shellfish fishery in some areas within or outside Chesapeake Bay. Because of the lack of specific expertise within the assembled group, no timeline or suggested approach is provided in this report. Both the importance of this issue, and if deemed important, the research required to address it, should be evaluated by researchers with appropriate expertise.

<u>Duration of Research:</u>

The consensus of the workshop is that most of the important research questions can be addressed sufficiently within a ***5-year time frame***, although duration of individual research topics range from short- to long-term reflecting the inherent complexity of the problems to be studied. This estimate of the time to substantially complete required research is ***consistent with the recommendation of the NAS report***. However, the NAS report assumes that information generated from triploid aquaculture will be available to supplement targeted research and contribute to data requirements.

The 5-year time frame is based on the number of issues that need to be addressed, the sequential nature of some research required, and the importance of experiments that utilize multiple age classes of animals. Not all studies can be conducted simultaneously—some very important studies will depend upon data and insights generated from others. Therefore, while the duration of an individual project may be described as a 1-2 year effort, full implementation of the assessment may extend longer. Some studies will require use of large numbers of individuals that are several years old. These animals are not currently available. We cannot make a decision to proceed based solely on responses of juvenile and small oysters if the goal is to evaluate risks, and to predict the potential to establish self-reproducing populations. It is also important to note that the estimated time frame depends upon a number of key pre-requisites including adequate funding, critical 'infrastructure' elements, production of sufficient numbers of *C. ariakensis* to conduct tests, and the need for peer review of both proposals and results used for management decisions.

Infrastructure Needs:

It is crucial to recognize that there are significant limits in the existing infrastructure capacity to address many of the issues raised here in a ‘rapid response’ mode. Many of the studies require that one or more stocks or species of oysters be used or that multiple cohorts of oysters be included. Presently, there are two research hatcheries at VIMS (Gloucester Point and Wachapreague) with the capability to produce large quantities of *C. ariakensis* under strict quarantine conditions, with two other facilities (the Kauffman Aquaculture Center, VIMS and the Aquaculture and Restoration Ecology Laboratory at Horn Point, UMCES) due to become fully operational within the next 12 months. Further, there is a relatively restricted supply of spawning stock for any of the *C. ariakensis* groups. Synergism between co-occurring studies can exist, with oysters from a single spawn serving several experiments; however, at some point the capacity of the existing quarantine hatchery facilities to supply research animals (diploids and triploids) for multiple experiments becomes limiting. This is especially true for studies that require specific seasonal supply of larval or juvenile oysters. There can be significant lag times associated with obtaining the stocks of choice, conditioning the brood stock and producing larvae. There is also a particularly acute limitation in the lack of infrastructure to support the production and maintenance of Specific Pathogen Free (SPF) oysters that are required for conducting most disease challenge studies. Maximizing the capacity of existing quarantined hatchery facilities to support a variety of research projects simultaneously will require investment in operating expenses and personnel. Development of biosecurity protocols for hatcheries and experimental facilities is also required to minimize the likelihood of an accidental introduction of diploid *C. ariakensis*. In addition to hatcheries, there will need to be an investment in infrastructure required to perform contained experiments (e.g., mesocosms, etc.) in order conduct the number of studies that will be required in as short a period of time as possible, and to perform comparative studies at several sites within and outside Chesapeake Bay. Limits currently exist on the capacity to rear large oysters and conduct experiments in quarantine systems in the Bay region. Modest investments in modifications and retrofitting of water outflow systems to insure that no live gametes, larvae or novel pathogens are released into Chesapeake Bay would allow research to be conducted at several sites. This would facilitate studies to compare and contrast *C. ariakensis* growth across the broad range of conditions found in the region and would enhance the scope and speed the pace of research on *C. ariakensis*.

The importance of disease monitoring in field studies with *C. ariakensis* and *C. virginica* was widely recognized by the workshop participants; however, the current disease diagnostic capacity in the region is limited by space, technology, trained personnel and operating expenses.

Failure to address these infrastructure limitations will have the inevitable consequence of slowing the pace and limiting the extent to which the recommended research can be conducted.

Peer Review:

There was broad consensus that allocation of funds should be based on a strong peer review process. Peer-review is always good practice, but is especially important in this case given the long-term implications of an introduction of *C. ariakensis* to Chesapeake Bay and the substantial investment that will be required to conduct research needed to inform the decision-making process. Peer review will help ensure the quality of research plans and provide a mechanism for selecting projects with the greatest likelihood of success in addressing critical issues. In addition, review panels of expert scientists, following well-accepted scientific peer review and panel protocols, will not only uphold the credibility of the funding process, but can provide managers with insights on how best to coordinate multiple projects within the context of key infrastructure limitations described previously. The inclusion of researchers from outside the Chesapeake Bay region with no personal or institutional stake in the outcome of funding decisions in proposal reviews and panels is important. Reviews can be conducted in an expedited fashion—hence the time it takes to conduct reviews should not be viewed as a barrier. Participants also agreed that review should be ongoing throughout the decision-making process. As results become available to the management community, they should be evaluated before being utilized. An ongoing technical review mechanism, perhaps in the form of a technical panel of experts, should be an integral part of the EIS process. ***Ultimately, the credibility of decisions made will depend upon the quality of the science that underlies those decisions, and strong peer review will ensure that quality is achieved.***

Table 1. Summary of research recommendations. Although questions are listed separately, some could be efficiently addressed simultaneously by strategically designing experiments. In the case of the ‘ecosystem services’ topic, however, it is likely that more than one study would be required to adequately address the problem.

Research questions and topics	Priority	Same or similar research recommended in NAS report?	Issues addressed:		
			Ability of *C. ariakensis* to thrive in Chesapeake Bay	Risks and benefits to *C. virginica* and other bivalves	Risks and benefits to ecosystem functions and services
What is the susceptibility of *C. ariakensis* to *Bonamia* spp. pathogens that could be present in Chesapeake Bay and other estuaries? (experiments)	Essential	X	X		
What is the potential for vertical transmission of Herpes viruses? (experiments)	High	X	X		
What is the susceptibility of *C. ariakensis* to *Perkinsus* sp. pathogens other than *P. marinus*? (experiments)	Moderate	X	X		
How will the use of triploid *C. ariakensis* in experiments affect the accuracy of predictions about diploids? (experiments)	High		X		
What is the stability and reproductive potential of triploid *C. ariakensis*? (experiments)	Moderate	X	X		
Is there genetic variation among *C. ariakensis* stocks from Asia related to important physiological and ecological traits that will affect the risks and benefits associated with introducing it to the Chesapeake Bay? (sampling and experiments)	High	X	X		
What are the growth, survival and feeding responses of *C. ariakensis* versus *C. virginica* under a range of conditions in Chesapeake Bay? (experiments)	High	X	X		

Research questions and topics	Priority	Same or similar research recommended in NAS report?	Issues addressed:		
			Ability of *C. ariakensis* to thrive in Chesapeake Bay	Risks and benefits to *C. virginica* and other bivalves	Risks and benefits to ecosystem functions and services
What are the vital reproductive rates and processes of *C. ariakensis v. C. virginica* under East Coast environmental conditions? (experiments)	High	X	X		
Do physiological and behavioral characteristics of *C. ariakensis* and *C. virginica* larvae differ in ways that would affect larval survival and dispersal? (experiments and model)	High	X	X		
What are the settlement cues and substrate preferences of *C. ariakensis*? (experiments)	High	X	X		
Population models to predict abundance and spread of *C. ariakensis* in Chesapeake Bay	High	X	X		
Are their habitat/strain combinations that would yield population growth and coexistence of both *C. ariakensis* and *C. virginica*? (model)	High		X		
Is there a difference in mortality rates of juvenile *C. ariakensis* and *C. virginica* in responses to dissolved oxygen concentrations , sedimentation and predation? (experiments)	High	X	X		
Will *C. ariakensis* increase disease transmission and prevalence for other bivalves? (experiments)	High			X	
Will *C. ariakensis* reduce disease transmission or prevalence for other bivalves? (experiments)	Moderate			X	
Models of disease dynamics	Moderate			X	

Research questions and topics	Priority	Same or similar research recommended in NAS report?	Issues addressed:		
			Ability of *C. ariakensis* to thrive in Chesapeake Bay	Risks and benefits to *C. virginica* and other bivalves	Risks and benefits to ecosystem functions and services
What is the risk posed to *C. virginica* by cross-species fertilization? (experiments or models)	High	X		X	
What is the likely extent, magnitude and outcome of competition between *C. ariakensis* and *C. virginica*? (experiments; data generated should be added to models)	High	X		X	
What is the timing of reproduction of *C. ariakensis* relative to that of *C. virginica*? (experiments)	High	X	X	X	
What is the likely effect of *C. ariakensis* on predation on *C. virginica?* (models and/or experiments)	Moderate			X	
What will the growth form(s) of *C. ariakensis* be in Chesapeake Bay **and** what are the consequences of those growth forms to other organisms? Will the growth form of *C. ariakensis* provide the habitat value formerly provided by *C. virginica*? (experiments)	High				X
How will *C. ariakensis* affect the abundance of oyster predators and competitors? (experiments and models)	Moderate				X
Will *C. ariakensis* affect biogeochemical cycling or plankton composition differently than *C. virginica?* (experiments)	Moderate				X
Will *C. ariakensis* accumulate human pathogens or *E. coli* to a greater degree than *C. virginica?*	Essential				

Acknowledgements:

All workshop participants (listed in Appendix III) made valuable contributions to the deliberations and recommendations described in this report. We are especially grateful to the people who traveled from outside the Bay region to provide needed advice and perspective. In addition, the workshop chairs would like to extend special thanks to: Robert Whitlatch, Roger Newell, Dave Bushek and James Kirkley for chairing workgroups; Mike Fritz, Merrill Leffler, and Jack Greer for facilitating discussions; Laura Sorabella, Angela Padaletti, and Stephanie Bonniwell for taking notes; the discussion leaders, facilitators and note-takers who helped draft workgroup recommendations, and Melissa Bugg without whose help the meeting could not have been set up and been successful on short notice.

Funding for this workshop was provided by the Chesapeake Bay Program and by the NOAA Chesapeake Bay Office. We also gratefully acknowledge the efforts of Mike Fritz and Jamie King.

Literature cited

NRC. 2004. Nonnative Oysters in the Chesapeake Bay. Committee on Nonnative Oysters in the Chesapeake Bay, National Research Council. 344 pages.

Appendix I
DRAFT: December 2003

MD/VA STRAWMAN PROPOSAL AND ALTERNATIVES FOR PROGRAMMATIC EIS

PROPOSAL

The State of Maryland and Commonwealth of Virginia propose to introduce the oyster species, *Crassostrea ariakensis*, into the tidal waters of Maryland and Virginia, beginning in 2005, for the purpose of re-establishing a naturalized, reproducing, and self-sustaining population of oysters. Diploid *C. ariakensis* will be propagated from the 3rd or later generation of the naturalized Oregon stock of this species, in accordance with the International Council for the Exploration of the Sea's (ICES) 1994 Code of Practices on the Introductions and Transfers of Marine Organisms. Deployment will generally occur first on State designated sanctuaries, where harvesting will be prohibited permanently, and on harvest reserve and special management areas, second, where only selective harvesting will be allowed, and last, on natural oyster bars, where traditional harvesting and aquaculture are allowed.

The States further propose to continue native oyster (*C. virginica)* restoration efforts in those areas of the Chesapeake Bay where conditions are most favorable for its survival by using the best available restoration strategies and stock assessment techniques, including the maintenance of the existing network of sanctuaries and harvest reserves, and supplementing natural recruitment of this species with hatchery produced spat on shell.

The objective of this proposal is to establish a self-sustaining oyster population that reaches a level of abundance in Chesapeake Bay comparable to stock sizes during the period 1920–1970. The benefits of a rehabilitated oyster resource include: improving water quality by filtering phytoplankton, suspended solids and organic particles from the water; providing important habitat for oysters, finfish, crabs and a diversity of other species; rehabilitating an oyster population capable of supporting an economically viable oyster industry; and preserving the Chesapeake Bay's communities and culture associated with working watermen.

PURPOSE AND NEED

Oysters are a keystone species in the Bay ecosystem. Native oyster populations have declined to less than one percent of their historic levels and do not show any signs of uniform and long-lasting resistance to the dominant parasites MSX and Dermo that are the primary causes for the decline. A need exists to restore the ecological role of oysters in the Bay through either native oyster restoration, or, in the absence of self-sustaining native oyster populations capable of overcoming the parasites and increasing oyster populations, a similar species that would restore the ecological role of a keystone species. This would be accomplished if it is determined that introduction of the 'Oregon' strain of

C. ariakensis would not harm the ecology of the Bay, would not introduce new diseases or parasites, would not interfere with restoration of native oysters, and would become self-sustaining and self-reproducing.

POSSIBLE ALTERNATIVES TO THE PROPOSED ACTION

Alternative 1 – No Action Alternative 1– Continue Present Course of Action: Continue Maryland's present Oyster Restoration and Repletion Programs, and Virginia's Oyster Restoration Program under current program and resource management policies and available funding using the best available restoration strategies and stock assessment techniques.

Alternative 2 – Expand Native Oyster Restoration Program: Expand and accelerate Maryland's Oyster Restoration and Repletion Programs, and Virginia's Oyster Restoration Program, including, but not limited to the development, production and deployment of disease resistant strain(s) of *C. virginica* (Eastern Oyster).

Alternative 3 – Harvest Moratorium: Implement a temporary harvest moratorium and oyster industry compensation (buy-out) program in Maryland and Virginia.

Alternative 4 Aquaculture: Establish and/or expand State-assisted, managed or regulated aquaculture operations in Maryland and Virginia using the native oyster species or suitable triploid non-native disease resistant oyster species.

Alternative5: Introduce and Propagate an Alternative Oyster Species (Other than *C. ariakensis*) or an Alternative Strain of *C. ariakensis*: Introduce and propagate in the State-sponsored, managed or regulated oyster restoration programs in Maryland and Virginia, a disease resistant oyster species other than *C. ariakensis*, or an alternative strain of *C. ariakensis,* in accordance with the International Council for the Exploration of the Sea's (ICES) 1994 Code of Practices on the Introductions and Transfers of Marine Organisms.

Alternative 6 – Combination of Alternatives.

Appendix II

Identifying and prioritizing research required to evaluate ecological risks, benefits and alternatives related to the potential introduction of *Crassostrea ariakensis* to Chesapeake Bay

What are the most important questions?
What data are needed to answer those questions?
What are the best approaches for gathering data, analyzing new and existing data, and modeling to get the information needed?
How long will it take, and how much will it cost?

Agenda

December 2, 2003: Day 1
Rhode Room

8:00	Registration and Coffee, Pastries and Fruit
8:30	Background and Workshop Goals Denise Breitburg, The Academy of Natural Sciences/Smithsonian Environmental Research Center
9:00	What We Do and Do Not Know About *Crassostrea ariakensis* Mark Luckenbach, Virginia Institute of Marine Sciences
9:30	Risk Analysis: Synthesizing Data to Predict Risks and Benefits of Alternatives Steve Bartell, Cadmus
10:00	Break (15 min)
10:15	Geographic Spread of Non-native Species Greg Ruiz, Smithsonian Environmental Research Center
10:45	Ecology and Invasive Species Robert Whitlatch, University of Connecticut and member of the NAS panel
11:15	Disease Ecology and Introduced Species (note that the original speaker was unable to attend and Greg Ruiz addressed this topic)

11:45	Economic Considerations James Kirkley, Virginia Institute of Marine Science
12:15	Lunch (provided)
1:15	Benefits and Problems of Oyster Introductions Elsewhere Roger Mann, Virginia Institute of Marine Science
1:45	NAS Research Recommendations Robert Whitlatch, University of Connecticut and member of the NAS panel
2:00	Key Points from the Morning Presentations and Charge to Workgroups Jonathan Kramer, University of Maryland Sea Grant Program
2:15 – 4:15	Workgroup Session 1: Brainstorming session – Identify questions and data needs. (See attached handout for workgroup locations)
4:15-5:00	Workgroup reports to group: What questions have been identified?
5:00	Adjourn (Dinner on your own)

December 3, 2003: Day 2
Rhode Room

8:00	Coffee, pastries and fruit
8:30 – 12:00	Workgroup Session 2: Prioritize and flesh out questions and data needs; recommend approaches,required funding and time requirements.

(See attached handout for workgroup locations)

12:00-1:00	Lunch (provided)
1:00- 2:00	Workgroup reports

2:00 – 3:30 Plenary Discussion: Workshop recommendations for a prioritized research plan to evaluate risks and benefits of alternative methods (including the introduction of C. ariakensis) for restoring oysters to Chesapeake Bay.

3:30 Adjourn

Workshop Breakout Groups

December 2-3, 2003

Group	Group Color Code	Location
Disease Issues and Other Stressors	Red	Selby Room
Population and Community Ecology/Biogeography and Coast Wide Spread	Blue	Chester Room
Physiology, Reproductive Biology and Ecological Genetics	Green	Glebe Room
Economics and Risk Assessment	Yellow	Rhode Room

APPENDIX III
WORKSHOP PARTICIPANTS

Identifying and Prioritizing Research Required to Evaluate Ecological Risks, Benefits, and Alternatives Related to the Potential Introduction of *Crassostrea ariakensis* to Chesapeake Bay

December 2-3, 3003
Annapolis, MD

Workshop Participants

George Abbe	Academy of Natural Sciences
Stan Allen	Virginia Institute of Marine Science
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Melanie Bishop	University of North Carolina
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Loren Coen	Marine Resources Research Institute
Melanie Davenport	Chesapeake Bay Commission
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Matt Hare	University of Maryland
Pauli Hayes	Virginia Sea Grant
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Jamie King	National Oceanic and Atmospheric Administration
Jim Kirkley	Virginia Institute of Marine Science
Jonathan Kramer	Maryland Sea Grant
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