ASC Bivalve Standard

Version 1.1

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Table of Contents

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Version control

Document version history:

Version:	Release date:	Effective date:	Remarks/changes:
v1.1	March 7th 2019	March 15th 2019	Update of the standard to meet ASC style requirements (e.g. Inclusion of structure of the standards, formatting and wording). Align the scope, 'about the ASC' and 'overview of the ASC system'. The content of the actual Standard, as defined by criteria / indicators / requirements under Principles [1-7], remains unchanged from version 1.0.
v1.0	January 2012	January 2012	Update of the Standard to meet ASC style requirements (e.g. inclusion of introduction chapters 'about the ASC' and 'overview of the ASC system', formatting and wording). The content of the actual Standard remained unchanged from version 0.1.
v0.1	May 2011	May 2011	Handover of the Standard by the Bivalve Aquaculture Dialogue Steering Committee to the Aquaculture Stewardship Council
v0.1	August 2010	August 2010	Original version developed and approved by the bivalve Aquaculture Dialogue Steering Committee under the original title "Bivalve Aquaculture dialogue Standards"

It is the responsibility of the user of the document to use the latest version as published on the ASC-website.

Available language(s)

This Bivalve Standard is available in the following language(s):

Version:	Available languages
v1.1 v1.0	English (official language)
v1.0	Japanese
v1.0	Spanish

In case of any inconsistencies and/or discrepancies between available translation(s) and the English version, the online English version (pdf-format) will prevail.

Copyright notice

ABOUT THE AQUACULTURE STEWARDSHIP COUNCIL (ASC)

The Aquaculture Stewardship Council (ASC) is an independent, not-for-profit organisation that operates a voluntary, independent third-party certification and labelling programme based on a scientifically robust set of standards.

The ASC standards define criteria designed to help transform the aquaculture[1] sector[2] towards environmental sustainability and social responsibility, as per the ASC Mission.

ASC Vision

A world where aquaculture plays a major role in supplying food and social benefits for mankind whilst minimising negative impacts on the environment.

ASC Mission

To transform aquaculture towards environmental sustainability and social responsibility using efficient market mechanisms that create value across the chain.

ASC Theory of Change

A Theory of Change (ToC) is an articulation, description and mapping out of the building blocks required to achieve the organisation's vision**.**

ASC has defined a ToC which explains how the ASC certification and labelling programme promotes and rewards responsible fish farming practices through incentivising the choices people make when buying seafood.

ASC's Theory of Change can be found on the ASC website.

[1] Aquaculture: Aquaculture is the farming of aquatic organisms, including fish, molluscs, crustaceans and aquatic plants. Farming implies some form of intervention in the rearing process to enhance production, such as regular stocking, feeding, protection from predators, etc. Farming also implies individual or corporate ownership of the stock being cultivated (FAO).

[2] Aquaculture sector: Represents a group of industries (e.g.: feed industry, farming industry, processing industry, etc.) and markets that share common attributes (i.e. aquaculture products).

THE ASC DOCUMENT AND CERTIFICATION SYSTEM

ASC is a full member of the ISEAL Alliance and implements a voluntary, independent third-party certification system[3] consisting of three independent actors:

I.	Scheme Owner	i.e. Aquaculture Stewardship Council
II.	Accreditation Body	i.e. Assurance Services International (ASI)
III.	Conformity Assessment Body (CAB)	i.e. accredited CAB's

Scheme Owner

ASC, as scheme owner:

- sets and maintains standards according to the ASC Standard Setting Protocol which is in compliance with the "ISEAL Code of Good Practice - Setting Social and Environmental Standards". The ASC standards are normative documents;

- sets and maintains Implementation Guidance which provides guidance to the Unit of certification (UoC) on how to interpret and best implement the indicators within the Standard;

- sets and maintains the Auditor Guidance which gives guidance to the auditor how to best assess a UoC against the indicators within the Standard;

- sets and maintains the Certification and Accreditation Requirements (CAR) which adheres at a minimum to the "ISEAL Code of Good Practice - Assuring compliance with Social and Environmental Standards". The CAR describes the accreditation requirements, assessment requirements and certification requirements. The CAR is a normative document.

These above listed documents are publicly available on the ASC-website.

Accreditation Body

Accreditation is the assurance process of assessing the Conformity Assessment Body (CAB) against accreditation requirements and is carried out by an Accreditation Body (AB). The appointed AB of ASC is Assurance Services International (ASI, "Accreditation Services International" prior to January 2019) which uses the CAR as normative document for the accreditation process.

Assessment findings of ASI-accreditation audits and an overview of current accredited CABs is publicly available via the ASI-website (http://www.accreditation-services.com).

[3] Third-party Certification System: Conformity assessment activity that is performed by a person or body that is independent of the person or organisation that provides the object, and of the user interests in that object (ISO 17000)

Conformity Assessment Body

The UoC contracts the CAB which employs auditor(s) that conduct a conformity assessment (hereafter 'audit') of the UoC against the relevant standard. The management requirements for CABs as well as auditor competency requirements are described in the CAR and assured through ASI accreditation.

ASC Audit and Certification Process

The UoC is audited at Indicator-level.

An ASC audit follows strict process requirements. These requirements are detailed in the CAR. Only ASI-accredited CABs are allowed to audit and certify a UoC against ASC standards. As scheme owner, ASC itself is not - and cannot be - involved in the actual audit and/or certification decision of a UoC. Granted certificates are the property of the CAB. ASC does not manage certificate validity.

Audit findings of all ASC audits, including granted certificates, are made publicly available on the ASC-website. These include the audit findings that result in a negative certification decision.

Note: in addition to the Standard's, there are certification requirements that apply to UoCs seeking certification; these requirements are detailed in the CAR.

ASC Logo use

ASC-certified entities shall only sell their product carrying the ASC Logo if a Logo Licence Agreement (LLA) has been signed. On behalf of the ASC, the Marine Stewardship Council (MSC) Licensing Team will issue logo license agreements and approve logo use on products. For more information see: ASC Logo.

Unauthorised logo display is prohibited and will be treated as a trademark infringement.

STRUCTURE OF ASC STANDARDS

A Standard is "*a document that provides, for common and repeated use, rules, guidelines or characteristics for products or related processes and production methods, with which compliance is not mandatory*".

ASC Standards are as follows designed:

- ASC Standards consist of multiple Principles – a Principle is a set of thematically related Criteria which contribute to the broader outcome defined in the Principle title;

- Each Principle consists of multiple Criteria – each Criterion defines an outcome that contributes to achieving the outcome of the Principle;

- Each Criterion consists of one or several Indicators – each Indicator defines an auditable state that contributes to achieving the Criterion outcome.

Both Principles and Criteria include Rationale statements providing a set of reasons (backed by reference notes if needed) as to why the Principle or Criterion is needed.

SCOPE AND UNIT OF CERTIFICATION

Linked to the ASC Vision, the Scope of the ASC Bivalve Standard (hereafter "the Standard") addresses the key negative environmental and social impacts associated with the Bivalve aquaculture industry. An ASC-certified farm contributes to the ASC Vision by reducing, mitigating or eliminating these negative impacts.

The Scope of the Standard is translated into seven Principles that apply to every UoC:

- Principle 1 - Obey the law and comply with all applicable legal requirements and regulations where farming operation is located.
- Principle 2 - Avoid, remedy or mitigate significant adverse effects on habitats biodiversity, and ecological processes.
- Principle 3 - Avoid adverse effects on the health and genetic diversity of wild populations.
- Principle 4 - Manage disease and pests in an environmentally responsible manner.
- Principle 5 - Use resources efficiently.
- Principle 6 - Be a good neighbor and conscientious coastal citizen.
- Principle 7 - Develop and operate farms in a socially and culturally responsible manner.

The Criteria within the Principles apply to every UoC

Unit of Certification (UoC)

The applicable UoC is determined by the CAB/ auditor and adheres to the Standard's Criteria UoC-requirements as outlined in the CAR.

Biological and geographic scope to which the Standard applies

The ASC Bivalve Standard applies globally to all locations and scales of filter-feeding bivalve aquaculture production systems. Bivalve aquaculture is defined as active husbandry of bivalve shellfish from seed to harvest within a defined area and with defined ownership of the shellfish being cultured.

How to read this document?

In the following pages, tables with indicators and their corresponding requirements are included. Within each criterion, requirements tables are followed by a rationale section that provides a brief overview of why the issues are important and how the proposed requirements address them.

Definitions are provided in footnotes.

The ASC Bivalve Standard will be supplemented by an auditor guidance document detailing the methodologies used to determine if the ASC Bivalve Standard is being met, as well as guidance for producers to achieve compliance to the ASC Bivalve Standard.

Metric Performance Levels

Several Indicators in the Standard require a Metric Performance Level (MPL). The applicable MPL is directly listed after the Indicator ("Requirement" section).

1. PRINCIPLE: OBEY THE LAW AND COMPLY WITH ALL APPLICABLE LEGAL REQUIREMENTS AND REGULATIONS WHERE FARMING OPERATION IS LOCATED

Issue: Principle 1 is intended to ensure that all farms aiming to be certified against the ASC Bivalve Standard meet their legal obligations as a baseline requirement. Adhering to the law will ensure that producers meet the most basic environmental and social requirements and will serve as a platform on which the effectiveness of the requirements will be based.

1.1 Criterion: All applicable legal requirements and regulations where farming operation is located

INDICATOR	REQUIREMENT
1.1.1 Evidence of compliance with all applicable legal requirements and regulations where the farming operation is located (e.g., permits, licenses, evidence of lease, concessions and rights to land and/or water use)	Yes

Rationale—Bivalve aquaculture operations must, at a minimum, adhere to national and local laws. The ASC Bivalve Standard may develop sustainability requirements beyond those required by law, but the baseline requirement for any aquaculture operation must be compliance with the legal obligations of the producing country. Laws that compel a farmer to take a certain action take precedent over voluntary requirements (e.g., mandatory control of an invasive species using methods otherwise not allowed under the requirements).

2. PRINCIPLE: AVOID, REMEDY OR MITIGATE SIGNIFICANT ADVERSE EFFECTS ON HABITATS, BIODIVERSITY, AND ECOLOGICAL PROCESSES

Issue: One of the main areas of potential environmental concern associated with bivalve aquaculture is intensity of production and its effect on the ecological communities that are in close proximity to farming operations. Since shellfish are farmed in dynamic coastal environments, the ecosystem effects of farming are difficult to measure in a way that can be applied consistently from farm to farm. To overcome this challenge, the Dialogue developed a tiered approach based on initial risk assessments followed by increasing levels of monitoring dependent on localized site-specific conditions. In addition, it was agreed that, in order to verify environmental sustainability, the requirements must also address the cumulative impact of multiple farms in a given area.

2.1 Criterion: Benthic effects for off-bottom and suspended-culture methods[4]

INDICATOR	REQUIREMENT
2.1.1 Acceptable levels of total 'free' sulfide in surficial sediment (0-2 centimeters from the surface) measured beneath the farm in comparison to control sites[5]	≤ 1500 µM, monitoring every five years is required ≥1500 µM and ≥ 3000 µM, monitoring every year is required
2.1.2 Unacceptable levels of total 'free' sulfide in surficial sediment measured beneath the farm in comparison to control sites	≥ 3000 µM
2.1.3 In cases where natural background sulfide levels exceed 3000 µM, the annual S concentrations should not significantly[6] exceed levels measured at reference sites located outside the farm[7]	Yes

[4] Farms utilizing in- and on-bottom husbandry practices are exempted from assessment for benthic organic enrichment. These standards specifically target off-bottom and suspended-culture activities that permit greater stocking biomass per area than can be achieved using bottom culture approaches. See Appendix I for additional rationale

[5] Sampling design and sulfide methodology are included with the standards as separate documents

[6] Statistical significance (i.e., 95% confidence interval)

[7] Farming activity is permitted in areas where the natural benthic environment is already heavily enriched with organic matter prior to the initiation of any shellfish aquaculture activities

2.1.4 Sulfide analysis may be replaced by direct analysis of benthic community structure (i.e., infaunal surveys) in areas where this biotic approach is preferred by the applicant or is already mandated by a regulatory body[8]	**Yes**
2.1.5 Allowance for bivalve aquaculture over areas that provide a particularly significant or essential biological or ecological function within the broader ecosystem[9]	None

Rationale—Bivalve aquaculture often results in increased organic deposition underneath and adjacent to farms. The accumulation and mineralization of this excess organic matter in sediments can cause stress on benthic organisms through oxygen depletion and the toxic effects of hydrogen sulfide (H2S). The impacts on benthic communities due to increased organic matter sedimentation, oxygen deficiency (hypoxia and anoxia) and toxic effects of H2S are well known (e.g., Pearson and Rosenberg 1978, Hargrave et al., 2008b) and can include changes in the size and structure of benthic infaunal communities. Various organic enrichment indicators and impact classification systems have been proposed in the scientific literature. Biotic indices for assessing benthic habitat environmental quality range from simple indicators of species richness to more complex statistical approaches. These classical methods of macrofauna analysis directly address our objective of assessing potential impacts on seabed biological communities. However, taxonomic descriptions and determinations of numerical abundance and biomass requires highly trained personnel working over extended periods and the associated costs are prohibitive for routine site assessments and monitoring purposes.

Total 'free' sulphide (S2-) in surficial (0-2 cm) sediments is a cost-effective indicator of the organic enrichment effects of shellfish aquaculture on benthic communities. In general, there is consistency between changes in various biological and geochemical variables and total S2- in surface sediments along organic enrichment gradients (see Hargrave et al., 2008a). Other metrics, such as redox potential, sediment oxygen demand, sediment organic content and benthic diversity indices were also considered. They were rejected because of measurement challenges, costs and/or inherent variation. More information on the rationale behind the total "free" sulfide measurement can be found in Appendix I.

In addition to measuring levels of total 'free' sulfide, bottom video/imaging is also a relatively cost-effective way to quickly determine whether or not sediments underneath a farm have already become hypoxic or if the benthic conditions underneath or adjacent to a farm may be especially sensitive to increased organic loading from biodeposition. If bottom video/imaging reveals non-depositional substrate and the absence of sensitive benthic habitat, there is a lower risk of adverse benthic effects from bivalve aquaculture operations.

[8] Biotic indicator decision thresholds need to be assessed to ensure equivalency with the thresholds identified for total 'free' sulfide given in requirement 2.1.1. There are several papers that have been published linking specific benthic sulfide levels to indices for benthic biodiversity. Please refer to the reference section for examples (e.g., Hargrave et. al. 2008)

[9] Areas containing biogenic structures that are not particularly adapted to sedimentation or organic enrichment (e.g., tubeworm mounds, bryozoans mounds, bivalve beds and reefs or sponge gardens that form a structure for other epifauna)

2.2 Criterion: Pelagic effects

INDICATOR	REQUIREMENT
2.2.1 The ratio of clearance time[10] (CT) over retention time[11] (RT) (If the area of all of the farms within a water body as defined in Appendix I, inclusive of the certification unit, is less than 10% of the total area of the water body, then requirements 2.2.1 and 2.2.2 need not apply)	>1
2.2.2 Where clearance time is less than retention time, the ratio of clearance time over primary production time[12]	>3
2.2.3 Equivalency with requirements 2.2.1 or 2.2.2 may be demonstrated, if a farm or group of farms is able to prove, through more comprehensive carrying capacity modeling that, in aggregate, they do not exceed the ecological carrying capacity of the applicable water body in which they are located	Yes

Rationale—There is potential for bivalve farming operations to exceed the ecological carrying capacity of the body of water in which they are located. Ecological carrying capacity has been defined as the stocking or farm density above which unacceptable ecological impacts begin to manifest (Inglis et al. 2000). This happens when the removal of phytoplankton by all bivalve farms in a water body, including the applicant site, outstrips the capacity of the ecosystem to replenish the supply, resulting in adverse conditions for wild and cultured populations. The ASC Bivalve Standard addresses this issue using relatively simple calculations that compare how long it takes a population of bivalves to clear a body of water (clearance time–CT) with how long it takes for tides to flush that body of water (retention time–RT). Please refer to Appendix I for the rationale and specific formulas for the carrying capacity measurement, including a protocol for defining applicable water body boundaries. When carrying capacity is exceeded, farmed areas should have or be part of a bay-scale management plan for addressing potential cumulative pelagic effects from multiple farms.

[10] Clearance time is the number of days required for the dominant bivalve stock(s) (wild and cultured) to clear the volume of the bay or regional water body (i.e., sites with no clear boundaries). The dominant species census should be based on the peak standing stock during the year. The calculation is based on published clearance rate data for the bivalve group (mussels, scallops, clams and oysters)

[11] Retention time is the number of days for tides to flush a volume of water equal to the volume of the bay or water body

[12] PPT is the number of days required for the replacement of the standing stock of phytoplankton in the bay (i.e., time-scale of phytoplankton population growth). PPT is the ratio of yearly averages of phytoplankton biomass (B) to phytoplankton primary production (PPP) within the system. B can be estimated from chlorophyll a measurements, published data or satellite predictions assuming a carbon to chlorophyll ratio of 50. PPP can be obtained from published results or model predictions.

2.3 Criterion: Critical habitat and species interactions

INDICATOR	REQUIREMENT
2.3.1 Allowance for harm to threatened/endangered species[13] or the habitat on which they depend	None

Rationale—Some bivalve shellfish farms are situated in areas with critical habitat essential for endangered species survival. In order to preserve local biodiversity, it is important that the ASC Bivalve Standard takes into account potential risks that bivalve aquaculture poses to critical habitats and species. For this reason, in the proposed requirements, farming operations will not be permitted to adversely affect endangered species or the habitat on which they depend. This is particularly applicable to shellfish operations that employ dredging as a means to harvest crops that are ready for market. Although we have not excluded bottom culture from potential certification, dredging will not be allowed if there is a significant risk to endangered species or the habitat on which they depend.

The Dialogue acknowledges that harvest methods, such as dredging (either with a "dry" dredge or with hydraulic jets that loosen the soil) or raking with hand rakes, will disturb the benthos and cause some degree of mortality to non-target organisms, such as worms and crabs. However, when a grower uses a dredge on their lease, they know exactly where to go and will harvest planted shellfish in an efficient and systematic fashion.

Most shellfish farming takes place in shallow coastal water with a sandy or silty bottom. The species that live in these waters are well-adapted to periodic disturbances from storms and wave action. (DeAlteris et al. 1999) Species in these environments tend to be opportunists that rapidly re-colonize disturbed bottom and are tolerant of high loads of suspended sediment. (Coen, 1995) Studies have shown that these environments will recover from dredge harvesting in a few weeks or months. Perhaps most significantly, shellfish farmers replant seed (and often replace shell) following harvest. They will allow that seed to grow undisturbed for many months (in some cases, up to six years), replacing and improving the firm substrate that provides important habitat for many species. It has been observed that cultured bottom is typically far more diverse and productive than nearby areas devoid of shellfish cultivation or areas that are regularly dredged by wild harvest fishermen. (DeAlteris et al. 2004)

2.4 Criterion: Environmental awareness

INDICATOR	REQUIREMENT
2.4.1 Evidence of environmental training, compliance to regional codes of practices or implementation of environmental management plans	Required

Rationale—The final measure to ensure that farming operations are not adversely affecting the ecological integrity of the area in which they are located is to make certain that farmers have the appropriate level of environmental awareness. This can be done by requiring farmers to have evidence of environmental training/education or to be in compliance with a set of environmental codes of practices and/or management plans.

[13] As defined by national law or as found in the International Union for Conservation of Nature Red List of Threatened Species.

3. PRINCIPLE: AVOID ADVERSE EFFECTS ON THE HEALTH AND GENETIC DIVERSITY OF WILD POPULATIONS

Issue: Bivalve aquaculture may pose risks to wild populations through introduced cultivated species and exotic pests and pathogens. When species are introduced into an area without a proper assessment of potential risks, they may cause increased predation and competition, disease, habitat destruction, genetic stock alterations and in some cases, extinction. Farms using hatchery seed to cultivate native species have the potential to affect the genetic diversity of proximate natural populations.

3.1 Criterion: Introduced pests and pathogens

INDICATOR	REQUIREMENT
3.1.1 Allowance for the illegal introduction of a non-native species, pest or pathogen attributable to the farm within 10 years prior to assessment	None
3.1.2 Documentation of compliance with established protocol or evidence of following appropriate best management practices for preventing and managing disease and pest introductions with seed and/or farm equipment	Required

Rationale—A leading cause of biodiversity loss in aquatic ecosystems is the introduction of exotic species. Historically, managers of shellfish resources frequently employed introductions of non-native species to counteract or reverse the impacts of overfishing and habitat degradation. These actions have caused profound changes in some coastal marine ecosystems. The ecological and genetic risks of shellfish introductions are well-characterized but so poorly quantified as to make generalizations or predictions of impacts impossible. (NRC 2004) For example, the Pacific oyster Crassostrea gigas has been introduced from its native home in Japan to all continents except Antarctica. (Mann 1979) Its ecological impacts range from not thus far detectable to displacement of native species. The present day risk from introductions associated with bivalve aquaculture may be overstated (Naylor et al. 2001), as no new, non-native bivalve species has been introduced for aquaculture purposes for several decades. Introductions by mechanisms other than bivalve aquaculture (e.g., via ballast water and the pet and live seafood trades) pose larger threats to marine biodiversity.

3.2 Criterion: Sustainable wild seed procurement

INDICATOR	REQUIREMENT
3.2.1 Excluding larval collection, evidence that purchased or collected wild seed is not harvested from an open-access, unregulated source	Required

Rationale—Translocations of native species among different geographic areas can pose risks to the genetic diversity of wild populations. This issue has been debated with respect to escapes from salmon net pen culture. However, salmon populations, unlike shellfish populations, are highly structured by homing and adaptations to natal freshwater spawning grounds. Marine shellfish, on the other hand, have widely dispersing planktonic larvae and typically show minimal genetic divergence over broad spatial scales. (Hedgecock et al. 2007a)

The issue of translocation probably arises most often in shellfish aquaculture with respect to sourcing of wild seed to stock farms. An environmental requirement for shellfish aquaculture operations that rely upon translocations of wild seed necessitates an assessment of the potential risk for overfishing the reproductive sustainability of the wild source stock. Therefore, if growers are transporting seed or spat collected from other regions or harvesting excessive amounts of seed locally, an assessment is necessary to determine whether or not the manner in which the wild seed is collected for grow-out adversely affects recruitment or demography of local bivalve populations. For this reason, farms that use wild seed from open-access, unregulated sources will be ineligible for certification.

3.3 Criterion: Introduced non-native cultivated species

INDICATOR	REQUIREMENT
3.3.1 Evidence of responsible[14] introduction of non-native cultivated species	Required

Rationale—Most growing areas already have stringent requirements regarding the introduction of exotic animals and plants into the environment, yet regulations and enforcement may be insufficient to prevent intentional or accidental introductions. Where introduction of a non-native bivalve species is allowed by law (e.g., a species identified on a “clean list” of non-harmful species), the best practice for reducing ancillary introductions is to follow the International Council for the Exploration of the Sea’s (ICES 2005) “Code of Practice.”

3.4 Criterion: Native species cultivation

INDICATOR	REQUIREMENT
3.4.1 For hatchery produced seed, documentation of efforts made to address genetic concerns specific to species and geographic region where the seed will be out-planted (See Appendix II for guidance)	Required

Rationale—Since a substantial and growing fraction of global shellfish aquaculture depends on hatchery-propagated seed, it is necessary to understand and ameliorate the potential risks. In addition to potentially diluting the genetic diversity of proximate wild populations, hatchery-based shellfish aquaculture may also affect the fitness or adaptedness of natural populations. One part of this risk,

[14] At a minimum, farms must have a permit(s) substantiating compliance with ICES guidelines for introduction of exotic species and certification to ICES requirements regarding parasites and pathogens.

from mixing of genetically divergent populations, is the same as that faced in translocations and, as discussed above, appears minimal for bivalve mollusks, owing to high gene flow among natural populations. The other part of this risk is the genetic change inevitably brought about by intentional or unintentional artificial selection ("domestication" selection) in the hatchery environment. For example, fine-mesh screens are used universally in shellfish hatcheries to cull small individuals from larval cultures. This practice may select for rapid larval development. If this trait were negatively correlated with post-settlement survival and growth and if, through widespread farming of hatchery seed, this selected hatchery stock were to swamp a local population, then the reproductive success of the wild population could, in principal, be reduced. Many traits could be subject to such domestication selection. Unfortunately, there are no data on the genetic impacts of hatchery practices; indeed, designing an experiment to measure genotype-by-environment interaction for larval traits across both hatchery and natural habitats would be challenging. Nevertheless, risks from hatchery enhancements on genetic diversity or adaptation are manageable with appropriate designs and monitoring. (Hedgecock and Coykendall 2007)

Effective size of hatchery stocks must be kept large to avoid inbreeding and random genetic changes. Other best practices that diminish the risk of genetic impacts of hatchery-based shellfish aquaculture are to use local broodstock, rotate broodstock within spawning seasons and between years, and avoid returning hatchery-propagated stock to the hatchery as broodstock. These practices reduce the possibility for cumulative genetic change, owing to domestication selection. However, practices designed to minimize differences between cultured and wild stocks also prevent domestication and genetic improvement of farmed stocks, which, in the long run, could lead to desirable increases in efficiency of aquaculture production.

One way to eliminate the risk of interaction between wild and hatchery stocks (thus permitting domestication and genetic improvement of bivalve molluscs to proceed) is to render farmed stocks sterile. Triploidy is commonly induced in shellfish to reduce reproductive effort, divert energy to growth and improve meat quality during the normal spawning season. (Allen and Downing 1986; Nell 2002) Because triploids are effectively sterile, their use in shellfish aquaculture dramatically reduces gene flow between farmed and wild native or naturalized stocks. Triploidy does not, however, afford long-term protection against the introduction of a non-native farmed species. (NRC 2004) Triploid seed is currently produced by fertilizing diploid eggs with sperm from tetraploid males. (Guo et al. 1996; NRC 2004) Bio-security of reproductively competent tetraploid stocks in the environment is an issue that is just beginning to be addressed. (Piferrer et al. 2009) Early experience with tetraploid Pacific oysters suggests that they are not robust enough, at present, to out-compete diploid stocks.

3.5 Criterion: Transgenic animals

INDICATOR		REQUIREMENT
3.5.1	Allowance for farming of transgenic[15] animals	None

Rationale—The farming of transgenic animals, in general, creates additional issues regarding genetic impacts on wild populations. For this reason, transgenic animals are not allowed under this requirement.

[15] Introduced genes from other species

4. PRINCIPLE: MANAGE DISEASE AND PESTS IN AN ENVIRONMENTALLY RESPONSIBLE MANNER

Issue: Management of diseases is a key issue in any form of intensive farming. The ASC Bivalve Standard strives for disease and pest management practices that have the lowest impact possible on the surrounding ecosystem.

4.1 Criterion: Disease and pest management practices

INDICATOR	REQUIREMENT
4.1.1 Allowance for the application of mutagenic, carcinogenic or teratogenic pesticides on the farm or farmed animals	None
4.1.2 Allowance for the application of chemicals that persist as toxins in the marine environment or on the farm or farmed animals	None
4.1.3 Only non-lethal management (e.g., exclusion, deterrents and removal) of critical species[16] that are pests or predators	Yes
4.1.4 Allowance for the use of leadline or lead sinkers on predator netting	None
4.1.5 Allowance for the use of explosives	None

Rationale—Some of the most challenging issues faced by shellfish farmers involve the control and management of diseases, predators, pests and fouling organisms. Most shellfish species are susceptible to a number of parasitic, bacterial and viral diseases. (Bower & McGladdery 1997) Low levels of sub-lethal infection are almost routine and mass mortalities are common. Shellfish are primitive organisms with rudimentary immune systems and, once they leave the hatchery, there is no economical way to deliver drugs or antibiotics to significant numbers of animals. Perhaps the best hope of controlling the spread of disease is through the use of management practices that call for the pathological inspection of animals to ensure that infected animals are not moved into areas that do not currently have endemic infections. Long-term selective breeding programs that mimic nature by amplifying the genetic tendencies for disease resistance are also showing promise in limiting the impacts of diseases that are already endemic.

Fouling control represents perhaps the greatest challenge for many shellfish farmers. The firm substrate offered by shell, ropes and the various containers that growers use to protect their crop from predators provides an ideal habitat for numerous fouling organisms that may include seaweeds, other shellfish, barnacles and many species of tunicates and bryozoans. Fouling organisms block the flow of food-rich water, often competing for food and frequently decreasing the quality, appearance and value of the end product. Fouling organisms can quickly colonize clean gear, more than doubling the weight

[16] As defined by national law or as found in the IUCN Red List of Threatened Species.

of culture gear in a few weeks. Some growers estimate that as much as 30 percent of their operating costs are related to fouling control. (Adams et al. 2009) Control measures include avoidance e.g., temporal or spatially keeping the crop away from the larval stages of the fouling organisms) mechanical removal (e.g.,scraping, brushing or power washing) and killing the fouling organisms (e.g., air drying or dipping in various caustic solutions such as brine, acetic acid or lime). Most of these solutions are components already found in seawater (salt or CaCo3) and, as long as they are handled and disposed of properly (allowing for appropriate dilution), there should be little impact to non-target organisms.

Pests and predators also pose a significant threat for shellfish farmers. Shellfish at high densities (especially juveniles) are a tempting treat for armies of crabs, starfish, fish, rays, predatory snails and diving birds. It is not uncommon for unprotected plantings to suffer near 100 percent mortalities in just a few weeks. Growers have developed a wide array of predator exclusion devices to protect their crops, ranging from mesh bags to rolls of netting similar to those used to protect fruit trees from birds. For birds, which in some cases are protected from lethal control measures by law, growers must rely on exclusion barriers or repellants such as lasers and noise, similar to land farmers. For more primitive predators, such as starfish, conchs and crabs, growers typically use a combination of barriers and trapping. New England oyster farmers have relied on dragging starfish “mops” (i.e., large weighted cotton ropes that entangle starfish which are then dipped in vats of boiling water) since the late 1800s. They also historically used applications of quicklime (CaO2) to control starfish and oyster drills (Urosalpinx cinerea). Many jurisdictions continue to mandate lethal control of starfish wherever they are encountered.

Since any action will have some measurable impact, it is important for these requirements to ensure that the impacts are localized, temporary and reversible. It also is important that the actions do not cause harm to endangered species or have a permanent impact on critical habitat.

5. PRINCIPLE: USE RESOURCES EFFICIENTLY

Issue: Although shellfish farming has one of the lowest carbon footprints of all intensive/semi-intensive food production systems, it is reasonable to expect shellfish farms to be efficient and demonstrate sustainable energy use. Also, proper waste management and pollution control are important in minimizing the impact that farming operations have on the environment.

5.1 Criterion: Waste management/pollution control

INDICATOR	REQUIREMENT
5.1.1 Evidence of waste reduction (e.g., reuse and recycling) programs	Yes
5.1.2 Evidence of appropriate storage and/or disposal of biological waste	Yes
5.1.3 Evidence of appropriate storage and/or disposal of chemical and hydrocarbon wastes	Yes
5.1.4 Spill prevention and response plan for chemicals/hydrocarbons originating from farming operations	Required

Rationale—Shellfish growers should also be responsible for waste disposal and protect against harmful chemical and hydrocarbon spills. Farming operations should have sufficient prevention and response plans in place and farm employees should have the training necessary to properly dispose of waste, and prevent and manage chemical and hydrocarbon spills.

5.2 Criterion: Energy efficiency

INDICATOR	REQUIREMENT
5.2.1 Evidence of energy use monitoring relative to production and ongoing effort to improve efficiency	Yes
5.2.2 Maintenance records for farm equipment (e.g., boats and generators) are up to date and available	Yes

Rationale—Climate change and the impacts associated with anthropogenic CO2 emissions represent the biggest environmental challenge facing current and future generations. Because of this, energy consumption used in food production has become a source of major public concern. Therefore, the requirements state that on-farm energy consumption should be monitored on a continual basis and that growers should develop means to improve efficiency and reduce consumption of energy sources, particularly those that are limited or carbon-based.

6. PRINCIPLE: BE A GOOD NEIGHBOR AND CONSCIENTIOUS COASTAL CITIZEN

Issue: Shellfish aquaculture often occurs in close proximity to communities that may be affected by farming activities. Conflict resulting from a lack of agreement over how coastal resources should be used can severely impact the social sustainability of a bivalve farming operation.

6.1 Criterion: Community relations and interaction

INDICATOR	REQUIREMENT
6.1.1 Visible floats must be of a uniform color, except where otherwise specified by law (if applicable to growing area)	Required
6.1.2 Uniform positioning and orientation of visible farm structures, except where specified by law (if applicable to growing area)	Required
6.1.3 Allowance for floats made out of open-cell Styrofoam	None
6.1.4 Noise, light and odor originating from the farm are minimized in areas where it may impact others (if applicable to growing area)	Required
6.1.5 Evidence of compliance with all applicable navigational rules and regulations	Required
6.1.6 Documented cleanup of receiving shoreline in response to gear loss based on local conditions	Required
6.1.7 Substantial gear (e.g., floats, cages, bags, predator nets and racks) is identifiable to farm (if applicable to growing area)	Yes
6.1.8 Provision of equipment for gear recovery (e.g., scoop nets and grapple hooks)	Required
6.1.9 A mechanism (e.g., insurance or an industry agreement to collect derelict gear) is in place for the decommissioning of abandoned farms	Yes
6.1.10 Conflict resolution protocol, including publicly available registry of complaints and evidence of due diligence to resolve them	Required

6.1.11 Evidence of outreach (e.g., meeting records, newsletters, consultation with communities and indigenous groups, or membership in association with documented outreach program)	Required
6.1.12 Evidence of acknowledgment of indigenous groups' rights (if applicable to growing area)	Required

Rationale—Conflicts may occur between producers and surrounding communities. It is the farmer's responsibility to minimize potential impacts by maintaining clean and orderly farm sites that do not impede navigation. Conflicts that arise between producers and surrounding communities shall be addressed through a verifiable conflict resolution policy in which complaints from communities are responded to and addressed in a timely manner. Community rights and interactions with farmers, groups of farmers and corporate farms are complex and often dynamic. The intent of these requirements is to enable communities to have a clear and transparent way of interacting with producers and for producers to interact with communities in a positive manner while responsibly maintaining their farm sites.

7. PRINCIPLE: DEVELOP AND OPERATE FARMS IN A SOCIALLY AND CULTURALLY RESPONSIBLE MANNER

Issue: Bivalve aquaculture should be undertaken in a socially responsible manner that ensures the operations benefit farm workers and local communities. The labor rights of individuals working on shellfish farms are important and farm working conditions should ensure that employees are treated and paid fairly. Appropriate farm conditions include no child labor, no forced labor and no discrimination. Complaint procedures and protection for whistle blowers are critical to achieving and maintaining fair and equitable working conditions. Socially responsible shellfish farming should ensure worker health and welfare through safe and hygienic working conditions with relevant training available for workers and managers. Please refer to Appendix III for extra guidance and definitions for the following social requirements.

7.1 Criterion: Child labor

INDICATOR		REQUIREMENT
7.1.1	Incidences of child[17] labor[18]	0

Rationale—Adherence to the child labor codes and definitions included in this section indicates alignment with what the International Labour Organisation (ILO) and international conventions generally recognize as the key areas for the protection of child and young workers.[19] Children are particularly vulnerable to economic exploitation, due to their inherent age-related limitations in physical development, knowledge and experience. Children need adequate time for education, development and play and, therefore, shall never be exposed to work or working hours that are hazardous[20] to their physical or mental well-being. To this end, the requirements related to what constitutes child labor are designed to protect the interests of children and young workers in certified aquaculture operations.

[17] A "child" is defined as any person less than 15 years of age. A higher age would apply if the minimum age law stipulates a higher age for work or mandatory schooling. If, however, the local minimum age law is set at 14, in accordance with developing country exceptions under ILO Convention 138, the lower age will apply.

[18] "Child labor" is defined as any work by a child younger than the age specified in the definition of a child, except for light work as provided for by ILO Convention 138, Article 7.

[19] A "young worker" is defined as any worker between the age of child, as defined above, and under the age of 18.

[20] "Hazardous work" is defined as work that, by its nature or circumstances in which it is carried out, is likely to harm the health or safety of workers.

7.2 Criterion: Forced, bonded or compulsory labor

INDICATOR	REQUIREMENT
7.2.1 Incidences of forced[21], bonded[22], or compulsory labor	0

Rationale—Forced labor—such as slavery, debt bondage and human trafficking—is a serious concern in many industries and regions of the world. Ensuring that contracts are clearly articulated and understood by employees is critical to determining that labor is not forced. The inability of a worker to freely leave the workplace and/or an employer withholding original identity documents of workers are indicators that employment may not be at-will. Employees shall always be permitted to leave the workplace and manage their own non-work time. Employers are never permitted to withhold original worker identity documents. Adherence to these policies shall indicate an aquaculture operation is not using forced, bonded or compulsory labor forces.

7.3 Criterion: Discrimination

INDICATOR	REQUIREMENT
7.3.1 Incidences of discrimination[23]	0

Rationale—Unequal treatment of employees, based on certain characteristics (such as sex or race), is a violation of a worker's human rights. Additionally, widespread discrimination in the working environment can negatively affect overall poverty and economic development rates. Discrimination occurs in many work environments and takes many forms. In order to ensure that discrimination does not occur at certified aquaculture farms, employers must prove their commitment to equality with an official anti-discrimination policy, a policy of equal pay for equal work, as well as clearly outlined procedures to raise, file and respond to a discrimination complaint in an effective manner. Evidence, including worker testimony, of adherence to these policies and procedures will indicate minimization of discrimination.

[21] "Forced" is all work or service that is extracted from any person under the menace of any penalty for which said person has not offered himself or herself voluntarily or for which such work or service is demanded as a repayment of debt. "Penalty" can imply monetary sanctions and physical punishment, such as loss of rights and privileges or restriction of movement (or withholding of identity documents)

[22] "Bonded" is when a person is forced by the employer or creditor to work to repay a financial debt to the crediting agency

[23] "Discrimination" is any distinction, exclusion or preference, which has the effect of nullifying or impairing equality of opportunity or treatment. Not all distinction, exclusion or preference constitutes discrimination. For instance, a merit or performance-based pay increase or bonus is not by itself discriminatory. Positive discrimination in favor of people from certain underrepresented groups may be legal in some countries.

7.4 Criterion: Health and safety

INDICATOR	REQUIREMENT
7.4.1 All health and safety related accidents and violations are recorded and corrective action is taken when necessary	Yes
7.4.2 Occupational health and safety training is available for all employees	Yes
7.4.3 Employer responsibility and proof of insurance (accident or injury) for employee medical costs in a job-related accident or injury, unless otherwise covered	Yes

Rationale—When an accident, injury or violation occurs, the company must record it and take corrective action to identify the root causes of the incident, remediate and take steps to prevent future occurrences of similar incidents. Consistent and effective employee training in health and safety practices is an important preventative measure. Finally, while many national laws require that employers assume responsibility for job-related accidents and injuries, not all countries require this and not all employees (e.g., in some cases, migrant and other workers) will be covered under such laws.

7.5 Criterion: Fair and decent wages

INDICATOR	REQUIREMENT
7.5.1 Payment of fair and decent wages	Yes

Rationale—Workers shall be paid fair and equitable wages. Company policies and practices shall also prohibit deductions in pay for disciplinary actions. Payments shall be made in a manner convenient to workers.

7.6 Criterion: Freedom of association and collective bargaining

INDICATOR	REQUIREMENT
7.6.1 Employees have access to freedom of association and collective bargaining	Yes

Rationale—Having the freedom to associate and bargain collectively is a critical right of workers because it allows workers to have a more balanced power relationship with employers when doing such things as negotiating fair compensation. Although this does not mean all workers of a certified

aquaculture operation must be in a trade union or similar organization, workers must not be prohibited from accessing such organizations when they exist. If they do not exist or are illegal, companies must make it clear that they are willing to engage in a collective dialogue through a representative structure freely elected by the workers.

7.7 Criterion: Non-abusive disciplinary practices

INDICATOR	REQUIREMENT
7.7.1 Incidences of abusive disciplinary practices occurring on the farm	0

Rationale—The rationale for discipline in the workplace is to correct improper actions and maintain effective levels of employee conduct and performance. However, abusive disciplinary actions can violate a worker's human rights. The focus of disciplinary practices shall always be on the improvement of the worker. A certified aquaculture operation shall never employ threatening, humiliating or punishing disciplinary practices that negatively impact a worker's physical and mental[24] health or dignity. Employers that support non-abusive disciplinary practices as described in Appendix III, as well as evidence from worker testimony, shall indicate that a certified aquaculture operation is not employing abusive disciplinary practices.

7.8 Criterion: Working hours

INDICATOR	REQUIREMENT
7.8.1 Incidences, violations or abuse of working hours and overtime laws or expectations (see Appendix III for details)	None

Rationale—Abuse of working hours is a widespread issue in many industries and regions. Workers subject to extensive overtime can suffer consequences in their work-life balance and are subject to higher fatigue-related accident rates. In accordance with better practices, employees in certified aquaculture operations are permitted to work—within defined guidelines—beyond normal work week hours but must be compensated at premium rates.[25] Requirements for time-off, working hours and compensation rates as described should reduce the impacts of overtime.

[24] Mental abuse: Characterized by the intentional use of power, including verbal abuse, isolation, sexual or racial harassment, intimidation or threat of physical force

[25] Premium rate: A rate of pay higher than the regular work week rate. Must comply with national laws, regulations and/or industry standards.

APPENDIX I: FORMULAS, SAMPLE CALCULATIONS AND ADDITIONAL BACKGROUND FOR PRINCIPLE 2

Bivalve culture and seabed organic enrichment

One of the primary ways that shellfish aquaculture may modify the ecosystem is by increasing the sedimentation of organic matter. By filtering suspended organic matter and changing the packaging to larger, more rapidly sinking particles (feces and pseudo-feces), shellfish can enhance the flux of organic material to the seabed. Studies on the effect of shellfish aquaculture seabed organic enrichment on benthic habitat and communities have provided a continuum of results from observations of no, or minimal, negative effects (Baudinet et al., 1990; Grenz et al., 1990; Hatcher et al., 1994; Grant et al., 1995; Shaw, 1998; Chamberlain et al., 2001; Crawford et al., 2003; Harstein & Rowden, 2004; Anderson et al., 2005; Mallet et al., 2006; Miron et al., 2005; Lasiak et al., 2006) to significant changes within farms (Dahlbeck and Gunnarsson, 1981; Mattsson and Linden, 1983; Kasper et al., 1985; Tenore et al., 1985; Jaramillo et al., 1992; Chililev and Ivanov, 1997; Mirto et al., 2000; Stenton-Dozey et al., 1999; 2001; Chamberlain et al., 2001; Christensen et al., 2003; Smith and Shackley 2004; Harstein and Rowden, 2004; Otero et al., 2006; Giles et al., 2006; Metzger et al., 2007; Cranford et al. 2009) and at the coastal ecosystem scale (Hargrave et al., 2008). The extent and magnitude of benthic effects is always site specific with vulnerability depending on factors controlling waste organic matter input (e.g., scale, duration and intensity of shellfish production, husbandry practices, seston concentration, and food utilization rate and efficiency) and hydrographic and physical factors controlling the assimilative capacity of the local environment (e.g., water depth, sedimentation rate, current and wind speed).

Bivalve biodeposition rates are related to individual feeding rates that depend, in part, on animal size and the species under culture. The principal factor determining the organic supply to the seabed is the total biomass of bivalves stocked within the farm. Suspended culture provides opportunities to greatly increase stocking biomass within an area relative to bottom culture and therefore represents a greater risk to benthic communities. The studies cited above that have shown significant negative effects on the sea bed were generally conducted in areas with suspended culture. Given the relatively low risk for benthic organic enrichment impacts posed by on- and in-bottom culture, these activities are exempted from the organic enrichment requirements. On-bottom culture is considered here to be limited to intertidal and sub-tidal husbandry practices which do not require bivalve holding structures that can aid in increasing the stocking biomass (e.g., poles and cages).

By comparing the level of total ‘free’ sulfides in the sediment beneath a farm to a nearby control site, the degree of organic enrichment can be assessed. Sediment organic enrichment classifications have been identified based on the known effects of changes in sediment sulphide on the biodiversity of macrofauna (see Hargrave et al., 2008b and cited references). The associated sulphide threshold values enable managers to distinguish between normal ranges of “background” concentrations from those indicative of benthic habitat degradation.

Relationships between biological variables are consistent with changes in sulphide levels as sediments are transformed from oxic to anoxic status. Impacts to benthic fauna biodiversity resulting from increased S concentrations can be significant and occur at low S levels. The transition from oxic to hypoxic conditions has been identified as occurring at 1500 µM S. This threshold represents a transition from “moderate” to “reduced” macrobenthic sulphide concentration and changes in the benthic macrofauna community structure (described by Hargrave et al., 2008b). A nomogram was used to show that various benthic enrichment classification schemes based on changes in different inter-related chemical and infauna biodiversity (defined by Pearson and Rosenberg, 1975) at which the mean number of taxa are reduced by approximately 50 to 60 percent relative to typical oxic conditions (Hargrave et al. 2008b). Anoxic sediments were characterized by S concentrations >6000 µM S. A transition within the hypoxic class of sediments at 3000 µM has been identified where less S-

tolerant taxa disappear but more tolerant opportunistic species have not increased in abundance. S levels above 3000 µM represent a condition that exerts “severe hypoxic stress” on benthic community structure (defined by Diaz and Rosenberg, 1995) and characterize a “polluted” sediment condition (defined by Pearson and Rosenberg, 1975) that poses a high risk to benthic habitat.

Table 1 below, illustrates the tiered assessment approach for the benthic effects of suspended bivalve culture. The sampling design and measurement protocols for the benthic assessment are included with the requirements as separate documents.

Table 1

METHOD	CLASSIFICATION	DECISION	CONDITION
Seabed video/ imaging and surficial sediment sulfide (S) concentration at farm sampling sites vs. reference sites	Non-depositional, coarse sediment (sand, cobble) or S ≤1500 µM)	Acceptable	Monitor every 5 years.
	Depositional, fine sediment and A) S >1500 and ≤ 3000 µM	Acceptable	Monitor every year and take management responses when necessary to maintain farm S levels within the range of natural variance measured at adjacent reference sites.
	B) S > 3000 µM	Unacceptable	Management response (e.g. site fallowing) necessary before farm is eligible for certification

If water renewal is faster than water clearance (CT>RT) it is expected that carrying capacity will not be exceeded. If CT<RT, cultured bivalves may be able to control the ecosystem and an additional assessment is required linking clearance time to primary production (PPT). The rationale for the Tier 2 calculation is that phytoplankton production in a bay can support sustainable aquaculture, up to a point, even when the bay is poorly flushed. Primary production time should be shorter than clearance time. Otherwise, the algae which shellfish feed on will quickly be depleted. In theory, the requirement could be CT/PPT>1 but in practice CT/PPT should be >3. This is based on empirical data from a series of estuaries and is a logical assumption due to the algal buffer stock required in order to realize a certain level of primary production, not to mention the occurrence of other unknown filter-feeder stocks in proximity to the shellfish farming operations (Smaal & Prins, 1993). It should be understood that this factor of 3 is a practical figure rather than an ecological fixed requirement. When CT/PPT ≤ 3, farms are no longer eligible for certification. If this is the case, bay-wide management plans that address the potential cumulative pelagic effects of multiple farms and reduce regional stocking levels are necessary in order to ensure that the ecological carrying capacity is not being exceeded.

A consideration of the "water body" is required for these calculations, and to address cumulative effects where zones of influence of adjacent farms overlap. In many instances, such as enclosed bays or inlets, the geographic boundaries of the area in which the farm is located may be obvious and considered as the water body. In other situations, such as meandering complex waterways or the open coast, there may be no clear boundaries. In these cases, there needs to be some estimation of the water body in which the farm sits so its zone of influence can be estimated in relation to carrying capacity, proximity to sensitive communities or foraging range of protected species.

There are several methods that can be used to estimate the water body or farm zone of potential influence, ranging from a full hydrodynamic model with or without explicit phytoplankton dynamics, to a simple estimation of the tidal excursion and residual currents using current meters, or, more cheaply, by drifters, or dye release. It is assumed that most offshore bivalve farms will be relatively large and possibly be owned by companies with greater resources than small, inshore farms in enclosed bays, so the use of current meters should not be an impediment.

Formulas and sample calculations

Estimating a Farm's Zone of Influence

As a general rule, estimating the zone of potential influence for a farm should give results showing it is less than the area of the enclosing bay, or it is limited to a circle around the farm calculated from the mean current and some time scale to allow for phytoplankton regrowth or turnover time. Under reasonable conditions, phytoplankton growth is in the order of 1-2 days. Therefore an approximation of the water body, based on zone of potential influence, is:

> Mean current speed at the farm x 2 tidal cycles (i.e., 25 hours if the M2 tide is dominant) x mean water depth (or depth of growing lines if the farm is in deep water)

Example calculations for a farm located some distance offshore are:

1. Mean current speed of 5 cm/s and water depth of 15 m. The zone of potential influence would, therefore, have a radius of 4.5 km, and the volume of the water body is 675,000 m^3.
2. Mean current speed of 2 cm/s, water depth of 30 m, but the growing lines only extend to 7 m depth. The zone of potential influence has a radius of 1.8 km, and the volume of the water body is 126,000 m^3.

CT (days) = V_t / (N x C)

Where

V_t is the total volume of the water body at high tide (liters)
N is number of bivalves
C is average clearance rate (liters/individual species/day) at harvest size

Retention Time (RT), *as used in indicator 2.2.1*

RT = -1 x P / ln (V_l / V_t)

Where

P is the tidal periodicity, the length of the tidal cycle (e.g. ~0.5 days for semidiuranl tides)
V_l is the total volume of the water body at low tide (liters)
V_t is the total volume of the water body at high tide

Note: For deep stratified culture areas (e.g., open ocean and fjords), this calculation should be limited to the surface mixed layer. In areas where water exchange is not dominated by tidal flushing (e.g., controlled primarily by river flow or wind forcing) an appropriate volume exchange should be calculated.

Primary Production Time (PPT), *as used in indicator 2.2.2*

PPT = B/PPP

Where

B is the yearly averages of phytoplankton biomass,
PPP is the phytoplankton primary production (PPP) within the system.

Note: B can be estimated from chlorophyll a measurements, published data or satellite predictions assuming a carbon to chlorophyll ratio of 50. PPP can be obtained from published results or model predictions. Some examples of available data resources include:

http://marine.rutgers.edu/opp/
http://www.science.oregonstate.edu/ocean.productivity/index.php

APPENDIX II: GUIDANCE FOR NATIVE SPECIES CULTIVATION

Genetic impacts of hatchery produced seed

It is possible that hatchery production of seed could negatively impact wild populations of cultivated species by altering their genetic composition over time in ways that compromise their long-term viability. Efforts should be made to address genetic concerns specific to species and the geographic region where the seed will be out-planted. This may include preserving diversity of broodstock and seed by 1) using local broodstock, 2) rotation of broodstock within spawning seasons and between years, and 3) avoiding the use of hatchery-propagated stock in the hatchery as broodstock. This may also include documentation that the scale of farming and the reproductive potential of crops (e.g., whether diploid or triploid, or considering age at harvest and age at first maturation) are well-below the size and reproductive potential of the natural population within a reasonable “dispersal kernel” from the farm. Compliance with this requirement would depend on the availability of local fisheries information and management. This may include documentation through common garden trials, for example, that performance (e.g., survival and growth) or characteristics (e.g., shell shape and color) of hatchery-propagated diploid seed have not diverged from that of wild seed. Compliance might involve a shared responsibility between hatcheries and farms. It may also include the production of sterile seed for out-planting from breeding programs that intentionally alter wild stocks for improved culture traits, such as growth, yield, survival and morphology. In the situation where restoration efforts in the geographic region of out-planting involve intentional divergence from wild stocks to produce disease resistant wild populations this may include documentation of cooperation with such efforts.

APPENDIX III: GUIDANCE FOR THE SOCIAL COMPONENT OF THE ASC BIVALVE STANDARD

The standards related to labor issues and work conditions on the farm were created with input from Social Accountability International (SAI), a recognized leader on labor issues. SAI also recommended the following guidance to accompany the social component of the ASC Bivalve Standard.

1. Child labor

Guidance

- Child workers under the age of 15 perform only light work (see definition of "light work" below) as long as it does not exceed 2 hours per day on a school day or holiday and the total number of hours spent on light work and on school does not exceed 7 hours/day.
- For employees aged 15-18 (defined as young workers), work should not conflict with schooling. The combination of daily transportation, school time and work time should not exceed 10 hours).
- Hazardous work is not performed by those below age 18. This includes heavy lifting disproportionate to their size, operating heavy machinery, working night shifts and exposure to any toxic chemicals.

Definitions

"Light work," as defined by ILO convention 138, article 7.1, is work is work that is 1) not likely to be harmful to a child's health or development and 2) not likely to prejudice their attendance at school, participation in vocational orientation or training programs or diminish their capacity to benefit from instruction received.

2. Forced, bonded, compulsory labor

Guidance

- Employer should never be permitted to withhold original identity documents.
- Contracts should be clearly stated and understood by employees and never lead to employee being indebted (e.g., employees paying for training programs).
- Employees should be free to leave the workplace when not working and manage their own non-work time.

Note: Extra care should be given to migrants and contractor/ subcontractor situations

3. Discrimination

Guidance

- Company shall not engage in or support discrimination in hiring, remuneration, access to training, promotion, termination or retirement based on caste, national origin, religion, disability, gender, sexual orientation, union membership, political affiliation or age.

- Company shall not interfere with employee rights to exercise or observe tenets or practices, or to meet needs related to race, caste, national origin, religion, disability, gender, sexual orientation, union membership or political affiliation.

4. Health and safety

Guidance

- Minimization of hazards and risks in the working environment, including documented procedures and policies to prevent workplace accidents and injuries. Emergency response procedures should exist and be known by employees.
- Documentation of occupational health and safety violations.
- Access to clean lavatories, potable water and sanitary facilities.
- Dormitories must be clean, safe, and meet the basic needs of employees.
- Insurance, if not otherwise provided, to cover employees who suffer accident or injury in the work environment. Special consideration must be given to migrant or foreign workers who may fall outside of local or national laws and legislation.
- Corrective action plan for accidents that have occurred.

5. Fair and decent wages

Guidance

- No deductions for disciplinary actions, wage and benefits are clearly articulated to employees.
- Wages and benefits are rendered in a manner convenient to employees (e.g., no travel, promissory notes, coupons, products or merchandise to replace cash, checks or electronic methods of payment).
- No labor-only contracting relationships or false apprenticeship schemes (see definitions of "labor-only contracting relationship" and "false apprenticeship" below)

Definitions

Labor-only contracting arrangement: The practice of hiring workers without establishing a formal employment relationship for the purpose of avoiding payment of regular wages or the provision of legally required benefits, such as health and safety protection.

False apprenticeship scheme: The practice of hiring workers under apprenticeship terms without stipulating terms of the apprenticeship and wages in the contract. It is a "false" apprenticeship if the purpose is to underpay people, avoid legal obligations or employ children.

6. Freedom of association and collective bargaining

Guidance

- Employers should respect the right of all personnel to form and join trade unions of their choice and to bargain collectively.
- When such situations are restricted under law, employers should facilitate parallel means of independent and free association and bargaining and ensure they are not the subject of discrimination. When rights are restricted, the company needs to make clear to workers that they are willing to engage workers in collective dialogue through representative structure and that they are willing to provide them with the opportunity to do so.

7. Non-abusive disciplinary practices

Guidance

- Absolutely no engagement in or support of corporal punishment, mental or physical coercion or verbal abuse. Fines or wage deductions are also not an acceptable method for disciplining workers.

8. Working hours and overtime

Guidance

- Auditors shall be aware of working hours and overtime requirements in local legislation. They can check time sheets and payroll and verify through worker interviews that workers are working legally allowed hours. Pay slips and pay records can confirm whether overtime hours are being paid at a premium. To verify that overtime is not the norm, interviews can be conducted and production records checked, as well as time sheets other records of working hours, for at least one year before. Some exceptions can be made for overtime not being voluntary, if there is a collective bargaining agreement in place that allows it.
- Employer shall comply with applicable laws and industry standards related to working hours. “Normal work week” can be defined by law but shall not on a regular basis (i.e., constantly or majority of the time) exceed 48 hours. Variations based on seasonality may apply.
- All overtime shall be paid at a premium and should not exceed 12 hours per week. Overtime work shall be voluntary. Exceptions to this last requirement can be made in cases where it is legal and in which there is a collective bargaining agreement in place which addressed this, in order to meet short-term business demands

APPENDIX IV: EXPERIMENTAL DESIGN FOR ASSESSMENT OF BENTHIC IMPACTS OF SUSPENDED BIVALVE CULTURE

1.0 Rationale

Before-After-Control-Impact (BACI) and Gradient Analysis (GA) models may be used for detecting environmental change or disturbance in benthic variables affected by suspended bivalve culture. The recommended experimental designs are consistent with the tiered assessment approach described in Section 2.1.1 for non-depositional areas of hard substrates using bottom video/imaging methods or measurements of total 'free' sulfide (S) or other indicators of organic enrichment in depositional areas where bottom samples can be collected. Changes in benthic habitat characteristics are assessed by comparing observations at a series of stations inside and outside of a farmed area either along transects or randomly placed as groups of sampling sites to assess temporal and spatial differences in measured variables. The design selected determines station locations, numbers and sampling frequency. The before-after (BA) test compares observations within a farmed area before and after culture lines are established. Alternatively, non-farm (control) and farm (impact) areas can be compared (CI design) to determine if bivalve culture has changed temporal variations in selected variables. If both BA and CI data from multiple sites are available, the BACI model detects environmental change associated with disturbance. Regression analysis is used to test for spatial trends in a GA model where changes in variables occur with increasing distance from a farm.

2.0 Tiered Assessment

A tiered assessment approach discussed in Section 2.1.1 is recommended for assessing effects of bivalve culture on benthic habitat conditions (Fig. 1). Collection of bottom video or other images along transects once every 5 years (Tier 1a assessment) is recommended for monitoring high energy, low risk areas where hard bottom prevents sample collection. Sampling in lower energy areas with depositional conditions represented by sand or mud bottom types may be carried out once every 5 years, annually or more frequently depending on the level of risk determined by mean or median S concentrations (Tier 1b and 2a and 2b assessments).

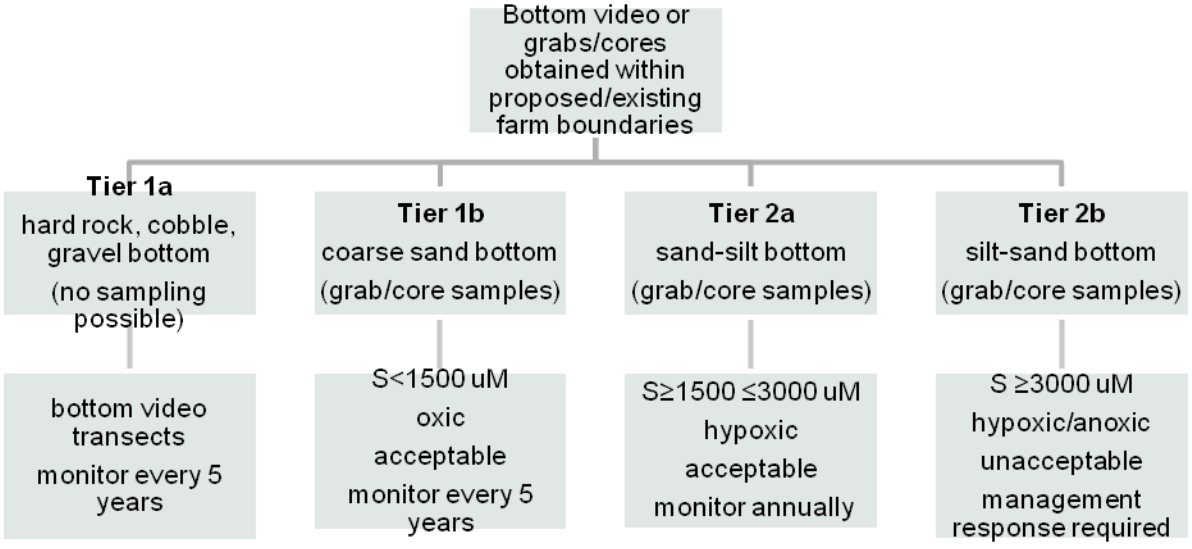

Fig. 1 Tiered assessment approach for assessing effects of bivalve culture on benthic habitat conditions (see Section 2.1.1).

3.0 Sampling Designs

3.1 Before-After Observations with and without a Control

Green (1979) described a simple approach for detecting environmental changes associated with human activity by making observations at one location before and after the activity has occurred. Underwood (1991, 1992), Smith et al. (1993) and Underwood (1994) presented a more complete experimental sampling design with observations at multiple sites in impacted and control areas (BACI) to determine if changes in measured variables are due to environmental disturbance and to account for natural variations.

A full application of the BACI method requires collection of replicate samples in time and space over different temporal scales at multiple sampling sites to determine if an 'event' has changed one or more measured variables. Observations are compared before and after the start of the activity potentially causing the disturbance. Multiple sites selected at random within impacted and control areas can be sampled with observations at the same locations before and after the activity has begun. Ideally sampling should occur at random times but seasonal effects can be minimized if sampling occurs at a fixed time during the year.

The approach has been used following the establishment of mussel culture to assess temporal variations in benthic macrofauna communities (Lasiak et al. 2006) and geochemical indicators for sediment organic enrichment (Cranford et al. 2009).

3.2 Control-Impact Observations

Often data from the before period is not available. If this is the case a control-impact (CI) model can be used to compare sites within and outside of farm boundaries in locations assumed to be unaffected by disturbance. A decision must be made to select sampling locations within impacted and control areas on a random or stratified basis. If a randomized design is used control sites must be a sufficient distance from the impacted area to represent natural 'background' variability (i.e. be unaffected by the events within the farm). The appropriate distance may be determined by sampling along transects with distance and directions determined by the velocity and direction of major currents (discussed below). The appropriate upstream or downstream locations for control sites relative to a bivalve culture area will vary with specific hydrographic conditions. In some studies benthic effects were only measureable directly under culture arrays (Grant et al. 1995, Crawford et al. 2003). However, modelling studies of the distribution of biodeposits from mussel aquaculture have shown that depending on current speed and water depth enhanced settling of particulate matter from cultures could occur up to distances of 30 to 90 m from a farm (Weise et al. 2009). The CI approach was used in an embayment-wide study in a shallow inlet with intensive mussel culture to show effects of organic enrichment in farm vs. non-farm areas (Hargrave et al. 2008).

A possible complication in applying the CI approach to suspended bivalve aquaculture is that plankton depletion by farms may occur over large spatial scales such that natural sedimentation rates outside of farms are decreased below natural values. This could result in lower organic loading outside of farm areas and decreased sulphide levels, confounding comparison between farm and control sites. This hypothesis was tested and rejected in a study of intensive mussel culture in shallow eutrophic embayments (Cranford et al. 2009). It was shown that sediment geochemistry at control sites located

as close as 10 m to farm boundaries did not change significantly before and after the expansion of mussel production, while the farm sites exhibited a significant increase in organic enrichment during this same period.

3.3 Gradient Observations

The BACI designs allow area-by-area (e.g. farm vs. non-farm) comparisons to detect environmental changes against a background of natural variability when there are defined boundaries for the impacted area. GA using sampling along transects provides an alternative design for assessing effects of bivalve aquaculture where boundaries between impacted and control areas may be poorly defined or variable between sites. Sampling along transects may be more sensitive for detecting spatial differences than a CI design if the disturbance is directional (Ellis and Schneider 1997). Sampling stations on transects should be located along the axis of the major current with either uniform spacing or at variable distances to reflect expected diminished effects with increasing distance from culture arrays. Crawford et al. (2003) provided an example of observations along transects to evaluate benthic effects of shellfish farms.

4.0 Station Location and Numbers

Since the power of statistical tests increases with sample size (Sokal and Rohlf 1995) numbers of observations should be as large as practically possible with equal numbers of observations at all locations being compared. As a compromise between monitoring costs and statistical power to detect differences between sites, triplicate samples at ten sites along transects or within farm and non-farm locations are recommended. Replication (3 samples x 5 sites, n=15) for each group of stations to be compared is required to account for variations in benthic conditions common in shallow coastal areas where bivalve aquaculture occurs.

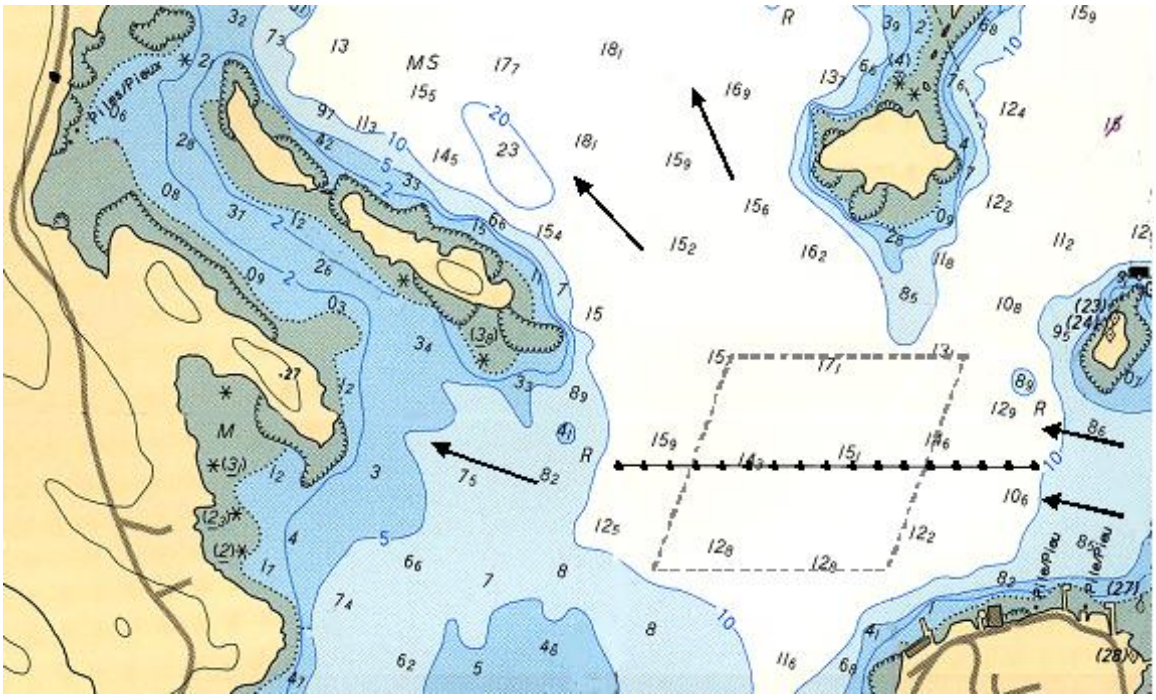

Fig. 2 Example of gradient sampling approach for Tier 1a assessment of benthic effects of bivalve aquaculture (Fig. 1). The video transect (solid line) or photographic transect (solid dots) is aligned along the axis of the major current (arrows) and runs through the farm boundaries (dotted lines).

Tier 1a assessments of non-depositional or hard bottoms using bottom imaging should be conducted using the GA approach with transects extending from inside to outside of a proposed or existing farm area. It is assumed that culture lines are positioned to maximize flow through the arrays to avoid 'shading' effects. Sampling transects to obtain bottom video requires that a boat travel across a farm site without interference by culture or mooring lines. The orientation of transects should as far as possible follow depth contours to minimize depth and sediment type variations. Bottom imaging with GPS navigation would be obtained continuously (video) or at random or regular intervals (still images) along the entire length of a transect running through and outside farm boundaries in both directions. Image analysis can then be applied to examine gradients in benthic conditions along the transect.

Bottom samples for the preliminary site assessment and subsequent Tier 1b, 2a and 2b monitoring programs may be collected using the GA approach sampling in either upstream or downstream directions by collecting samples at known distances from the farm boundary (Fig 3). Station spacing can be uniform or increase with distance from the edge of a farm with triplicate surface (0-2 cm) sediment samples collected at each of the five sites along a transect.

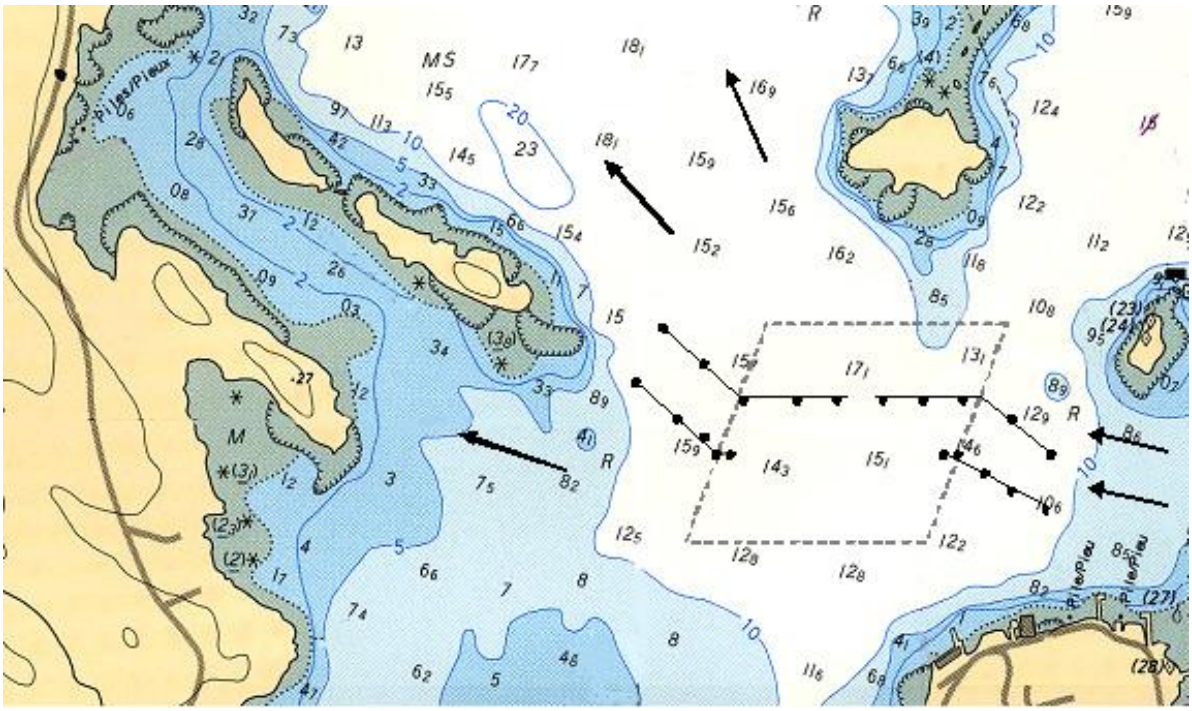

Fig. 3 Four examples of gradient sampling approach for Tier 1b, 2a and 2b assessment of benthic effects of bivalve aquaculture (Fig. 1). The bottom sampling sites (solid dots) are aligned along the axis of the major current (arrows) and run through the farm boundaries (dotted lines). Samples for total 'free' sulfide measurements would be collected in triplicate from five stations along a single transect either upstream or downstream from the farm. Station spacing could be uniform or increase with distance from the farm edge. Transects inside and outside the boundaries of the culture area should have generally similar depths and sediment types.

Bottom samples for the preliminary site assessment and subsequent Tier 1b, 2a and 2b monitoring programs may also be collected using a random control-impact sampling approach (CI and BACI) by collecting samples in either upstream or downstream directions at known distances from the farm boundary. Triplicate surface (0-2 cm) sediment samples can be collected from five randomly located stations within farm and non-farm areas (Fig. 4). Control sites are located in an area assumed not to be influenced by the cultured stock (e.g. a sufficient distance from farm boundaries to be unaffected by increased sedimentation of bio-deposits). Depths and bottom substrates in the farm and non-farm areas should be similar to avoid confounding effects of depth and sediment type on S concentrations.

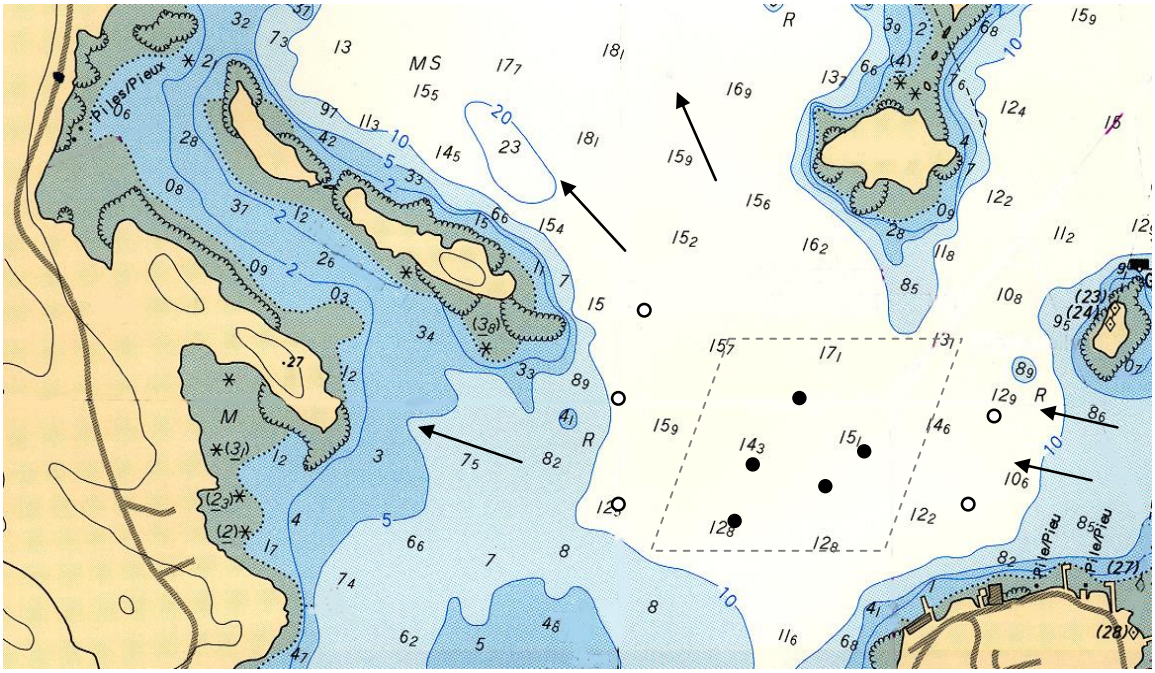

Fig. 4 Example of random sample station locations for Tier 1b, 2a and 2B assessments of benthic effects of bivalve aquaculture. Total 'free' sulfide measurements would be made on triplicate samples of surface (0-2 cm) sediment collected at five stations with generally similar depths and sediment types located randomly within (solid dots) and outside (open circles) of the farm boundary.

5.0 Statistical Analyses

Gradient Sampling Approach: Data from samples at five or more locations along each transect can be compared using linear and non-linear regression methods to test for significant gradients with distance

and goodness-of-fit. Common statistical packages allow *p* values to be calculated with assumptions that independent residuals have equal variance. Bartlett's test can be used to test for equal variance across groups of samples.

Random Sampling Approach: A one-sample one-tailed t-test can be used to determine if the mean S concentration within a group of stations where bottom samples can be collected exceeds threshold S concentrations (1500 and 3000 µM) to determine if a Tier 1b, 2a or 2b assessment is required. The precision of the test depends on the number of observations, assumes random sampling with independent observations and a normal distribution. This may not be the case when sample size is small (n=15). The null hypothesis (mean S concentration at the culture site is equal to mean values at control sites) is rejected when $p<0.05$.
A Wilcoxon signed-rank test can be used to compare median values between samples in same location are compared before and after culture activities have begun. The non-parametric test is recommended due to the small numbers of samples being compared. The null hypothesis is that the distribution functions of median values of two related sample groups are the same (before-after observations have a median difference of zero).

When sample size is small (n<20) a Mann-Whitney U test can be used to determine if median values between two groups of samples are significantly different. The test determines if samples from two independent groups have identical distribution functions and median values. There is no requirement for a normal distribution. Time can be used as the grouping variable in a BA comparison to test the null hypothesis that the temporal change in median S concentrations in two groups of stations is the same. The test can also be applied in a CI design to compare median values of S within farm and non-farm areas using site as the grouping variable. The null hypothesis is that observations do not differ significantly between the two areas. If BA and CI comparisons are available with multiple sampling times and locations in control and farm sites before and after culture operations begin, a BACI can be performed using an ANOVA model with site, time and site x time interaction. An ANOVA, however, requires confirmation that the data are normally distributed and the statistical power is reduced when sample size is small.

APPENDIX V: METHODS FOR REDOX (EhNHE) AND 'FREE' SULFIDE MEASUREMENTS IN MARINE SEDIMENTS

1.0 Collection of sediment samples

1. At sites <20 m depth divers can be used to push open ended acrylic core tubes into the sediment to maintain the sediment-water interface as undisturbed as possible. A gentle twisting motion prevents sediment compaction and keeps the sediment surface inside level with that outside the core during insertion.
2. Upper and lower ends of a core are closed with rubber stoppers or plastic caps to prevent water leakage.
3. At deeper depths grabs (e.g. Van Veen, 0.25 m^2) can be taken. If sediment does not completely fill the grab a reasonably undisturbed sample of the upper sediment layers can be obtained.
4. Holes of sufficient diameter to allow insertion of a cut-off syringe are drilled at 2 cm distances in a spiral fashion down the length of a core tube and covered with duct tape allow sediment subsamples to be withdrawn from different depths.
5. After retrieval of a core, kept upright and handled gently to minimize disturbance of the sediment surface, a sharp blade is used to cut into the duct tape (X) over each hole starting at the top.
6. Sampling in a sequential down-core fashion prevents disturbance of deeper layers when more shallow depths are sampled first.
7. A 5-ml cut-off plastic syringe is used as a subcorer filled by slowly withdrawing the fully inserted plunger as the body of the syringe is pushed horizontally into the core.
8. A mixed sample from surface sediment in a grab can be obtained in the same way by inserting the syringe obliquely to 2 cm depth. The plunger is partially withdrawn as the open-end of the syringe barrel is slowly pushed into the sediment. The procedure is repeated until the barrel is fully withdrawn and the syringe filled with mixed sediment from the 0-2 cm layer with no air bubbles.
9. Syringes must be closed with tight fitting (air-tight) plastic caps and stored on ice or refrigerated (5 ºC).
10. Analyses for redox potentials (Eh_{NHE}) and dissolved ('free') sulfides (HS^-, H_2S, $S^=$) (S) should be carried out within 4 to 6 hr but samples may be stored for up to 72 hr if refrigerated or held on ice without being frozen.

2.0 Redox (Eh_{NHE}) potentials

2.1 Materials

1. An ion specific electrode (ISE) meter (e.g. Orion 4-Star pH/ISE, model #1215001) or any mV meter with a connector suitable for attachment of the redox electrode.
2. An oxidation reduction potential (ORP) Pt electrode combined with an internal reference electrode (e.g. Orion 96-78BNWP) with a cable and appropriate connector for attachment to the ISE meter. The electrode should have a thin Pt disc (rather than a pin), be refillable (not gel-filled) with an epoxy body (to avoid breakage).
3. A 4 M KCL filling solution (e.g. Orion solution #900011, KCL saturated with Ag/AgCl) is recommended for redox electrodes used in marine sediments.
4. Redox reference solutions may be purchased from some ISE electrode manufacturers or standards such as Zobells solutions can be prepared from reagents (see below).
5. Cleaning strips can be used for polishing Pt electrodes (available for Orion ISE electrodes) or a fine powdered detergent can be used as an abrasive.

2.2 Zobell Eh standard solutions

1. **Zobell Standard A:** weigh 2.11 g of $K_4Fe(CN)_6 \cdot 3H_2O$ (potassium hexacyanoferrate (II) trihydrate) and 0.825 g of $K_3Fe(CN)_6$ (potassium hexacyanoferrate (III)) into a 50 ml volumetric flask, add ~25 ml of distilled water to dissolve the solids and dilute to 50 ml.
2. **Zobell Standard B:** weigh 0.21 g $K_4Fe(CN)_6 \cdot 3H_2O$, 0.825 g $K_3Fe(CN)_6$ and 1.695 g of $KF \cdot 2H_2O$ (potassium fluoride dehydrate) into a 50 ml volumetric flask, add ~25 ml of distilled water to dissolve the solids and dilute to 50 ml.
3. Fresh ZoBell's solutions must be at least 24 h old before use. Solutions are stable at room temperature for several months when stored in air-tight glass-stopper flasks.

2.3 Assessing Pt electrode performance

1. Pt electrodes stored dry must be activated by adding the filling solution at least 24 hr before determining performance in redox standards.
2. A prepared electrode should stabilize rapidly (<30 sec) due to the strong oxidation-reduction coupled reaction in a standard solution.
3. With a 4 M KCL filling solution Zobell solution A should have a potential of +234 ± 9 mV and solution B +300 ± 9 mV at 20 ºC.
4. After a day's use the Pt tip of the electrode should be cleaned with a detergent or abrasive strip followed by rinsing with distilled water. For long term storage (more than a week) the filling solution can be removed and the probe stored dry.

2.4 EhNHE measurements

1. Samples in 5 ml syringes allow two 2 ml sub-samples of sediment to be analyzed, either as duplicates or with the second sample used for other analyses (e.g. water content, grain size, organic matter).
2. Prior to analysis 2 ml of sediment is pushed from a syringe into a small (50 ml) beaker. Syringe markings can be used to determine the extruded volume.
3. Temperature measurements of the subsample should be made immediately and the Pt electrode placed in the sample to ensure full contact between the Pt tip and wet sediment.
4. mV readings should stabilize within 1-2 min. If redox conditions are not controlled by single oxidation-reduction reactions, as in oxic sediments, there is often a slow, continuous drift of electrode potentials (Whitfield 1969). An arbitrary time (3-4 min) can be chosen to record mV readings if they do not stabilize in less than this time. Potentials in reduced sediments usually stabilize more rapidly due to redox conditions being controlled primarily by the reversible half-cell reaction [$HS^-aq. \leftrightarrow S^o$ rhomb + $H^+aq. + 2e^-$] (Berner 1963).
5. Measured mV potentials are corrected to be relative to the normal hydrogen electrode (Eh_{NHE}) by addition of a potential characteristic for the filling solution used and the sample temperature. (Table 1)

Table 1 Reference electrode potentials (mV) relative to the normal hydrogen electrode at different temperatures and filling solution concentrations to be added to Pt electrode potential to determine Eh_{NHE}. From Wildish et al. (1999).

Temperature (°C)	1.5 M KCL Orion #900001	4 M (saturated) KCL Orion #900011
5	254	219
10	251	214
15	249	209
20	244	204
25	241	199
30	238	194

3.0 'Free' sulfides

3.1 Materials

1. A portable ISE meter (e.g.Orion 4-Star pH/ISE, model #1215001) or any mV meter with a connector suitable for the AgS electrode.
2. An Orion $Ag^+/S^=$ combination electrode (Orion #96-16BNWP) or similar electrode with a thin disc of Ag (rather than a pin) at the electrode tip. The electrode should be refillable with an epoxy body and have a suitable connector to allow attachment to the ISE meter.
3. If the Orion 96-16 Sure-flow combination electrode is used, Optimum Results™ A solution (Orion #900061) is recommended as the filling solution for precise $S^=$ measurements with optimum temperature and response time (Thermo Electron Corp. 2003).

3.2 Sulfide anti-oxidant buffer (SAOB) solution

1. SAOB solution can be purchased (e.g. from Orion as Sulfide Anti-oxidant Buffer (SAOB II) Reagent Pack #941609) or prepared from separate reagents.
2. 20.0 g of NaOH and 17.9 g EDTA buffer (ethylenediaminetetraacetic acid disodium salt dehydrate) are placed in a 250 ml volumetric flask and diluted to volume with distilled water.
3. Allow the solution to cool to room temperature before use. The solution is stable for up to 7 days if stored in a refrigerator.
4. Just before standards or samples are to be analyzed 8.75 g of L-ascorbic acid is added to the 250 ml of the SAOB solution. The mixture is unstable and must be used within 3 hr.
5. The SAOB with ascorbic acid is added to standards and wet sediment samples in a 1:1 volume ratio.

3.3 Sulfide standards

1. A stock solution of 0.1 M Na_2S is prepared by weighing 2.402 g $Na_2S•9H_2O$ into a 100 ml volumetric flask and diluting to 100 ml with de-oxygenated (N_2–bubbled) distilled water. Large crystals should be ground to a fine consistency using a mortar and pestle. Use rubber gloves and weigh the reagent on a balance in a fume-hood.

2. Although solutions of $Na_2S \bullet 9H_2O$ are unstable and easily oxidized on exposure to air (Barica 1973), the concentrated 0.1 M stock solution can be stored refrigerated in a dark, air-tight bottle for up to 48 hours.
3. A decreasing concentration series is prepared by transferring 10 ml of the concentrated stock solution into a volumetric flask and diluting to 100 ml with 90 ml of de-oxygenated distilled water.
4. The procedure is repeated sequentially using 10 ml aliquots of each standard and 90 ml of de-oxygenated water (e.g. 10 ml of 10,000 μM $S^=$ standard solution is transferred to a volumetric flask and diluted to 100 ml to prepare 1000 μM $S^=$).
5. Diluted standards are unstable and must be used for calibration of an electrode as soon as possible.

3.4 $Ag^+/S^=$ electrode calibration

1. A dry $Ag^+/S^=$ combination electrode must be activated by adding the filling solution at least 24 hr before use.
2. A set of standards (e.g. 100, 1000, 10000, 100000 μM $S^=$) is prepared to span the range expected in samples.
3. Standards should be at the same temperature as samples.
4. The $Ag^+/S^=$ combination electrode tip should be gently cleaned using an abrasive strip or detergent solution before each calibration.
5. Calibration of the $Ag^+/S^=$ combination electrode should be carried out working from the lowest to highest concentration in a standard series.
6. Standards are diluted 1:1 with equal volumes of SAOB (with ascorbic acid added) (e.g. 2 ml standard + 2 ml SAOB).
7. The ISE meter should be used in the direct measurement mode to record mV potentials after they stabilize (usually <2 min).
8. The theoretical slope constant for the inverse linear relation between $\log_{10} S^=$ and mV potential is approximately -28 mV (Thermo Electron Corp. 2003).
9. The slope of the calibration curve is slightly temperature sensitive with theoretical values between -28.1 and -29.1 at 10 and 20 ºC, respectively. In practice slope coefficients vary (-26 to -34) depending on electrode characteristics.
10. Electrodes should be calibrated at least once a day or during a day before and after analysis of a set of samples.

3.5 Sulfide measurements

1. Electrochemical potentials are temperature sensitive and standards and samples should be the same temperature (±1 °C).
2. SAOB is added to the sediment (1:1 volume) immediately following redox measurements.
3. The $Ag^+/S^=$ electrode is positioned such that the tip is fully immersed in the SAOB-sediment mixture.
4. Alkaline conditions (pH>12) created by SAOB will dissolve solid phase metal-sulfide complexes causing $S^=$ concentrations to increase over time as particulate phase sulfides (FeS and pyrite) are solubilized. The effect can be minimized by recording potentials as soon as possible when drift has stabilized (1-2 min).
5. The stable mV reading is used in the calibration curve regression to calculate μM $S^=$.
6. $Ag^+/S^=$ electrodes can be wiped clean and rinsed with distilled water between the analysis of successive samples.
7. The reference electrode filling solution should be drained and the chamber rinsed with distilled water if the electrode is to be stored for more than one week.

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