Interstate Shellfish Sanitation Conference

2023 Biennial Meeting

Task Force I Report

Baton Rouge, Louisiana

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March 18-23, 2023
Baton Rouge Marriott

Proposal No. **13-107**

Submitter	Robert Rheault
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Address Line 1	1623 Whitesville Road
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Proposal Subject	Sources of Seed for Aquaculture
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter VI. Shellfish Aquaculture
Text of Proposal/ Requested Action	.03 Seed Shellstock Seed may come from any growing area, or from any growing area in any classification, provided that: A. The source of the seed is sanctioned by the Authority B. Seed from growing areas ~~or growing areas~~ in the restricted or prohibited classification have acceptable levels of poisonous or deleterious substances; and C. Seed from growing areas ~~or growing areas~~ in the prohibited classification are cultured for a minimum of ~~six (6) months~~ one month while average daily water temperatures are above 50 degrees F.
Public Health Significance	Shellfish seed collected or cultured in certain growing areas that are in the prohibited classification have been shown through repeated sampling to be free of deleterious substances (John Mullen RI DOH, unpub. data, Rheault unpubl. data, Rice unpub. data, Leavitt unpub. data). A period of one month is typically adequate to purge viral and bacterial contaminants provided water temperatures are high enough to maintain active metabolic activity (above 60 degrees F or 15 degrees C) (Richards 1988). Once the Authority is satisfied that adequate sampling has demonstrated that the seed have "acceptable levels of deleterious substances", then a 30 day period of culture in open waters should be adequate to allow purging of bacterial and viral contaminants to ensure that public health is protected. The Authority retains the right to deny seed collection and culture in any area, or to require additional testing for deleterious substances, or to require longer periods to purge contaminants as necessary. The original intent of this section was to provide for purging of viral and bacterial contamination prior to harvest for consumption on the assumption that deleterious substances were at acceptable levels prior to moving the seed to grow out areas The six-month requirement was implemented as a short-hand way to ensure that seed were grown for at least one month when water temperatures exceeded 60 degrees F. It makes little sense to require relay times in excess of one month for seed that are typically more than six months from harvest size when shellstock relay times as short as two weeks are common. References Cited: Richards, G. (1988), Microbial Purification of Shellfish: A Review of Depuration

	and Relaying, J. Food Protection 51(3)218-251. Supporting Information: RI DOH metals data (oyster seed grown in Billington Cove Marina) Unpublished data from Rd. Dale Leavitt (clam seed grown in Warwick Cove Marina)
Cost Information	This change should facilitate record keeping and documentation efforts required to ensure that seed from prohibited waters do not get harvested until bacterial and viral contamination has been purged.
Action by 2013 Task Force I	Recommended referral of Proposal 13-107 to an appropriate committee as determined by the Conference Chairman.
Action by 2013 General Assembly	Adopted recommendation of 2013 Task Force I on Proposal 13-107.
Action by FDA May 5, 2014	Concurred with Conference action on Proposal 13-107.
Action by 2015 Aquaculture Facility Inspection Committee	Recommended the following: (1) Referral of Proposal 13-107 back to Committee as appointed by the Conference Chair. (2) The charge of the Committee be expanded to include updating and revising the Aquaculture Chapter of the Model Ordinance to reflect current practices and methods and submit proposals for the next Annual Meeting.
Action by 2015 Task Force I	Recommended adoption of Aquaculture Facility Inspection Committee recommendations on Proposal 13-107.
Action by 2015 General Assembly	Adopted recommendation of Task Force I on Proposal 13-107.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 13-107.
Action by 2017 Aquaculture Facilities Inspection Committee	Recommended adoption of Proposal 13-107 as substituted. Section I. Definitions Replace definition 9. in Section I of the Model Ordinance as follows: 9. Aquaculture means cultivating shellfish in controlled conditions for human consumption. Cultivation includes propagation and growing of shellfish. These activities may occur in natural or man-made water bodies. These activities include seed production, cultivation in natural water bodies when shellfish are held off the bottom such as the use of racks, bags, or cages, and when shellfish are held in man-made water bodies such as the use of tanks, ponds, or raceways. These activities do not include depuration, wet storage or the broadcasting of spat or seed shellfish being left to mature the same as wild shellfish. Modify definition 93. in Section I of the Model Ordinance as follows: (93) Prohibited means a classification used to identify a growing area where the harvest of shellstock for any purpose, except depletion or gathering or nursery culture of seed for aquaculture, is not permitted. Section IV. Chapter IV. Shellstock Growing Areas Change @03 E. (2)(a) to read: (2) General. The Authority shall:

(a) Not permit the harvest of shellstock from any area classified as prohibited, except for the harvest of shellstock for the gathering of seed or nursery culture for aquaculture or the depletion of the areas classified as prohibited; and

Replace Chapter VI. Aquaculture in its entirety as follows:

Chapter VI. Aquaculture
Requirements for the Authority

[Note: The Authority must meet the requirements of this section even if the Authority does not formally adopt this section in regulation.]
@ .01 General.

A. Activities which have been determined to pose a significant public health concern and need regulation outlined in this Chapter include, but are not limited to:
(1) Seed production in waters classified as Prohibited or Unclassified;
(2) Aquaculture that attracts birds or mammals; and
(3) Land based aquaculture
B. The Authority shall:
(1) Approve the written operational plan for operations as outlined in @.01A above.
(2) Inspect operations outlined in @.01A above at least annually; and
(3) At a minimum inspect operator records to verify that appropriate permits are up to date and operational plans required in @ .01 A(1). are being implemented.
(4) Consistent with Chapter IV @ .01 (D)(1)(e) when aquaculture as defined in the Model Ordinance attracts birds or mammals their presence should be considered for possible adverse effects on growing area water quality

@ .02 Seed Shellstock.

A. The Authority shall establish the maximum seed size for each species of shellfish that can be produced in prohibited waters. In determining the maximum seed size Authorities shall establish sizes that require a minimum of 120 days of growing to reach market size.
B. The Authority shall establish appropriate corrective actions for when seed exceeds the maximum seed size when it has been produced in waters classified as prohibited.
C. All sources of seed produced or collected in prohibited waters shall be sanctioned by the Authority.

Requirements for the Harvester/Dealer

.01 Exceptions.

Hatcheries and nurseries rearing larvae and/or seed that are located in:
A. Approved or conditionally approved growing areas are exempt from these requirements.
B. Restricted or Conditionally Restricted would be exempt from these requirements but subject to relay requirements in Chapter V for seed that exceeds

the maximum seed size established by the Authority.

.02 General.

A. Any person who performs aquaculture as defined in the Model Ordinance or operates an aquaculture facility to raise shellfish for human consumption shall obtain:

(1) A permit from the Authority for the activity and functioning of his facility;

(2) A harvester's license; and

(3) Certification as a dealer, where necessary.

B. Shellfish aquaculture as defined in the Model Ordinance shall be practiced only in strict compliance with the provisions of the permit issued by the Authority for the aquaculture activity. Authorization shall be based on the operator's written operational plan.

C. Prior to beginning his activity, an operator shall obtain the permission of the Authority for use of his facility.

D. Any shellfish seed raised in aquaculture that exceeds the maximum seed size established by the Authority shall be subjected to relaying or depuration prior to direct marketing if the culture area or facility is located in or using water which is in:

(1) The closed status of the conditionally approved classification;

(2) The restricted classification;

(3) The open status of the conditionally restricted classification; or

E. Only drugs sanctioned by the FDA shall be used for shellfish treatment.

F. Harvesting, processing, storage, and shipping requirements for shellfish raised in a land-based aquaculture facility or a seed rearing facility or system that exceeds the maximum seed size established by the Authority shall be the same as the requirements for shellfish specified in Chapters V., VII., VIII., IX., X., XI., XII., XIII. and XIV.

G. Complete and accurate records shall be maintained for at least two (2) years by the operator of the aquaculture facility and shall include the:

(1) Source of shellfish, including seed if the seed is from growing areas which are not in the approved or conditionally approved classification;

(2) Water source, its treatment method, if necessary, and its quality in land based systems.

.03 Seed Production in Water Classified as Prohibited or Unclassified.

Seed may come from any growing area, or from any growing area in any classification, provided that:

A. The source of the seed if from waters classified as prohibited or unclassified is sanctioned by the Authority; and

B. Operational Plan. Each aquaculture site that cultures seed in waters classified as prohibited or unclassified shall have a written operational plan. The plan shall be approved by the Authority prior to its implementation and shall include:

(1) A description of the design and activities of the culture facility;

(2) The specific site and boundaries in which shellfish aquaculture activities will be conducted;

(3) The types and locations of any structures, including rafts, pens, cages,

nets, or floats which will be placed in the waters;
(4) The species of shellfish to be cultured and harvested;
(5) Procedures to assure that no poisonous or deleterious substances are introduced from the seed production activities;
(6) Corrective actions for addressing seed exceeding the maximum seed size as defined by the Authority.

.04 Aquaculture that attracts birds or mammals.

A. Operational Plan. Each aquaculture site that the Authority determines may attract sufficient birds and/or mammals that their waste presents a human health risk shall have a written operational plan. The plan shall be approved by the Authority prior to its implementation and shall include:
(1) A description of the design and activities of the culture facility;
(2) The specific site and boundaries in which shellfish aquaculture activities will be conducted;
(3) The types and locations of any structures, including rafts, pens, cages, nets, or floats which will be placed in the waters;
(4) The species of shellfish to be cultured and harvested;
(5) Procedures to assure that no poisonous or deleterious substances are introduced from the aquaculture activities;
(6) Maintenance of the required records

.05 Land Based Aquaculture.

A. Operational Plan. Each facility shall have a written operational plan. The facility must obtain approval from the Authority prior to its implementation and shall include:
(1) A description of the design and activities of the culture facility;
(2) The specific site and boundaries in which shellfish culture activities will be conducted;
(3) The types and locations of any structures, including rafts, pens, cages, nets, tanks, ponds, or floats which will be placed in the waters;
(4) The species of shellfish to be cultured and harvested;
(5) Procedures to assure that no poisonous or deleterious substances are introduced into the activities;
(6) A program of sanitation, maintenance, and supervision to prevent contamination of the shellfish products;
(7) A description of the water source, including the details of any water treatment process or method;
(8) A program to maintain water quality, which includes collection of microbial water samples and their method of analysis and routine temperature and salinity monitoring. The bacterial indicator monitored shall be the same as used for monitoring growing areas;
(9) If applicable, collection of data concerning the quality of food production (algae or other) used in the artificial harvest system; and
(10) Maintenance of the required records.

B. Each land-based facility conducting aquaculture as defined by the Model Ordinance shall maintain the following records while the aquaculture activity continues.
(1) Construction and remodeling plans for any permitted aquaculture

facility;
(2) Aquaculture operational plans; and
(3) Aquaculture permits.

C. Water Systems.
(1) If the land-based aquaculture system is of continuous flow through design, water from a growing area classified as approved, or in the open status of the conditionally approved classification at all times shellfish are held, may be used without treatment.

D. Water Quality.
(1) Shellstock cultured in a closed or recirculating system that exceeds the maximum seed size shall meet the requirements for water quality and testing in Chapter VII C. .04 (3) (a), (b), (c), and (d) may be used in direct marketing.
(2) Shellstock cultured in a closed or recirculating system that exceeds the maximum seed size and does not meet the requirements of Section D. (1) shall be relayed or depurated consistent with Chapter IV prior to direct marketing.

.06 Polyculture Systems.

A polyculture system shall:

A. Meet all requirements in Section .05 Land Based Systems;
B. Provide information concerning all sources of and species of all organisms to be cultivated, cultured, and harvested;
C. Include in its operational plan requirements to:
(1) Monitor for human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances that might be associated with polyculture activities; and
(2) Subject all harvested shellstock to relaying or depuration if human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances exist at levels of public health significance.

Move Chapter VI Section .07 to a new Chapter:

Chapter XVII Shellfish Gardening

@ .01 Shellfish Gardening.

If a State recognizes shellfish gardening the Authority:
A. Shall permit or register shellfish gardening activities.
B. Shall establish permit or registration conditions and determine classification of waters where shellfish gardening can take place prior to its implementation.
C. Shall provide information to the shellfish gardener on the risk of consuming shellfish from private docks, piers, and shellfish floats attached to piers or docks and from waters not classified and open to harvest for direct consumption.
D. May require that the shellfish gardener maintain records on the disposition of the shellfish product and provide these records to the Authority.

	@ . 02 Requirements for the Shellfish Gardener. A. Shellfish gardening shall be practiced only in strict compliance with the provisions of the permit issued by the Authority for the oyster/shellfish gardening activity. B. Shellfish gardeners shall document that they understand the risks associated with consumption for shellfish grown from docks or private piers. C. If required by the Authority, shellfish gardeners shall keep accurate records on the fate or final destination of all shellfish grown at their shellfish garden site and provide these records to the Authority upon request.
Action by 2017 Task Force I	Recommended adoption of Aquaculture Committee recommendation on Proposal 13-107 as amended. Section I. Definitions Replace definition 9. in Section I of the Model Ordinance as follows: 9. Aquaculture means cultivating shellfish in controlled conditions for human consumption. Cultivation includes propagation and growing of shellfish. These activities may occur in natural or man-made water bodies. These activities include seed collection, production, cultivation in natural water bodies when shellfish are held off the bottom such as the use of racks, bags, or cages, and when shellfish are held in man-made water bodies such as the use of tanks, ponds, or raceways. These activities do not include depuration or, wet storage. ~~or the broadcasting of spat or seed shellfish being left to mature the same as wild shellfish.~~ Modify definition 93. in Section I of the Model Ordinance as follows: (93) Prohibited means a classification used to identify a growing area where the harvest of shellstock for any purpose, except depletion or gathering or nursery culture of seed for aquaculture, is not permitted. Section IV. Chapter IV. Shellstock Growing Areas Change @03 E. (2)(a) to read: (2) General. The Authority shall: (a) Not permit the harvest of shellstock from any area classified as prohibited, except for the harvest of shellstock for the gathering of seed or nursery culture for aquaculture or the depletion of the areas classified as prohibited; and Replace Chapter VI. Aquaculture in its entirety as follows: Change @03 E. (2)(a) to read: (2) General. The Authority shall: (a) Not permit the harvest of shellstock from any area classified as prohibited, except for ~~the harvest of shellstock for~~ the gathering of seed or nursery culture for aquaculture or the depletion of the areas classified as prohibited; and Chapter VI. Aquaculture Requirements for the Authority [Note: The Authority must meet the requirements of this section even if the

Authority does not formally adopt this section in regulation.]

@ .01 General.

A. Aquaculture ~~A~~activities which may~~have been determined to~~ pose a significant public health concern and are regulated~~need regulation outlined~~ in this Chapter include, but are not limited to:

(1) Seed production in waters classified as Prohibited or Unclassified;

(2) Aquaculture structures that attracts birds or mammals; and

(3) Land based aquaculture

B. The Authority shall:

(1) Approve the written operational plan for operations as outlined in @.01A above.

(2) Inspect operations outlined in @.01A above at least annually; and

(3) At a minimum inspect operator records to verify that appropriate permits are up to date and operational plans required in @ .01 A(1). are being implemented.

(4) Consistent with Chapter IV @ .01 (D)(1)(e) when aquaculture as defined in the Model Ordinance attracts birds or mammals their presence should be considered for possible adverse effects on growing area water quality

@ .02 Seed Shellstock.

A. The Authority shall establish the maximum seed size for each species of shellfish that can be produced in prohibited waters. In determining the maximum seed size Authorities shall establish sizes that require a minimum of 120 days of growing to reach market size.

B. The Authority shall establish appropriate corrective actions for when seed exceeds the maximum seed size when it has been produced in waters classified as prohibited.

C. All sources of seed produced or collected in prohibited waters shall be sanctioned by the Authority.

Requirements for the Harvester/Dealer

.1 Exceptions.

Hatcheries and nurseries rearing larvae and/or seed that are located in:

A. Approved or conditionally approved growing areas are exempt from these requirements.

B. Restricted or Conditionally Restricted would be exempt from these requirements but subject to relay requirements in Chapter V for seed that exceeds the maximum seed size established by the Authority.

.2 General.

A. Any person who performs aquaculture as defined in the Model Ordinance or operates an aquaculture facility to raise shellfish for human consumption shall obtain:

(1) A permit from the Authority for the activity and functioning of his facility;

(2) A harvester's license; and

(3) Certification as a dealer, where necessary.

B. Shellfish aquaculture as defined in the Model Ordinance shall be practiced only in strict compliance with the provisions of the permit issued by the Authority for the aquaculture activity. Authorization shall be based on the operator's written operational plan.

C. Prior to beginning his activity, an operator shall obtain the permission of

	the Authority for use of his facility. D. Any shellfish seed raised in aquaculture that exceeds the maximum seed size established by the Authority shall be subjected to relaying or depuration prior to direct marketing if the culture area or facility is located in or using water which is in: (1) The closed status of the conditionally approved classification; (2) The restricted classification; (3) The open status of the conditionally restricted classification; or E. Only drugs sanctioned by the FDA shall be used for shellfish treatment. F. Harvesting, processing, storage, and shipping requirements for shellfish raised in a land-based aquaculture facility or a seed rearing facility or system that exceeds the maximum seed size established by the Authority shall be the same as the requirements for shellfish specified in Chapters V., VII., VIII., IX., X., XI., XII., XIII. and XIV. G. Complete and accurate records shall be maintained for at least two (2) years by the operator of the aquaculture facility and shall include the: (1) Source of shellfish, including seed if the seed is from growing areas which are not in the approved or conditionally approved classification; (2) Water source, its treatment method, if necessary, and its quality in land based systems. .3 Seed Production in Water Classified as Prohibited or Unclassified. Seed may come from any growing area, or from any growing area in any classification, provided that: A. The source of the seed if from waters classified as prohibited or unclassified is sanctioned by the Authority; and B. Operational Plan. Each aquaculture site that cultures seed in waters classified as prohibited or unclassified shall have a written operational plan. The plan shall be approved by the Authority prior to its implementation and shall include: (1) A description of the design and activities of the culture facility; (2) The specific site and boundaries in which shellfish aquaculture activities will be conducted; (3) The types and locations of any structures, including rafts, pens, cages, nets, or floats which will be placed in the waters; (4) The species of shellfish to be cultured and harvested; (5) Procedures to assure that no poisonous or deleterious substances are introduced from the seed production activities; (6) Corrective actions for addressing seed exceeding the maximum seed size as defined by the Authority. .4 Aquaculture that attracts birds or mammals. A. Operational Plan. Each aquaculture site that the Authority determines may attract sufficient birds and/or mammals that their waste presents a human health risk shall have a written operational plan. The plan shall be approved by the Authority prior to its implementation and shall include: (1) A description of the design and activities of the culture facility; (2) The specific site and boundaries in which shellfish aquaculture activities will be conducted; (3) The types and locations of any structures, including rafts, pens,

cages, nets, or floats which will be placed in the waters;
(4) The species of shellfish to be cultured and harvested;
(5) Procedures to assure that no poisonous or deleterious substances are introduced from the aquaculture activities;
(6) Maintenance of the required records

.5 Land Based Aquaculture.

A. Operational Plan. Each facility shall have a written operational plan. The facility must obtain approval from the Authority prior to its implementation and shall include:
(1) A description of the design and activities of the culture facility;
(2) The specific site and boundaries in which shellfish culture activities will be conducted;
(3) The types and locations of any structures, including rafts, pens, cages, nets, tanks, ponds, or floats which will be placed in the waters;
(4) The species of shellfish to be cultured and harvested;
(5) Procedures to assure that no poisonous or deleterious substances are introduced into the activities;
(6) A program of sanitation, maintenance, and supervision to prevent contamination of the shellfish products;
(7) A description of the water source, including the details of any water treatment process or method;
(8) A program to maintain water quality, which includes collection of microbial water samples and their method of analysis and routine temperature and salinity monitoring. The bacterial indicator monitored shall be the same as used for monitoring growing areas;
(9) If applicable, collection of data concerning the quality of food production (algae or other) used in the artificial harvest system; and
(10) Maintenance of the required records.

B. Each land-based facility conducting aquaculture as defined by the Model Ordinance shall maintain the following records while the aquaculture activity continues.
(1) Construction and remodeling plans for any permitted aquaculture facility;
(2) Aquaculture operational plans; and
(3) Aquaculture permits.

C. Water Systems.
(1) If the land-based aquaculture system is of continuous flow through design, water from a growing area classified as approved, or in the open status of the conditionally approved classification at all times shellfish are held, may be used without treatment.

D. Water Quality.
(1) Shellstock cultured in a closed or recirculating system that exceeds the maximum seed size shall meet the requirements for water quality and testing in Chapter VII C. .04 (3) (a), (b), (c), and (d) may be used in direct marketing.
(2) Shellstock cultured in a closed or recirculating system that exceeds the maximum seed size and does not meet the

requirements of Section D. (1) shall be relayed or depurated consistent with Chapter IV prior to direct marketing.

.6 Polyculture Systems.

A polyculture system shall:

A. Meet all requirements in Section .05 Land Based Systems;
B. Provide information concerning all sources of and species of all organisms to be cultivated, cultured, and harvested;
C. Include in its operational plan requirements to:
   (1) Monitor for human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances that might be associated with polyculture activities; and
   (2) Subject all harvested shellstock to relaying or depuration if human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances exist at levels of public health significance.

Move Chapter VI Section .07 to a new Chapter:

Chapter XVII Shellfish Gardening

@ .01 Shellfish Gardening.

If a State recognizes shellfish gardening the Authority:
A. Shall permit or register shellfish gardening activities.
B. Shall establish permit or registration conditions and determine classification of waters where shellfish gardening can take place prior to its implementation.
C. Shall provide information to the shellfish gardener on the risk of consuming shellfish from private docks, piers, and shellfish floats attached to piers or docks and from waters not classified and open to harvest for direct consumption.
D. May require that the shellfish gardener maintain records on the disposition of the shellfish product and provide these records to the Authority.

@ . 02 Requirements for the Shellfish Gardener.

A. Shellfish gardening shall be practiced only in strict compliance with the provisions of the permit issued by the Authority for the oyster/shellfish gardening activity.
B. Shellfish gardeners shall document that they understand the risks associated with consumption for shellfish grown from docks or private piers.
C. If required by the Authority, shellfish gardeners shall keep accurate records on the fate or final destination of all shellfish grown at their shellfish garden site and provide these records to the Authority upon request.

Recommends a committee be appointed by the Conference Chair to review and revise existing guidance documents related to the Aquaculture Chapter.

Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-107.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 13-107.
Action by 2019 Aquaculture Committee	In 2017 the Conference adopted the new language of Proosal 13-107 to modify the requirements of Chapter VI. The Conference further directed the development of guidance for Chapter VI. The Aquaculture Committee was charged with the development of a Guidance Document. That work was not completed. The Chapter VI language that was adopted in 2017 is not included in the 2019 Task Force II report. The Aquaculture Committee recommended referral of the Guidance Document request included in Proposal 13-107 to an appropriate committee as determined by the Conference Chairperson with further instruction that the committee be convened before the Spring Executive Board meeting to begin development of a guidance document for the revised Aquaculture Chapter.
Action by 2019 Task Force I	Recommended adoption of the Aquaculture Committee recommendation on Proposal 13-107.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 13-107.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 13-107.
Action by 2023 Aquaculture Committee	Recommended: 1. Adoption of revised Chapter VI. Guidance language Section IV Guidance Documents – Chapter VI. Shellfish Aquaculture or Section III Public Health Reasons and Explanations NSSP guidance documents provide the public health principles supporting major components of the NSSP and its Model Ordinance, which includes the requirements of the program. For the most up to date and detailed listing of requirements, the reader should consult the most recent edition of the Model Ordinance. **Introduction** This chapter provides guidance on NSSP standards intended to address human health hazards specifically associated with molluscan shellfish aquaculture activities covered under Chapter VI. of the NSSP Model Ordinance requirements. Additional information concerning the disease-causing potential of molluscan shellfish can be found in the NSSP Model Ordinance Guidance Documents: *Guidance for Developing Marine Biotoxin Contingency Plan, Sanitary Survey and the Classification of Growing Waters*, and *Shellstock Relay*. For the purposes of the NSSP Model Ordinance, Aquaculture is defined as the cultivation of bivalve shellfish in controlled conditions for human consumption. This includes cultivation of molluscan shellfish in natural water bodies or man-made systems. Aquaculture can also include the cultivation of molluscan shellfish with non-molluscan species in a common aquaculture system known as polyculture. Bivalve shellfish raised in open water aquaculture operations are generally subject to the same potential for contamination as naturally occurring bivalve shellfish populations. As a result, there is substantial overlap in the sanitary controls within the NSSP Model Ordinance for bivalve shellfish harvested from aquaculture operations

and those harvested from naturally occurring populations. There are potential human health concerns specific to land-based or recirculating aquaculture that may require the implementation of operation specific management measures. Activities such as relaying, wet storage, depuration, growing area classification and tagging, are regulated under their respective NSSP Model Ordinance chapters. Aquaculture activities regulated underChapter VI. of the NSSP Model Ordinance are those unique to aquaculture operations and have the potential to pose a significant public health concern if not properly managed. As outlined in Chapter VI @.01A, these include, but are not limited to:

(1) Natural seed collection and/or the rearing of larvae and seed shellfish in growing areas and/or hatcheries and nurseries in, or using, waters classified as Prohibited or Unclassified;

(2)Aquaculture activities that include off-bottom structures that may attract bird and/or mammal congregations to the extent that their waste may present a human health risk; and,

(3) Land-based aquaculture operations and/or Poly Culture.

Hatcheries and Nurseries- Exemptions and Exceptions to Chapter VI

Chapter VI. makes certain exemptions and exceptions for hatcheries and nurseries rearing larvae and/or seed that are located in, or draw water from, growing areas in the Approved or Conditionally Approved classifications. Hatcheries and nurseries rearing larvae and/or seed that are located in, or draw water from, growing areas in the the Restricted or Conditionally Restricted classification, are also exempt from these requirements if seed does not exceed the maximum seed size established by the Authority under Chapter VI @ .02 (A) or if they adhere to the relay requirements in Chapter V for seed that exceeds the maximum seed size established by the Authority per Chapter VI @ .02 (A).

Requirements for the Authority

To meet the requirements for shellfish aquaculture in Chapter VI, the Authority must have an adequate legal basis, and established procedures, to regulate aquaculture activities outlined in Chapter VI @.01A that occur within their jurisdiction. At a minimum, this includes oversight over the issuance of permits, the [1]review and approval of operational plans for any operations conducting activities in Chapter VI @.01A., and the ability to inspect such operations at least annually to verify that appropriate permits are up to date and operational plans are being implemented. It may also be necessary, based on the aquaculture operations practiced in a jurisdiction, for the Authority to impose additional control measures or recordkeeping requirements upon aquaculture practitioners in the form of regulation, policies, and/or enforceable permit conditions or operational plans Discussion of additional Authority imposed control measures and associated responsibilities are found under their respective subheading.

Requirements for the Harvester/Dealer (Aquaculture Operator)

It is the responsibility of the operator of an aquaculture facility to verify compliance with NSSP MO requirements, and associated local rules and regulations, and to obtain the permission of the Authority prior to conducting any of the aquaculture activities outlined in Chapter VI. The operator of an aquaculture facility may also be

[1]

required to conduct record keeping and implement control measures as outlined in regulation, permit conditions, and/or their operational plan as necessary based on individual aquaculture practices and the requirements of the Authority. It is important to note that in many states the Authority does not require formal operational plans, rather the required elements of operational plans listed below are included in permit application materials and as regulations and/or enforceable permit conditions. Discussion of additional harvester control measures and responsibilities are found under their respective subheading.

Seed Production in Water Classified as Prohibited or Unclassified

When adequate controls are implemented, natural seed collection and/or the rearing of larvae and seed shellstock in growing areas and/or hatcheries and nurseries located in, or using, waters classified as prohibited or unclassified, provides aquaculturists the opportunity to access shellstock resources or utilize areas or waters for seed production that would otherwise not be available for the production of shellstock intended for direct human consumption. Often areas that are unclassified or classified as prohibited due to real or potential pollution (such as marinas, boat yards, etc.) are ideal locations for hatchery or nursery operations due to their proximity to physical infrastructure (docks and piers, freshwater, electricity) and other factors (i.e. protection to wave action, ease of access, security, etc.) important to hatchery and nursery production.

The harvesting of shellstock from unclassified areas or areas in the prohibited classification is not allowed for any purpose, except depletion, gathering of seed or hatchery and nursery production. The use of prohibited or unclassified waters for the gathering of natural seed and/or hatchery and nursery production is acceptable because these operations do not produce shellstock for direct consumption; rather, the seed produced/gathered is moved to Restricted, Conditionally Restricted, or Approved areas in for grow-out prior to harvest for consumption. Research has shown that shellstock has the ability to purge itself of microbial pathogens and certain chemical contaminants over time when moved to clean saline water. In addition, limited exposure during early life stages to lipophilic or other contaminants that cannot be easily purged from shellstock does not constitute a public health hazard if the shellstock are moved to clean waters while these contaminants still represent a small constituent of the total shellstock tissue mass. As a result, seed from prohibited or unclassified areas does not pose a risk to public health provided the Authority ensures they are relocated to suitable waters and provided adequate time for the reduction of contaminants and growth prior to harvest for consumption. *For more information see Section IV Guidance Documents – Chapter II. Growing Areas.*

Maximum Seed Size

Section II Chapter VI @ .02 requires the Authority to sanction (permit) all sources of seed produced or collected in unclassified or prohibited waters, and to establish a maximum seed size for each species of shellfish that are produced in unclassified or prohibited waters. The Authority must set the maximum seed size to ensure a minimum of 120 days of growing to reach market size following movement from unclassified or prohibited waters to waters in other classifications. This period of growth is intended to ensure any potential contaminants accumulated in seed shellstock tissues while being reared in unclassified or prohibited waters will represent a small constituent of the total tissue mass at harvest. 120 days also provides sufficient time for the purging of any bacterial or viral pathogens.

A maximum seed size may be established via regulation, enforceable permit conditions, or within an individual aquaculture operations enforceable operational

plan. To determine the appropriate maximum seed size for each species, the Authority may choose to rely on existing locally appropriate data or conduct species specific studies. Growth rates vary across and within regions and can be influenced by a number of environmental factors (i.e. temperature, food availability and quality), genetics (i.e. triploid vs. diploid), and culture practices (i.e. stocking density, on-bottom vs off-bottom). It is also common to see differential growth rates between individual shellfish within a single nursery system. Some hatchery and nursery activities are considered self-limiting with regards to the size of shellstock they can support (i.e. spat on shell, etc.). In such systems, shellstock are likely to be moved to clean waters and remain there for far longer than 120 days prior to harvest. For wild seed collection and other types of nursery activities (upwellers, floating nursery bags, etc.), operators may wait to move shellstock to clean waters until they are close to the maximum seed size. In these cases operators must closely monitor growth rates to ensure shellstock does not exceed the maximum seed size and trigger the need for corrective actions.

The NSSP MO requires the Authority and operator to establish appropriate corrective actions, as required in Chapter VI .03 (B), for when seed that has been produced in waters classified as prohibited or unclassified exceeds the maximum size. With few exceptions, the seed will generally need to be destroyed or moved to a restoration site sanctioned by the Authority. It is critical that the Authority and aquaculture operators work together to ensure the establishment of a maximum seed size that is consistent with production practices and local environmental conditions, and ensures the minimum 120 days prior to harvest to prevent unnecessary loss of shellstock. Corrective actions may be established via regulation, enforceable permit conditions, or within an individual aquaculture operation's enforceable operational plan. If corrective actions are required, it is recommended that the operation and/or Authority adjust practices and/or reevaluate permit conditions and/or the operational plan to prevent further violation of maximum seed size requirements.

An important factor in determining the maximum seed size is if the Authority has established a market or legal harvest size for each species produced in waters classified as prohibited or unclassified. In states where a minimum enforceable market (AKA harvest) size is in place, it may be possible to establish a relatively larger maximum seed size and have sufficient confidence, and a legal basis, to ensure seed shellstock originating from waters classified as prohibited or unclassified will not be harvested prior to the required 120 days, without requiring additional record keeping, segregation, or other measures. In cases where a state does not have an established minimum market size, and are relying on long established market standards to base the determination of an appropriate maximum seed size, it is likely a conservative maximum seed size, and/or additional measures such as record keeping, segregation, or other measures will be required as an enforceable permit condition or enforceable element of an operational plan to provide verifiable compliance with the 120 day requirement. Alternatively, the Authority may allow an operator to adopt a minimum harvest size as an element of their enforceable operational plan and possibly forgo or reduce the need for record keeping, segregation, or other measures.

Operational Plan

The NSSP MO Section II Chapter VI .03 requires aquaculture operations that collect or culture seed in waters classified as prohibited or unclassified develop a written operational plan and receive approval by the Authority prior to its implementation; such a plan shall at a minimum include:

(1) A description of the design and activities of the culture facility;

(2)The specific site and boundaries in which shellfish aquaculture activities will be conducted;

(3)The types and locations of any structures, including rafts, pens, cages, nets, or floats which

will be placed in the waters;

(4)The species of shellfish to be cultured and harvested;

(5) Procedures to assure that no poisonous or deleterious substances are introduced from the seed production activities; and,

(6)Corrective actions for addressing seed exceeding the maximum seed size as defined by the
Authority.

If the information for items #1-4 is provided in permit application materials or as a condition on permits, these may be substituted for inclusion in a formal operational plan. Item #5 is often codified in state regulation, and adherence is agreed upon by the operator when signing their permit(s). In other cases, written operational plans containing elements, or the entirety, of the information required in #1-6 may be used to supplement other documentation provided by the permit holder or applicant to satisfy this requirement. In some instances additional information, such as an operator/Authority agreed upon minimum harvest size, segregation and record keeping protocols for shellstock relocated from prohibited areas, or other elements specific to managing human health risks associated with individual operations and as required by the Authority must be submitted. Any form of enforceable written record of the required information in #1-6, and agreed upon by the Authority, is sufficient to meet the intent of Chapter VI @ .03.

Facility Inspection

If an operation plan is determined to be required for an aquaculture site, the authority must inspect the operation at least annually. The inspection is intended to ensure the operation is adhering to the operational plan, and verify that appropriate permits and any reporting, if required, are up to date.

Aquaculture activities that include off-bottom structures that may attract bird and/or mammal congregations to the extent that their waste may present a human health risk

Microbial contamination from nonpoint pollution sources such as wildlife waste in growing areas represents a public health risk. Wildlife such as birds and/or mammals have been documented to host Campylobacter spp., Salmonella spp., Listeria, E. coli, Vibrio cholerae, Aeromonas spp., Enterococcus spp., and other zoonotic enteric viruses and bacteria within their digestive tract and feces. A number of these pathogens have a low infectious dose, and have the potential for survival and growth during harvest, processing, transportation and storage (Stelma et at. 1991). A detailed summary of zoonotic pathogens of concern to shellfish sanitation is provided in Stelma et at. 1991. While human enteric pathogens can be isolated in the intestinal tracts of a number of species of birds and/or mammals that inhabit coastal and marine

waters, the level of risk to shellfish consumers from wildlife waste is not fully understood; however, it is believed to be less than that related to human sources (Stelma 1991). This largely because for pathogens introduced from wildlife waste to result in human infections they must be a strain that is pathogenic to humans and must be ingested at an infectious dose (Smith et al. 2021). The vast majority of enteric pathogen strains isolated from wildlife waste and growing area waters subject to nonpoint wildlife derived pollution have not been associated with reported human infections, and the majority likely do not have the ability to cause illness in humans (Smith et al. 2021; Stelma, 1991).

The use of floating and off-bottom gear, mainly for oyster culture, has increased in recent years due to the benefits these methods provide aquaculturists to avoid sensitive benthic habitats, and for ease of handling, maintenance, and improved growth rates and survival. However, floating and exposed off-bottom aquaculture gear can provide a roosting platform for various types of birds and/or mammals and become a feeding and defecating site, when these congregations reach sufficient numbers they can present public health concerns.

Increased fecal coliform loading due to congregations of birds and mammals on or around aquaculture structures may result in degradation of water quality to the extent that growing areas no longer meet NSSP criteria outlined in Chapter IV, resulting in growing areas closures, a downgrade in water quality, or potentially a recall of harvested products. Waste associated with congregations of birds and/or mammals on floating and exposed off-bottom aquaculture gear has recently been associated with increased fecal coliform levels in shellfish growing areas and shellfish meats in New York, in some cases requiring growing area closures, and sampling of growing areas and oysters held in floating aquaculture gear prior to reopening of affected areas and farms (NYSDEC). Such actions have had significant adverse impacts on aquaculture operators and highlighted the need to identify potential water quality impacts associated with congregations of birds and mammals on or around aquaculture structures prior to them reaching the level of public health concern.

In addition to concerns associated with water quality degradation, shellstock held in or near structures that serve as a roosting platform for various types of birds and/or mammals may accumulate bird or mammal fecal matter that could serve as a vector for human infections when shellfish are consumed. In the U.S. reports of outbreaks and sporadic infections linked to wildlife contamination of molluscan shellfish are rare, but have been documented. In October, 2021, an investigation indicated that eight people became ill after consuming raw oysters harvested from a small coastal pond in Rhode Island. The illnesses were associated with *Campylobacter jejuni* bacterial contamination linked to the presence of flocks of birds congregating on floating aquaculture gear (RIDOH).

The recent incidence of shellfish derived human infections and water quality issues associated with bird congregations on floating and off bottom gear has prompted management measures focused on mitigating human health concerns related to wildlife congregations on aquaculture sites. Under the growing area classification responsibilities at Chapter VI. *Shellstock Growing Areas* the Authority is required to consider the presence of wild animals or resident and migrating bird populations for possible adverse effects on growing areas, and to identify and evaluate all actual or potential sources of pollution which may affect the growing area during routine water quality sampling, sanitary surveys, triennial, and annual evaluations. Under aquaculture specific provisions in Chapter VI.@04, the Authority is required to evaluate aquaculture sites to determine if the aquaculture operation and the associated culture gear may attract sufficient numbers of birds and/or mammals to

the extent that their waste presents a human health risk. If the Authority determines a human health risk may exist or develop, the Authority must require the operator to submit a written operational plan, including mitigation or deterrent measures to minimize the potential pollution impact of birds and/or mammals, to the Authority for approval prior to its implementation. The two separate, yet interrelated, requirements provide a means for the Authority to evaluate risk associated with proposed aquaculture operations and, if necessary, institute deterrent or mitigation measure before they are approved, and a means to evaluate risk associated with existing aquaculture sites on a routine basis via observations and results from water quality sampling, sanitary surveys, triennial, and annual evaluations.

Risk Determination of Aquaculture Operations

Any aquaculture operation utilizing floating gear or other structures that may serve as a roosting or resting platform for birds or mammals (e.g. work floats, pilings, etc.) has the potential to attract bird and mammal congregations. However, the presence of wildlife, or their waste, on aquaculture gear alone is generally not sufficient to determine if a human health risk may be present. Positioning sampling stations in proximity to aquaculture sites provides a means to evaluate risk associated with existing operations (*See Chapter VI. Shellstock Growing Areas for more information on pollution source sampling)*[1] . Shellstock sampling from existing sites may also provide an indication of potential risk; however, it is important to note fecal coliform counts do not differentiate between human pathogenic and non-pathogenic strains of bacteria, and we currently do not have an estimate of the correlation of human enteric pathogens with coliforms in wildlife waste; although, the risk is considered to be less than that from human derived sources (Stelma, 1991; Smith et al. 2021). Further, there are no bacteriological standards for shellstock meats within the Model Ordinance so an understanding of background levels would likely be necessary to support interpretation of shellfish sampling results.

When evaluating proposed sites the Authority can consider a number of site related factors that may influence whether bird and/or mammal congregations on aquaculture gear may present a risk to human health. These factors include evaluating existing information on the seasonal or year round abundance, type, and behavior of wildlife (e.g. feeding, nesting, migration, etc.), within the growing area where a site is being proposed. An evaluation of site specific hydrodynamic information for the growing area where a site is proposed to be located can also help inform the potential level of risk. Factors such as stratification, tidal magnitude, water depth, current velocity, and wave action can influence the extent to which wildlife waste may become an issue. Areas with minimal currents or flushing may be more susceptible to water quality impacts from smaller congregations of wildlife than those with high current velocities and flushing. Sites proposed within proximity to other facilities that may attract birds and mammals could also increase the risk of gear to serve as roosting platforms for existing populations of birds or mammals in the area. Operation design is also a major consideration for determining if a proposed aquaculture operation may present a risk to human health. The type, extent, and density of exposed gear on the site can impact flushing around gear arrays, and either reduce or increase fecal loading associated with bird and/or mammal waste. Other operation specific practices can be adapted to reduce the potential for a human health concern to develop. For example, floating gear is often used during the nursery and intermediate stages of culture. In areas where the potential risk of human health concerns are high, shellstock may be able to be moved from floating or exposed gear to submerged gear or planted on bottom for a period of time prior to harvest. In addition, the implementation of proactive deterrent measures may provide the Authority with confidence that issues can be avoided before they reach a level of

human health concern.

The approach the Authority employs to meet the requirements of Chapter VI.04 will generally be based on the availability of resources to conduct required water quality sampling at existing aquaculture sites, the availability of resources and existing information needed to evaluate risks associated with proposed sites, and the Authority's confidence that bird and/or mammal congregations on aquaculture gear, and the resulting waste, may or may not present a human health risk based on their evaluation and observations. The information necessary to support an evaluation of risk for new and existing aquaculture operations may be derived from a number of sources such as growing area classification information, external sources, and/or information provided by the aquaculture operator within application materials or other reporting to the Authority. To the extent possible, aquaculture operators should detail to the Authority within their application materials, or other reporting, any site selection criteria or operational design specifics intended to minimize the potential pollution impact of birds and/or mammals they are proposing to proactively employ. This will help the Authority determine which of the following approaches to meet the requirements of Chapter VI.04 they will employ.

1. **Monitoring approach**- If the Authority determines that sufficient evidence does not exist to preemptively require new or existing aquaculture operators to adopt mitigation or deterrent measures, they may choose to continue to monitor the growing area in compliance with growing area classification requirements in Chapter IV. The monitoring should be conducted in a manner that would allow the Authority to identify and address potential human health concerns associated with bird and/or mammal congregations on aquaculture gear, prior to them reaching a level of public health significance. This strategy may require adjusting water quality sampling stations and sampling frequency around aquaculture operations, shellstock meat sampling, microbial source tracking or other forms of directed pathogen sampling, and/or other monitoring or reporting measures as appropriate. In these cases, the Authority and operators should consider the development of procedures to rapidly institute operational plans including deterrent and/or mitigation measures should a concern be identified. The Authority should document any bird and/or mammal congregations on aquaculture sites during aquaculture site inspections, routine water quality monitoring, annual and triennial reviews, and sanitary surveys, and consider adjusting sampling/monitoring frequency around any observed seasonal, or other, trends in wildlife activity.

2. **Preemptive approach**- If the Authority determines that sufficient evidence of a public health concern associated with the use of floating gear exists, or that insufficient resources exist to increase monitoring around new aquaculture operations, they may choose to preemptively require aquaculture operators to provide an operational plan and institute bird and/or mammal mitigation and/or deterrent measures. Alternatively, the Authority may implement industry-wide or operation specific mitigation (e.g. submergence requirements) and/or deterrent measures to minimize impacts from birds and/or mammals via regulation, enforceable permit conditions and/or policies. The Authority should continue to document any bird and/or mammal congregations on aquaculture sites during aquaculture site inspections, routine water quality monitoring, annual and triennial

reviews, and sanitary surveys, and monitor water quality within proximity to aquaculture facilities to evaluate efficacy of measure outlines within operation plans.

Operational Plan

Under Chapter VI.04, if the Authority determines that the aquaculture operation and the associated culture gear may attract sufficient numbers of birds and/or mammals to the extent that their waste presents a human health risk, the operator is required to enact mitigation measures as a component of an operational plan. The plan shall be approved by the Authority prior to its implementation and include:

1. A description of the design and activities of the culture facility;

2. The specific site(s) and boundaries in which the shellfish aquaculture activities will be conducted;

3. The types and locations of any structures, including rafts, pens, cages, nets, or floats which will be placed in the waters;

4. The species of shellfish to be cultured and harvested;

5. Procedures to assure that no poisonous or deleterious substances are introduced from the aquaculture activities;

6. A description of the mitigation or deterrent measures to minimize the potential pollution impact of birds and/or mammals; and

7. Maintenance of the required records.

If the information for items #1-4 & 6-7 is provided in permit application materials or on final permits, these may be substituted for inclusion in a formal operational plan. Likewise, #5 is often codified in state and/or federal regulation, and adherence is agreed upon by the operator when signing their permit(s) or by law. In other cases written operational plans containing elements, or the entirety, of the information required in #1-7 may be submitted. Any form of enforceable written record of these items is sufficient to meet the intent of Chapter VI @ .04. To meet the requirements of #6, if necessary, the written operational plan or application materials should clearly describe any operational, maintenance, handling and/or sanitary practices for the aquaculture gear and shellfish that will be conducted to prevent contamination of the growing area from waste attributed to congregations of birds and/or mammals on aquaculture structures. This may include a written description, sketches and/or photos of deterrents or mitigation measures to be used. Strategies may include a suite of deterrents (i.e. kites, ~~cannons~~, sprinklers, spikes etc.) or mitigation measures (e.g. submerging gear and shellfish prior to harvest, relocating floating gear to areas with significant flow, seasonal harvest restrictions, configuring farm sites to maximize flushing, etc.) that will address human health concerns related to year-round or seasonal congregations of birds and/or mammals. In addition, plans should address evaluation of the efficacy of deterrent and/or mitigation measures, and potential triggers that would require changing or adapting deterrent or mitigation measures to address new bird or mammal species and/or behavioral changes, and amendments should be made to the plan, as needed, based on changes to the culture operation, gear, and/or reduced efficacy of the approved deterrents and/or mitigation measures

employed by the aquaculture operator.

Facility Inspection

If an operation plan is determined to be required for an aquaculture site, the authority must inspect the operation at least annually. The inspection is intended to ensure the operation is adhering to the operational plan, verify that appropriate permits are up to date, and that control measures to prevent possible adverse public health effects from birds or mammals are effective. In addition, the Authority should continue to document any bird and/or mammal congregations on aquaculture sites during, routine water quality monitoring, sanitary surveys, triennial, and annual evaluations, and continue monitor water quality within proximity to aquaculture facilities to evaluate efficacy of mitigation and/or deterrent measure outlined within operation plans. The Authority should consider the development of written protocols associated with evaluating the effectiveness of the deterrents and/or mitigation strategies. If the Authority or Operator documents large congregations of birds and/or mammals on aquaculture gear, and/or an accumulation of fecal matter, an evaluation of the efficacy of current control measures may be necessary to determine if additional control measures are needed.

Polyculture and Land-based Aquaculture Considerations

Polyculture and land-based monoculture operations must be under adequate control to assure the shellstock product harvested will be acceptable for human consumption. The Authority must establish detailed procedures for issuing permits for shellfish aquaculture, approving culturing facilities and boundaries, controlling of harvesting, sampling of shellstock, monitoring environmental parameters, keeping records, imposing quarantine measures, controlling the use of animal drugs to stimulate growth or treat diseases, and developing other control measures as may be necessary.

The Authority should work with FDA in its review of the plans for a land based aquaculture operation. Of particular concern in land-based systems is the use of a closed or recirculating water system. Potential exists for shellstock contamination through the failure of the water treatment system to sufficiently disinfect the water to control levels of human pathogens that might be introduced through the water supply or other means. There is also potential for the increased concentration of poisonous and deleterious substances such as animal drugs or antifouling agents in the water supply and subsequently the shellstock over time.

Prior to the harvest of shellstock from land-based systems for sale in interstate commerce, the aquaculturist must demonstrate that the water in the land-based system meets the NSSP Model Ordinance criteria for direct sale of shellstock to the consumer. If the water supply does not meet those criteria, the aquaculturist must subject the shellstock to relaying or depuration prior to sale. For more information related to Relay or Depuration, see Chapters V and XV, respectively.

The cultivation of shellfish with other species in a common aquaculture system is known as polyculture. There are some additional public health concerns related to polyculture. Greater potential may exist for contamination of oysters, clams, mussels and scallops with human pathogens and animal drugs in polyculture. However, the extent of that potential is not known. The extensive use of tanks, sea enclosures, floating rafts, ponds, etc. in polyculture makes the oysters, clams, mussels or scallops highly vulnerable to pollution from various sources, including their association with the other species present in the polyculture operation. The usage of anti-fouling

agents (tributyltin, copper, etc.), hormones, and antibiotics in finfish aquaculture has evoked concern about its environmental effects and potential threat to human health through bioaccumulation in shellfish. Therefore, a conservative approach to polyculture is provided in the NSSP Model Ordinance requirements.

.05 Land Based Aquaculture

a. Need for polyculture and land-based monoculture operations to be under sanitary control. Potential increased consumer risk due to land-based operations.

b. Public health concerns of polyculture elaborated on

c. Conservative approach suggested

d. Authority must establish procedures for issuing permits, approving culturing sites and boundaries, controlling harvest, sampling of shellstock, monitoring environmental parameters, e. Authority encouraged to work with FDA for review of land-based aquaculture operation plans

.06 Polyculture Systems

A polyculture system shall: A. Meet all requirements in Section .05 Land Based Systems; B. Provide information concerning all sources and species of all organisms to be cultivated, cultured, and harvested; and C. Include in its operational plan requirements to: (1)Monitor for human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances that might be associated with polyculture activities; and (2)Subject all harvested shellstock to relaying or depuration if human pathogens, unacceptable levels of animal drugs, and other poisonous or deleterious substances exist at levels of public health significance.

Facility Inspections

If an operation plan is determined to be required for an aquaculture site, the authority must inspect the operation at least annually. The inspection is intended to ensure the operation is adhering to the operational plan, verify that appropriate permits and any reporting, if required, are up to date.

Citations

Stelma, G.N. and L.J. McCabe. 1990. Non-point pollution from animal sources and shellfish sanitation. J. of Food Protection. Vol. 55, No. 8, pp.649 -656.

Smith, O.M., Snyder, W.E. and Owen, J.P. (2020), Are we overestimating risk of enteric pathogen spillover from wild birds to humans?. Biol Rev, 95: 652-679. https://doi.org/10.1111/brv.12581

The Rhode Island Department of Health (RIDOH) Potters Pond Closed to Shellfish Harvesting. [(accessed on 1 July 2022)]; Available online:

	https://www.ri.gov/press/view/42081 2) Allow the Aquaculture Committee to continue to refine Chapter VI. Guidance on aspects related to managing human health concerns from bird and mammal congregations on aquaculture gear.
Action by 2023 Task Force I	Recommends adoption of the Aquaculture Committee recommendations on Proposal 13-107 with the word "cannon" struck. If the information for items #1-4 & 6-7 is provided in permit application materials or on final permits, these may be substituted for inclusion in a formal operational plan. Likewise, #5 is often codified in state and/or federal regulation, and adherence is agreed upon by the operator when signing their permit(s) or by law. In other cases written operational plans containing elements, or the entirety, of the information required in #1-7 may be submitted. Any form of enforceable written record of these items is sufficient to meet the intent of Chapter VI @ .04. To meet the requirements of #6, if necessary, the written operational plan or application materials should clearly describe any operational, maintenance, handling and/or sanitary practices for the aquaculture gear and shellfish that will be conducted to prevent contamination of the growing area from waste attributed to congregations of birds and/or mammals on aquaculture structures. This may include a written description, sketches and/or photos of deterrents or mitigation measures to be used. Strategies may include a suite of deterrents (i.e. kites, ~~cannons~~, sprinklers, spikes etc.) or mitigation measures (e.g. submerging gear and shellfish prior to harvest, relocating floating gear to areas with significant flow, seasonal harvest restrictions, configuring farm sites to maximize flushing, etc.) that will address human health concerns related to year-round or seasonal congregations of birds and/or mammals. In addition, plans should address evaluation of the efficacy of deterrent and/or mitigation measures, and potential triggers that would require changing or adapting deterrent or mitigation measures to address new bird or mammal species and/or behavioral changes, and amendments should be made to the plan, as needed, based on changes to the culture operation, gear, and/or reduced efficacy of the approved deterrents and/or mitigation measures employed by the aquaculture operator.

Proposal No. **13-111**

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Proposal Subject	DSP PPIA Kit for Determination of Okadaic Acid Toxins Group (OA, DTX1, DTX2) in Molluscan Shellfish
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas .11 Approved NSSP Laboratory Tests Marine Biotoxin Testing
Text of Proposal/ Requested Action	The DSP PPIA kit be approved as a Marine Biotoxin Laboratory Test Method.
Public Health Significance	Okadaic acid (OA) and its analogues, DTX1, DTX2, together with their ester forms are known as the group of OA-toxins. These toxins, lipophilic and heat stable, are produced by dinoflagellates and can be found in various species of shellfish, mainly in filter feeding bivalve molluscs. The OA-toxins group causes Diarrheic Shellfish Poisoning (DSP), which is characterized by symptoms such as diarrhea, nausea, vomiting and abdominal pain. These symptoms may occur in humans shortly after consumption of contaminated bivalve molluscs such as mussels, clams, scallops or oysters. Inhibition of serine/threonine phosphoprotein phosphatases is assumed to be responsible for these toxic effects. Recently in the Pacific Northwest harvest areas, outbreaks of DSP have occurred.
Cost Information	Refer to Para D.1. of the Checklist
Action by 2013 Laboratory Methods Review and Quality Assurance Committee	Recommended referral of Proposal 13-111 to an appropriate committee as determined by the Conference Chairman and directed the Executive Office send a letter to the submitter requesting additional information as provided by the Laboratory Methods Review and Quality Assurance Committee.
Action by 2013 Task Force I	Recommended adoption of Laboratory Methods Review and Quality Assurance Committee recommendation on Proposal 13-111.
Action by 2013 General Assembly	Adopted recommendation of 2013 Task Force I on Proposal 13-111.
Action by FDA May 5, 2014	Concurred with Conference action on Proposal 13-111.
Action by 2015 Laboratory Methods Review Committee	Recommended referral of Proposal 13-111 to an appropriate committee as determined by the Conference Chair until additional data are received.
Action by 2015 Task Force I	Recommended adoption of Laboratory Methods Review Committee recommendation on Proposal 13-111.
Action by 2015 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-111.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 13-111.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 13-111.

Action by 2017 Laboratory Committee	Recommended referral of Proposal 13-111 to an appropriate committee as determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 13-111
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-111.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 13-111.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 13-111 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended adoption of the Laboratory Committee recommendation for Proposal 13-111.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 13-111.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 13-111.
Action by 2023 Laboratory Committee	Recommended no action on Proposal 13-111. Rationale: ISSC Constitution, Bylaws, and Procedures – Procedure XV, Section 7, Subdivision A, states that "the method submitter has eighteen months from the date of the written request from the ISSC to provide the information/data necessary to complete the evaluation of the method. If there is no response from the submitter within this timeframe, the Laboratory Committee will recommend no action on the Proposal."
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 13-111.

Proposal No. **13-114**

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Proposal Subject	Receptor Binding Assay (RBA) for Paralytic Shellfish Poisoning (PSP) Toxicity Determination
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas. 11 Approved NSSP Laboratory Tests
Text of Proposal/ Requested Action	4. Approved Limited Use Methods for Marine Biotoxin Testing This submission presents the 'Receptor Binding Assay (RBA) for Paralytic Shellfish Poisoning (PSP) Toxicity Determination' for consideration as an NSSP Approved Limited Use Method. The RBA is a competition-based assay that employs radiolabeled saxitoxin (3H-STX) to compete with PSP toxins present in standards/samples for binding sites on natural receptors in the assay. Following incubation with the receptors, unbound 3H-STX is removed and the remaining labeled toxin is measured with a scintillation counter. The amount of remaining 3H-STX is inversely proportional to standard/sample toxicity. The RBA offers a high-throughput, sensitive, and quantitative alternative to the mouse bioassay (MBA), which has been the long-standing reference method for PSP toxicity. Further, the RBA eliminates the use of live animals for detection of these toxins. While the RBA still uses receptors prepared from animals, the number of animals required for analysis is significantly reduced. Using native receptors as the analytical recognition elements for the assay allows for a composite measure of overall toxicity, as opposed to toxin concentrations measured by liquid chromatographic methods that require conversion factors of equivalent toxicity to calculate the overall toxicity. The RBA has undergone AOAC single- and multi-laboratory validation and is designated through AOAC as an Official Method of Analysis (OMA 2011.27). Results from those studies, and additional data, are included in this proposal submission for the RBA to be considered for approval as an NSSP Approved Limited Use Method for Marine Biotoxin Testing.
Public Health Significance	Paralytic shellfish poisoning intoxications result from the consumption of seafood (primarily bivalve molluscs) contaminated with neurotoxins known as paralytic shellfish toxins (PSTs). This suite of toxins binds to voltage-gated sodium channels and may result in paralysis if enough toxin is consumed. In extreme cases when respiratory support is not available to the patient, the intoxication may prove fatal. Since the toxins cannot be destroyed during cooking and there is no way to remove the toxins from seafood, the best control strategy is to ensure that contaminated product never reaches the market. To protect public health, harvesting closures are implemented when toxicity exceeds the guidance level of 80 micrograms saxitoxin equivalents per 100 grams of shellfish tissue. As such, accurate analytical methods are needed to monitor shellfish toxicity for making decisions regarding opening and closing shellfish growing areas accordingly.

	Acceptance of the RBA as an NSSP Approved Limited Use Method for PSP toxicity determination would provide monitoring and management programs with an additional tool that can be used for monitoring toxin levels and making regulatory decisions. Not only does the RBA eliminate the need for live animals for PSP testing, it is also more sensitive than the MBA, thereby providing an early warning system for monitoring programs as toxin levels begin to rise.
Cost Information	The estimated cost for a full 96-well plate assay is ~$95.00. Including standards and samples with triplicate measurements (as well as three dilutions per sample to ensure the unknown samples fall within linear range of assay), the cost per sample for quantitative results would be ~$13.60. If running multiple plates or in screening mode, sample costs would be reduced. Further, the filter plates used in the RBA differ from ELISA plates in that all reagents are added to each well as needed rather than already being a component of the plate, making it more practical and cost-effective to analyze samples when there is less than a full plate.
Action by 2013 Laboratory Methods and Quality Assurance Review Committee	1. Recommended approval of this method as an alternative to the mouse bioassay for PSP in mussels. 2. Recommended approval of this method for Limited Use for clams and scallops for the purpose of screening and precautionary closure for PSP. 3. Recommended referral of this proposal to an appropriate committee as determined by the Conference Chairman to address this method in oysters. 4. Recommended Executive Office sends a letter to submitter to request a checklist for evaluation of labs using this method with said checklist to be submitted within three (3) months.
Action by 2013 Task Force I	Recommended adoption of Laboratory Method Review and Quality Assurance Committee recommendation on Proposal 13-114.
Action by 2013 General Assembly	Adopted recommendation of 2013 Task Force I on Proposal 13-114.
Action by FDA May 5, 2014	Concurred with Conference action on Proposal 13-114.
Action by 2015 Laboratory Methods Review Committee	Recommended referral of Proposal 13-114 to an appropriate committee as determined by the Conference Chair until additional data for oyster matrix are received.
Action by 2015 Task Force I	Recommended adoption of Laboratory Methods Review Committee recommendation on Proposal 13-114.
Action by 2015 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-114.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 13-114.
Action by 2017 Laboratory Committee	Recommended referral of Proposal 13-114 to an appropriate committee as determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 13-114.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-114.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 13-114.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 13-114 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 13-114.

Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 13-114.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 13-114.
Action by 2023 Laboratory Committee	Recommended no action on Proposal 13-114. Rationale: Original submitter is no longer able to pursue this proposal and no other laboratory is available at this time.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 13-114.

Proposal No. **15-109**

Submitter	Alison Sirois and Jackie Knue
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Proposal Subject	PSP HPLC-PCOX Species Expansion
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II Growing Areas .11 Approved NSSP Laboratory Tests
Text of Proposal/ Requested Action	4. Approved Limited Use Methods for Marine Biotoxin Testing PCOX This submission presents data to support the use of PCOX method for Quahogs (M. mercenaria and A. icelandica), Surf Clams (S. solidissima), Geoducks (P. generosa), Butter Clams (S. giganteus), Little Neck Clams (P. stamineais), and Razor Clams (S. patula) for regulatory paralytic shellfish toxin (PST) testing. Results of the 2009 Interstate Shellfish Sanitation Conference (ISSC) proposal 09-104 concluded the PCOX method approved for official use as a Type IV method; subsequently after single laboratory validation (SLV) and collaborative studies, ISSC proposal 13-309 accepted PCOX method as an AOAC official method of analysis (OMA) in 2013. Currently PCOX is an "Approved for Limited Use" method for mussel, clam, oyster and scallop. SLV work will be presented for quahogs, surf clams, geoducks, butter clams, little neck clams, and razor clams that demonstrates comparable performance characteristics for these species as with mussels, clams, oysters, and scallops using the PCOX method. The cost and challenges associated with maintaining both the MBA and PCOX methods for these species are high; differing laboratory skill sets are required and state laboratories have limited budgets and staff resources. Additionally, the recent shortage of the NIST saxitoxin standard used for MBA proficiencies is of concern if laboratories are expected to maintain MBA for verification purposes for these species. The requested action is being made and data presented for the purpose of inclusion of quahogs, surf clams, geoducks, butter clams, little neck clams, and razor clams as approved species (by addition to the footnote that includes mussels, clams, oysters, and scallops or as the ISSC deems appropriate) within the NSSP Guide Section IV Guidance Documents Chapter II. Growing Areas .11 Laboratory Tests Methods Table, Methods for Marine Biotoxin Testing with Biotoxin Type: Paralytic Shellfish Poisoning (PSP), Application: Growing Area Survey & Classification Sample Type: Shellfish And Application: Controlled Relaying Sample Type: Shellfish.

Public Health Significance	The PCOX method was developed to provide a rapid, high throughput chemical assay that would eliminate the need to sacrifice animals, AOAC mouse bioassay (MBA), for toxin detection. There is a worldwide move to replace assays that use live animals as test subjects. Laboratories currently using PCOX for regulatory PST testing have found that the lower detection limits of the PCOX method allow for better early warning therefore better management of PST closures and significantly improved public health decision-making. The addition of the proposed species will allow regulatory laboratories to move away from the costliness of maintaining MBA and eliminate the need to sacrifice animals as well as improve management of species specific closure decision–making.
Cost Information	Total consumable costs for the analysis is estimated at $10/sample. A chemistry laboratory will usually be equipped with an LC system and a post column reactor to carry out the analysis. Total capital costs for the instrumentation required for the analysis is approximately $120,000. Although the upfront investment for instrumentation is high, the removal of care, maintenance, and cost of mice quickly offsets this expenditure.
Action by 2015 Laboratory Method Review Committee	Recommended referral of Proposal 15-109 to an appropriate committee as determined by the Conference Chair for evaluation of data and until additional data are received.
Action by 2015 Task Force I	Recommended adoption of 2015 Laboratory Method Review Committee recommendation on Proposal 15-109.
Action by 2015 General Assembly	Adopted recommendation of Task Force I on Proposal 15-109.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 15-109.
Action by 2017 Laboratory Committee	Recommended referral of Proposal 15-109 to an appropriate committee as determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 15-109.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 15-109.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 15-109.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 15-109 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 15-109.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 15-109.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 15-109.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 15-109 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 15-109.

Proposal No. **15-112**

Submitter	Executive Board
Affiliation	Interstate Shellfish Sanitation Conference (ISSC)
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Proposal Subject	Direct Plating Method for trh
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas .11 Approved NSSP Laboratory Tests
Text of Proposal/ Requested Action	This method was developed by Jessica Jones (FDA Gulf Coast Seafood Laboratory) and is being submitted by the ISSC Executive Board. The Executive Board granted interim approval to this method on March 13, 2015. The Executive Board is submitting this proposal to comply with Article V. Section 1. of the ISSC Constitution, Bylaws, and Procedures. Submitted by method developer Jessica Jones (FDA Gulf Coast Seafood Laboratory)

M	Vibrio Indicator Type:	Application: PHP Sample Type: Shucked	Application : Reopening
EIA[1]	*Vibrio vulnificus (V.v.)*	X	
MPN[2]	*Vibrio vulnificus (V.v.)*	X	
SYBR Green 1 QPCR-MPN[5]	*Vibrio vulnificus (V.v.)*	X	
MPN[3]	*Vibrio parahaemolyticus (V.p.)*	X	
PCR[4]	*Vibrio parahaemolyticus (V.p.)*	X	
Direct Plating[6]	*trh+ Vibrio parahaemolyticus (V.p.)*	X	X

Footnotes:

1 EIA procedure of Tamplin, et al, as described in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, 1992.

2 MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, followed by confirmation using biochemical analyses or by the DNA -alkaline phosphatase labeled gene probe (vvhA).

3 MPN format with confirmation by biochemical analysis, gene probe methodology as listed in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, or a method that a State can demonstrate is equivalent.

4 PCR methods as they are listed in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, or a method that a State

	can demonstrate is equivalent. [5] *Vibrio vulnificus*, ISSC Summary of Actions 2009. Proposal 09-113, Page 123. [6] Direct plating method for *trh* as described in Nordstrom et al., 2006.
Public Health Significance	Scientific evidence suggests that the presence of the *trh* gene in *V. parahaemolyticus (V.p.)* is correlated with higher virulence. Additionally, at the 2013 conference, proposal 13-202 was adopted which requires testing for the presence of trh prior to reopening of growing areas closed as a result of *V.p.* illnesses [Chapter II @.01.F(5)]. Currently, there are no NSSP approved methods for enumeration of *trh*. This method is a needed option for testing following *V.p.* illness closures.
Cost Information	This method costs ~$5 per test for laboratory consumables, supplies, and reagents. Most equipment needed for testing is standard microbiology equipment, but purchase of a specialized water bath or environmental chamber may be necessary at a cost of ~$3,000-$5,000. Additional costs for a laboratory would vary based on their operational overhead and labor.
Action by 2015 Laboratory Methods Review Committee	Recommended referral of Proposal 15-112 to an appropriate committee as determined by the Conference Chair to further review the data submitted.
Action by 2015 Task Force I	Recommended adoption of 2015 Laboratory Methods Review Committee recommendation on Proposal 15-112.
Action by 2015 General Assembly	Adopted recommendation of Task Force I on Proposal 15-112
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 15-112.
Action by 2017 Laboratory Committee	Recommended referral of Proposal 15-112 to an appropriate committee as determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Lab Committee recommendation on Proposal 15-112.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 15-112.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 15-112.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 15-112 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 15-112.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 15-112.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 15-112.
Action by 2023 Laboratory Committee	Recommended no action on Proposal 15-112. Rationale: The DNA probe necessary for this method is no longer available.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 15-112.

Proposal No. **15-114**

Submitter	Executive Board
Affiliation	Interstate Shellfish Sanitation Conference (ISSC)
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Proposal Subject	Pre-Proposal for Male-Specific Coliphage Enumeration in Wastewater by Direct Double-Agar Overlay Method
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas .11 Approved NSSP Laboratory Tests
Text of Proposal/ Requested Action	The submitter of the pre-proposal requests approval to submit a full proposal to the ISSC for approval of the analytical method for use in the NSSP. Submitted by the developer Kevin Calci (FDA Gulf Coast Seafood Laboratory) Proposed Use of the Method: This method is applicable for the enumeration of MSC wastewater influent, effluent and sewage contaminated surface waters. The method will directly determine the quantity of MSC in wastewater to provide information of the viral reduction efficiencies of wastewater treatment plants. Method is also applicable for the analysis of surface source waters as part of a shoreline survey. Description of Method: This method employs E. coli HS (pFamp) RR as a male-specific coliphage host in a direct double agar overlay for the quantification of plaque forming units. All sample volumes are plated in triplicate. Briefly, 2.5ml of sample is mixed with 2.5ml of soft agar and 0.2ml of Famp host and then poured onto bottom agar petri plate. One ml of the sample is serially diluted down to 1:10 and 1:100. Those two dilutions are then plated by placing 2.5ml of sample is mixed with 2.5ml of soft agar and 0.2ml of Famp host and then poured onto bottom agar petri plate. The plates are incubated at 35-37°C for 16-20 h. Under indirect light the plaque forming units are counted. The working range of the 9 plate method would be 14pfu/1OOml to 1.0 x 106 pfu/1 OOml.
Public Health Significance	Scientific consensus at the MSC informational meeting supported the use of MSC to evaluated wastewater treatment plant viral reduction efficiency to better inform the SSCA's conditional management plans impacted by wastewater treatment plant operations. This method would identify a consistent and accurate measure of MSC load in wastewater influent, effluent and surface waters.
Cost Information	
Action by 2015 Laboratory Methods Review Committee	Recommended referral of Proposal 15-114 to an appropriate committee as determined by the Conference Chair to await SLV data.
Action by 2015 Task Force I	Recommended adoption of 2015 Laboratory Methods Review Committee recommendation on Proposal 15-114.
Action by 2015 General Assembly	Adopted recommendation of Task Force I on Proposal 15-114.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 15-114.
Action by 2017	Recommended referral of Proposal 15-114 to an appropriate committee as

Laboratory Committee	determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 15-114.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 15-114.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 15-114.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 15-114 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 15-114.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 15-114.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 15-114.
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 15-114 as amended.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 15-114.

Submitter	US Food & Drug Administration (FDA)
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Proposal Subject	Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS) Method for the Determination of Diarrhetic Shellfish Poisoning (DSP) Toxins in Shellfish.
Specific NSSP Guide Reference	Section IV. (Guidance Documents), Chapter II. (Growing Areas), Section .14 (Approved Laboratory Tests), Table 2 (Approved Methods for Biotoxin Testing) and Table 4 (Approved Limited Use Methods for Marine Biotoxin Testing)
Text of Proposal/ Requested Action	The intention is for this method to be an Approved Method for Marine Biotoxin Testing for clams and that it should appear in Section IV. (Guidance Documents), Chapter II. (Growing Areas), Section .14 (Approved Laboratory Tests), Table 2 (Approved Methods for Marine Biotoxin Testing) under the new heading: Biotoxin Type: Diarrhetic Shellfish Poisoning (DSP), and the applications should be (1) Growing Area Survey and Classification and (2) Controlled Relaying with the sample type of Shellfish for both. In addition, the method should also be included in Table 4 (Approved Limited Use Methods for Biotoxin Testing) for mussels and oysters. Additional validation will be submitted later in order to move mussels and oysters also to Table 2.
Public Health Significance	Method will be used to control hazard from Diarrhetic Shellfish Poisoning (DSP) in shellfish. No methods for DSP are currently listed in the NSSP yet shellfish harvesting closures have occurred due to these toxins in Texas since 2008, in the Pacific Northwest since 2011, and in the New England region since 2015. Regulatory laboratories in these regions are currently using best available science of LC-MS/MS according to the EU reference SOP for LC-MS/MS determination of lipophilic shellfish toxins.
Cost Information	Capital equipment purchases: $500,000. Consumable cost per sample: $10.00
Research Needs Information	
a. Proposed specific research need/ problem to be addressed	No methods are currently approved for use to control DSP hazard under the NSSP. The EU has adopted LC-MS/MS as the reference method for all of the lipophilic shellfish toxins, including DSP. This method is a modified version of the EU LC-MS/MS method optimized specifically for DSP.
b. Explain the relationship between proposed research need and program change recommended in the proposal	The proposal will provide full SLV data for the detection of DSP toxins in clams. Therefore it would be considered an Approved Method for clams (Table 2). Based on the immediate need for this method, it was felt that the submission should be made with the available data for clam with the intention of subsequent validation for mussels and oysters, for which only preliminary data is provided here. Therefore, the method should be considered for Approved Limited Use at this time for mussel and oyster and be included in Table 4 for these matrices.
c. Estimated cost	$10,000
d. Proposed sources of funding	FDA internal funding
e. Time frame anticipated	Submission of all materials in order to be reviewed prior to the 2017 bi-annual ISSC meeting.
Action by 2017 Laboratory Committee	Recommended the following: 1) Adoption of Proposal 17-103 as an Approved Method for clams

	2) Referral of Proposal 17-103 to an appropriate committee as determined by the Conference Chair to determine the appropriateness of the method for mussels and oysters.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendations on Proposal 17-103.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-103.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-103.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 17-103 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 17-103.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 17-103.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 17-103.
Action by the 2023 Laboratory Committee	Recommended referral of Proposal 17-103 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 17-103.

Proposal No. **17-106**

Submitter	Pacific Rim Shellfish Sanitation Association
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Proposal Subject	Matrix Expansion for the Receptor Binding Assay (RBA) for Paralytic Shellfish Poisoning (PSP) Toxicity Determination to Allow Use with Geoduck
Specific NSSP Guide Reference	Section IV, Chapter II.14 -- NSSP Approved Laboratory Tests (p. 261 Table 2. Approved Methods for Marine Biotoxin Testing -- footnote 2, and/or p. 263 Table 4. Limited Use Methods for Marine Biotoxin Testing -- footnote 5)
Text of Proposal/ Requested Action	This submission presents the 'Matrix Expansion for the Receptor Binding Assay (RBA) for Paralytic Shellfish Poisoning (PSP) Toxicity Determination to Allow Use with Geoduck' for consideration as an NSSP Approved Method for Marine Biotoxin Testing for PSP in Geoduck. The RBA is a competition-based assay that employs radiolabeled saxitoxin (3H-STX) to compete with PSP toxins present in standards/samples for binding sites on natural receptors in the assay. Following incubation with the receptors, unbound 3H-STX is removed and the remaining labeled toxin is measured with a scintillation counter. The amount of remaining 3H-STX is inversely proportional to standard/sample toxicity. The RBA offers a high-throughput, sensitive, and quantitative alternative to the mouse bioassay (MBA), which has been the long-standing reference method for PSP toxicity. Further, the RBA eliminates the use of live animals for detection of these toxins. While the RBA still uses receptors prepared from animals, the number of animals required for analysis is significantly reduced. Using native receptors as the analytical recognition elements for the assay allows for a composite measure of overall toxicity, as opposed to toxin concentrations measured by liquid chromatographic methods that require conversion factors of equivalent toxicity to calculate the overall toxicity. The RBA has undergone AOAC single and multi-laboratory validation and is designated through AOAC as an Official Method of Analysis (OMA 2011.27). The RBA is currently an NSSP Approved Method for Marine Biotoxin Testing for PSP in mussels as well as a NSSP approved for Limited Use Method for clams and scallops for the purpose of screening and precautionary closure for PSP (ISSC 2015 Summary of Actions Proposal 13-114). Here we provided results from a single laboratory validation study for use of RBA with the matrix geoduck (*Panopea*) viscera for submission for the RBA to be considered for approval as an NSSP Approved Method for Marine Biotoxin Testing for PSP.
Public Health Significance	Paralytic shellfish poisoning intoxications result from the consumption of seafood (primarily bivalve molluscs) contaminated with neurotoxins known as paralytic shellfish toxins (PSTs). This suite of toxins binds to voltage-gated sodium channels and may result in paralysis if enough toxin is consumed. In extreme cases when respiratory support is not available to the patient, the intoxication may prove fatal. Since the toxins cannot be destroyed during cooking and there is no way to remove the toxins from seafood, the best control strategy is to ensure that contaminated

	product never reaches the market. To protect public health, harvesting closures are implemented when toxicity exceeds the guidance level of 80 micrograms saxitoxin equivalents per 100 grams of shellfish tissue. As such, accurate analytical methods are needed to monitor shellfish toxicity for making decisions regarding opening and closing shellfish growing areas accordingly. Acceptance of the RBA as an NSSP Approved Method for Marine Biotoxin Testing for PSP toxicity determination in geoduck (*Panopea*) would provide monitoring and management programs with an additional tool that can be used for monitoring toxin levels and making regulatory decisions. Not only does the RBA eliminate the need for live animals for PSP testing, it is also more sensitive than the MBA, thereby providing an early warning system for monitoring programs as toxin levels begin to rise.
Cost Information	For the assay: The estimated cost per 96-well plate assay is ~$95.00. Including standards and samples with triplicate measurements (as well as three dilutions per sample[ranging from 3.5-600 µg STX eq 100 g-1] to ensure the unknown samples fall within linear range of assay), the cost per sample for quantitation would be ~$13.60. If running multiple plates or in screening mode, sample costs would be reduced. (Van Dolah 2013) For proposal: The cost of RBA work for geoduck matrix expansion is covered by and existing grant awarded to the Sitka Tribe of Alaska. Naturally contaminated samples from Washington and Alaska are pulled from regular samples tested by the respective state agencies that are part of routine shellfish testing. Therefore, there is no additional cost or funding necessary for the proposal.
Research Needs Information	
a. Proposed specific research need/ problem to be addressed	Paralytic shellfish poisoning (PSP) is a foodborne illness caused by ingestion of contaminated shellfish. The paralytic shellfish toxin, saxitoxin (STX), and its analogs are potent neurotoxins responsible for PSP. Marine dinoflagellates and freshwater cyanobacteria produce STX. The STX can accumulate in filter-feeding bivalve mollusks to levels that are toxic to humans. Symptoms of PSP include: tingling and numbness of the perioral area and extremities, drowsiness, incoherence, loss of motor control, and following high dose consumption, respiratory paralysis. In 1965 the mouse bioassay (MBA) was adopted as an official AOAC method for STX determination. The MBA has been the only method available for PSP testing for the last five decades. Both North American and European regulatory agencies have expressed the desire to transition to a more humane PSP testing method that does not require the use of live animals and is not subject to the matrix effects documented for the MBA (Turner 2012). Recently, the NSSP approved a post-column oxidation liquid chromatographic (PCOX) method and a receptor binding assay (RBA) as alternatives to the MBA. The PCOX method is approved for full use; whereas, the RBA is approved for limited use (the RBA is only approved for shellfish matrices evaluated in the single lab and multi-lab validation studies). Both the PCOX and RBA are sensitive quantitative assays for STX detection, and they do not require the use of live animals. The RBA is approved for regulatory testing of mussels as an alternative to the MBA and is approved for limited use as a screening tool for clams and scallops, but is not yet approved for use with geoduck (*Panopea*) due to a lack of data. Geoduck

	are a major commercial product, with large dive fisheries in Southeast Alaska and the Puget Sound that require STX testing. This proposal requests consideration for the NSSP RBA approval to be expanded to include geoduck. The proposal provides data from a single laboratory validation (SLV) of the RBA for geoduck testing as support for this request.
b. Explain the relationship between proposed research need and program change recommended in the proposal	This method is intended for use as an NSSP Approved Limited Use Method for screening for PSP toxicity in shellfish. The RBA serves as an alternative to the MBA in these applications, offering a measure of composite toxicity with high throughput and the elimination of live animal testing. (Van Dolah 2013) This application is for the addition of geoduck to the list of matrices approved for use with the RBA. There is an acknowledged need for this method in NSSP. A significant portion of the Washington and Alaska state shellfish industries are comprised of the harvest of geoduck. Approval of the RBA for use with geoduck would provide an alternative to (1) the MBA, which uses live animals, and (2) the PCOX HPLC method, which requires costly equipment and skilled personnel and offers low throughput. Acceptance of the RBA as an NSSP Approved Method for Marine Biotoxin Testing for PSP toxicity determination in geoduck would provide monitoring and management programs with an additional tool that can be used for monitoring toxin levels and making regulatory decisions. Not only does the RBA eliminate the need for live animals for PSP testing, it is also more sensitive than the MBA. **References**: Van Dolah 2013. ISSC application: Receptor Binding Assay (RBA) for Paralytic Shellfish Poisoning (PSP)Toxicity Determination. Van Dolah et al. 2012. Determination of paralytic shellfish toxins in shellfish by receptor binding assay: collaborative study. J AOAC Int. May-Jun;95(3):795-812. Van Dolah et al. 2009. Single-laboratory validation of the microplate receptor binding assay for paralytic shellfish toxins in shellfish. J AOAC Int. Nov-Dec;92(6):1705-13. Ruberu et al. 2012. Evaluation of variability and quality control procedures for a receptor-binding assay for paralytic shellfish poisoning toxins. Food Addit Contam Part A Chem Anal Control Expo Risk Assess.29(11):1770-9. Turner et al. 2012. Investigations into matrix components affecting the performance of the official bioassay reference method for quantitation of paralytic shellfish poisoning toxins in oysters. Toxicon : official journal of the International Society on Toxicology 59, 215-230. OMA 2011.27. AOAC Official Method 2011.27 Paralytic shellfish toxins (PSTs) in shellfish, receptor binding assay. In Official Methods of Analysis of AOAC International. http://www.eoma.aoac.org.
c. Estimated cost	
d. Proposed sources of funding	This research was performed by the Sitka Tribe of Alaska using funds from an ANA ERE grant

e. Time frame anticipated	
Action By 2017 Laboratory Committee	Recommended referral to an appropriate committee as determined by the Conference Chair.
Action By 2017 Task Force I	Recommended adoption of the Laboratory Committee recommendation on Proposal 17-106.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-106.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-106.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 17-106 to an appropriate committee as determined by the Conference Chairperson.
Action by 2019 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 17-106.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 17-106.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 17-106.
Action by the 2023 Laboratory Committee	Recommended referral of Proposal 17-106 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 17-106.

Proposal No. **17-108**

Submitter	Titan Fan, Ph.D
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Proposal Subject	Detection of ASP biotoxins in *Mytilus edulis* (Blue Mussel) shellfish by ELISA for Domoic Acid
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas, Table 2.
Text of Proposal/ Requested Action	SLV Proposal supporting the use of Beacon Domoic Acid Plate Kit as fit for purpose as an Approved NSSP Method for quantification of ASP toxins in Marine Biotoxin Monitoring Programs.
Public Health Significance	Shellfish consumption can pose a mammal and bird health risk (1) when toxins produced by cyanobacteria present in water and shellfish growing areas, concentrate in shellfish meat due to their filter feeding system. A Closed Status for any growing areas with shellfish tissue levels of ASP of 2 mg/100 g (20 ppm) or more have been established to protect the consumer from exposure (2). The most common clinical signs of acute toxicity are gastrointestinal distress, confusion and neurological symptoms, disorientation, memory loss, coma and death (3). (1). M.Fernanda, F, Mazzillo, C. Pomeroy, J.Kuo, P. Ramondi,R. Prado, M.Silver. 2010. Aquatic Biol. 9:1-12. (2). NSSP Guide for the Control of Molluscan Shellfish: 2015 Rev. Sec.IV Chp. II., p 231. (3). Kathi A. Lefebvre, Alison Robertson, Toxicon, Vol. 56, Issue 2, 15 Aug. 2010, p. 218-230.
Cost Information	The price per sample is eight to nine dollars dependent upon the number of samples tested during one ELISA run, and/or the volume of kits purchased. There is an ELISA Plate Reader requirement. They can range in price from a low cost unit at approximately $2,600 to a higher cost of $15,000 USD unit depending upon complexity.
Action By 2017 Laboratory Committee	Recommended referral of Proposal 17-108 to an appropriate committee as determined by the Conference Chair.
Action By 2017 Task Force I	Recommended adoption of the Laboratory Committee on Proposal 17-108.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-108.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-108.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 17-108 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 17-108.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 17-108.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 17-108.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 17-108 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 17-108.

Proposal No. **17-110**

Submitter	U.S. Food and Drug Administration (FDA)
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Proposal Subject	Alkaline Phosphatase Probe Method for *Vibrio vulnificus* and *Vibrio parahaemolyticus* Detection in Oysters - Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV Guidance Documents Chapter II Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt the text of the attached checklist for the probe method for detecting *Vibrio vulnificus* (Vv) and *Vibrio parahaemolyticus* (Vp) in oysters and to append the checklist to the list of NSSP Laboratory Evaluation Checklists at the end of .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists.
Public Health Significance	Currently, there is no checklist adopted by the ISSC for the probe method for detecting Vv and Vp in oysters. The attached checklist provides the quality assurance and method requirements that laboratory evaluation officers will use to evaluate laboratories implementing this method in support of the NSSP. The checklist documents the number of critical, key or other nonconformities and how overall laboratory status for the method is determined.
Cost Information	NA
Action By 2017 Laboratory Committee	Recommended Proposal 17-110 be referred to an appropriate committee as determined by the Conference Chair.
Action By 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 17-110.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-110.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-110.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 17-110 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended adoption of the Laboratory Committee recommendation on Proposal 17-110.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 17-110.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 17-110.
Action by 2021 Laboratory Committee	Recommended adoption of Proposal 17-110 as amended with Interim Approval by the Executive Board
Action by 2021 ISSC Executive Board	Granted Interim Approval in effect until the Conference convenes at the 2023 ISSC Biennial Meeting.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 17-110.

Proposal No. **17-116**

Submitter	U.S. Food and Drug Administration (FDA)
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Proposal Subject	Sanitary Control of Molluscan Shellfish Harvested From Federal Waters
Specific NSSP Guide Reference	Section I Purposes & Definitions Section II Model Ordinance Chapter IV Shellstock Growing Areas Section II Model Ordinance Chapter VI Shellfish Aquaculture
Text of Proposal/ Requested Action	Insert the following definition for Federal Waters in Section I Purposes & Definitions as follows: **Federal Waters** means the waters that fall outside of State and local jurisdiction but within U.S. sovereignty (typically 3-200 nautical miles offshore). Federal waters include the territorial sea and exclusive economic zone. Insert the language below for Section II Model Ordinance Chapter IV Shellstock Growing Areas @.01 Sanitary Survey. E. Sanitary surveys for Federal waters will be the responsibility of FDA. Sanitary surveys will be conducted in accordance with Chapter IV @.01, as applicable. @.03 Growing Area Classification. F. FDA is responsible for the classification of growing areas in Federal waters. Federal waters are classified as Approved for shellfish harvesting unless such areas are known to be polluted (i.e., microbiological, chemical, and marine biotoxin hazards) and involve commercial shellfish resources . Insert the language below for Section II Model Ordinance Chapter VI Shellfish Aquaculture just after the text in @.03and prior to Shellfish Gardening @.04 Aquaculture in Federal Waters A. Federal Agency Responsibilities. Once the appropriate permits for the construction of the aquaculture facility have been obtained, (1) NOAA is responsible for establishing a contract, in consultation with FDA, with the aquaculture facility describing requirements of the NSSP including (a) the frequencywith which NOAA will audit the aquaculture facility and vessels, (b) testing requirements of the aquaculture facility, and (c) the generation of product identification for traceability (i.e., tag numbers); and (2) FDA is responsible for reviewing the aquaculture facility operational plan prior to the start of operations, as well as the annual inspection of records, to ensure adherence to NSSP requirements. FDA is also responsible for the classification of the growing area(s) associated with the aquaculture facility.

	@.~~04~~05 Shellfish Gardening Insert the language below for Section II Model Ordinance Chapter VI Shellfish Aquaculture just after .07 .08 Requirements for the Harvester in Aquaculture in Federal Waters A. Prior to beginning any aquaculture activities, the person who performs aquaculture or operates an aquaculture facility to raise shellfish in Federal waters for human consumption shall obtain the appropriate permission(s) from Federal agencies as described in @.04. B. Operational Plan. Each aquaculture facility shall have a written operational plan as described for Land Based Aquaculture in Section II Chapter VI .05(A). The operational plan shall also include: (1) Description of harvest, tagging, handling, storage, transportation, and landing procedures; (2) Description of a marine biotoxin management and contingency plan (Section II Chapter IV @.04) to include marine biotoxin sampling consistent with Section II Chapter IV @.04(a)(5) and ensure product segregation and control until biotoxin results confirm the shellfish do not contain biotoxins equal to or exceeding criteria established in Section IV Chapter II .08.; (3) Description of a contingency in the event of an emergency situation or condition (e.g., sewage or oil spills); and (4) Procedures for implementing product recalls. C. Each aquaculture facility obtain review from the FDA to ensure adherence to NSSP requirements prior to its implementation. If the aquaculture facility makes changes to the operational plan, they shall obtain a new review from the FDA to ensure adherence to the NSSP requirements.
Public Health Significance	Currently, the NSSP Guide does not explicitly cover requirements for the sanitary control of molluscan shellfish harvested from U.S. Federal waters. The lack of standards for this activity has impeded the harvest of shellfish, notably aquaculture, from Federal waters to date. FDA's policy on the classification of growing areas in offshore Federal waters as described in Verber 1977 was followed in drafting the Proposal. Adding specific language to the Model Ordinance on the appropriate requirements for this activity will facilitate safe and sanitary access to additional shellfish resources.
Cost Information	N/A
Action By 2017 Task Force I	Recommended adoption of Proposal 17-116 on an interim basis with a sunset date of November 1, 2021 and that during this period a committee be appointed to evaluate aquaculture activities in federal waters.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-116.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-116.
Actions by 2019 Federal Waters Committee	Recommended the adoption of the following proposals: 19-202,19-203, 19-214, 19-223, 19-228, 19-229, 19-120

	The Committee was provided a task list developed by the Federal Waters Subcommittee which includes a number of regulatory actions necessary to provide a framework for incorporating shellfish from Federal Waters into the NSSP.
Action by 2019 Task Force I	Recommended Proposal 17-116 be referred to an appropriate committee as determined by the Conference Chairperson with further instruction to identify the specific sanitary survey criteria requirements to be used by FDA.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 17-116.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 17-116.
Action by 2023 Federal Waters Committee	Recommends deletion of the sunset date of November 1, 2021 from Proposal 17-116.
Action by Task Force I	Recommends adoption of the Federal Waters Committee recommendation on Proposal 17-116.

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Proposal Subject	Conditionally Conforming Laboratory Status
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter I. Shellfish Sanitation Program Requirements for the Authority @.03 B. 1. b. Section II. Model Ordinance Chapter III. Laboratory @.01 Section II. Model Ordinance Chapter XV. Depuration .03 J. (4)
Text of Proposal/Requested Action	The requested action is to create a NSSP laboratory status of conditionally conforming. This status is based on a demonstrated proficiency of laboratory method performance. Laboratories that are found to conditionally conform for a laboratory analysis may support the NSSP. **MO Chapter 1.@.03 B. 1. b.** v. Performance Evaluation: Conditionally Conforms. Tto be deemed conditionally conforming under the NSSP, a laboratory must meet one of the following laboratory performance criteria: (a) Complete an appropriate ISSC Accepted SLV; or (b) Complete a Method Verification Study, Section IV. Chapter II. .20 that successfully transfers; or (c). Successfully complete a proficiency and/or inter-laboratory study approved by the FDA Shellfish LEO or State certified Shellfish LEO. (d) This laboratory status will remain in effect until an technical FDA Shellfish LEO or FDA certified State Shellfish LEO Evaluation occurs as in @.03 B. **MO Chapter III. @.01 Quality Assurance** A. NSSP Conformance Required for all laboratories supporting the NSSP. All laboratory analyses shall be performed by a laboratory found to conform, conditionally conform or provisionally conform by the FDA Shellfish LEO or FDA certified State Shellfish LEO in accordance with the requirements established under the NSSP. **MO Chapter XV. .03 J. (4)** (a) Are analyzed by a laboratory which has been evaluated and found to conform or conditionally conform to the NSSP pursuant to the requirements in Chapter III, using an NSSP-Approved Method;

Public Health Significance	A technical Laboratory evaluation, as outlined in MO Chapter 1.@.03B.1.b.ii, is conducted to verify that conditions are present *in the laboratory* which **should** result in the accurate outcome of method data. A performance evaluation **verifies** that the method data produced *by the laboratory and for all analysts* is accurate. A technical evaluation does not examine the quality of a laboratory's method data for validity, standardization or for individual analysts. If a laboratory has successfully passed a proficiency study, SLV or MV, and statistically confirmed method data results, the laboratory can be assumed to have technically performed the method correctly. Under current interpretation a laboratory may have completed and had accepted by the conference a method SLV with accompanying checklist yet not be able to support the NSSP with data until a FDA Shellfish LEO or FDA certified State Shellfish LEO conducts a technical inspection at their laboratory using the laboratory's own checklist. If a laboratory has proven its ability to perform a method, then the laboratory should be able to conditionally support the NSSP with data. A cooperative goal of the NSSP, FDA and the SSCA is to assure that a laboratory's data is accurate, verified and standardized. Method based performance evaluations confirm data which results in standardization across laboratories. Method based performance evaluations statistically verify data accuracy. Performance Evaluations therefore support the legal defensibility of the laboratory's Laboratory Quality Management System.
Cost Information	Cost of conducting SLV, MV or Proficiency Participation
Action by 2019 Laboratory Committee	Recommended no action on Proposal 19-101. Rationale: This issue is addressed by Proposal 19-301.
Action by 2019 Task Force I	Recommended adoption of Proposal 19-101 as submitted.
Action by 2019 General Assembly	Recommended referral of Proposal 19-101 to an appropriate committee as determined by the Conference Chair.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-101.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 19-101 to an appropriate committee as determined by the Conference Chairperson
Action by 2023 Task Force I	Recommends referral of Proposal 19-101 as amended to the Laboratory Committee with the provision that a recommendation for interim approval be provided at the 2023 Fall Executive Board Meeting. **MO Chapter 1.@.03 B. 1. b.** vi.Performance Evaluation: Conditionally Conforms. Tto be deemed conditionally conforming under the NSSP, a laboratory must meet one of the following laboratory performance criteria: (a) Complete an appropriate ISSC Accepted SLV; or (b) Complete a Method Verification Study, Section IV. Chapter II. .20 that successfully transfers; or ~~(c). Successfully complete a proficiency and/or inter-laboratory study approved by the FDA Shellfish LEO or State certified Shellfish LEO.~~ (c~~d~~) This laboratory status will remain in effect until an technical FDA Shellfish LEO or FDA certified State Shellfish LEO Evaluation occurs as in @.03 B. **MO Chapter III. @.01 Quality Assurance** A. NSSP Conformance Required for all laboratories supporting the NSSP. All

	laboratory analyses shall be performed by a laboratory found to conform, conditionally conform or provisionally conform by the FDA Shellfish LEO or FDA certified State Shellfish LEO in accordance with the requirements established under the NSSP. **MO Chapter XV. .03 J. (4)** (a) Are analyzed by a laboratory which has been evaluated and found to conform or conditionally conform to the NSSP pursuant to the requirements in Chapter III, using an NSSP-Approved Method;

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Proposal Subject	Laboratory approval for sample analysis with no Model Ordinance defined method or action level
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter III. Laboratory @.01 Quality Assurance (A)
Text of Proposal/ Requested Action	Chapter III. @.01 A. NSSP Conformance Required~~. for all laboratories supporting the NSSP~~. All laboratory analyses for compliance with classification requirements that require a specific method, actions level, and use defined in the Model Ordinance shall be performed by a laboratory found to conform or provisionally conform by the FDA Shellfish LEO or FDA certified State Shellfish LEO in accordance with the requirements established under the NSSP.
Public Health Significance	This proposed amendment to Chapter III, @.01 (A) updates the requirement related to the use of data analyzed by a laboratory that has not been certified by the FDA Shellfish LEO or FDA certified State Shellfish LEO and potentially used for regulatory purposes. The amendment allows state shellfish authorities to use non FDA approved laboratories when methods and action levels have not been defined in the Model Ordinance. Washington state has developed an extensive array of partnerships aimed at evaluating pollution conditions around shellfish growing areas primarily related to microbiological conditions and remediating any impacts identified. Local and state government agencies, tribes, and wastewater treatment plant operators collect data that may be used by the Shellfish Authority to manage the status of shellfish harvesting areas. Sampling activities from sewage spills, agricultural manure discharges, failing septic systems, and treatment loss at wastewater treatment plants have resulted in temporary closures of harvest areas. In turn, data collected from partner agencies has been used to identify when the pollution issue has been resolved and when the growing area can be opened. All sample analysis is completed by laboratories inspected by state regulatory agencies but have not evaluated for conformance by the FDA Shellfish LEO or FDA certified State Shellfish LEO. Washington state periodically uses laboratory analysis to determine if shellfish and shellfish harvesting areas are impacted by poisonous and deleterious substances. Shellfish closures or consumption advisories may be implemented based on this

	data. There are currently no laboratories approved by FDA Shellfish LEO for the analysis of poisonous and deleterious substances. The proposal assures that an FDA approved laboratory is required when laboratory methods and action levels are defined in the Model Ordinance and data may be used for regulatory action (marine water quality, marine biotoxins, Male Specific Coliphage). This proposal will give state shellfish authorities the flexibility to adapt to ongoing environmental conditions and make appropriate public health decisions based on laboratory data.
Cost Information	
Action by 2019 Task Force I	Recommended referral of Proposal 19-105 to an appropriate committee as determined by the Conference Chair
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-105.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-105.
Action by 2023 Supplemental Lab Data	Recommended adoption of Proposal 19-105 as amended. Chapter III. @.01 A. NSSP Conformance Required for all laboratories supporting the NSSP. For any toxin, pathogen, bacteria, virus, or other contaminant for which there is an action level specified in the NSSP and an Approved NSSP Method or Approved Limited Use Method of detection, ~~A~~all laboratory analyses ~~for compliance with classification requirements that require a specific method, actions level, and use defined in the Model Ordinance~~ generating data to support regulatory decisions shall be performed by a laboratory found to conform or provisionally conform by the FDA Shellfish LEO or FDA certified State Shellfish LEO in accordance with the requirements established under ~~the NSSP~~ Chapter I @.03 B. 1. (1) If there is a toxin, pathogen, bacteria, virus, or other contaminant for which the NSSP has no Approved NSSP Method or Approved Limited Use Method, the Authority may use a non-evaluated laboratory to generate data utilizing the best science available. In these circumstances, the Authority shall follow the procedures and guidelines defined in Chapter III @.02 Methods. (2) Shellfish growing area closures may be made using data generated in non-evaluated laboratories.
Action by 2023Task Force I	Recommends adoption of the Supplemental Lab Data Committee recommendation on Proposal 19-105.

Proposal No. 19-108

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Proposal Subject	Aquaculture Seed Shellstock
Specific NSSP Guide Reference	Section II Model Ordinance, Chapter VI. Shellfish Aquaculture, Requirements of the Authority @.02
Text of Proposal/ Requested Action	**@ .02 Seed Shellstock** A. The Authority shall establish the maximum seed size for each species of shellfish that can be produced in prohibited waters. In determining the maximum seed size Authorities shall establish sizes that require a minimum of 60~~120~~ days of growing with water temperatures over 50 degrees F to reach market size. B. For states that have not established a minimum market size, the Authority shall establish record-keeping protocols to track seed sourced from prohibited waters to ensure seed have at least 60 days of growing with water temperatures above 50 degrees F before sale for human consumption. C. ~~B.~~ The Authority shall establish appropriate corrective actions for ~~when~~ seed that exceeds the maximum seed size when it is being cultured in ~~has been produced in~~ waters classified as prohibited. D. ~~C.~~ All sources of seed produced or collected in prohibited waters shall be sanctioned by the Authority.
Public Health Significance	Existing language does not describe how the Authority should establish maximum seed size in states that have no minimum market size. Further the existing language does not require that shellfish from prohibited waters are held in waters above 50 degrees to ensure that the animals are metabolically active. Shellfish seed collected or cultured in prohibited waters have been shown through repeated sampling not to accumulate heavy metals at levels that exceed EPA alert levels. (John Mullen RI DOH, unpub. Data, Rheault unpubl. Data, Rice unpub. Data, Leavitt unpub. Data). A period of one month is typically adequate to purge bacterial contaminants provided water temperatures are high enough to maintain active metabolic activity (above 50 degrees F or 10 degrees C) (Richards 1988). Several studies have demonstrated that viral contamination in relayed or depurated shellfish is reduced to non-detect levels in 30-40 days (McLeod et. Al. 2017 and Choi and Kingsley 2016). The Authority has the option to deny seed culture in any area, or to require additional testing for deleterious substances, or to require longer purge periods as they deem necessary based on potential sources of contaminants. References Cited: Richards, G. (1988), Microbial Purification of Shellfish: A Review of Depuration and Relaying, J. Food Protection 51(3)218-251. C. McLeod et. Al. (2017) Depuration and Relaying: A Review on Potential

	Removal of Norovirus from Oysters. Comprehensive Reviews in Food Science and Food Safety, Vol.16, pp. 692-706 Choi, C. and D. H. Kingsley. Temperature-Dependent Persistence of Human Norovirus within Oysters (Crassostrea virginica). Food and Environmental Virology, 8:141-147. 2016. Supporting Information: RI DOH metals data ☹ oyster seed grown in Billington Cove Marina) Unpublished data from Rd. Dale Leavitt: (clam seed grown in Warwick Cove Marina)
Cost Information	Proposal would not impact the enforcement costs for the authority and would simplify management for growers.
Action by 2019 Task Force I	Recommended referral of Proposal 19-108 to an appropriate committee as determined by the Conference Chairperson.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-108.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-108.
Action by 2023 Aquaculture Committee	Recommended no action on Proposal 19-108. Rationale: There is not sufficient data or need for action.
Action by 2023 Task Force I	Recommends referral of Proposal 19-108 to an appropriate committee as determined by the Conference Chairperson.

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Proposal Subject	Point source approved standard station locations.
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.02 Microbiological Standards E.(3)I.
Text of Proposal/ Requested Action	I Sample station locations shall be adjacent to actual or potential sources of pollution and adequate in terms of number and spatial distribution to support the conclusion that the growing area is characterized by water quality meeting the approved classification bacteriological requirements.
Public Health Significance	Stations in waters classified as approved are frequently not adjacent to pollution sources. Stations represent a miniscule portion of points within a growing area. The stations should be located so that it is reasonable to believe that, if a station were established at any point in the area where no station currently exists, that new station would yield bacteriological data meeting the relevant bacteriological standard consistent with the classification.
Cost Information	No cost.
Action by 2019 Task Force I	Recommended referral of Proposal 19-110 to an appropriate committee as determined by the Conference Chairperson.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-110.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-110.
Action by 2023 Growing Area Classification Committee	Recommends no action on Proposal 19-110. Rationale: The proposed language is redundant with MO Section II, Chapter IV @.02 B, "Water Sample Stations. The Authority shall assure that the number and location of sampling stations is adequate to effectively evaluate all pollution sources."
Action by 2023 Task Force I	Recommends adoption of the Growing Area Classification Committee recommendation on Proposal 19-110.

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Proposal Subject	Nonpoint source approved standard station locations.
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.02 Microbiological Standards F.(6)(b)(i).
Text of Proposal/ Requested Action	(i) Sample station locations ~~are~~ shall be adequate ~~to produce the data to effectively evaluate all nonpoint sources of pollution~~in terms of number and spatial distribution to support the conclusion that the growing area is characterized by water quality meeting the approved classification bacteriological requirements;
Public Health Significance	The Model Ordinance Chapter IV.@.02B indicates "The Authority shall assure that the number and location of sampling stations is adequate to effectively evaluate all pollution sources." That includes all nonpoint sources of pollution so there is no need to state that requirement within IV.@.02F. Stations represent a miniscule portion of potential points within a growing area. The stations should be located so that it is reasonable to believe that, if a station were established at any point in the area where no station currently exists, that new station would yield bacteriological data meeting the relevant bacteriological standard consistent with the classification.
Cost Information	No cost.
Action by 2019 Task Force I	Recommended referral of Proposal 19-112 to an appropriate committee as determined by the Conference Chairperson
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-112.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-112.
Action by Growing Area Classification Committee	Recommends no action on Proposal 19-112. Rationale: The proposal language is redundant with MO Section II, Chapter IV @.02 B, "Water Sample Stations. The Authority shall assure that the number and location of sampling stations is adequate to effectively evaluate all pollution sources."
Action by 2023 Task Force I	Recommends adoption of the Growing Area Classification Committee recommendation on Proposal 19-112.

Proposal No. 19-115

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Proposal Subject	Emergency Conditions/closed status to reflect Chapter II use of harvest area
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification A. General (1) and (5)
Text of Proposal/ Requested Action	@.03 Growing Area Classification c. General. Each growing area shall be correctly classified as approved, conditionally approved, restricted, conditionally restricted, or prohibited, as provided by this Ordinance. (1) Emergency Conditions. A growing area or a portion of a growing area (harvest area) shall be placed in the closed status under Section @.03 A. (5) when unpredicted pollution conditions exist ~~which were not included in the database used to classify the area~~. If it is determined that an emergency condition or situation exists, then the growing area or harvest area will be immediately (within twenty-four (24) hours) placed in the closed status. (a) If the growing area or harvest area is already closed due to resource conservation under existing fishery laws or regulation, the area is considered to be in the closed status. If the authority choses to uses this approach, an MOU detailing coordination and, communication between agencies and patrol shall be required. ~~(a)~~(b) If no harvest areas are impacted by Emergency Conditions, placement into the closed status is not required. (2)…….. (3)..... (4) …… (5) Status of Growing Areas. The status of a growing area is separate and distinct from its classification and may be open, closed or inactive for the harvesting of shellstock. Supporting information for all changes in the status of growing areas shall be documented by a written record in the central file. (a) Open Status. Except for an area in the prohibited classification, any correctly classified growing area is normally open for the purposes of harvesting shellstock, subject to the limitations of its classification. (b) Closed Status. Any classified growing area or harvest area may be closed for a limited or temporary period

	because of: (i) An emergency condition or situation; (ii) The presence of biotoxins in concentrations of public health significance; (iii) Conditions stipulated in the management plan of conditionally approved or conditionally restricted areas; (iv) Failure of the Authority to complete a written sanitary survey or triennial review evaluation report; or (v) The requirements for biotoxins or conditional area management plans as established in Section @.04 and Section @.03, respectively, are met. I Reopened Status. A growing area or harvest area temporarily placed in the closed status as provided in (b) above, shall be returned to the open status only when:
Public Health Significance	Closed status following an emergency situation can include an entire growing area or a harvest area within the growing area; This change is consistent with Chapter II where, if appropriate, only a harvest area is closed due to an outbreak and not necessarily the entire growing area. In addition, the text stating conditions that were not included in the data base makes no sense related to emergency conditions and actually state the obvious. Deletion of that statement clarifies this part of the MO.
Cost Information	There should be no need to close an area that has no shellfish resource or is already closed by existing regulation. If this proposal is accepted by the Conference, it would save money for any state that is required to post closures in the newspaper (public notice); For Maryland the cost is ~$1500, so it would represent a significant savings.
Action by 2019 Task Force I	Recommended referral of Proposal 19-115 to an appropriate committee determined by the Conference Chair
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-115.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-115.
Action by 2023 Growing Area Classification Committee	Recommends adoption of Proposal 19-115 as amended. @.03 Growing Area Classification c. General. Each growing area shall be correctly classified as approved, conditionally approved, restricted, conditionally restricted, or prohibited, as provided by this Ordinance. (1) Emergency Conditions. A growing area or a portion of a growing area (harvest area) shall be placed in the closed status under Section @.03 A. (5) when ~~unpredicted~~ pollution conditions exist which were not included in the data used to classify the area. If it is determined that an emergency condition or situation exists, then the growing area or harvest area will be immediately (within twenty-four (24) hours) placed in the closed status. ~~(a) If the growing area or harvest area is already closed due to resource conservation under existing fishery laws or regulation, the area is considered to be in the closed status. If the authority choses to uses this approach, an MOU detailing coordination~~

	~~and, communication between agencies and patrol shall be required.~~ ~~(b) If no harvest areas are impacted by Emergency Conditions, placement into the closed status is not required.~~ (2) …….. (3)..... (4) …… (5) Status of Growing Areas. The status of a growing area is separate and distinct from its classification and may be open, closed or inactive for the harvesting of shellstock. Supporting information for all changes in the status of growing areas shall be documented by a written record in the central file. (a) Open Status. Except for an area in the prohibited classification, any correctly classified growing area is normally open for the purposes of harvesting shellstock, subject to the limitations of its classification. (b) Closed Status. Any classified growing area or harvest area may be closed for a limited or temporary period because of: (i) An emergency condition or situation; (ii) The presence of biotoxins in concentrations of public health significance; (iii) Conditions stipulated in the management plan of conditionally approved or conditionally restricted areas; (iv) Failure of the Authority to complete a written sanitary survey or triennial review evaluation report; or (v) The requirements for biotoxins or conditional area management plans as established in Section @.04 and Section @.03, respectively, are met. I Reopened Status. A growing area or harvest area temporarily placed in the closed status as provided in (b) above, shall be returned to the open status only when:
Action by 2023 Task Force I	Recommends adoption of the Growing Area Classification Committee recommendation on Proposal 19-115.

Proposal No. 19-116

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Proposal Subject	Adding a time frame to the limited or temporary period an area can be remain under a closed status prior to being reclassified.
Specific NSSP Guide Reference	Section II, Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification A. (5) (b).
Text of Proposal/ Requested Action	(b) Closed Status. Any classified growing area may be closed for a limited or temporary period, not to exceed more than one year prior to a reclassification because of: (i) An emergency…; (ii) The presence…; (iii) Conditions stipulated…; (iv) Failure of…; or (v) The requirements….
Public Health Significance	The M. O. Chapter IV @.03 A. (5) (b) states that any classified growing area may be closed for a limited or temporary period because of: (i) through (vi). The time frame "limited or temporary period "is not defined in the "Guide". The authority is required by @.03 A. (1) to place a growing area in the closed status ..." under Section @.03 A. (5) when pollution conditions exist which were not included in the database used to classify the area. If it is determined that an emergency condition or situation exists, then the growing area will be immediately (within 24 hours) placed in the closed status." Once the area is in the closed status, harvesting, attempting to harvest, possession, or sale of shellfish from the closed area is prohibited. A time limit of up to but not to exceed one year from the time the area was placed in the closed status allows the authority time with defined maximum to determine the source /cause(s) of a pollution or contamination problem before initiating a reclassification while still protecting public health by virtue of the area being in a closed status. The proposed change will not lessen public health protection.
Cost Information	Does not add any cost and may actually save administrative cost by averting multiple reclassifications in the process of sorting out the final correct classification.
Action by 2019 Task Force I	Recommended referral of Proposal 19-116 to an appropriate committee as determined by the Conference Chairperson.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-116.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-116.
Action by 2023 Growing Area Classification Committee	Recommends adoption of Proposal 19-116 as amended: (b) Closed Status. Any classified growing area may be closed ~~for a limited or temporary period, not to exceed more than one year prior to a reclassification~~ because of: (i) An emergency…; (ii) The presence…; (iii) Conditions stipulated…; (iv) Failure of…; or

	(v) The requirements….
Action by 2023 Task Force I	Recommends adoption of the Growing Area Classification Committee recommendation on Proposal 19-116.

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Proposal Subject	Marine Biotoxin Control - Public Health Reasons
Specific NSSP Guide Reference	Section III. Public Health Reasons and Explanations, Model Ordinance Chapter IV. Shellstock Growing Areas, @.04
Text of Proposal/ Requested Action	**. @.04 Marine Biotoxin Control** ***Marine Biotoxins*** Unlike human pathogens, marine biotoxins occur naturally in aquatic environments. Toxins are produced by certain micro-algae (also called phytoplankton), including dinoflagellates and others. Shellfish are filter feeders and may ingest and concentrate toxic phytoplankton from the water column when present in shellfish growing waters. Toxins are accumulated in the viscera and/or other tissues of shellfish and are transferred to humans when the shellfish are eaten (Gordon et al., 1973). Marine biotoxins are a public health concern for many reasons; for example, marine biotoxins: • May build up in shellfish in concentrations up to 100 times greater than in surrounding waters; • Are not normally destroyed by cooking or processing; • Cannot be detected by taste; and • Can cause illness and death if consumed in sufficient concentrations. In most cases, the toxin has no effect on the shellfish itself, and how long each shellfish vector remains toxic depends on the individual species in question. Additionally, there are non-traditional and emerging vectors of these toxins that also are potentially toxic foods. One example is that pufferfish, typically associated with tetrodotoxin, may also contain saxitoxin (e.g., puffers from coastal waters of Florida). Toxic dinoflagellates or diatoms are single-cell marine plants that are indigenous to most coastal and estuarine waters on the Atlantic, Gulf, and Pacific coasts of America, as well as in many other parts of the world. Dinoflagellates and diatoms in their vegetative stage flourish ("bloom") seasonally when water conditions are favorable. Blooms of these organisms can occur unexpectedly and rapidly, or may follow predictable patterns. Because dinoflagellates occur naturally, their presence in the water column does not necessarily constitute a health risk. In fact, traces of their toxin in shellfish meat does not necessarily mean they are hazardous. Toxicity depends on concentration (dose) in the shellfish. Red tide refers to the discoloration of seawater caused by blooms of marine algae. Red tides are not always red. They occur in many colors, including amber, brown,

purple, red, and pink. The relationship between red tides and biotoxin poisoning is widely misunderstood, and many people mistakenly believe that shellfish are safe to eat if no red tide is visible. While red tide can be related to harmful algae, it is helpful to remember that:

- Toxic blooms may be other colors, such as blue-green;
- Marine biotoxin poisoning can happen when there is no discoloration of the water; and
- Several marine algae that pose no public health risk to humans can turn the water red.

Diseases and Outbreaks

All humans are susceptible to shellfish poisoning. A disproportionate number of shellfish-poisoning cases occur among tourists or others who are not native to the location where the toxic shellfish are harvested, and fishermen and recreational harvesters. This may be due to disregard for either official quarantines or traditions of safe consumption.

Diagnosis of shellfish poisoning is based entirely on observed symptomatology and recent dietary history. Human ingestion of contaminated shellfish results in a wide variety of symptoms, depending on the toxin(s) present, their concentrations in the shellfish, and the amount of contaminated shellfish consumed.

Marine Biotoxin Plans – Management & Contingency

The suitability of some growing areas for shellfish harvesting is periodically influenced by the presence of marine biotoxins, such as those responsible for PSP, NSP, ASP, DSP and AZP. The occurrence of these toxins is often unpredictable, and the potential for them to occur exists along most coastlines of the United States and other countries having shellfish sanitation Memoranda of Understanding (MOU) agreements with the United States.

For this reason, even when the authority has no history or reason to expect toxin-producing phytoplankton in their growing areas, every shellfish-producing authority must have a contingency plan that defines administrative procedures, laboratory support, sample collection procedures, and patrol procedures to be implemented on an emergency basis in the event of the occurrence of shellfish toxins. For producing authorities where there is historic occurrence of toxin-producing phytoplankton and toxicity in shellfish from their growing areas, the authority must develop a management plan.

Most authorities will have a combination of management and contingency plans - management plans to address those growing areas with historic occurrence of certain toxin-producing phytoplankton, and contingency plans to address toxin-producing phytoplankton in growing areas in the event of such emergence. As an example, an authority may have statewide historical occurrence of PSP toxin-producing phytoplankton, for which it develops a management plan; however, because of a lack of illness outbreak or historical evidence of phytoplankton that produce ASP, NSP, DSP, and AZP toxins, the authority also develops a contingency plan that addresses how the authority will manage the emergence of those particular toxins.

Guidance for the development of contingency and management plans is found at Ch IV @.04.

Shellfish Meat Analyses

Laboratory methods to detect marine biotoxins in shellfish include:

- Animal bioassay;
- Biochemical;
- Rapid test kits; and
- Chemical analytical methods.

The mouse bioassay historically has been the most universally applied technique for examining shellfish toxins. Other bioassay procedures have been developed and are becoming more generally applied. In recent years, considerable effort has been appli to development of chemical analyses to replace or provide alternatives to in-vivo (liv animal) bioassays.

Marine biotoxin testing methods fall into two categories in the NSSP:

1. **Approved** (Section IV. Guidance Documents Chapter II Growing Areas .14 Table 2.)
   Approved methods are those methods that have undergone ISSC evaluation and have been adopted into the NSSP (for certain species) for regulatory decisions, including reopening a growing area after a closure.

2. **Approved Limited Use** (Section IV. Guidance Documents Chapter II Grow Areas .14 Table 4.)
   Approved limited use methods (sometimes referred to as rapid or screening methods) are testing methods that have been evaluated by the ISSC and foun fit for purpose for the NSSP, thereby providing confidence in those methods specific screening purposes. **Most limited use methods may be used for specific screening purposes, the results of which an authority may use t close a growing area; however, an approved method must be utilized to reopen an area following a closure.**

For analyses of toxins for which no method has been adopted into the NSSP, best available science is employed.

Toxin Profiles (PSP, DSP, NSP, ASP, AZP)

Paralytic Shellfish Poisoning (PSP) Toxin	
Cause	Saxitoxins are produced by the dinoflagellates of the genus *Alexandrium* (formerly *Gonyaulax*). The dinoflagellate *Pyrodinium bahamense* is also a producer of saxitoxins.
Analogs	Water-soluble alkaloid neurotoxins that are collectively referred to as saxitoxins or paralytic shellfish toxins (PSTs). To date 57 analogs have been identified, although not all are always present, and they vary greatly in overall toxicity. In addition to saxitoxin (the parent compound), monitoring laboratories typically analyze for approximately 12 other analogs that may contribute measurably to toxicity.
Occurrences	Historically, *Alexandrium* blooms have occurred between

	April and October along the Pacific coasts from Alaska to California and in the Northeast from the Canadian Provinces to Long Island Sound (US Public Health Service, 1958); but these patterns may be changing. The blooms, which may or may not result in discoloration of seawater, generally last only a few weeks and most shellfish (with the exceptions of some species of clams and scallops, which retain the toxin for longer periods) clear themselves rapidly of the toxin once the bloom dissipates.
Predictability	Toxic blooms of these dinoflagellates can occur unexpectedly or follow predictable patterns.
Action Level	0.8 ppm (80 µg/100 g) saxitoxin equivalents. Selective species closures are allowed under the NSSP. In shellfish growing areas where low levels of PSP routinely occur, harvesting for thermal processing purposes is allowed. Thermal processing is defined by FDA regulation 21 CFR 113. Thermal processing will not entirely destroy PSP content of the shellfish; therefore, the Authority must develop and implement procedures to control harvesting and transportation of shellfish intended to be processed.
Action Level Origin	The regulatory limit was set in the 1930s (Wekell, 2004). The minimum concentration of PSP toxin that will cause intoxication in susceptible persons is not known. Epidemiological investigations of PSP in Canada, however, have indicated 200 to 600 micrograms of PSP toxin will produce symptoms in susceptible persons. A death has been attributed to the ingestion of a probable 480 micrograms of PSP toxin. Investigations indicate that lesser amounts of the toxin have no deleterious effects on humans.
Monitoring	Monitoring programs for analysis of PSP toxins include: • Samples submitted by industry with a MOU. • Samples collected by shellfish authority personnel. • Sentinel species monitoring.
Shellfish Lab Methods	The mouse bioassay is still the most widely accepted detection method for the saxitoxins around the world and has been shown to adequately protect the public's health. In 2009, the Interstate Shellfish Sanitation Conference approved a post-column oxidation HPLC-PCOX method, making it the newest regulatory method available for PSP toxins in the U.S. The receptor binding assay, a competition assay whereby radiolabeled saxitoxin competes with unlabeled saxitoxin for a finite number of available receptor sites as a measure of native saxitoxin concentrations in a sample, was also approved as an official AOAC method in 2011.
Disease	Paralytic Shellfish Poisoning
Mortality	Death has been reported to occur as soon as 3 to 4 hours after consumption.
Onset	Symptoms can generally occur within 30 minutes of

		consuming contaminated seafood, although reports have indicated that symptoms can even ensue within a few minutes, if high enough toxin concentrations are present.
	Symptoms, Illness Course	Predominantly neurologic and include tingling of the lips, mouth, and tongue; numbness of extremities; paresthesias; weakness; ataxia; floating/dissociative feelings; nausea; shortness of breath; dizziness; vomiting; headache; and respiratory paralysis. Medical treatment consists of providing respiratory support, and fluid therapy can be used to facilitate toxin excretion. For patients surviving 24 hours, with or without respiratory support, the prognosis is considered good, with no lasting side effects. In fatal cases, death is typically due to asphyxiation. In unusual cases, death may occur from cardiovascular collapse, despite respiratory support, because of the weak hypotensive action of the toxin.
	General Food Associations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
	Outbreak Examples	In New England in 1972, shellfish suddenly became toxic in a previously unaffected portion of the coastline, which resulted in many illnesses (Schwalm, 1973). Despite widespread PSP closures, poisoning events still occur and are generally associated with recreational harvest. For example, in July 2007, a lobster fisherman harvested mussels from a floating barrel off Jonesport, Maine (an area that was currently open to shellfish harvesting), and he and his family ate them for dinner. All four consumers became ill with PSP symptoms, and three of them were admitted to the hospital. It was apparent that the barrel of mussels had originated further up the coast in an area that had been banned to commercial harvest (DeGrasse, 2014).
	Diarrhetic Shellfish Poisoning (DSP) Toxin	
	Cause	Certain *Dinophysis spp*. and *Prorocentrum spp*. produce okadaic acid and dinophysis toxins that cause DSP.
	Analogs	A group of lipid-soluble polyether toxins that includes okadaic acid, the dinophysistoxins, and a series of fatty acid esters of okadaic acid and the dinophysistoxins (collectively known as DSTs) (Uchida, 2018).
	Occurrence	DSP toxin-producing phytoplankton have been documented to occur off the coasts of Washington (Trainer et al., 2013) and Texas (Deeds et al., 2010) as well as off the coast in the northeast (e.g., Massachusetts [Tong et al., 2014], Maine, and Connecticut). Known global distribution of DSTs also includes Japan, Europe, Asia, Chile, Canada, Tasmania, and New Zealand (Trainer, 2013). In 2008, a large portion of the Texas Gulf Coast was closed to the harvesting of oysters due to the presence of okadaic acid in

			excess of the FDA guidance level. Although no illnesses were reported in 2008, these were the first closures in the U.S. due to confirmed toxins.
		Predictability	Dinoflagellates are known to thrive in stratified systems and Dinophysis has particular adaptive strategies to cope with freshwater plumes (Trainer, 2013).
		Action Level	0.16 ppm total okadaic acid equivalents (i.e., combined free okadaic acid, dinophysistoxins, acyl-esters of okadaic acid and dinophysistoxins)
		Action Level Origin	Established by FDA in 2011 for total (esterified plus non-esterified OA + DTXs (with no guidance for PTXs and YTXs) (Trainer, 2013).
		Monitoring	Production of DSTs has been confirmed in several *Dinophysis* species, including *D. fortii, D. acuminata, D. acuta, D. norvegica, D. mitra, D. rotundata, D. ovum, D. sacculus, D. caudate, and D. tripos*, and in the benthic dinoflagellates *Prorocentrum lima, P. concavum* (or *P. maculosum*), *P. micans, P. minimum*, and *P. redfieldii*. One other *Dinophysis* species, *D. hastate*, is also suspected to produce toxins (Trainer, 2013). Precautionary closures initiated based on cell abundance are not useful, but observations show promise in providing early warning to DSP events (Trainer, 2013).
		Shellfish Lab Methods	Until recently, DSP was managed by mouse bioassay and/or monitoring shellfish growing waters for the presence of *Dinophysis* organisms. Unfortunately, the dose-survival times for the DSP toxins in the mouse assay vary considerably, and fatty acids interfere with the assay, giving false-positive results. A suckling mouse assay has been developed and used for control of DSP. This assay measures fluid accumulation after injection of the shellfish extract. In 2017 an LCMS/MS method for quantifying DTXs in clams was approved in the NSSP. For other species, the best available science is recommended.
		Disease	Diarrhetic Shellfish Poisoning
		Mortality	This disease generally is not life-threatening.
		Onset	Onset of the disease, depending on the dose of toxin ingested, may be as little as 30 minutes to 3 hours.
		Symptoms, Illness Course	DSP is primarily observed as a generally mild gastrointestinal disorder; i.e., nausea, vomiting, diarrhea, and abdominal pain, accompanied by chills, headache, and fever. Symptoms may last as long as 2 to 3 days, with no chronic effects.
		General Food Associations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
		Outbreak Examples	Although there have been numerous outbreaks of diarrhetic shellfish poisoning around the world, until recently there were no confirmed cases of DSP in the U.S. that were due to domestically harvested shellfish (Trainer, 2013). In 2011, approximately 60 illnesses occurred in British Columbia, Canada, and 3 illnesses occurred in Washington State due to consumption of DSP-contaminated mussels. Subsequent

		harvesting closures and product recalls were issued (Lloyd, 2013).
	Neurotoxic Shellfish Poisoning (NSP) Toxin	
	Cause	NSP is caused by brevetoxins produced by the dinoflagellates of the genus *Karenia* (formerly *Gymnodinium*).
	Analogs	Comprised of more than 10 lipid-soluble cyclic polyethers. A number of analogs and metabolites have been identified. NSP-causing toxins in shellfish include intact algal brevetoxins and their metabolites (collectively known as NSTs). In addition to brevitoxins, numerous other *Karenia spp*. Found in the Gulf of Mexico and around the world regularly associated with blooms produce hymnodimine, karlotoxins, and other potent toxins (Watkins, 2008).
	Occurrence	In Gulf coast areas, toxicity in shellfish has been associated with red tide outbreaks caused by massive blooms of the toxic dinoflagellate, *Karenia brevis* (formerly *Ptychodiscus brevis*). Naturally occurs in Gulf of Mexico, Caribbean Sea, and along New Zealand coasts; it regularly produces blooms along the coasts of Florida and Texas. Blooms may cause ocean to appear red, brown, or simply darkened and are usually accompanied by massive fish kills and mortalities in marine mammals and sea birds (Watkins, 2008). Dupuration time of brevetoxins in shellfish varies, but is typically within two to eight weeks, although reports of much longer retention (nearly one year post bloom) have been documented (Watkins, 2008).
	Predictability	*Karenia* blooms show no indication of regular recurrence and shellfish generally take longer to eliminate the toxin. Blooms were once considered to be sporadic and seasonal, but historical records demonstrate these blooms have occurred in Florida almost annually in the years since the 1940s. Although more frequent in late summer and early fall, Florida blooms have been documented in almost every month of the year and may disperse in a matter of weeks, or may be present for many months at a time; in 2006, a bloom off the coast of Sarasota lasted over 12 months. Occurrence and magnitude of blooms are unpredictable.
	Action Level	0.8 ppm (20 mouse units/100 g tissue or 80 µg/100 g tissue) brevetoxin-2 equivalents The cell count of members of *Karenia brevis* in the water column exceeds 5,000 cells per liter of water.
	Action Level Origin	Uncooked clams from a batch eaten by a patient in Florida with NSP symptoms were found to contain 118 mouse units per 100 grams of shellfish meat. However, consumption of even a few contaminated shellfish may result in poisoning and the severity of the disease may be dependent on many factors, including dose, bodyweight, underlying medical conditions, and the age of the victim as well as possibly the toxin mixture of the particular bloom (Watkins, 2008).

Monitoring	Water cell counts and tissue samples.
Shellfish Lab Methods	Toxicity of shellfish exposed to the dinoflagellate *Karenia brevis* has been historically assessed by mouse bioassay in the U.S.; however, mouse bioassay is not very specific for NSP toxins (Watkins, 2008). Efforts are underway to validate *in-vitro* methods for detection of brevetoxins in shellfish. For example, rapid, sensitive ELISA test kits already are commercially available for this purpose. Biomarkers of brevetoxin contamination in shellfish have been identified by using LC/MS. Structural confirmation of these metabolites and brevetoxins in shellfish can be made by LC/MS, a method that offers high sensitivity and specificity. A method for detection, identification, and quantification of brevetoxins is HPLC-MS. Radioimmunoassay (RIA) and Receptor Binding Assay (RBA) are also under current use (Watkins, 2008). Available detection methods are not equal in their ability to measure naturally-produced brevetoxins, and most methods are hampered by the absence of specific reference standards for brevetoxin congeners (Watkins, 2008).
Disease	Neurotoxic Shellfish Poisoning
Mortality	No fatalities have been reported, but hospitalizations occur.
Onset	Onset of this disease occurs within a few minutes to a few hours. A mean time to onset of 3-4 hours has been reported in the few documented outbreaks (Watkins, 2008).
Symptoms, Illness Course	Both gastrointestinal and neurological symptoms characterize NSP, including tingling and numbness of lips, tongue, and throat; muscular aches; dizziness; diarrhea; and vomiting. Respiratory distress has been recorded. Duration is fairly short, from a few hours to several days. Recovery is complete, with few after-effects.
General Food Associations	Oysters and clams.
Outbreak Examples	The most common public health problem associated with *Karenia* blooms is respiratory irritation; however, neurotoxic shellfish poisonings associated with *Karenia brevis* blooms have been reported in Florida (US Center for Disease Control, 1973). Until NSP toxins were implicated in more than 180 human illnesses in New Zealand in 1992/1993 due to consumption of cockles and green shell mussels, NSP was considered to be an issue only in the U.S. Outbreaks of NSP are rare where programs for monitoring *K. brevis* blooms and shellfish toxicity are implemented. An NSP outbreak involving 48 individuals occurred in North Carolina in 1987 (Morris, 1991). A series of NSP cases occurred along the southwest coast of Florida, in 2006, after people consumed recreationally-harvested clams from waters unapproved for shellfish harvesting (Watkins, 2008).
Amnesic Shellfish Poisoning (ASP) Toxin	

Cause	ASP is caused by domoic acid that is produced by diatoms of the genus *Pseudonitzchia*.
Analogs	The neurotoxin domoic acid is a water-soluble, non-protein, excitatory amino acid. Isomers of domoic acid have been reported, but are less toxic than domoic acid itself. Excitatory amino acid (EAA) analogues of glutamate.
Occurrence	During a 1991-1992 incident in Washington and a 2015 event on the west coast from Washington to California, high toxin levels persisted for several months (Liston, 1994; McCabe et al. 2016). There was also an extensive event in the Northeast from Maine to Rhode Island in 2016, with different regions showing varying toxicity and species dominance within the bloom. The event started in late September in eastern Maine and ended in October; however, Rhode Island experienced another bloom in February of 2017. During 1991 and 1992, there was a spread of domoic acid producing organisms throughout the world including the detection of high numbers of the diatom *Pseudonitzschia pseudodelcatissima* in Australia and *Pseudonitzschia pseudoseratia* in California. Domoic acid has also been recovered from shellfish in Washington and Oregon.
Predictability	Blooms of *Pseudonitzschia* are of varying intensity, duration and extent. Environmental factors associated with ASP in shellfish are currently unknown.
Action Level	20 ppm domoic acid
Action Level Origin	In 1987 in eastern Canada, DA poisonings sickened individuals, leading to Health Canada's establishment of the regulatory limit. (Wekell, 2004)
Monitoring	Monitoring programs for ASP toxin are designed around the shellfish species of interest.
Shellfish Lab Methods	The mouse bioassay for domoic acid is not sufficiently sensitive and does not provide a reliable estimate of potency. The NSSP approved regulatory method for detecting domoic acid in seafood is a reversed-phase HPLC method with ultraviolet (UV) detection. There is also an AOAC approved ELISA for the detection of domoic acid.
Disease	Amnesic Shellfish Poisoning
Mortality	All fatalities, to date, have involved elderly patients.
Onset	The toxicosis is characterized by onset of gastrointestinal symptoms within 24 hours; neurologic symptoms occur within 48 hours.
Symptoms, Illness Course	ASP is characterized by gastrointestinal disorders (vomiting, diarrhea, abdominal pain) and neurological problems (confusion, short-term memory loss, disorientation, seizure, coma). Human clinical signs of domoic acid toxicity are reported as mild gastrointestinal symptoms, from an oral dose of 0.9-2.0 mg domoic acid (DA)/kg body weight. Neurologic effects, such as seizure and disorientation, are reported from an oral dose of 1.9-4.2 mg DA/kg body weight. The toxicosis

		is particularly serious in elderly patients, and includes symptoms reminiscent of Alzheimer's disease.
	General Food Associations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
	Outbreak Examples	The first human domoic acid poisoning events were reported in 1987, in Canada (Perl, 1990). While domoic acid exposure still exists, there have been no documented ASP cases since 1987, following implementation of effective seafood toxin-monitoring programs (Pulido, 2008).
		Azaspiracid Shellfish Poisoning (AZP) Toxin
	Cause	*Azadinium spp*. is the producer of azaspiracids, which cause AZP.
	Analogs	The lipid-soluble toxin azaspiracid and several derivatives (AZAs). More than 30 AZA analogs have been identified, with three analogs routinely monitored in shellfish (AZA1, AZA2, and AZA3).
	Occurrence	Coastal regions of western Europe, as well as NW Africa and eastern Canada.
	Predictability	Detected between mid-summer and mid-winter from northern/western European waters, but in certain cases, the presence of AZAs in phytoplankton does correspond to the timing of shellfish contamination, yet toxin levels in bivalves can remain elevated for 8 – 12 months following initial exposure.
	Action Level	160 µ/kg shellfish meat
	Action Level Origin	Estimation of consumption of a single portion of shellfish and through estimate of an Acute Reference Dose. Derived from epidemiological observations caused by a mixture of naturally occurring analogs (AZA 1, 2, and 3). Based on methods available in 2001.
	Monitoring	Range of species in which AZAs have been detected includes mussels (*M. edulis; M. galloprovincialis*), oysters (*Crossostrea gigas, Ostrea edulis*), scallops (*Pecten maximus*), clams (*Tapes philipinarum, Ensis siliqua, Donax* spp.), and cockles (*Cerastroderma edule*). AZAs have also been found in crustaceans. Monitoring programs will benefit from major research efforts to identify the causative organism(s) because there is often, but not always, a correlation between the presence of potentially toxigenic phytoplankton species and the subsequent accumulation of toxins in shellfish.
	Shellfish Lab Methods	AZAs are not routinely monitored in shellfish harvested in the U.S., but, in the EU, the mouse bioassay has been used. As for many of the lipophilic toxins, the mouse assay is not adequately sensitive or specific for public- health purposes. *In-vitro* assays and analytical methods are now available to assess the toxicity of AZA-contaminated shellfish and to confirm the presence of AZA analogs in shellfish. These methods are in various stages of validation for regulatory use

	around the world. LC/MS is used as a confirmatory method for AZA, providing unambiguous structural confirmation of AZA analogs in shellfish samples.
Disease	Azaspiracid Shellfish Poisoning
Mortality	No known fatalities to date.
Onset	Symptoms appear in humans within hours of eating AZA-contaminated shellfish.
Symptoms, Illness Course	Symptoms are predominantly gastrointestinal disturbances resembling those of diarrhetic shellfish poisoning and include nausea, vomiting, stomach cramps, and diarrhea. Illness is self-limiting, with symptoms lasting 2 or 3 days.
General Food Associations	Detected in mussels, oysters, scallops, clams, cockles, and crabs.
Outbreak Examples	The first case of AZP was detected in the Netherlands in 1995, where 8 people became ill after consuming mussels. From 1997 – 2000, approximately 80 individuals reported illnesses from mussels and scallops harvested from Ireland, Italy, France, and United Kingdom (Twiner, 2008). There have been no confirmed cases of AZP in the U.S. from domestically-harvested product. In 2008, the first recognized outbreak of AZP in the U.S. was reported, but was associated with a mussel product imported from Ireland (Klontz et al. 2009).

Resources

The 2012 version of FDA's Bad Bug Book, Foodborne Pathogenic Microorganisms and Natural Toxins, is a comprehensive resource from which a great deal of information has been used for the toxin profiles in the table above. It is accessible at https://www.fda.gov/media/83271/download

For more discussion of chemical structures and properties, methods of analysis, source organisms and habitat, occurrence and accumulation in shellfish, toxicity of toxins, prevention of intoxication, cases and outbreaks, and regulations and monitoring, see the FAO Paper 80: Marine Toxins. This may be accessed as follows:

Paralytic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e05.ht
Diarrhetic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0e.ht
Neurotoxic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0o.ht
Amnesic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0n.ht
Azaspiracid Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0p.ht
References	http://www.fao.org/3/y5486e/y5486e0t.htm

The FDA online course, Shellfish Growing Areas, introduces participants to requirements and procedures under the NSSP to ensure that shellfish are harvested from safe waters. The course contains a significant section addressing marine biotoxins. The course may be accessed at https://www.accessdata.fda.gov/ORAU/ShellfishGrowingAreas/SGA_summa

ry.htm.

Additional information from the Centers for Disease Control and Prevention, Morbidity and Mortality Weekly Report (MMWR) contains illness reports related to these toxins. This may be accessed at https://www.cdc.gov/mmwr/index.html.

NIH/PubMed: Various Shellfish-Associated Toxins provides a list of research abstracts in the National Library of Medicine's MEDLINE database.

The specific seafood with which each toxin generally is associated is included in the profiles above to help readers link symptoms to potential sources. However, all shellfish (filter-feeding mollusks, as well as the carnivorous grazers that feed on these mollusks (such as whelk, snails, and, in some cases, even lobster and octopus), may become toxic in areas where the source algae are present.

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~~Marine biotoxins may be ingested by molluscan shellfish feeding on toxic~~ dinoflagellates. Dinoflagellates in their vegetative stage flourish seasonally when water conditions are favorable. Toxic blooms of dinoflagellates or diatoms can occur unexpectedly or may follow predictable patterns. PSP, NSP ~~and Domoic Acid poisoning, also known as ASP are the three (3) types of~~ poisonings most commonly associated with oysters, clams, mussels and ~~scallops in the United States.~~

Cases of paralytic shellfish poisoning, including several fatalities resulting from poisonous shellfish, have been reported from both the Atlantic and Pacific coasts. The minimum quantity of poison, which will cause intoxication in the susceptible person, is not known. Epidemiological investigations of paralytic shellfish poisoning in Canada have indicated 200 to 600 micrograms of poison will produce symptoms in susceptible persons. A death has been attributed to the ingestion of a probable 480 micrograms of poison. Investigations indicate ~~that lesser amounts of the poison have no deleterious effects on humans.~~ Growing areas should be closed at a level to provide an adequate margin of ~~safety, since in many instances, toxicity levels will change rapidly.~~

A review of the literature and research dealing with the source of the poison, ~~the occurrences, and distribution of poisonous shellfish physiology and toxicology, characteristics of the poison, and prevention and control of poisoning has been prepared.~~

In Gulf coast areas, toxicity in shellfish has been associated with red tide outbreaks caused by massive blooms of the toxic dinoflagellate, *Karenia brevis* (formerly *Ptychodiscus brevis*). Toxic symptoms in mice suggest a type of NSP ~~rather than symptoms of PSP. The most common public health problem~~ associated with *Karenia brevis* blooms is respiratory irritation; however, NSP associated with *Karenia brevis* blooms have been reported in Florida. Uncooked clams from a batch eaten by a patient with neurotoxic symptoms were found to ~~contain 118 mouse units per 100 grams of shellfish meat.~~

Toxic dinoflagellates or diatoms are indigenous to most coastal and estuarine waters on the Atlantic, Gulf, and Pacific coasts of America, as well as in many ~~other parts of the world. Blooms of these organisms can occur unexpectedly~~ and rapidly. This phenomenon occurred in New England in 1972 when shellfish suddenly became toxic in a previously unaffected portion of the coastline and resulted in many illnesses. During 1991 and 1992, there was a spread of domoic acid producing organisms throughout the world including the detection of high numbers of the diatom *Pseudo-nitzschia pseudo-delcatissima* in Australia and *Pseudo-nitzschia pseudo-seratia* in California. Domoic acid was also recovered from shellfish in Washington and Oregon. All shellfish producing States or ~~MOU countries must have a contingency plan that defines administrative~~ procedures, laboratory support, sample collection procedures, and patrol procedures to be implemented on an emergency basis in the event of the

~~occurrence of shellfish toxins. A model State contingency plan for control of marine biotoxins is provided in the NSSP Model Ordinance Guidance Documents, *Guidance for Developing Marine Biotoxin Contingency Plans* (ISSC/FDA, 2017).~~

~~All States or MOU countries must monitor toxin levels to establish a baseline~~ historical reference. Thereafter, States or MOU countries where shellfish toxins are likely to occur must monitor toxin levels on a routine basis to meet the approved area requirements for direct market harvesting. Experience with ~~monitoring for shellfish toxins suggests that an effective program should include the following:~~

~~Sampling stations should be located at sites where past experience has shown toxin is most likely to appear first.~~

Samples should be collected of shellfish species which are most likely to reveal the early presence of toxin and which are most likely to show the highest toxin levels. For example, mussels have been found to be useful for early PSP ~~detection.~~

~~The frequency and period for collection of samples should be based upon historical patterns. This assumes several years of baseline data in order to establish stations and sampling plans.~~

~~An information network should be established between the health and marine resource communities and the Authority. Any toxin-like illnesses related to shellfish and environmental phenomena such as algal blooms, fish kills, or bird~~ kills, which might indicate the early stages of an increase in toxin levels, should ~~be rapidly communicated over the network.~~

~~Sampling stations and frequency of sampling should be increased when monitoring data or other information suggests that toxin levels are increasing.~~

Sample collection, sample transportation, and sample analysis procedures should be developed so that in an emergency sample results will be known ~~within twelve (12) hours.~~

When monitoring data or other information indicates that toxin levels have ~~increased to the quarantine levels, growing area closures must be immediately~~ implemented. The determination of which growing areas should be closed should include consideration of the rapidity with which toxin levels can increase ~~to excessive levels and the inherent delays in the State sample collection procedures. It may be appropriate to close growing areas adjacent to known~~ toxic areas until increased sampling can establish which areas are toxin free and ~~that toxin levels have stabilized.~~

Shellfish growing areas closed because marine biotoxins have exceeded quarantine levels may be reopened for growing after a sufficient number of samples and other environmental indices, if used, have established that the level of toxin will remain below quarantine levels for an extended period. For example, experience has shown that appropriate reopening criteria include a

minimum of three (3) samples collected over a period of at least fourteen (14) days. These samples should show the absence of PSP or levels below 80 ~~micrograms per 100 grams.~~

~~**A.** Contingency Plan.~~

The suitability of some areas for harvesting shellstock is periodically influenced by the presence of toxigenic micro-algae. Recent increases in toxigenic micro-~~algae distribution dictate that a more comprehensive series of public health~~ controls be adopted. The need exists to make contingency plans to address the contamination of a growing area by toxigenic micro-algae or a disease outbreak caused by marine biotoxin. This contingency plan must describe administrative ~~procedures, laboratory support, sample collection procedures, and patrol procedures to be implemented on an emergency basis in the event of the~~ occurrence of marine biotoxin in shellstock. The primary goal of this planning ~~should be to ensure that maximum public health protection is provided in~~ growing areas subject to marine biotoxin contamination. For a discussion of marine biotoxin disease and its management in shellfish growing areas, see the ~~NSSP Model Ordinance Guidance Documents: *Guidance for Developing Marine Biotoxin Contingency Plan* (ISSC/FDA, 2017).~~

~~**B.** Marine Biotoxin Monitoring.~~

The primary purpose of a marine biotoxin-monitoring program is to prevent illness or death among the shellfish consuming public. The monitoring program should use the "indicator station" and "critical species" concepts to develop an ~~early warning system to prevent harvest of biotoxin contaminated shellstock.~~ For a full discussion, see the NSSP Model Ordinance Guidance Documents: ~~*Guidance for Developing Marine Biotoxin Contingency Plan* (ISSC/FDA, 2017).~~

~~**C.** Closed Status of Growing Areas.~~

~~In the event of a toxigenic micro-algae bloom, shellstock growing areas shall~~ be placed in the closed status for harvesting to prevent human consumption of ~~biotoxin contaminated shellfish. The biotoxin level governing the need to~~ place the growing area in the closed status will vary depending on the species ~~of toxigenic micro-algae and the species of bivalve shellfish. Since the ability~~ to concentrate biotoxins varies among species, it is possible for one (1) species in a growing area to have safe levels of biotoxin while another species in the ~~same growing area will have dangerous biotoxin concentrations. In this~~ situation, the Authority may permit the harvesting of one (1) species with no adverse public health consequences while prohibiting the harvest of another species. In these situations, the Authority must closely monitor the growing ~~area and develop a sufficient database for use in making this determination.~~

The Authority must develop criteria, which must be met before a growing area can be returned to the open status for harvesting. These criteria should integrate public health, conservation, and economic considerations. The criteria should also employ a sufficient number of samples and other environmental indices, if used, to establish that the level of toxin will remain, for an extended period of

	~~time, at levels safe for human consumption. For additional discussion concerning biotoxin contamination of shellstock, see the NSSP Model Ordinance Guidance Documents: *Guidance for Developing Marine Biotoxin Contingency Plan* (ISSC/FDA, 2017).~~ ~~**D.** Heat Processing.~~ ~~Heat treatment can reduce the toxicity of some biotoxins. When heat treatment is used, the Authority must require that the processor provide adequate demonstration of the destruction of the biotoxin and adequate controls to assure that the end product is safe for human consumption.~~ ~~**E.** Records.~~ Good record keeping is essential to the successful management of a Marine Biotoxin Contingency Plan. Appropriate records of monitoring data, evaluation reports, and closure and reopening notices should be compiled and Recommended referral of Propossl 19-123 to an appropriate committee as esignated by the Conference Chair ~~maintained by the Authority. This~~ information is important in defining the severity of the problem, as well as for ~~a retrospective evaluation of the adequacy of the entire control program.~~
Public Health Significance	Marine biotoxins can cause injury, illness, or death. More clearly presented information will assist NSSP participants in understanding the public health reasons for marine biotoxin contingency and management plans.
Cost Information	None
Action by 2019 Task Force I	Recommended referral of Proposal 19-123 to an appropriate committee as determined by the Conference Chair.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-123.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-123.
Action by 2023 Biotoxin Committee	Recommends adoption of Proposal 19-123 as substituted. **@.04 Marine Biotoxin Control** **Marine Biotoxins Overview** Shellfish are filter feeders and, therefore, can concentrate toxic phytoplankton from the water column when present in shellfish growing waters. The toxins produced by certain species of phytoplankton can cause illness and death in humans. Toxins are accumulated in the viscera and/or other tissues of shellfish, and human exposure occurs when the shellfish are eaten (Gordan *et al.,* 1973). In most cases, the toxin has no effect on the shellfish itself, and toxin retention times vary by shellfish species. These toxins are not ~~normally~~ destroyed by cooking or processing and cannot be detected by taste. The presence of toxic phytoplankton in the water column or traces of their toxin in shellfish meat does not necessarily constitute a health risk, as toxicity is dependent on toxin concentration ~~(dose)~~ in the shellfish and amount of shellfish consumed (dose). To protect the consumer, the Authority must evaluate the concentration of toxin present in the shellfish ~~or the toxic phytoplankton concentration in the water column~~ against the levels established in the NSSP Model Ordinance to determine what action, if any, should be taken. ~~In most cases, the toxin has no effect on the shellfish itself, and toxin retention times~~

~~vary by shellfish species. Additionally, there are non-traditional and emerging food trends that can cause toxin poisoning. One example is that pufferfish, typically associated with tetrodotoxin, may also contain saxitoxin (e.g., puffers from coastal waters of Florida).~~

Toxic dinoflagellates and~~or~~ diatoms are single-cell marine ~~plants~~ algae that are indigenous to most coastal and estuarine waters on the Atlantic, Gulf, and Pacific coasts of America, as well as in many other parts of the world. Dinoflagellates and diatoms in their vegetative stage ~~flourish~~ proliferate ("bloom") seasonally when water conditions are favorable. Blooms of these organisms can occur unexpectedly and accumulate rapidly; or may follow predictable patterns.

~~Red tide refers to the discoloration of seawater caused by blooms of marine algae. Red tides are not always red. They occur in many colors, including amber, brown, purple, blue-green, red, and pink.~~ The relationship between red tides and biotoxin poisoning is widely misunderstood. Red tide refers to the discoloration of seawater caused by blooms of marine algae.~~, and many people mistakenly believe that shellfish are safe to eat if no red tide is visible.~~ While red tide ~~can~~ may be related to harmful algae, it is helpful to remember that:

- Harmful algal blooms (HABs) may be other colors (e.g. brown and green)~~, including amber, brown, purple, blue, green, and pink~~;
- Marine biotoxin poisoning can happen when there is no discoloration of the water; and
- Several marine algae species that pose no public health risk can cause water discoloration.

Diseases and Outbreaks Overview

Humans are susceptible to shellfish poisoning and although relatively few intoxications have been reported in the United States, fatalities have occurred (CDC 2022, Backer et al, 2015, Newell et al, 2022). Monitoring of water or shellfish for toxins to prevent commercial distribution of contaminated products is protective of public health, however, illnesses may also occur following self-harvest of shellfish (Watkins et al. 2008, Newell et al, 2022). Lack of awareness of closures or monitoring status, disregard for official quarantines, or failure to follow traditions associated with safe consumption might increase the risk of such illnesses.

Diagnosis of shellfish poisoning is generally based on observed symptoms and recent dietary history. Unconsumed shellfish might also be tested for algal toxins (Coleman et al, 2018). Human ingestion of contaminated shellfish results in a wide variety of symptoms, depending on the toxin(s) present, their concentrations in the shellfish, and the amount of contaminated shellfish consumed (CDC Yellow Book, 2020).

~~All humans are susceptible to shellfish poisoning, although intoxication from commercially harvested product is extremely rare.~~ Instead, a~~A~~ disproportionate ~~number of shellfish poisoning cases occur among tourists or others who are not native to the location where the toxic shellfish are harvested, and fishermen, as well as fishers~~ and recreational harvesters. ~~This may be due to lack of awareness or disregard for either official quarantines or traditions of safe consumption.~~

~~Diagnosis of shellfish poisoning is~~ generally ~~based entirely on observed symptomatology and recent dietary history. Human ingestion of contaminated shellfish results in a wide variety of symptoms, depending on the toxin(s) present,~~

~~their concentrations in the shellfish, and the amount of contaminated shellfish consumed.~~

There are five (5) types of shellfish poisonings which are specifically addressed in the NSSP Model Ordinance: paralytic shellfish poisoning (PSP), neurotoxic shellfish poisoning (NSP), amnesic shellfish poisoning or domoic acid poisoning (ASP), diarrhetic shellfish poisoning (DSP) and azaspiracid shellfish poisoning (AZP). ~~ASP (also known as domoic acid poisoning), DSP and AZP.~~ Of these five (5) types of shellfish poisoning, PSP, NSP and ASP are the most dangerous. PSP and ASP can cause death at sufficiently high exposures. In addition, ASP can cause lasting neurological damage. DSP and AZP cause similar symptoms mostly related to diarrhea and abdominal pain.

Paralytic Shellfish Poisoning (PSP)

~~PSP is caused by saxitoxins produced primarily by the certain dinoflagellates of the genus *Alexandrium* (formerly *Gonyaulax*). The dinoflagellate *Pyrodinium bahamense* is also a producer of saxitoxins.~~ PSP is caused by saxitoxins produced by certain dinoflagellates of the genus *Alexandrium* (formerly *Gonyaulax*), ~~and~~ *Pyrodinium bahamense, and Gymnodinium catenatum*. Potential symptoms of PSP are numerous and can include tingling or numbness in the face, hands, and feet;~~,~~ weakness;~~,~~ slurred speech;~~,~~ difficulty swallowing;~~,~~ shortness of breath;~~,~~ nausea;~~,~~ vomiting;~~,~~ dizziness;~~,~~ headache and high blood pressure. Onset of symptoms is typically rapid (i.e. 30 minutes or less), and death from asphyxiation can occur in some cases (Etheridge 2010 and references therein).

Historically, *Alexandrium* blooms have occurred between April and ~~October~~ December along the Pacific coast~~s~~ from Alaska to California and in the Northeast from the Canadian Provinces to Long Island Sound (U.S. Public Health Service, 1958~~);~~), but these patterns may be ~~changing~~evolving. The blooms generally last only a few weeks, and most shellfish (except for some species of clams and scallops which retain the toxin for longer periods) clear themselves rapidly of the toxin once the bloom dissipates. Toxic blooms can occur unexpectedly or follow predictable patterns.

For example, i~~I~~n New England in 1972, shellfish suddenly became toxic in a previously unaffected portion of the coastline~~,~~ which resulted in many illnesses (Schwalm, 1973). ~~Despite widespread PSP closures, poisoning events still occur and are generally associated with recreational harvest.~~ In another case, ~~For example,~~ in July 2007, a lobster fisherman harvested mussels from a floating barrel off Jonesport, Maine (an area that was ~~currently~~ open to shellfish harvesting)~~,~~ and he and his family ate them for dinner. All four consumers became ill with PSP symptoms~~,~~ and three of them were admitted to the hospital. After further investigation, ~~It~~ it became~~was~~ apparent that the barrel of mussels had originated further up the coast in an area that had been banned to commercial harvest (DeGrasse, 2014).

In 2002, the first saxitoxin event to occur in Florida waters was identified as a result of illnesses caused from consumption of pufferfish caught from the Indian River Lagoon in the Titusville area (Landsberg, 2006). This led to investigating *Pyrodinium bahamense* presence in the lagoon system as this species could cause shellfish toxicity. Shellfish meat samples collected in the Indian River Lagoon for *Pyrodinium bahemense* were found to test positive for saxitoxin. Initial shellfish samples collected showed only trace amounts of saxitoxin. As a result, the State of Florida integrated a monitoring program for PSP in the state's biotoxin management plan as it relates to molluscan shellfish. Over the years Florida has had growing area closures due to PSP

but no illnesses due to shellfish consumption. Historically, *Pyrodinium bahamense* blooms have occurred between April and October along the east and west coasts of Florida.

Neurotoxic Shellfish Poisoning (NSP) – ~~TABLE DISCUSSION ON NSP UNTIL NEXT MEETING AS MORE INFROMATION IS NEEDED. NONE OF THE SUGGESTED CHANGES BELOW HAVE BEEN INCORPORATED.~~

From the Carolinas through the Gulf coast states~~In the United States,~~ NSP is caused by brevetoxins that are primarily produced by the dinoflagellate *Karenia brevis*~~s of the genus *Karenia*~~ (formerly of the genus *Gymnodinium*). ~~From the Carolinas through the Gulf coast states, toxicity in shellfish has been associated with red tide outbreaks caused by massive blooms of the toxic dinoflagellate, *Karenia brevis*.~~ The most common public health problem associated with *Karenia* blooms is respiratory irritation; however, neurotoxic shellfish poisonings associated with *Karenia brevis* blooms have been reported in Florida (Center for Disease Control, 1973 [a] and [b]). Onset of symptoms can occur within 18 hours of exposure, although an average onset time has been noted as three to four hours following consumption (Grattan et al 2016). Gastrointestinal symptoms are commonly reported, but neurological symptoms such as numbness and tingling in the face, hands, and feet;~~,~~ partial limb paralysis;~~,~~ slurred speech;~~,~~ loss of coordination; and even reversal of hot and cold sensations have also occurred (Watkins et al 2008).~~It regularly produces blooms along the coasts of Florida and Texas. Blooms may cause ocean~~ If seawater is colored, it may to ~~appear red, brown, or simply darkened and are usually accompanied by massive fish kills and mortalities in marine mammals and sea birds (Watkins, 2008).~~

Karenia brevis ~~blooms show no indication of regular recurrence and shellfish generally take longer to eliminate the toxin. B~~blooms were once considered to be sporadic and seasonal, but historical records demonstrate these blooms have occurred in Florida almost annually in the years since the 1940s. They now regularly occur along the Gulf Coast between Florida and Texas, and a~~A~~lthough more ~~-~~frequent in late summer and early fall, Florida blooms have been documented in almost every month of the year and may disperse in a matter of weeks, or may be present for many months at a time~~; in 2006, a bloom off the coast of Sarasota lasted over 12 months~~. Occurrence and magnitude of blooms are unpredictable. If seawater is colored during a bloom, it may appear red, brown, or simply darkened, and blooms are usually accompanied by fish kills and mortalities in marine mammals and sea birds (Watkins, 2008).

Amnesic Shellfish Poisoni~~o~~ng (ASP)~~ – TABLE DISCUSSION ON ASP UNTIL NEXT MEETING AS MORE INFROMATION IS NEEDED about toxin producers and confirm not duplicating language from 19-124.~~

ASP is caused by domoic acid, which is produced by certain diatoms of the genus *Pseudo-nitzschia*. *Pseudo-nitzschia australis* and *Pseudo-nitzschia multiseries* are commo~~m~~n toxin producers on the west coast and in the Northeast, while members of the *Pseudo-nitzschia pseudodelicatissima*-complex are common toxin producers in the Gulf of Mexico. However, there are multiple potential toxic species in each region, and *Pseudo-nitzschia cuspidata~~e~~* has resulted in at least one (1) west coast and one (1) Bay of Fundy closure. ~~—~~

~~—~~

Acute exposure to domoic acid can cause nausea, diarrhea, headaches,

confusion/disorientation, seizures, and most severely, permanent short-term memory loss, coma, or death (Lefebvre and Robertson 2010, Shumway et al 2018). Onset of these symptoms can occur within 24 to 48 hours of consumption (Perl et al 1990, Grattan et al 2016). The effects of chronic, low-level exposure to domoic acid through shellfish consumption are still being studied, but potential impacts include impairment of fetal development, memory deficits, and kidney damage (Grattan et al. 2018 and Funk et al. 2014).

~~Paragraph to be drafted. (Bryant)~~

The factors which influence domoic acid production are not well understood but may include irradiance levels, photoperiod length, salinity, trace metals including iron and copper, the presence of marine bacteria, and decreased or halting cellular growth (Doucette et al. 2008, Lelong et al. 2014, Cusack et al. 2002). Nutrient limitations are suggested to influence species diversity which, at times, may favor toxin-producing species but studies are also underway to determine if nutrient limitations may influence domoic acid production (Thorel et al. 2017).

~~The effects of chronic, low-level consumption of domoic acid are being studied and may lead to the impairment of fetal development, memory deficits, and kidney damage (Grattan et al. 2018 and Funk et al. 2014).~~

Blooms of *Pseudo-nitzschia* are of varying intensity, duration and extent. During a 1991-1992 incident in Washington and a 2015 event on the west coast from Washington to California, high toxin levels persisted for several ~~months~~ years (Liston, 1994; McCabe et al. 2016). There was also an extensive event in the Northeast from Maine to Rhode Island in 2016, with different regions showing varying toxicity and species dominance within the event~~bloom~~. The event started in late September in eastern Maine and ended in October; however, Rhode Island experienced another bloom in February of 2017. ~~The NSSP Model Ordinance requires that growing areas be placed in the closed status when the domoic acid concentration is equal to or exceeds 20 parts per million raw shellfish.~~

Diarrhetic Shellfish Poisoning (DSP)

~~DSP is caused by okadaic acid and related congeners (e.g., *dinophysis* toxins) produced primarily by dinoflagellates of the genus *Dinophysis*. Eight species of the genus *Dinophysis* are toxigenic (*D. acuminata, D. acuta, D. caudata, D. fortii, D, norvegica, D. ovum, D. sacculus, D. tripos*). All eight species are present on the U.S. east coast and Gulf of Mexico; five species (*D. acuminata, D. acuta, D. fortii, D. norvegica, D. tripos*) are present on the U.S. west coast. The dinoflagellate *Prorocentrum lima* and two species of *Phalacroma (P. rotundatum* and *P. mitra*) also produce DSP toxins. (Anderson, 2021) *Procentrum lima and Phalacroma rotundatum* are present on the U.S. east coast, west coast and Gulf of Mexico. *Phalacroma mitra* is present in the U.S. Gulf of Mexico.~~

~~A 2016 *Dinophysis norvegica* bloom in a Maine salt pond led to the identification of a toxin previously unknown to occur in shellfish, dihydrodinophysistoxin-1. Studies are occurring to determine the potency of the new toxin relative to regulated DSP toxins.~~

~~**Note: will be obtaining specific language**~~

Diarrhetic Shellfish Poisoning (DSP) is caused by okadaic acid and related congeners (e.g., dinophysis toxins) produced primarily by dinoflagellates of the genus *Dinophysis*. Typical symptoms of DSP include abdominal pain, nausea and vomiting, diarrhea, headache, fever, and chills, with a short onset time and symptoms lasting up to three days (Lloyd 2013, US National Office for HABs 2019). Eight *Dinophysis* species known to occur in U.S. waters, including *D. acuminata, D. acuta, D.*

caudata, D. fortii, D. norvegica, D. ovum, D. sacculus, and *D. tripos*, as well as the dinoflagellate *Prorocentrum lima* and two species of *Phalacroma* (*P. rotundatum* and *P. mitra*) are all known to produce toxins (Reguera et al 2014). All eight *Dinophysis* species are present on the U.S. east coast and Gulf of Mexico, while five species (*D. acuminata, D. acuta, D. fortii, D. norvegica*, and *D. tripos*) are present on the U.S. west coast. *Prorocentrum~~.~~ lima* and *Phalacroma~~.~~ rotundatum* are present in U.S. east coast, west coast, and Gulf of Mexico waters, while *Phalacroma~~.~~ mitra* has only been found in the Gulf of Mexico. DSP toxin profiles vary by species and strain (Anderson 2021).

A 2016 *Dinophysis norvegica* bloom in a Maine salt pond led to the identification of a toxin previously unknown to occur in shellfish, dihydrodinophysistoxin-1 (Deeds et al 2020). ~~As of 2021, studies are occurringbeing carried out to determine the potency of the new toxin relative to regulated DSP toxins.~~

Although there have been numerous outbreaks of DSP around the world, no confirmed cases of DSP in the U.S. that were due to domestically harvested shellfish occurred prior to 2011 (Trainer 2013). A cluster of DSP illnesses, with DSP toxins confirmed in blue mussels (Mytilus edulis), occurred in Washington state in July 2011 (3 persons; Lloyd 2013) and in British Columbia, Canada in July-August 2011 (62 persons; Taylor 2013). Subsequent harvesting closures and product recalls were issued. DSP toxins have been detected at levels exceeding the guidance level~~FDA regulatory limit~~ in the Eastern oyster (*Crassostrea virginica*; Texas; Campbell 2010; Deeds 2010)~~;~~, the Pacific oyster (*Crassostrea gigas*), varnish clam (*Nuttalia obscurata*), ~~and~~ manila clam (*Venerupis philippinarum*) (Washington; Trainer 2013),~~;~~ California mussels (*Mytilus californianus*) from Washington and Monterey Bay, CA (Trainer 2013; Schultz 2019)~~;~~, and various commercial and non-commercial shellfish species from New York, Massachusetts, and Maine~~, Delaware, and Maryland~~ waters (Hattenrath-Lehmann et al 2013, Deeds et al 2020, Trainer et al. 2013 ~~Wolny et al 2020,~~ Anderson 2021).~~.;~~ ~~andThey~~DSP toxins have also been detected in non-commercial shellfish ~~in~~during research studies in Mid-Atlantic states. (Hattenrath-Lehmann et al 2013, Wolny et al 2020, Anderson 2021).

~~**Discussion tabled in 6/1/21 at this point**~~

~~Certain *Dinophysis* spp. and *Prorocentrum* spp. produce okadaic acid and dinophysis toxins that cause DSP. DSP toxin-producing phytoplankton have been documented to occur off the coasts of Washington (Trainer et al. 2013) and Texas (Deeds et al. 2010) as well as off the coast in the Northeast (e.g., Massachusetts [Tong et al. 2015]). Dinoflagellates are known to thrive in stratified systems and *Dinophysis* has adaptive strategies to cope with freshwater plumes (Trainer, 2013).~~

~~Although there have been numerous outbreaks of diarrhetic shellfish poisoning around the world, until recently there were no confirmed cases of DSP in the U.S. that were due to domestically harvested shellfish (Trainer, 2013). In 2011, approximately 60 illnesses occurred in British Columbia, Canada, and three illnesses occurred in Washington State due to consumption of DSP-contaminated mussels. Subsequent harvesting closures and product recalls were issued (Lloyd, 2013).~~

Azaspiracid Shellfish Poisoning (AZP)

AZP is caused by azaspiracids produced by certain dinoflagellates of the genus *Azadinium and Amphidoma*. Compared to the other biotoxins discussed, AZP has

been much less studied globally and within the United States, with only limited monitoring data available. Azaspiracids have been detected in seawater on both the wWest cCoast, in Washington (Puget Sound) (Trainer et al. 2013, Kim et al. 2017, Anderson et al. 2021) and the eEast cCoast, in Virginia (Chesapeake Bay and VA coastal bays) (Onofrio et al. 2021). Harvesting closures in the United States have not been documented due to AZP toxins. Toxic blooms are known to occur in coastal regions of western Europe (James et al. 2002, Tillman et al. 2017) and northwestern Africa (Taleb et al. 2006) .

~~*Azadinium* spp. is the producer of azaspiracids, which cause AZP. While AZP has occurred in the U.S., the contaminated shellfish was imported (Klontz et al. 2009). Harvesting closures in the U.S. have not been documented due to AZP toxins. Toxin blooms are known to occur in coastal regions of western Europe as well as northwestern Africa and eastern Canada.~~
Symptoms of AZP are similar to those noted with DSP, and include nausea, vomiting, cramps, and diarrhea, with symptoms typically persisting for two to three days from onset (Furey et al 2010, Shumway et al 2018).

The first case of AZP was detected in the Netherlands in 1995, where eight people became ill after consuming mussels harvested at Killary Harbour, Ireland (McMahon and Silke 1996). From 1997 ~~–~~through 2000, approximately 80 individuals reported illnesses from mussels and scallops harvested from Ireland, Italy, France, and the United Kingdom (Twiner, 2008). There have been no confirmed cases of AZP in the U.S. from domestically harvested product. In 2008, the first recognized outbreak of AZP in the U.S. was reported but was associated with a mussel product imported from Ireland (Klontz et al. 2009).

Marine Biotoxin Plans – Management & Contingency

The suitability of some growing areas for shellfish harvesting is periodically influenced by the presence of marine biotoxins. The occurrence of these toxins is often unpredictable, and the potential for them to occur exists along most coastlines of the United States and other countries ~~having~~ with shellfish sanitation ~~Memoranda of Understanding (MOU) agreements~~ arrangements with the United States. The unpredictability in occurrence of toxic blooms was demonstrated in New England in 1972 ~~when shellfish suddenly became toxic in a previously unaffected portion of the coastline and resulted in many illnesses~~ (Schwalm, 1973).

For this reason, even when the authority has no history or reason to expect toxin-producing phytoplankton in their growing areas, every shellfish-producing authority must have a contingency plan that defines administrative procedures, laboratory support, sample collection procedures, and patrol procedures to be implemented on an emergency basis in the event of the occurrence of shellfish toxins. For producing authorities where there is historic occurrence of toxin-producing phytoplankton and toxicity in shellfish from their growing areas, the authority must develop a management plan for those toxin groups.

Most authorities will have a combination of management and contingency plans~~-management plans~~. Management plans are used to address those growing areas with historic occurrence of certain toxin-producing phytoplankton~~,~~ and contingency plans are used to address toxin-producing phytoplankton in growing areas in the event of such emergence. As an example, an authority may have statewide historical occurrence of PSP toxin-producing phytoplankton, for which it develops a management plan; however, because of a lack of illness outbreak or historical evidence of phytoplankton that produce ASP, NSP, DSP, and AZP toxins, the

authority also develops a contingency plan that addresses how the authority will manage the emergence of those toxins.
~~those toxins.~~

Guidance for the development of contingency and management plans is found in Section IV Guidance Documents, Chapter II Growing Areas @.02.

Resources

U.S. National Office for Harmful Algal Blooms, https://Hab.whoi.edu

Food and Drug Administration, Marine Biotoxin Management for Molluscan Shellfish V1_2, https://collaboration.fda.gov/biotoxins/?elq=f3a546ff4e224fca89660b1cf26461f9&elqCampaignId=5608&elqTrackId=de384479b4e8416997f078b1277d4578&elqaid=6833&elqat=1&utm_campaign=Seafood+Safety+Update+-+Marine+Biotoxin+Video&utm_medium=email&utm_source=govdelivery

Woods Hole Oceanographic Institution, Anderson Lab, https://www2.whoi.edu/site/andersonlab/

U.S. Center for Disease Control, Harmful Algal Bloom Overview, https://www.cdc.gov/habs/illness-symptoms-marine.html
National Oceanic and Atmospheric Administration, Harmful Algal Bloom Overview, https://www.noaa.gov/what-is-harmful-algal-bloom

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Paralytic Shellfish Poisoning (PSP)[1,2,3]	
Toxin	Saxitoxins
Causative Organism(s)	*Alexandrium sp.* ; *Pyrodinium bahamense*; *Gymnoinium catenatum*
Historic Geographic Range (US)	Alexandrium sp.~~Northeast~~ Atlantic coast from New York to Maine; Pacific coast from Alaska to California; *Pyrodinium bahamense*- Gulf; and Atlantic coasts of Florida; *Gymnodinium catenatum* - Gulf coast
Onset/Duration	Onset within 30 minutes; Duration of a few hours to a few days
Major Symptoms	Tingling or numbness in face, hands, and feet; weakness; slurred speech; difficulty swallowing; shortness of breath; nausea; vomiting; dizziness; headache; high blood pressure. Death from asphyxiation can occur.

Neurotoxic Shellfish Poisoning (NSP)[1,4,5]	
Toxin	Brevetoxins
Causative Organism(s)	*Karenia brevis*
Historic Geographic Range (US)	Gulf ~~Coast~~ coast and ~~east~~ Atlantic coast of Florida; One instance ~~in~~ on the Atlantic coast of North Carolina
Onset/Duration	Onset within three~~3~~ to four~~4~~ hours or up to 18 hours; Duration of two to three days
Major Symptoms	Gastrointestinal symptoms; numbness and tingling in the face, hands, and feet; partial limb paralysis; slurred speech; loss of coordination; reversal of hot and cold sensations

Amnesic Shellfish Poisoning (ASP)[1,3,6]	
Toxin	Domoic Acid
Causative Organism(s)	*Pseudonitzschia sp.*
Historic Geographic Range (US)	~~Northeast~~ Atlantic coast from New York to Maine; Gulf ~~Coast~~coast; Pacific coast from Alaska to California

Onset/Duration	Onset within 24-48 hours; Duration of certain symptoms can be months to years or permanent
Major Symptoms	Nausea, diarrhea, headache, confusion/disorientation, seizures. Can cause short-term memory loss, coma, or death.

Diarrhetic Shellfish Poisoning (DSP)[1,3,7]	
Toxin	Okadaic Acid
Causative Organism(s)	*Dinophysis sp. ; Prorocentrum lima ; Phalacroma rotundatum* and *mitra*
Historic Geographic Range (US)	~~East~~ Atlantic coast from Virginia to Maine; Gulf ~~Coast~~coast; Pacific ~~Coast~~ coast from Washington to California
Onset/Duration	Onset from 30 minutes to 15 hours; Duration up to three days
Major Symptoms	Abdominal pain; nausea; vomiting; diarrhea; headache; fever and chills

Azaspiracid Shellfish Poisoning (AZP)[1,3,8]	
Toxin	Azaspiracids
Causative Organism(s)	*Azadinium sp.*
Historic Geographic Range (US)	No known occurrences
Onset/Duration	Onset within hours; Duration up to three days
Major Symptoms	Abdominal pain; nausea; vomiting; diarrhea

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Action by 2023 Task Force I	Recommends adoption of the Biotoxin Committee recommendation on Proposal 19-123.

Proposal No. 19-124

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Proposal Subject	Marine Biotoxin Control – Guidance Document
Specific NSSP Guide Reference	Section IV Guidance Documents Chapter II. Growing Areas Chapter IV. Shellstock Growing Areas .02
Text of Proposal/ Requested Action	**.02 Guidance for Developing Marine Biotoxin Contingency and Management Plans.** Regardless of whether a growing area has a history of toxin-producing phytoplankto being able to detect occurrences and take appropriate action to prevent contaminated product from entering commerce is an important part of marine biotoxin control. There are two types of plans defined in the NSSP MO for the control of marine biotoxins: a *contingency plan* and a *management plan*. The *contingency plan* is primarily for reactive management to an illness outbreak or emergence of a toxin-producing phytoplankton in a growing area that has not historically occurred before. The contingency plan is only appropriate for a shellfish Authority that has no history or reason to expect toxin-producing phytoplankton in th growing areas. The primary goal of the contingency plan is to detect emerging toxins and to outline response activities necessary to prevent additional illnesses (if illness already occurred) and protect the public's health. The *management plan* is primarily for proactive management of marine biotoxins in growing areas with a history of toxin-producing phytoplankton and toxicity in shellfi and/or a previous illness event or outbreak. A management plan is required for a shellfish authority that has a history of toxin-producing phytoplankton, toxicity in shellfish and/or an illness event or outbreak attributed to their growing areas. A shellfish authority might have a management plan for certain marine biotoxins, lik PSP toxins, but a contingency plan for toxins like AZP toxins. **General Plan Elements** Whether the authority is developing a plan to manage biotoxins, or a contingency pla for the unexpected, the plan should address the following elements: • Statutory and/or Regulatory Authorities • Resource/Growing Areas and Species • Communication • Control & Response • Growing Area Reopening Criteria • Recordkeeping • Post Event Actions

- Plan Testing, Post Event Activities

Recommended General Plan Guidelines

**Statutory and/or Regulatory Authorities*

The authority should prepare a summary of the laws and regulations in the state (or MOU country) that allow the authority to promptly and effectively take actions to prevent or remove potentially toxic shellfish from commerce in the event of a marine biotoxin event, including:

1. close a growing area to harvest;
2. embargo shellfish that has not entered commerce;
3. prevent harvesting of contaminated species;
4. provide for embargo and/or recall of any potentially toxic shellfish already o the market; and
5. withdraw interstate shipping permits.

**Resource/Growing Areas and Species*

As is the case in several aspects of the NSSP MO, the plan should include a list or reference to a list of locations of classified shellfish growing areas and the species present in the area. This is especially important if the authority intends to implement species-specific biotoxin closures as part of the plan.

**Communication*

Information-sharing among government and non-government agencies is critical as p of an effective biotoxin plan, whether contingency or management. As such, the authority should establish and formalize channels of communication with appropriat partner agencies (e.g., wildlife, epidemiology, local health, public safety, public heal and environmental), research or academic organizations (e.g., marine biologists), adjacent shellfish control authorities, industry, and other similar partners in advance any serious biotoxin event.

Information to be communicated includes that which is relevant to early warning as as control and response, including:

1. abnormal environmental phenomenon that may be associated with a shellfish growing area (e.g., bird, fish, or marine mammal die-offs or abnormal behavior, or water discoloration);
2. occurrences of toxic phytoplankton blooms;
3. toxin-like illness reports in humans;
4. growing area closures (specifically, disseminating information on occurrences and/or toxicity in shellfish meats to adjacent states, industry and local health agencies);
5. coordination of control activities taken by state and federal agencies or departments and district, regional, or local health authorities (e.g., patrol legal actions); and
6. consumer educational outreach during growing area closure periods.

This aspect of the plan may include references to Memoranda of Understanding and tables that outline each partner's roles and responsibilities, and procedures that defin how agencies will maintain contact lists. Model press releases, email notifications, a similar templates may also be useful.

**Control and Response Activities*

An authority's plan should include the following elements to address control and response activities:

1. Growing Area Closure Criteria

   An authority's plan (either contingency or management) should define the circumstances under which the authority will place a growing area in the clo status due to marine biotoxin contamination. The criteria should integrate pu health and economic considerations. Principle considerations include

   * The rapidity with which toxin levels can increase to excessive levels
   * Inherent delays in sample collection and results;
   * The number of samples required to initiate action;
   * The size of the area to be closed, including a safety zone (it may be appropriate to close harvesting areas adjacent to known toxic areas u increased sampling can establish which areas are toxin free and that toxin levels have stabilized); and
   * The type of harvesting restrictions to be invoked (all species or spec species).

   The biotoxin level governing the need to place the growing area in the close status may vary depending on the species of phytoplankton and the species o bivalve shellfish. Since the ability to concentrate biotoxins varies among species, it is possible for one species in a growing area to have safe levels of biotoxin while another species in the same growing area will have dangerou biotoxin concentrations. In this situation, the authority may allow the harves of one species with no adverse public health consequences while prohibiting harvest of another species. In these situations, the authority must closely monitor the growing area and develop a sufficient database for use in makin this determination.

2. Administrative Actions

   The authority should specify the administrative procedures, including timeframes, necessary to place growing areas in the closed status, identify potentially contaminated shellfish products, determine the distribution of the products, and initiate embargo and/or recall activities.

3. Other Control Activities.

   If the authority's statutes or regulation do not allow for a certain administrati action and/or the authority must seek a court order or other legal action, the authority should define the procedures and timeframes, where applicable.

   The authority should also refer to, or describe patrol activities relative to growing area closures due to marine toxins.

**Growing Area Reopening Criteria*

	The authority's plan should describe how the authority determines that shellfish for commercial harvest in a growing area are safe for harvest and distribution into commerce for human consumption following an event. The protocol should reflect th authority's consideration of the public's health, and economic consequences. A system of representative samples and other environmental indices are typically use to establish detoxification curves indicating that the level of toxin or cell counts have decreased to acceptable levels. Several authorities require that three (3) samples collected over a period of fourteen (14) days show results below the quarantine limit before reopening the affected area. **Routine Monitoring Program* A routine surveillance monitoring program (also referred to as an early warning phytoplankton and/or shellfish-monitoring program) is recommended as part of a marine biotoxin control plan to detect the presence of a "bloom." In describing this program, the authority should include: 1. Geographic Distribution of Primary Sampling Stations For both phytoplankton and shellfish monitoring plans, primary sampling stations (also referred to as indicator or sentinel stations) should be located a sites where toxin is most likely to first appear, based either on past experienc or knowledge of site conditions. The geographic distribution for collection o samples should take into consideration the randomness of toxic algal blooms For these reasons, several years of baseline data are often necessary in order establish stations. To facilitate knowledge transfer, it is advisable that the authority describe its rationale in selecting sampling sites. 2. Determination of Species to be Sampled For a monitoring plan, sampling design should always take into account wha commercially-harvested species are present in the growing area and samples should be collected of species which are most likely to reveal the early prese of toxin and are most likely to show the highest toxin levels. For example, mussels have been found to be useful for early detection of an event. 3. Frequency and Timing of Sample Collection 4. Just as location of sampling sites should be carefully considered, the authorit should establish the frequency and period for collection of samples in order t identify an event as early as possible. Historical occurrences and fluctuation coastal phytoplankton populations due to the influence of meteorological an hydrographic events are important considerations. For example, a large rain storm may cause nutrient loading in coastal waters and trigger a toxic phytoplankton bloom or a hurricane may drive offshore phytoplankton bloo onshore. As well, uptake rates for various species of shellfish being tested is critical in terms of timing. 5. Sample Collection Procedures 6. Sample collection, sample transportation, and sample analysis procedures should be developed and predictable timeframes established between collection and results. The Authority should ensure that in an emergency, such as a suspected biotoxin illness, the normal timeframe can be compressed and sample results known as quickly as possible. It is important to consider emergency coverage schedules for staff and lab availability outside of normal office hours during harmful algal bloom events.

7. Identification of Laboratories/Analysts;
Biotoxin sample results must be provided by an NSSP conforming lab that is utilizing an approved or limited use method. For checklist requirements and additional guidance regarding laboratory evaluation for conformance, see Chapter II Growing Areas. For NSSP requirements, see Section II MO, Cha I Shellfish Sanitation Program, @.03(B).

The Authority should consider where they can access sample processing for biotoxins that occur or may occur within their jurisdiction, and identify alternative laboratory support, should that support become necessary.

8. Description of Testing Methods, Which May Include Approved Limited Use and Approved Methods
To control marine biotoxins, the authority must evaluate the concentration o toxin present in the shellfish. In the case of NSP, phytoplankton must be monitored as well as shellfish. Approved and limited use methods are listed the NSSP Guidance Documents.

9. Establishment of Appropriate Screening Levels
Though the NSSP establishes the toxin levels in shellfish at which a growing area must be closed, many programs implementing early warning systems include phytoplankton cell counts. Additionally, shellfish toxin levels that a below the regulatory levels may trigger emergency or expanded testing, or precautionary closures. Growing areas should be closed at a level that provid an adequate margin of safety, since in many instances, toxicity levels will change rapidly and the time between sampling and results should be conside Precautionary closures can be made in order to prevent the harvest of potentially toxic shellfish while sample results are being collected and processed.

10. Procedures to Expand Sampling if Toxin Levels or Cell Counts Indicate a Harmful Algal Bloom.
When an early warning system detects increased toxicity/cell counts or other information suggests that toxin levels are increasing, it is important that the authority have procedures to promptly expand sampling to additional station and/or increase the frequency of sampling for marine biotoxins. The procedu should include plans for obtaining the additional resources necessary to implement the expanded sampling and laboratory analysis program.

If a plan consists of water sampling for phytoplankton cell counts as surveillance, the authority should identify its plan to be able to initiate an emergency shellfish sampling program

**Recordkeeping*

Records generated as part of a marine biotoxin program may be important in definin the severity of an event, as well as for retrospectively evaluating the adequacy of the entire control program.

The NSSP requires certain biotoxin-related records be maintained. As such, authorit plan should define records to be generated, reviewed, and maintained. Required reco include:

* Monitoring data, including shellfish and phytoplankton and water sample analyses results, relating to levels of marine biotoxins in each growing area;
* Closure and reopening notices;
* Investigation-related documents, including sample results;
* Recall-related records, including public warnings, notification to other states involved in the recall, FDA, and ISSC, recall status reports in accordance with Section II, Chapter II Risk Assessment and Risk Management, @.01(I); and
* Evaluation reports, which may include analyses of trends and detoxification curves.

An authority may also consider maintaining

- Records of reported illnesses that include data on the incidence of illness and appropriate case history data; and
- Pertinent environmental observations.

Whenever possible, the authority's servicing laboratory should archive shellfish homogenates for additional analysis.

**Plan Testing, Post Event Activities*

The authority should test the plan periodically to ensure prompt implementation in th event it is needed. As well, the authority should routinely review data post-event to improve aspects of the authority's plan. Because historical information plays such a critical role in the authority's plan, authorities are highly encouraged to document rationale for significant changes.

Heat Processing.

In shellfish growing areas where low levels of PSP routinely occur, harvesting for thermal processing purposes may be an alternative to consider. Thermal processing, as defined by applicable FDA regulations (21 CFR 113), will reduce the toxin concentration of certain toxins in the shellfish via dilution, not destruction.

If thermal processing is practiced, the authority must develop and implement procedures to control the harvesting and transportation of the affected shellfish to the processing plant; and must require that the processor provide adequate demonstration of the destruction of the biotoxin and adequate controls to assure that the end product is safe for human consumption.

~~NSSP guidance documents provide the public health principles supporting major~~ components of the NSSP and its Model Ordinance, which includes the requirement ~~the program . NSSP *Model Ordinance* requirements apply only to interstate~~ commerce although most states apply the requirements intrastate. For the most up ~~date and detailed listing of requirements, the reader should consult the most recent edition of the Model Ordinance.~~

~~**Introductin**~~

~~Shellfish are filter feeders and, therefore, they have the ability to concentrate toxic~~ phytoplankton from the water column when present in shellfish growing waters. T ~~toxins produced by certain species of phytoplankton can cause illness and death in~~ humans. Toxins are accumulated in the viscera and/or other tissues of shellfish and ~~are transferred to humans when the shellfish are eaten (Gordan *et al.*, 1973). These~~ toxins are not normally destroyed by cooking or processing and cannot be detected taste. The presence of toxic phytoplankton in the water column or traces of their to in shellfish meat does not necessarily constitute a health risk, as toxicity is depende on concentration (dose) in the shellfish. To protect the consumer, the Authority m evaluate the concentration of toxin present in the shellfish or the toxic phytoplankto concentration in the water column against the levels established in the NSSP Mode ~~Ordinance to determine what action, if any, should be taken.~~

While there is a wide range of methodologies developed for screening and confirmat of toxic phytoplankton and their toxins, methods must be adopted into the NSSP if th are to be implemented for the confirmation of toxins for making decisions to reopen growing areas. Additionally, there are screening methods that have been evaluated b the ISSC and found fit for purpose for the NSSP, thereby providing confidence in th methods for specific screening purposes. Toxin methods fall into two categories in t ~~NSSP: Approved Methods for Marine Biotoxin Testing (Section IV. Guidance~~ Documents Chapter II Growing Areas .14 Table 2.) and Approved Limited Use Methods for Marine Biotoxin Testing (Section IV. Guidance Documents Chapter II ~~Growing Areas .14 Table 4.). These methods range from mouse bioassays to immunochromatography and other antibody based platforms to chemical analytical~~ methods such as high performance liquid chromatography (HPLC). Information available in the referenced Tables above provides references for the methods and, as applicable, and limitations placed on the use of the method within the NSSP. For to that have no method adopted into the NSSP, best available science is employed.

There are five (5) types of shellfish poisonings which are specifically addressed in th ~~NSSP Model Ordinance: Paralytic Shellfish Poisoning (PSP), Neurotoxic Shellfish~~ Poisoning (NSP), Amnesic Shellfish Poisoning (ASP), also known as Domoic Acid poisoning, Diarrhetic Shellfish Poisoning (DSP) and Azaspiracid Shellfish Poisoning (AZP). Of these five (5) types of shellfish poisoning, PSP, NSP and ASP are the mo ~~dangerous PSP and ASP can cause death at sufficiently high concentrations. In addition, ASP can cause lasting neurological damage. PSP is caused by saxitoxins~~ produced by the dinoflagellates of the genus *Alexandrium* (formerly *Gonyaulax*). Th dinoflagellate *Pyrodinium bahamense* is also a producer of saxitoxins. NSP is caus ~~by brevetoxins produced by the dinoflagellates of the genus *Karenia* (formerly *Gymnodinium*). ASP is caused by domoic acid and is produced by diatoms of the~~ genus Pseudonitzchia. Certain *Dinophysis* spp. and *Prorocentrum* spp. produce okadaic acid and dinophysis toxins that cause DSP. *Azadinium* spp. is the producer o azaspiracids, which cause AZP.Both *Alexandrium* and *Karenia* can produce "red tide i.e. discolorations of seawater caused by blooms of the algae; however, they may als ~~reach concentrations that may result in toxic shellfish without imparting any water~~ discoloration. Toxic blooms of these dinoflagellates can occur unexpectedly or follo ~~predictable patterns. The unpredictability in occurrence of toxic blooms was demonstrated in New England in 1972 when shellfish suddenly became toxic in a~~ previously unaffected portion of the coastline and resulted in many illnesses (Schwal

1973). Historically, *Alexandrium* blooms have occurred between April and October along the Pacific coasts from Alaska to California and in the Northeast from the Canadian Provinces to Long Island Sound (U.S. Public Health Service, 1958); but th patterns may be changing. The blooms generally last only a few weeks and most shellfish (with the exception of some species of clams and scallops, which retain the toxin for longer periods) clear themselves rapidly of the toxin once the bloom dissipates. NSP has occurred from the Carolinas and extends throughout the Gulf Coast states. It shows no indication of regular recurrence and shellfish generally tak longer to eliminate the toxin (Liston, 1994). DSP and AZP cause similar symptoms mostly related to diarrhea and abdominal pain. DSP toxin-producing phytoplankton have been documented to occur off the coasts of Washington (Trainer et al. 2013) an Texas (Deeds et al. 2010) as well as off the coast in the northeast (e.g., Massachuset [Tong et al. 2015]).While AZP has occurred in the U.S., the contaminated shellfish w imported (Klontz et al. 2009). Harvesting closures in the U.S. have not been documented due to AZP toxins.

The minimum concentration of PSP toxin that will cause intoxication in susceptible persons is not known. Epidemiological investigations of PSP in Canada, however, ha indicated 200 to 600 micrograms of PSP toxin will produce symptoms in susceptible persons. A death has been attributed to the ingestion of a probable 480 micrograms PSP toxin. Investigations indicate that lesser amounts of the toxin have no deleterio effects on humans. Shellfish growing areas should be closed at a PSP toxin level, w provides an adequate margin of safety, since in many instances PSP toxicity levels c change rapidly.

The NSSP Model Ordinance requires that growing areas be placed in the closed statu when the PSP toxin concentration is equal to or exceeds the action level of 80 micrograms per 100 grams of edible portion of raw shellfish (FDA, 1977; FDA, 198

In shellfish growing areas where low levels of PSP routinely occur, harvesting for thermal processing purposes may be an alternative to consider. Thermal processing as defined by applicable FDA regulations (21 CFR 113) will reduce PSP toxin concentration of the shellfish via dilution, not destruction. If thermal processing is practiced, the Authority must develop and implement procedures to control the harvesting and transportation of the affected shellfish to the processing plant.

In Gulf coast areas, toxicity in shellfish has been associated with red tide outbreaks caused by massive blooms of the toxic dinoflagellate, *Karenia brevis*. The most common public health problem associated with *Karenia* blooms is respiratory irritation; however, neurotoxic shellfish poisonings associated with *Karenia brevis* blooms have been reported in Florida (Center for Disease Control, 1973 [a] and [b] Uncooked clams from a batch eaten by a patient with neurotoxic symptoms were found to contain 118 mouse units per 100 grams of shellfish meat. The NSSP Mod Ordinance mandates that growing areas be placed in the closed status when any NS toxin is found in shellfish meat at or above 20 MU per 100 grams of shellfish, or w the cell counts for members of the genus *Karenia* in the water column equal or exc 5,000 cells per liter of water.

ASP is caused by domoic acid, which is produced by diatoms of the genus *Pseudonitzachia*. Blooms of *Pseudonitzachia* are of varying intensity, duration and

extent.. During the 1991-1992 incident in Washington and the 2015 event on the w coast from Washington to California, high toxin levels persisted for several months (Liston, 1994; McCabe et al. 2016). There was also an extensive event in the Northeast from Maine to Rhode Island in 2016, with different regions showing var toxicity and species dominance within the bloom. The event started in late Septem in eastern Maine and ended in October; however, Rhode Island experienced anothe bloom in February of 2017.The NSSP Model Ordinance requires that growing area placed in the closed status when the domoic acid concentration is equal to or excee 20 parts per million raw shellfish.

The suitability of some growing areas for shellfish harvesting is periodically influenced by the presence of marine biotoxins such as those responsible for PSP, NSP, ASP, DSP and AZP. The occurrence of these toxins is often unpredictable, a the potential for them to occur exists along most coastlines of the United States and other countries having shellfish sanitation Memoranda of Understanding (MOU) agreements with the United States. As a result, states or countries with MOUs with the U.S. need to have management plans and/or contingency plans to address shellf borne intoxications.

Controlling Marine Biotoxins in Shellfish

There are two types of plans defined in the NSSP MO for the control of marine biotoxins

The contingency plan must describe administrative procedures, laboratory support, sample collection procedures, and patrol procedures to be implemented on an emergency basis in the event of the occurrence of shellfish toxicity (Wilt, 1974) The primary goal of this planning should be to ensure that maximum public health protection is provided. To achieve this goal the following objectives should be met

*An early warning system should be developed and implemented.
*Procedures should be established to define the severity of occurrences.
*The state or MOU country should be able to respond effectively to minimize illness.
*Adequate intelligence and surveillance information should be gathered a evaluated by the Authority.
*Procedures should be instituted to return the Biotoxin contaminated areas to th open status of their growing area classification.

Under the certification provisions of the NSSP, FDA and receiver states should hav the assurance that shellfish producing states or MOU countries are taking and can t adequate measures to prevent harvesting, shipping, and consumption of toxic shellf

To provide this assurance, the NSSP requires the Authority to develop and adopt a marine Biotoxin contingency plan for all marine and estuarine shellfish growing ar

The Authority's plan should specify how each of the objectives listed above will be accomplished. This document provides recommended guidelines to be used in preparing a plan to meet these objectives.

Recommended Contingency Plan Guidelines

- The process for precautionary closures:

- A sampling plan that considers water samples to evaluate t extent and intensity of the bloom
- A sampling plan that considers species specific shellfish sampling
- Access to screening tests; both rapid and approved method
- Trained staff to carry out sample collection and testing if necessary
- A reopening criteria

The Marine Biotoxin Management Plan

The marine biotoxin management plan is primarily for proactive management of marine biotoxins based on a history of toxin-producing phytoplankton and toxicity shellfish and/or a previous illness event or outbreak. The management plan must describe an early warning system, administrative procedures, laboratory support, sample collection procedures, patrol procedures to be implemented and reopening criteria (Wilt, 1974). A management plan is required for a shellfish Authority that a history of toxin-producing phytoplankton, toxicity in shellfish and/or an illness ev or outbreak attributed to their growing areas. A shellfish Authority might have a management plan for certain marine biotoxins like PSP toxins but a contingency pl for toxins like AZP toxins. The primary goal of the management plan should be to prevent illnesses from toxic shellfish and ensure that maximum public health protection is provided. To achieve this goal the following objectives should be met

- An early warning system should be developed and implemented.
- Procedures should be established to define the severity of occurrences.
- The Authority should be able to respond effectively to minimize illness.
  - Adequate intelligence and surveillance information should be gather and evaluated by the
  - Authority.
  - Procedures should be instituted to return the biotoxin contaminated area the open status of their
  - growing area classification.

** Provide an early warning system:*

1. Communication procedures should be established with other appropriate agencies to rapidly report to the Authority any abnormal environmental phenomenon that might be associated with shellfish growing areas such as bird or fish kills, water discoloration or abnormal behavior of shellfish or marine scavengers.
2. The Authorities should establish procedures for health agencies to report an toxin-like illnesses.
3. An early warning phytoplankton and/or shellfish-monitoring program shoul be implemented.

   These monitoring programs should use the "key station" (for both phytoplankton and shellfish monitoring) and "critical species" concepts (fo shellfish monitoring).

   * Sampling stations should be located at sites where past experience ha shown toxin is most likely to appear first.

   * When monitoring shellfish, samples should be collected of species

~~which are most likely to~~
reveal the early presence of toxin and which are most likely to show th
highest toxin levels. For example, mussels have been found to be usefu
~~for early PSP detection.~~
~~* The frequencies and periods for collection of samples should be established recognizing the randomness of PSP blooms. This assumes~~
several years of baseline data in order to establish stations and samplin
~~plans.~~
~~* Frequency of sampling should be adequate to monitor for fluctuation~~
~~coastal phytoplankton populations.~~

4. Cha nnels of communication concerning shellfish toxicity should be establis with other states, countries (in the case of MOU countries), FDA, and other responsible officials. A marine Biotoxin control official should be designa ~~by the Authority to receive and distribute all marine Biotoxin related information. Consultation with adjacent jurisdictions, marine biologists and other environmental officials might also be useful (Felsing, 1966; Quayle, 1969; Prakash *et al*., 1971).~~

~~* *Define the severity of the problem:*~~

~~1. A procedure should be established to promptly expand the sampling~~ program for marine Biotoxins in the event of increased toxicity/cell count ~~any indicator monitoring stations identified within the plan. Sampling~~ stations and frequencies of sampling should be increased when monitoring data or other information suggests that toxin levels are increasing. ~~procedure should include plans for obtaining the additional resources necessary to implement the expanded sampling and laboratory analysis program.~~

~~2. Information should be available concerning the location of commercial~~
~~3. shellfish resource areas and species present in the state.~~
Criteria should be developed to define the circumstances ~~under which grow areas will be placed in the closed status because of marine Biotoxin~~ contamination. The criteria should integrate public health, conservation, a economic considerations. Principal items of concern include consideration ~~the rapidity with which toxin levels can increase to excessive levels, the inherent delays in sample collection and results, the number of samples~~ required to initiate action, the size of the area to be closed (including a safe ~~zone), and the type of harvesting restrictions to be invoked (all species or~~ specific species). It may be appropriate to close harvesting areas adjacent t known toxic areas until increased sampling can establish which areas are to ~~free and that toxin levels have stabilized.~~

~~4.~~ Procedures should be established to prom ~~or lots might be potentially contaminated, and to determine the distribution of these products or lots.~~

~~* *Respond effectively to minimize illness:*~~

1. A summary should be provided citing the laws and regulations in the state (

MOU country) that promptly and effectively allow the Authority to restrict harvesting, withdraw interstate shipping permits, and to embargo/recall any potentially toxic shellfish already on the market in the event of a marine Biotoxin event. The plan should clearly define the timeframe involved in taking appropriate legal action.

2. The administrative procedures necessary to place growing areas in the close status, to withdraw interstate certification of dealers, and to embargo and recall shellfish should be delineated. The timeframe necessary to accompli these actions should also be specified.
3. A plan should be developed which will define what type of patrol program necessary to properly control harvesting in toxin contaminated growing are The program should be tested to ensure prompt implementation in the even is needed.
4. Procedures should be developed to promptly disseminate information on th occurrences of toxic phytoplankton blooms to the industry and local health agencies. It is helpful to establish relationships and procedures with other agencies such as the state CDC and Poison Control and authorities in advan of any serious biotoxin event.
5. Procedures should be established to coordinate control activities taken by st and federal agencies or departments and district, regional, or local health authorities.

** Return growing areas to the open status of their NSSP classification:*

1. Once a growing area is placed in the closed status because of marine Biotox contamination, a procedure should be instituted to gather data necessary to decide when the area can be returned to the open status of its classification. system of representative samples to establish detoxification curves should b part of this procedure.
2. The Authority should develop a set of criteria that must be met before a growing area can be returned to the open status. These criteria should integrate public health, conservation, and economic considerations, and employ a sufficient number of samples and other environmental indices, if used, to establish that the level of toxin or cell counts are below the closure level. For example, experience has shown that appropriate reopening criter for PSP include a minimum of three (3) samples collected over a period of least fourteen (14) days. These samples should show the absence of PSP o levels below 80 micrograms per 100 grams of shellfish tissue.
3. A program of consumer education should be continued as long as any area remains in the closed status because of marine Biotoxin contamination.

References
Title 21 CFR Part 7
References

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Public Health Significance	Marine biotoxins can cause injury, illness, or death. More clearly presented guidance will assist control authorities in developing marine biotoxin contingency and management plans.
Cost Information	None
Action by 2019 Task Force I	Recommended referral of Proposal 19-124 to an appropriate committee as determined by the Conference Chairperson.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-124.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-124.
Action by 2023 Biotoxin Committee	Recommended adoption of 19-124 as substituted. **.02 ~~Guidance for Developing~~ Marine Biotoxin Control Guidance~~Plans~~** NSSP guidance documents provide Authorities with information and best practices on how to implement the components of the Model Ordinance ~~the public health principles supporting major components of the NSSP and its Model Ordinance, which includes the requirements of the program~~. NSSP *Model Ordinance* requirements apply only to interstate commerce although most States apply the requirements intrastate. For the most up to date and detailed listing of requirements, the reader should consult the most recent edition of the Model Ordinance. An overview of marine biotoxins including associated biological vectors, diseases, historic outbreaks, and emerging trends can be found in Section III Public Health Reasons and Explanations Chapter IV. @.04 Marine Biotoxin Control. ~~**Introduction**~~

~~Shellfish are filter feeders and, therefore, they have the ability to concentrate toxic phytoplankton from the water column when present in shellfish growing waters. The toxins produced by certain species of phytoplankton can cause illness and death in humans. Toxins are accumulated in the viscera and/or other tissues of shellfish and human exposure occurs when the shellfish are eaten (Gordan *et al.*, 1973). These toxins are not normally destroyed by cooking or processing and cannot be detected by taste. The presence of toxic phytoplankton in the water column or traces of their toxin in shellfish meat does not necessarily constitute a health risk, as toxicity is dependent on concentration (dose) in the shellfish. To protect the consumer, the Authority must evaluate the concentration of toxin present in the shellfish or the toxic phytoplankton concentration in the water column against the levels established in the NSSP Model Ordinance to determine what action, if any, should be taken.~~

~~There are a wide range of methodologies developed for screening and confirmation of toxic phytoplankton and their toxins. Only methods adopted into the NSSP can be implemented for the purpose of confirming toxin concentration levels and making decisions to reopen growing areas. Additionally, some screening methods have been evaluated by the ISSC and found fit for purpose for the NSSP, thereby providing confidence in their use for specific screening purposes. Toxin methods fall into two (2) categories in the NSSP: Approved Methods for Marine Biotoxin Testing (Section IV. Guidance Documents Chapter II Growing Areas .14 Table 2.) and Approved Limited Use Methods for Marine Biotoxin Testing (Section IV. Guidance Documents Chapter II Growing Areas .14 Table 4.). These methods range from mouse bioassays to immunochromatography and other antibody based platforms to chemical analytical methods such as high performance liquid chromatography (HPLC). Information available in the referenced Tables above provides references for the methods and, as applicable, what limitations are placed on the use of the method within the NSSP. For toxins that have no method adopted into the NSSP, best available science is employed.~~

~~There are five (5) types of shellfish poisonings which are specifically addressed in the NSSP Model Ordinance: PSP, NSP, ASP (also known as Domoic Acid poisoning), DSP and AZP. Of these five (5) types of shellfish poisoning, PSP, NSP and ASP are the most dangerous. PSP and ASP can cause death at sufficiently high exposures. In addition, ASP can cause lasting neurological damage. PSP is caused by saxitoxins produced by the dinoflagellates of the genus *Alexandrium* (formerly *Gonyaulax*). The dinoflagellate *Pyrodinium bahamense* is also a producer of saxitoxins. NSP is caused by brevetoxins produced by the dinoflagellates of the genus *Karenia* (formerly *Gymnodinium*). ASP is caused by domoic acid and is produced by diatoms of the genus *Pseudo-nitzchia*. Certain *Dinophysis* spp. and *Prorocentrum* spp. produce okadaic acid and dinophysis toxins that cause DSP. *Azadinium* spp. is the producer of azaspiracids, which cause AZP.~~

~~Both *Alexandrium* and *Karenia* can produce "red tides", i.e. discolorations of seawater caused by blooms of the algae; however, they may also reach concentrations that cause toxic shellfish without imparting any water discoloration. Toxic blooms of these dinoflagellates can occur unexpectedly or follow predictable patterns. The unpredictability in occurrence of toxic blooms was demonstrated in New England in 1972 when shellfish suddenly became toxic in a previously unaffected portion of the coastline and resulted in many illnesses (Schwalm, 1973). Historically, *Alexandrium* blooms have occurred between April and October along the Pacific coasts from Alaska to California and in the~~

~~Northeast from the Canadian Provinces to Long Island Sound (U.S. Public Health Service, 1958); but these patterns may be changing. The blooms generally last only a few weeks and most shellfish (with the exception of some species of clams and scallops which retain the toxin for longer periods) clear themselves rapidly of the toxin once the bloom dissipates. Occurrence of *Karenia* blooms extends from the Carolinas south throughout the Gulf Coast States. DSP and AZP cause similar symptoms mostly related to diarrhea and abdominal pain. DSP toxin producing phytoplankton have been documented to occur off the coasts of Washington (Trainer et al. 2013) and Texas (Deeds et al. 2010) as well as off the coast in the Northeast (e.g., Massachusetts [Tong et al. 2015]).While AZP has occurred in the U.S., the contaminated shellfish was imported (Klontz et al. 2009). Harvesting closures in the U.S. have not been documented due to AZP toxins.~~

~~The minimum concentration of PSP toxin that will cause intoxication in susceptible persons is not known. Epidemiological investigations of PSP in Canada, however, have indicated 200 to 600 micrograms of PSP toxin will produce symptoms in susceptible persons. A death has been attributed to the ingestion of a probable 480 micrograms of PSP toxin. Investigations indicate that lesser amounts of the toxin have no deleterious effects on humans. Shellfish growing areas should be closed at a PSP toxin level, which provides an adequate margin of safety, since in many instances PSP toxicity levels can change rapidly.~~

~~The NSSP Model Ordinance requires that growing areas be placed in the closed status when the PSP toxin concentration is equal to or exceeds the action level of 80 micrograms per 100 grams of raw shellfish (FDA, 1977; FDA, 1985).~~

~~In shellfish growing areas where low levels of PSP toxin routinely occur, harvesting for thermal processing purposes may be an alternative to consider. Thermal processing as defined by applicable FDA regulations (21 CFR 113) will reduce the PSP toxin concentration of the shellfish via dilution, not destruction. If thermal processing is practiced, the Authority must develop and implement procedures to control the harvesting and transportation of the affected shellfish to the processing plant.~~

~~In Gulf coast areas, toxicity in shellfish has been associated with red tide outbreaks caused by massive blooms of the toxic dinoflagellate, *Karenia brevis*. The most common public health problem associated with *Karenia* blooms is respiratory irritation; however, neurotoxic shellfish poisonings associated with *Karenia brevis* blooms have been reported in Florida (Center for Disease Control, 1973 [a] and [b]).~~
~~Uncooked clams from a batch eaten by a patient with neurotoxic symptoms were found to contain 118 mouse units per 100 grams of shellfish meat. The NSSP Model Ordinance mandates that growing areas be placed in the closed status when any NSP toxin is found in shellfish meat at or above 20 MU per 100 grams of shellfish.~~

~~ASP is caused by domoic acid, which is produced by diatoms of the genus *Pseudo-nitzschia*. Blooms of *Pseudo-nitzschia* are of varying intensity, duration and extent. During a 1991-1992 incident in Washington and a 2015 event on the west coast from Washington to California, high toxin levels persisted for several months (Liston, 1994; McCabe et al. 2016). There was also an extensive event in the Northeast from Maine to Rhode Island in 2016, with different regions showing varying toxicity and species dominance within the bloom. The event started in late September in eastern Maine and ended in October; however, Rhode Island experienced another bloom in February of 2017. The NSSP Model Ordinance~~

~~requires that growing areas be placed in the closed status when the domoic acid concentration is equal to or exceeds 20 parts per million raw shellfish.~~

~~The suitability of some growing areas for shellfish harvesting is periodically influenced by the presence of marine biotoxins such as those responsible for PSP, NSP, ASP, DSP and AZP. The occurrence of these toxins is often unpredictable, and the potential for them to occur exists along most coastlines of the United States and other countries having shellfish sanitation Memoranda of Understanding (MOU) agreements with the United States. As a result, States or countries with MOUs with the U.S. need to have management plans and/or contingency plans to address shellfish-borne intoxications.~~

~~**Controlling Marine Biotoxins in Shellfish**~~

~~Under the certification provisions of~~In accordance with the NSSP, FDA and receiver States should have assurance that shellfish producing States or ~~MOU~~ countries ~~of~~with shellfish sanitation arrangements are taking and can take adequate measures to prevent harvesting, shipping, and consumption of toxic shellfish. To provide this assurance, the NSSP requires the Authority to develop and adopt either a marine biotoxin contingency plan and/or a marine biotoxin management plan for a specific list of biotoxins that covers each marine and estuarine shellfish growing area. Single plans can be developed for a whole state or can cover particular growing areas or toxins. An Authority may have an area with a contingency plan for some biotoxins and a management plan for others, a contingency plan for all biotoxins, or a management plan for all biotoxins.~~There are two (2) types of plans defined in the NSSP MO for the control of marine biotoxins.~~ A contingency plan is developed by an Authority that has no history or reason to expect toxin-producing phytoplankton in their growing areas. A marine biotoxin management plan is developed by an Authority that has historic occurrence of toxin-producing phytoplankton and toxicity in shellfish from their growing areas.

The Marine Biotoxin Contingency Plan
Section II. MO Ch IV. Shellstock Growing Areas @.04 Marine Biotoxin Control (A)

Purpose

The purpose of a contingency plan is for the Authority to be prepared to mitigate risk and protect public health if an unanticipated biotoxin event occurs in a classified shellfish growing area. Examples of an unanticipated biotoxin event include an illness outbreak or an emergence of a toxin-producing phytoplankton in a growing area where it has not historically occurred.~~The contingency plan is primarily for reactive management to an illness outbreak or an emergence of a toxin-producing phytoplankton in a growing area that has not historically occurred before. The contingency plan must describe administrative procedures, laboratory support, sample collection procedures, patrol procedures to be implemented on an emergency basis and reopening criteria (Wilt, 1974). The contingency plan is only appropriate for a shellfish Authority that has no history or reason to expect toxin-producing phytoplankton in their growing areas. The primary goal of the contingency plan should be to ensure that maximum public health protection is provided. To achieve this goal the following elements should be included:~~

The Model Ordinance requires that a contingency plan:

1. Address the toxins that cause each of the following illnesses (except those

addressed in a biotoxin management plan): PSP, ASP, NSP, DSP, and AZP.

a. Even if the toxin has never been known to occur in the area or it is biologically unlikely to occur in the area, it still must be addressed.

2. Define the administrative procedures and resources necessary to: initiate an emergency shellfish sampling program; close growing areas and embargo shellfish; prevent harvesting of contaminated species; provide for product recall; disseminate information; coordinate control actions; and establish reopening criteria.

a. It is important to note that the Model Ordinance does not require an Authority to take any actions following the development of a contingency plan, unless the Authority elects to include specific actions in their plan such as phytoplankton or biotoxin sampling protocols. Instead, this plan should define the procedures an Authority would follow in the event of a bloom or illness outbreak, as well as how the Authority would go about acquiring the resources needed to implement those procedures.

Contingency Plan Content Guidance

Element	**Recommended Plan Contents**
Emergency Sampling Program	• Identify area(s), phytoplankton, and/or shellfish for sampling • A procedure to promptly expand this sampling program, including increasing sampling stations and sampling frequency, in the event of increased toxicity/cell counts at any indicator monitoring stations identified within the plan • Identify partner sampling agencies available • Identify laboratory support, including capacity, method(s), contract(s) o In some circumstances, the Authority may have the laboratory support available in-house, but in other circumstances may have to identify alternate NSSP labs to conduct the necessary methods o If there is no approved method available, the Authority should identify an appropriate method for analysis following the procedures described in MO Chapter III @.02 Methods • Describe training for samplers • Identify financial resources available, request processes, and necessary approvals • Though not required by the Model Ordinance, it may be appropriate for the Authority to implement an early warning system within the contingency plan, as described in the *Marine Biotoxin Management Plan* section below

	Close Growing Areas and Prevent Harvest of Contaminated Species	• Identify the legal authority to close areas and restrict harvesting • Protocols to initiate closures, taking into consideration public health, economic, and conservation concerns. The rapidity with which toxin levels can increase, inherent delays in sample collection and results, the number of samples required to initiate action, the size of the area to be closed (including a safety zone), and the type of harvesting restrictions to be invoked (all species or specific species) should all be considered ~~It may be appropriate to include adjacent harvesting areas in an initial closure until increased sampling can establish which areas are toxin free and that toxin levels have stabilized.~~ • Describe the mechanism to quickly notify growers, harvesters, and dealers of closures • Describe protocols to notify patrol entities of emergency closures, as well as patrol procedures necessary to prevent harvest from closed areas
	Embargo Shellfish	• Identify the legal authority to embargo, detain, quarantine, or otherwise prevent the movement of shellfish in commerce and to withdraw interstate shipping permits • Describe procedures for embargoing shellfish, including the identification of affected lots and distribution networks, as well as any associated forms, tags, or other administrative tools used • Describe the mechanism for destruction of embargoed product if it is found to be adulterated, or the mechanism to release embargoed product if it is found to be free of contamination
	Coordinate Control Actions	• Describe the mechanism to notify partner state, local, federal, and/or tribal agencies to avoid duplicative efforts and streamline response
	Product Recall	• Identify the legal authority to recall product that may already be in commerce • Identify agency protocols for implementing a product recall • It may be helpful to develop templates and forms for recall, if they are not already in place
	Disseminate Information to Partners	• Establish relationships and procedures to notify:

		○ ISSC and FDA Shellfish Specialist~~, if needed~~ ○ Adjacent states or states that may have received adulterated product ○ Local, tribal, and state public health and safety ~~-~~partners ○ Poison control partners • Describe efforts to inform and educate the public about risks associated with biotoxins • It may be useful to develop information sheets or fillable templates for notifications, press releases, partner agency communications, or other routinely used documents • Develop a model communications plan or agree in advance on lead communications points of contact
	Reopening Criteria	• Establish how the authority will determine when an area can be reopened based on data ○ Reopening criteria should integrate public health, conservation, and economic considerations ○ To establish a detoxification curve or other environmental indices, such as phytoplankton concentration trends, more than one (1) sample collected at different times is needed.~~-~~ For example, some states collect two (2) samples over seven (7) days and others collect three (3) samples over fourteen (14) days. These samples should show the absence of biotoxins or levels below the closure guidance level ○ If species-specific sampling regimes are employed, then each species that exceeded the quarantine threshold must be tested independently to reopen • Identify laboratory support, including capacity, methods, and contracts ○ In some circumstances, the Authority may have the laboratory support available in-house, but in other circumstances, may have to identify alternate NSSP labs to conduct the lab methods approved for reopening ○ If there is no approved method available, the Authority should identify an appropriate method for analysis following the procedures described in MO Chapter III @.02 Methods

Additional Considerations:

a. If an Authority has a management plan and/or protocols such as patrol manuals or existing MOUs that are relevant and appropriate, the Authority may reference those documents within its contingency plan.

b. Relationships with academia, government, non-government, and industry partners can be extremely helpful in identifying the presence of previously unseen phytoplankton or biotoxins. It can be helpful to develop and maintain a general list of contact people or organizations that can collaborate on phytoplankton and biotoxin monitoring efforts.

c. The Model Ordinance also requires that certain records be maintained during and following an event. It is recommended that the contingency plan include details on record maintenance.

i. Appropriate records of illnesses should be compiled and maintained by the Authority. These records should include data on the incidence of illness and appropriate case history data. This information may be important in defining the severity of the problem, as well as for a retrospective evaluation of the adequacy of the entire control program.

ii. Records of shellfish sample results from toxin testing should include analysis of trends, detoxification curves, phytoplankton and water sample analyses, and pertinent environmental observations.

iii. Whenever possible, the Authority should archive shellfish or shellfish homogenates for additional analysis.

~~• A process for immediate precautionary closures;~~

~~• A sampling plan that considers water samples to evaluate the extent and intensity of the toxic phytoplankton distribution;~~

~~• A sampling plan that considers species-specific shellfish sampling;~~

~~• Access to biotoxin tests: both screening and approved methods;~~

~~• Trained staff to carry out sample collection and testing if necessary; and~~

~~Reopening criteria.~~

~~Under the certification provisions of the NSSP, FDA and receiver States should have the assurance that shellfish producing States or MOU countries are taking and can take adequate measures to prevent harvesting, shipping, and consumption of toxic shellfish. To provide this assurance, the NSSP requires the Authority to develop and adopt a marine biotoxin contingency plan for all marine and estuarine shellfish growing areas. The Authority's plan should specify how each of the objectives listed above will be accomplished. This document provides recommended guidelines to be used in preparing a plan to meet these objectives.~~

The Marine Biotoxin Management Plan

Section II. MO Ch IV. Shellstock Growing Areas @.04 Marine Biotoxin Control (B)

Purpose

The marine biotoxin management plan is ~~primarily~~ required for proactive management of marine biotoxins ~~for~~ in growing areas with a history of toxin-producing phytoplankton, toxins in shellfish at or above the guidance level in their

growing areas, ~~and toxicity in shellfish~~ and/or a previous illness ~~event or~~ outbreak. Similar to a contingency plan, the Model Ordinance requires that a~~The~~ management plan ~~must describe an early warning system,~~define the administrative procedures and resources necessary to: close growing areas and embargo shellfish; prevent harvesting of contaminated species; provide for product recall; disseminate information; coordinate control actions; and establish reopening criteria. Please refer to the Contingency Plan Content Guidance above for recommendations on how to develop these portions of the management plan

Additionally, the Model Ordinance requires that:

1. For any areas covered by a management plan, the Authority must maintain a toxin-producing phytoplankton and/or shellfish sampling program.
2. The management plan includes procedures to ensure that all shellfish harvested from growing areas or portion(s) of growing areas placed in the controlled access status will meet all conditions of harvest restrictions prior to being placed in distribution.

~~Strategies for meeting these requirements are described below.~~

~~., laboratory support, sample collection procedures, patrol procedures to be implemented and reopening criteria (Wilt, 1974). A management plan is required for a shellfish Authority that has a history of toxin producing phytoplankton, toxicity in shellfish and/or an illness event or outbreak attributed to their growing areas. A shellfish Authority might have a management plan for certain marine biotoxins like PSP toxins but a contingency plan for toxins like AZP toxins. The primary goal of the management plan should be to prevent illnesses from toxic shellfish and ensure that maximum public health protection is provided. To achieve this goal the following elements should be included:~~

- ~~An early warning system should be developed and implemented.~~
- ~~Procedures should be established to define the severity of occurrences.~~
- ~~The Authority should be able to respond effectively to minimize risk of illness.~~
- ~~Adequate intelligence and surveillance information should be gathered and evaluated by the Authority.~~

~~Procedures should be instituted to return the biotoxin contaminated areas to the open status of their growing area classification.~~

~~**Recommended Contingency Plan Guidelines**~~

~~**Implement an** **Early Warning System**~~

~~** Provide an early warning system:*~~

It is recommended that any Authority with a management plan should have an early warning system in place (https://www.fao.org/3/cc4794en/cc4794en.pdf). Early warning systems may include additional phytoplankton and/or shellfish monitoring efforts conducted by the Authority and/or by use of a network of observers and partnerships as well as communications with other organizations to identify environmental or biological warning signs.

- Establish relationships and ~~Communication~~ communication procedures with resource agencies ~~should be established with other appropriate agencies~~ to rapidly report to the Authority any abnormal environmental ~~phenomenon~~ phenomena that might be associated with shellfish growing areas, such as bird or fish kills, water discoloration or abnormal behavior of shellfish or marine scavengers.
- Establish relationships and communication~~The Authorities should establish~~ procedures for health agencies to report any toxin-like illnesses.

- ~~An early warning phytoplankton and/or shellfish monitoring program should be implemented. These monitoring programs should use the "primary station" (for both phytoplankton and shellfish monitoring) and "critical species" concepts (for shellfish monitoring).~~
- Primary ~~Sampling~~ sampling stations ~~(primary stations)~~ should be located at sites where ~~past~~ experience has shown toxins or blooms are ~~is~~ most likely to appear first.
- ~~When~~ If monitoring shellfish, samples should be collected of species which are most likely to reveal the early presence of toxin and which are most likely to show the highest toxin levels ~~(critical species)~~. For example, in some circumstances, mussels have been found to be useful for early detection.
- Sampling design should always consider what species are present in the growing area and commercially harvested.
    - ~~The frequencies and geographic distribution for collection of samples should be established recognizing the randomness of toxic algal blooms. This assumes several years of baseline data in order to establish stations and sampling plans.~~
    - ~~Frequency and geographic distribution of sampling should be adequate to monitor for fluctuations in coastal phytoplankton populations and the influence of meteorological and hydrographic events. For example, a large rain storm may cause nutrient loading in coastal waters and trigger a toxic phytoplankton bloom or a hurricane may drive offshore phytoplankton blooms onshore.~~
    - ~~Channels of communication concerning shellfish toxicity should be established with other States, countries (in the case of MOU countries), FDA, and other responsible officials. A marine biotoxin control official should be designated by the Authority to receive and distribute all marine biotoxin related information. Consultation with adjacent jurisdictions, marine biologists and other environmental officials is also useful (Felsing, 1966; Quayle, 1969; Prakash *et al*., 1971).~~

~~*Define the severity of the problem:*~~

1. ~~A procedure should be established to promptly expand the sampling program for marine biotoxins in the event of increased toxicity/cell counts at any indicator monitoring stations identified within the plan. Sampling stations and frequencies of sampling should be increased when monitoring data or other information suggests that toxin levels are increasing. The procedure should include plans for obtaining the additional resources necessary to implement the expanded sampling and laboratory analysis program.~~
2. ~~Information should be available concerning the location of commercial shellfish resource areas and species present in the State.~~
3. ~~Criteria should be developed to define the circumstances under which growing areas will be placed in the closed status because of marine biotoxin contamination. The criteria should integrate public health, conservation, and economic considerations. Principal items of concern include consideration of the rapidity with which toxin levels can increase to excessive levels, the inherent delays in sample collection and results, the number of samples required to initiate action, the size of the area to be closed (including a safety zone), and the type of harvesting restrictions to~~

be invoked (all species or specific species). It may be appropriate to close harvesting areas adjacent to known toxic areas until increased sampling can establish which areas are toxin free and that toxin levels have stabilized.

4. Procedures should be established to promptly identify which shellfish products or lots might be potentially contaminated, and to determine the distribution of these products or lots.

Respond effectively to minimize illness:

1. A summary should be provided citing the laws and regulations in the State (or MOU country) that promptly and effectively allow the Authority to restrict harvesting, withdraw interstate shipping permits, and to embargo/recall any potentially toxic shellfish already on the market in the event of a marine biotoxin event. The plan should clearly define the timeframe involved in taking appropriate legal action.
2. The administrative procedures necessary to place growing areas in the closed status, to withdraw interstate certification of dealers, and to embargo and recall shellfish should be delineated. The timeframe necessary to accomplish these actions should also be specified.
3. A plan should be developed which will define what type of patrol program is necessary to properly control harvesting in toxin contaminated growing areas. The program should be tested to ensure prompt implementation in the event it is needed.
4. Procedures should be developed to promptly disseminate information on the occurrences of toxic phytoplankton blooms to the industry and local health agencies. It is helpful to establish relationships and procedures with other agencies such as the State CDC and Poison Control and Authorities in advance of any serious biotoxin event.
5. Procedures should be established to coordinate control activities taken by State and Federal agencies or departments and district, regional, or local health authorities.

** Gather follow-up data:*

1. Appropriate records of illnesses should be compiled and maintained by the Authority. These records should include data on the incidence of illness and appropriate case history data. This information may be important in defining the severity of the problem, as well as for a retrospective evaluation of the adequacy of the entire control program.
2. Records of shellfish sample results from toxin testing should include analysis of trends, detoxification curves, phytoplankton and water sample analyses, and pertinent environmental observations.

Whenever possible the Authority should archive shellfish homogenates for additional analysis.

** Return growing areas to the open status of their NSSP classification:*
*

1. Once a growing area is placed in the closed status because of marine biotoxin contamination, a procedure should be instituted to gather data necessary to decide when the area can be returned to the open status of its classification. A system of representative samples to establish detoxification curves should be part of this procedure.
2. The Authority should develop a set of criteria that must be met before a

~~growing area can be returned to the open status. These criteria should integrate public health, conservation, and economic considerations, and employ a sufficient number of samples and other environmental indices, if used, to establish that the level of toxin or cell counts are below the closure level. For example, experience has shown that appropriate reopening criteria for PSP include a minimum of three (3) samples collected over a period of at least fourteen (14) days. These samples should show the absence of PSP or levels below 80 micrograms per 100 grams of shellfish tissue.~~

~~A program of consumer education should be continued as long as any area remains in the closed status because of marine biotoxin contamination.~~

Marine Biotoxin Management Strategies

It is necessary to recognize that different marine biotoxin management strategies are essential to address specific risks as well as geographic and logistical conditions. Marine biotoxin management strategies must include an appropriate number of samples to adequately address the specific risks. The Authority initiating biotoxin management plans should employ sampling in accordance with the strategies below until a baseline dataset of at least 36 samples per growing area or hydrographically linked waterbodies is developed (i.e. 36 phytoplankton samples for a phytoplankton strategy or 36 shellfish samples for a shellfish-related strategy). These samples should cover representative environmental conditions and a time span of at least three (3) years. Once this baseline dataset is developed and trends are established, the Authority may consider ~~modifying~~ reducing sample numbers, ~~and~~ frequency, and lot testing and/or increasing harvest days allowed in the marine biotoxin management plan in accordance with the strategies below.

All marine biotoxin management plans must establish, at a minimum, the below criteria:

- screening levels,
- methods,
- laboratory(s)/analyst(s),
- a representative sampling plan,
- representative sample locations (stations),
- representative sampling frequency; and
- a dataset that supports management decisions.

A. Phytoplankton monitoring: this strategy involves a routine program for sampling growing area waters for the presence of phytoplankton ~~species~~ documented or suspected to produce marine biotoxins. This complementary management strategy that enhances predictive capabilities of anticipating toxicity in shellfish must be used in combination with other management strategies.
The level of monitoring required will vary based on the historical database available to inform the sampling strategy (i.e., growing areas with a long history of defined temporal and spatial patterns of toxin-producing phytoplankton may have a more targeted approach to sampling, requiring less monitoring than for growing areas where temporal and spatial patterns have not been determined). ~~A dataset with at least 36 samples per growing area or hydrographically linked waterbodies for a time span of at least three (3) years of phytoplankton counts, comparing with the onset of shellfish toxicity when toxic phytoplankton are present, should be developed before the biotoxin monitoring plan may be modified.~~

Phytoplankton monitoring can be applied to all growing areas where collecting, transporting and processing water samples is logistically feasible, taking into consideration effects of zooplankton grazing and durability of various cell types to temperature and transport. This management strategy may be applied to aquaculture or wild harvest. Appropriate venues for this management strategy include but are not limited to; easily accessible wild harvest areas and aquaculture sites in state waters or aquaculture sites in federal waters.

~~The marine biotoxin management plan that incorporates this strategy must establish:~~

- ~~appropriate screening levels,~~
- ~~appropriate methods,~~
- ~~appropriate laboratory(s)/analyst(s),~~
- ~~an appropriate sampling plan,~~
- ~~appropriate sample locations (stations),~~
- ~~appropriate sampling frequency; and~~
- ~~a sufficient dataset to support management decisions.~~

The phytoplankton monitoring strategy shall be used together with one (1) or more of the other biotoxin management strategies. If it were used as the sole management strategy, phytoplankton monitoring would likely misrepresent the actual risk of marine biotoxins. Cell counts, as measured per liter of water, are often used to trigger additional testing of shellfish in biotoxin monitoring programs. These cell count criteria can only be established with a robust dataset; therefore, new monitoring programs should employ low cell count criteria to trigger shellfish toxicity samples to establish or refine the cell concentrations responsible for toxins accumulating in shellfish.

When an early warning system such as phytoplankton monitoring detects increased toxicity/cell counts or other information suggests that toxin levels are increasing, it is important that the Authority have procedures to promptly expand sampling to additional stations and/or increase the frequency of sampling for marine biotoxins. The procedures should include plans for obtaining the additional resources necessary to implement the expanded sampling and laboratory analysis program. If a plan consists of water sampling for phytoplankton cell counts as surveillance, the Authority should identify its plan to be able to initiate shellfish sampling.

Considerations should be made for how sampling is conducted such as phytoplankton net tows, filtered surface water, or whole water samples. The depth of water sampled should also be considered and evaluated for all species of phytoplankton being targeted. Some species of phytoplankton are known to display diurnal, vertical migration patterns within the water column, while other species are known to occur in dense patches.

Laboratory and field methods may include, but are not limited to light microscopy, flowcytometry, DNA fingerprinting, rapid toxin detection tests, and PCR assays. Analysts should be trained in each method employed and consideration should be given to complimentary methods of analysis such as light microscopy with phytoplankton identification confirmed by a rapid test at least in the initial phases of the monitoring program.

An appropriate sampling plan, station location, and sampling frequency should all factor in the location and type of the resource being monitored,

the species of phytoplankton anticipated or observed, and the environmental conditions that might result in a rapid bloom or trigger the production of toxicity in an existing population. Primary sampling stations (also referred to as indicator or sentinel stations) should be located at sites where toxic phytoplankton are most likely to first appear, based either on experience or knowledge of site conditions. The geographic distribution for collection of samples should take into consideration the randomness of toxic algal blooms. Establishing the frequency and period for collection of samples to identify an event as early as possible is an important consideration. Historical occurrences and fluctuations in coastal phytoplankton populations due to the influence of meteorological and hydrographic events are also significant. For example, a large rain storm may cause nutrient loading in coastal waters and trigger a toxic phytoplankton bloom, or a hurricane may drive an offshore phytoplankton bloom onshore. To facilitate knowledge transfer, it is advisable that the authority describe its rationale in selecting sampling sites.

B. Routine shellfish toxicity monitoring: this strategy involves a routine program for sampling and testing shellfish meats for the presence of marine biotoxins. Unless species-specific shellfish testing is conducted, the highest risk species (e.g. species that metabolizes toxin most quickly) occurring in the growing area shall be used. Many biotoxin monitoring programs have found mussels to be the best sentinel species. This strategy may be used alone or in combination with other management strategies.

The level of monitoring required will vary based on the historical database available to inform the sampling strategy (i.e., growing areas with a long history of defined temporal and spatial patterns of shellfish toxicity may have a more targeted approach to sampling, requiring less monitoring than for growing areas where temporal and spatial patterns have not been determined). ~~A dataset with at least 36 samples per growing area or hydrographically linked waterbodies across representative environmental conditions for a span of at least three (3) years shall be developed before the biotoxin monitoring plan may be modified.~~ Until the Authority is confident they understand the risk posed by marine biotoxins in the growing area, sampling should be as robust as possible, and managers should consider that harmful algal blooms can change dramatically from year to year.

This management strategy can be applied to all growing areas where collecting, transporting and processing shellfish samples is feasible. This management strategy can be applied to aquaculture or wild harvest. Appropriate venues for this management strategy include but are not limited to, easily accessible wild harvest areas and aquaculture sites in state waters or wild harvest areas and aquaculture sites in federal waters.

~~The marine biotoxin management plan that incorporates this strategy must establish:~~

- ~~appropriate screening levels,~~
- ~~appropriate methods,~~
- ~~appropriate laboratory(s)/analyst(s),~~
- ~~an appropriate sampling plan,~~
- ~~appropriate sample locations (stations),~~
- ~~appropriate sampling frequency; and~~
- ~~a sufficient dataset to support management decisions.~~

The routine shellfish toxicity monitoring strategy may be used independently or together with one (1) or more of the other biotoxin management strategies. If used as the sole management strategy, predicting future toxicity levels in shellfish and the appropriate sampling frequency can be difficult. Long-term databases can provide valuable historic information on the timing of toxicity occurring in shellfish as well as toxicity elimination from shellfish. Shellfish toxin levels that are below the regulatory levels may trigger emergency or expanded testing, or precautionary closures. Growing areas should be placed in the closed status at a level that provides an adequate margin of safety, since in many instances, toxicity levels will change rapidly and the time between sampling and results should be considered. Precautionary closures can be made to prevent the harvest of potentially toxic shellfish while sample results are being collected and processed.

Consideration should be given to the different species of shellfish present in a growing area, the intensity and duration of harmful algal blooms and the uptake and elimination rates of specific toxins from all species of shellfish harvested from the growing areas (e.g., sea scallops).
Methods shall be used in accordance with Section IV. Guidance Documents Chapter II Growing Areas.14 or Section II. Chapter III. @.02 C. Additionally, the Authority should identify laboratories that can perform approved methods for marine biotoxins and identify laboratory capacity.

An appropriate sampling plan, station location and sampling frequency should factor in the location and type of the resource being monitored, the species of shellfish harvested in the growing area and environmental conditions that might affect toxin uptake, such as water temperatures. Primary sampling stations (also referred to as indicator or sentinel stations) should be located at sites where toxin is most likely to first appear, based either on past experience or knowledge of site conditions. The geographic distribution for collection of samples should take into consideration the randomness of toxic algal blooms. Establishing the frequency and period for collection of samples to identify an event as early as possible is an important consideration.

Sample collection, sample transportation, and sample analysis procedures should be developed, and predictable timeframes established between collection and results. The Authority should ensure that in an emergency, such as a suspected biotoxin illness, the normal timeframe can be compressed, and sample results known as quickly as possible. It is important to consider emergency coverage schedules for staff and lab availability outside of normal office hours during harmful algal bloom events.

When an early warning system detects increased toxicity/cell counts or other information suggests that toxin levels are increasing, it is important that the Authority have procedures to promptly expand sampling to additional stations and/or increase the frequency of sampling for marine biotoxins. The procedures should include plans for obtaining the additional resources necessary to implement the expanded sampling and laboratory analysis program.

C. Pre-harvest shellfish toxicity testing: this strategy involves sampling and testing shellfish meats for the presence of marine biotoxins in the intended harvest area specifically in advance of harvesting. This strategy, if used

independent of any other strategy, shall permit harvest in specific geographic locations and for short durations. This strategy may also be used in combination with other management strategies and should be considered as a complementary strategy while developing datasets for alternative management strategies (e.g. pre-harvest shellfish toxicity testing in combination with phytoplankton monitoring which can evolve into a robust shellfish toxicity monitoring strategy).

This strategy requires representative samples that cover the spatial distribution of the area to be harvested. The duration of permitted harvest following sampling will vary based on the species being tested and the historical database available to inform the sampling strategy. ~~A dataset with at least 36 samples per harvest area shall be developed before the biotoxin monitoring plan may be modified.~~ Without at least 36 samples per harvest area over the span of at least three (3) years, the short duration of permitted harvest shall not exceed three (3) days from the time of shellfish collection for toxicity testing to harvest. The dataset could then be used to modify the duration of permitted harvest.

This management strategy can be applied to harvest areas where collecting, transporting and processing shellfish samples is feasible. This management strategy can be applied to aquaculture or wild harvest. Appropriate venues for this management strategy include but are not limited to; easily accessible and remote wild harvest areas and aquaculture sites in state and federal waters. If toxicity in excess of the established threshold in Section II. Chapter IV. @.04 C. is detected, the growing area must be either be placed in the closed or controlled access status.

A~~The~~ marine biotoxin management plan that incorporates this strategy must also establish:

- ~~appropriate screening levels,~~
- ~~appropriate methods,~~
- ~~appropriate laboratory(s)/analyst(s),~~
- ~~an appropriate sampling plan,~~
- ~~appropriate sampling frequency,~~
- a defined harvest area, and;
- appropriate duration for permitted harvesting subsequent to sampling.

This strategy is specifically for permitting harvest following shellfish testing. The duration of permitted harvesting will depend on the species being tested, the risk of increasing toxicity and the timing of additional sampling. Samples must be representative of the harvest area.
Methods shall be used in accordance with Section IV. Guidance Documents Chapter II Growing Areas .14 or Section II. Chapter III. @.02 C.

D. Shellfish lot testing: this strategy involves sampling and testing shellfish meats for the presence of marine biotoxins on a lot basis after harvest. This strategy may be combined with a pre-harvest shellfish toxicity testing strategy, the results of which permit harvest. Lot testing may also be used on a case by case basis to clear product harvested immediately prior to a biotoxin closure if the Authority determines it is necessary.

This strategy requires representative samples for each lot of harvested shellstock. Lot testing shall be permitted in growing areas in the Controlled Access Status and require Restricted Shellstock tags. The conditions for the

area in Controlled Access Status shall be defined in the harvest permit and may include holding shellstock until lot tests are available. ~~A dataset with at least 36 samples per harvest area over the span of at least three (3) years shall be developed before the biotoxin monitoring plan may be modified.~~

This management strategy can be applied to all growing areas where harvest occurs. This management strategy can be applied to aquaculture or wild harvest. Appropriate venues for this management strategy include but are not limited to; easily accessible and remote wild harvest areas and aquaculture sites in state and federal waters.

~~The~~ A marine biotoxin management plan that incorporates this strategy must also establish:

- ~~appropriate screening levels,~~
- ~~appropriate methods,~~
- ~~appropriate laboratory(s)/analyst(s),~~
- ~~an appropriate sampling plan,~~
- ~~appropriate sampling frequency, and;~~
- representative number of samples per lot.

Methods shall be used in accordance with Section IV. Guidance Documents Chapter II Growing Areas.14 or Section II. Chapter III. @.02 C.

E. Pre-harvest shellfish toxicity screening and lot testing: this strategy requires pre-harvest shellfish toxicity screening of the intended harvest area coupled with shellfish lot testing upon landing or receipt at the initial certified dealer.

This strategy shall permit harvest from intended harvest areas in the Controlled Access Status and require Restricted Shellstock tags. The conditions for the area in Controlled Access Status shall be defined in the harvest permit and may include holding shellstock until lot tests results are available. ~~A dataset with at least 36 samples taken monthly per harvest area spanning at least three (3) years shall be developed before the biotoxin monitoring plan may be modified.~~ In the absence of an adequate dataset, the initial number and frequency of pre-harvest and lot samples must be sufficient to conduct an evaluation of risk in the intended harvest area. The initial number of samples must be adequate to address the size of the intended harvest area and the amount of shellfish harvested. Single samples are not adequate for evaluation of risk. Should initial samples indicate minimal toxin levels or the absence of toxins, sampling can be reduced but must be conducted at least monthly or as often as necessary to monitor risk.

This management strategy can be applied to all growing areas where harvest occurs. This management strategy can be applied to aquaculture or wild harvest. Appropriate venues for this management strategy include but are not limited to; easily accessible and remote wild harvest areas and aquaculture sites in state and federal waters.

~~The~~ A marine biotoxin management plan that incorporates this strategy must also establish:

- ~~appropriate screening levels,~~
- ~~appropriate methods,~~
- ~~appropriate laboratory(s)/analyst(s),~~
- ~~an appropriate sampling plan,~~
- ~~appropriate sampling frequency,~~

- a defined harvest area, and;
- representative number of samples.

~~Methods shall be used in accordance with Section IV. Guidance Documents Chapter II Growing Areas.14 or Section II. Chapter III. @.02 C.~~

Heat Processing

In shellfish growing areas where low levels of biotoxins routinely occur, harvesting for thermal processing, referred to in the Model Oridinance as heat processing, purposes may be an alternative to consider. Thermal processing as defined by applicable FDA regulations (21 CFR 113) may reduce the biotoxin concentration of the shellfish via dilution, not destruction. While thermal processing has been demonstrated more for PSP toxins (Berenguer et al., 1993; Vieites et al. 1999; Dong et al., 2022), there are limited studies for the reduction of ASP and DSP toxins (McCarron et al 2008 and Vidal et al 2009). If thermal processing is practiced, the Authority must develop and implement procedures to control the harvesting and transportation of the affected shellfish to the processing plant, as well as end product testing of processed shellfish.

Shellfish Meat Analyses and Toxin Profiles
Section II. Chapter III. @.02 C
Section IV. Guidance Documents Chapter II Growing Areas.14

There are a wide range of methodologies developed for screening and confirmation of toxic
phytoplankton and their toxins. Only methods adopted into the NSSP can be implemented for the purpose of confirming toxin concentration levels in shellfish and making decisions to reopen growing areas. Additionally, some screening methods have been evaluated by the ISSC and found fit for purpose for the NSSP, thereby providing confidence in their use for specific screening purposes.

Toxin analyses methods fall into two (2) categories in the NSSP:

1. Approved Methods for Marine Biotoxin Testing (Section IV. Guidance Documents Chapter II Growing Areas .14 Table 2.); and
2. Approved Limited Use Methods for Marine Biotoxin Testing (Section IV. Guidance Documents Chapter II Growing Areas .14 Table 4.).

The methods within these categories range from mouse bioassays to immunochromatography and other antibody-based platforms to chemical analytical methods such as high-performance liquid chromatography (HPLC). The mouse bioassay historically has been the most universally applied technique for examining shellfish toxins. Other bioassay procedures have been developed and are becoming more generally applied. In recent years, considerable effort has been applied to development of chemical analyses to replace or provide alternatives to in-vivo (live animal) bioassays. For toxins that have no method adopted into the NSSP, best available science is employed and emergency use adoption may be considered following the requirements described in Model Ordinance Chapter III. Laboratory @ .02 Methods.

The following table provides ~~a survey of the laboratory methods available~~

information for each toxin covered by the NSSP.

Paralytic Shellfish Poisoning (PSP) Toxins	
Analogs	Water-soluble alkaloid neurotoxins that are collectively referred to as saxitoxins or paralytic shellfish toxins (PSTs). To date 57 analogs have been identified, although not all are always present, and they vary greatly in overall toxicity (Wiese et al., 2010). In addition to saxitoxin (the parent compound), monitoring laboratories typically analyze for approximately 12 other analogs that may contribute measurably to toxicity.
Guidance Level	0.8 ppm (80 µg saxitoxin equivalents /100 g tissue).
Origin	The regulatory limit was set in the 1930s (Wekell et al., 2004). The minimum concentration of PSP toxin that will cause intoxication in susceptible persons is not known. Epidemiological investigations of PSP in Canada, however, have indicated 200 to 600 micrograms of PSP toxin will produce symptoms in susceptible persons. A death has been attributed to the ingestion of a probable 480 micrograms of PSP toxin. Investigations indicate that concentrations of less than 200 ug of the toxin have no deleterious effects on humans.
Shellfish Lab Methods	The mouse bioassay is still the most widely accepted detection method for the saxitoxins around the world and has been shown to adequately protect the public's health. In 2009, the Interstate Shellfish Sanitation Conference approved a post-column oxidation HPLC-PCOX method, making it the newest regulatory method available for PSP toxins in the U.S. The receptor binding assay (RBA), a competition assay whereby radiolabeled saxitoxin competes with unlabeled saxitoxin for a finite number of available receptor sites, provides a measure of overall PSP toxicity in a sample (Van Dolah et al. 2009). The RBA was approved for mussels and approved limited use for clams and scallops in 2014.
General Molluscan ShellfishAssociations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
Neurotoxic Shellfish Poisoning (NSP) Toxins	
Analogs	Comprised of more than 10 lipid-soluble cyclic polyethers. Several analogs and metabolites have been identified. NSP-causing toxins in shellfish include intact algal brevetoxins and their metabolites (collectively known as neurotoxic shellfish toxins or NSTs) (Plakas and Dickey, 2010).
Guidance Level	0.8 ppm (20 mouse units/100 g tissue or 80 µg brevetoxin-2 equivalents /100 g tissue)
Origin	Uncooked clams from a batch eaten by a patient in Florida with NSP symptoms were found to contain

		118 mouse units per 100 grams of shellfish meat. However, consumption of even a few contaminated shellfish may result in poisoning and the severity of the disease may be dependent on many factors, including dose, bodyweight, underlying medical conditions, and the age of the victim as well as possibly the toxin mixture of the particular bloom (Watkins, 2008).
	Shellfish Lab Methods	The approved NSSP method for NSP toxins is the mouse bioassay. The MARBIONC ELISA is approved for limited use. Efforts are underway to validate *in vitro* methods for detection of brevetoxins in shellfish. The methods that follow may be used for screening purposes. For example, rapid, sensitive ELISA test kits already are commercially available for this purpose. Biomarkers of brevetoxin contamination in shellfish have been identified by using LC/MS. Structural confirmation of these metabolites and brevetoxins in shellfish can be made by LC/MS, a method that offers high sensitivity and specificity. A method for detection, identification, and quantification of brevetoxins is HPLC-MS. Radioimmunoassay (RIA) and Receptor Binding Assay (RBA) are also under current use (Watkins, 2008). Available detection methods are not equal in their ability to measure naturally produced brevetoxins, and most methods are hampered by the absence of specific reference standards for brevetoxin congeners (Watkins, 2008).
	General Molluscan Shellfish Associations	Oysters and clams.
	Amnesic Shellfish Poisoning (ASP) Toxin	
	Analogs	The neurotoxin domoic acid is a water-soluble, non-protein, excitatory amino acid. Isomers of domoic acid have been reported but are less toxic than domoic acid itself.
	Guidance Level	20 ppm (2mg domoic acid/100 g tissue)
	Origin	In 1987 in eastern Canada, domoic acid poisonings sickened individuals, leading to Health Canada's establishment of the regulatory limit (Wekell, 2004).
	Shellfish Lab Methods	The NSSP approved method for detecting domoic acid in seafood is a reversed-phase HPLC method with ultraviolet (UV) detection. The Reveal 2.0 ASP is an approved limited use method. There is an AOAC approved ELISA for the detection of domoic acid which may be used for screening purposes.
	General Molluscan Shellfish Associations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
	Diarrhetic Shellfish Poisoning (DSP) Toxins	
	Analogs	A group of lipid-soluble polyether toxins that includes okadaic acid (OA), the dinophysistoxins (DTXs), and a series of fatty acid esters of okadaic acid and the dinophysistoxins (collectively known as DSTs) (Uchida, 2018).
	Guidance Level	0.16 ppm (0.16 mg total okadaic acid equivalents/kg tissue). Total okadaic acids equivalents equal

	combined free okadaic acid, dinophysistoxins, acyl-esters of okadaic acid and dinophysistoxins.
Origin	Established by FDA in 2011 for total (esterified plus nonesterified okadaic acid and the dinophysistoxins (Trainer, 2013).
Shellfish Lab Methods	Until recently, DSP was managed by mouse bioassay and/or monitoring shellfish growing waters for the presence of *Dinophysis* organisms. Unfortunately, the dose-survival times for the DSP toxins in the mouse assay vary considerably, and fatty acids interfere with the assay, giving false-positive results. A suckling mouse assay has been developed and used for control of DSP. This assay measures fluid accumulation after injection of the shellfish extract. In 2017 an LCMS/MS method for quantifying dinophysistoxins in clams was approved in the NSSP. For other species, the best available science is recommended.
General Molluscan Shellfish Associations	Mussels, clams, cockles, oysters, and scallops (excluding the scallop adductor muscle).
Azaspiracid Shellfish Poisoning (AZP) Toxins	
Analogs	The lipid-soluble toxin azaspiracid and several derivatives (AZAs). More than 30 AZA analogs have been identified, with three analogs routinely monitored in shellfish (AZA1, AZA2, and AZA3).
Guidance Level	0.16 ppm (160 µg azaspiracid-1 equivalents/kg tissue)
Origin	Estimation of consumption of a single portion of shellfish and through estimate of an Acute Reference Dose. Derived from epidemiological observations caused by a mixture of naturally occurring analogs (AZA 1, 2, and 3). Based on methods available in 2001.
Shellfish Lab Methods	AZAs are not routinely monitored in shellfish harvested in the U.S., but, in the EU, the mouse bioassay has been used. As for many of the lipophilic toxins, the mouse assay is not adequately sensitive or specific for public- health purposes. *In vitro* assays and analytical methods are now available to assess the toxicity of AZA-contaminated shellfish and to confirm the presence of AZA analogs in shellfish. These methods are in various stages of validation for regulatory use around the world. LC/MS is used as a confirmatory method for AZA, providing unambiguous structural confirmation of AZA analogs in shellfish samples. Currently, there is no NSSP method for AZP toxins.
General Molluscan Shellfish Associations	Detected in mussels, oysters, scallops (excluding the scallop adductor muscle), clams, and cockles.

Resources

The 2012 version of FDA's Bad Bug Book, Foodborne Pathogenic Microorganisms and Natural Toxins, is a comprehensive resource from which a great deal of information has been used for the toxin profiles in the table above. It is accessible at https://www.fda.gov/media/83271/download

For more discussion of chemical structures and properties, methods of analysis, source organisms and habitat, occurrence and accumulation in shellfish, toxicity of toxins, prevention of intoxication, cases and outbreaks, and regulations and monitoring, see the FAO Paper 80: Marine Toxins. This may be accessed as follows:

- **Paralytic Shellfish Poisoning** http://www.fao.org/3/y5486e/y5486e05.html
- **Diarrhetic Shellfish Poisoning** http://www.fao.org/3/y5486e/y5486e0e.html
- **Neurotoxic Shellfish Poisoning** http://www.fao.org/3/y5486e/y5486e0o.html
- **Amnesic Shellfish Poisoning** http://www.fao.org/3/y5486e/y5486e0n.html
- **Azaspiracid Shellfish Poisoning** http://www.fao.org/3/y5486e/y5486e0p.htm
- **References** http://www.fao.org/3/y5486e/y5486e0t.htm

The FDA online course, Shellfish Growing Areas, introduces participants to requirements and procedures under the NSSP to ensure that shellfish are harvested from safe waters. The course contains a significant section addressing marine biotoxins. The course may be accessed at https://www.accessdata.fda.gov/ORAU/ShellfishGrowingAreas/SGA_summary.htm.

Additional information from the Centers for Disease Control and Prevention, Morbidity and Mortality Weekly Report (MMWR) contains illness reports related to these toxins. This may be accessed at https://www.cdc.gov/mmwr/index.html.

NIH/PubMed: Various Shellfish-Associated Toxins provides a list of research abstracts in the National Library of Medicine's MEDLINE database. The specific seafood with which each toxin generally is associated is included in the profiles above to help readers link symptoms to potential sources. However, all shellfish (filter-feeding mollusks, as well as the carnivorous grazers that feed on these mollusks (such as whelks, snails, and, in some cases, even lobsters and octopi), may become toxic in areas where the source algae are present.

Model Ordinance, Public Health Reasons & Explanations, Guidance, and Appendices References

- Section I. Purposes & Definitions
- Section II. Model Ordinance –Chapter III. Laboratory @.02 Methods (C) and (D)
- Section III. Public Health Reasons and Explanations– Chapter IV. Shellstock Growing Areas @.04 Marine Biotoxin Control (A)
- Section IV. Guidance Documents– Chapter II. Growing Areas @.03 Determining the Size of a Closed Area as a Result of Illnesses
- Section IV. Guidance Documents– Chapter II. Growing Areas @.04 Determining the Harvesting Periods Associated with Implicated Product for Identifying Shellfish to Be Included in the Recall
- Section IV. Guidance Documents - Chapter II. Growing Areas @.05 Determining the Scope of Implicated Product for Conducting a Recall
- Section IV. Guidance Documents – Chapter II. Growing Areas @.08 Action Levels, Tolerances and Guidance Levels for Poisonous or Deleterious Substances in Seafood
- Section IV. Guidance Documents – Chapter II. Growing Areas @.12 Growing Area Patrol and Enforcement
- Section IV. Guidance Documents – Chapter II. Growing Areas @.13 Control of Shellfish Harvesting

- Section IV. Guidance Documents – Chapter II. Growing Areas @.14 Approved NSSP Laboratory Tests
- Chapter XVI. Recalls, Closures, and Special Events Checklist & Appendices

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Action by 2023 Task Force I	Recommends adoption of the Biotoxin Committee recommendation on Proposal 19-124.

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<tr><td>Proposal Subject</td><td>Laboratory Method for Vibrio parahaemolyticus and Vibrio vulnificus Enumeration and Detection Through MPN and Real-Time PCR</td></tr>
<tr><td>Specific NSSP Guide Reference</td><td>Section IV Guidance Documents Chapter II Growing Areas .14 Approved NSSP Laboratory Tests</td></tr>
<tr><td>Text of Proposal/ Requested Action</td><td>5. Approved Methods fir Vibrio Enumeration

<table>
<tr><th></th><th>Vibrio Type:</th><th>Application : PHP Sample Type:</th><th>Application : Reopening</th></tr>
<tr><td>EIA[1]</td><td>Vibrio vulnificus (V.v.)</td><td>X</td><td></td></tr>
<tr><td>MPN[2]</td><td>Vibrio vulnificus (V.v.)</td><td>X</td><td></td></tr>
<tr><td>SYBR Green 1 QPCR-MPN[5]</td><td>Vibrio vulnificus (V.v.)</td><td>X</td><td></td></tr>
<tr><td>MPN[3]</td><td>Vibrio parahaemolyticus (V.p.)</td><td>X</td><td></td></tr>
<tr><td>PCR[4]</td><td>Vibrio parahaemolyticus (V.p.)</td><td>X</td><td></td></tr>
<tr><td>MPN-Real Time PCR[6]</td><td>tdh+ and trh+ Vibrio parahaemolyticus (V.p.)</td><td>X</td><td>X</td></tr>
<tr><td>MPN-Real Time PCR[7]</td><td>Vibrio parahaemolyticus (V.p.)</td><td>X</td><td>X</td></tr>
<tr><td>MPN-Real Time PCR[9]</td><td>Vibrio parahaemolyticus (V.p.) and Vibrio vulnificus (V.v.)</td><td>X</td><td>X</td></tr>
<tr><td>Direct Plating Method[8]</td><td>Vibrio parahaemolyticus (V.p.)</td><td>x</td><td>X</td></tr>
</table>
Bacteriological Analytical Manual, 7th Edition, 1992.

2 MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, followed by confirmation using biochemical analyses</td></tr>
</table>

	or by the DNA -alkaline phosphatase gene probe for vvhA as described by Wright et al., or a method that a State can demonstrate is equivalent. [3] MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, followed by confirmation using biochemical analyses or the DNA-alkaline phosphatase gene probe for tlh as described by McCarthy et al., or a method that a State can demonstrate is equivalent. [4] MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, and as described in the "Direct Plating Procedure for the Enumeration of Total and Pathogenic *Vibrio parahaemolyticus* in Oyster Meats" developed by FDA, Gulf Coast Seafood Laboratory, or a method that a State can demonstrate is equivalent. [5] *Vibrio vulnificus*, ISSC Summary of Actions 2009. Proposal 09-113, Page 123. [6] MPN-Real Time PCR Method for the tdh and trh Genes for Total *V. parahaemolyticus* as described in Kinsey et al., 2015. ISSC 2015 Summary of Actions Proposal 15-111, Page 397. [7] MPN-Real Time PCR Method for the *tlh* gene for total *V. parahaemolyticus* as described in Kinsey et al., 2015. ISSC 2015 Summary of Actions Proposal 15-113, Page 418 [8] Direct Plating Procedure in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, and as described in the 'Direct Plating Procedure for the Enumeration of Total and Pathogenic *Vibrio parahaemolyticus* in Oyster Meats' developed by FDA, Gulf Coast Seafood Laboratory. [9] MPN-Real Time PCR Method for *Vibrio parahaemolyticus* and *Vibrio vulnificus.* Washington State Department of Health, Food and Shellfish Bacteriology Laboratory.
Public Health Significance	The purpose of this method is to provide laboratories supporting the NSSP the ability to rapidly quantify *Vibrio parahaemolyticus (Vp)* and *Vibrio vulnificus (Vv)* from oysters using a high throughput real-time PCR assay. Rapid and early detection of these pathogens, complying with the required quantitative detection guidelines suggested by the ISSC, will help the shellfish industry market oysters for consumption that are within regulatory limits for these pathogens. This method once approved would add a testing method of MPN Real-Time PCR for *Vibrio vulnificus* and it would be an alternative to the *Vibrio parahaemolyticus* MPN Real-Time PCR methods already approved in the 2017 Model Ordinance.
Cost Information	The cost for this method is approx. $155 per sample. This estimate is based on recurring costs of consumables, reagents, and supplies needed for routine testing. It does not include indirect materials considered to be standard microbiology equipment such as analytical balance, PCR workstation, DNA purification system, refrigerator, pipettes, etc.
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-128 to an appropriate committee as determined by the Conference Chair.

Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-128.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-128.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-128.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 19-128 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-128.

Submitter	Leonora Porter - Spokesperson
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Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist – Reagent Water Quality
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas, .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists, 1. NSSP Laboratory Evaluation Checklist for Microbiology.
Text of Proposal/ Requested Action	The requested action is to adopt the modified text and update the reference in Section 1.7 Media Preparation for checklist item 1.7.6.
Public Health Significance	The suggested change addresses the importance of accurate information used in laboratory Quality Assurance Programs (QAPs) for recommended limits for the quality of reagent water used for microbiology testing by correcting the maximum acceptable limits for conductivity and resistivity testing based on the most current ***Standard Methods*** Edition. For 26 years, the incorrect units of measure for conductivity and resistivity have been printed in laboratory reference materials: ***Standard Methods for the Examination of Water and Wastewater***, 1992, 18th Edition; ***Standard Methods***, 2012, 22nd Edition; and ***Standard Methods***, 2017, 23rd Edition. The QA information is finally corrected in the ERRATA, dated 5/29/18 for ***Standard Methods*** 23rd Edition. The material states "In Section 9020, Table 9020:II (p. 9-14), the recommended Maximum Acceptable Limit for Conductivity Test should be "<2 µmhos/cm (µSiemens/cm) at 25°C." The incorrect "resistance" statement from the 18th Edition is removed in the 22nd and 23rd Editions of ***Standard Methods***. The resistivity (also called specific resistance) is the reciprocal of the conductivity, not resistance. A resistivity recommendation can be found in the Reagent Grade Water section.
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-131 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-131.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-131.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-131.
Action by 2023 Laboratory Committee	Recommended no action on Proposal 19-131. Rationale: There is no justification for changing the resistivity value in Line Item 1.7.6.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-131.

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Proposal Subject	Microbiology Laboratory Evaluation Checklist - Working Thermometers
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas, .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists, 1. NSSP Laboratory Evaluation Checklist for Microbiology
Text of Proposal/ Requested Action	The requested action is to adopt the modified text of the NSSP microbiology checklist, section 1.4 Laboratory Equipment, item 1.4.24:
Public Health Significance	The laboratory's goal is to ensure high-quality data using accepted scientific practices. The designated changes incorporate recommended best practices from a current recognized scientific publication. These types of acknowledged practices are used to develop a laboratory's Quality Assurance Program (QAP). The *verification* of working thermometers is now suitably referenced to support past and present practices in program laboratories and *recommends a rejection component (new)*. The newer/current reference material is cited to strengthen confidence in the acceptability of past practices for "checking" accuracy in working temperature monitoring devices. ***Standard Methods,*** 23rd Edition, states "Annually, or preferably semiannually, **verify** the accuracy of all working temperature-sensing devices (e.g., liquid-in-glass thermometers, thermocouples, and temperature-recording instruments) at the use temperature(s). To do this, compare each device's measurements to those of a certified NIST temperature-sensing device or one traceable to NIST and conforming to NIST specifications. Discard temperature-sensing devices that differ by >1ºC from the reference device."
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-132 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-132.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-132.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-132.
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 19-132 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-132.

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Proposal Subject	Microbiology & PCR Laboratory Evaluation Checklists - Working Thermometers
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas, .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists, NSSP Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt modified working thermometer language for these two NSSP laboratory evaluation checklists items. The modification is to remove the word "calibrated" and add thermometer accuracy requirements.
Public Health Significance	There are currently no NSSP accuracy criteria established for Liquid-in-Glass thermometers. This proposal establishes uncertainty requirements that should be considered prior to purchase since all thermometers and temperature recording devices are not created equally. Quality Assurance and Standardization are integral to the validity of the NSSP laboratory. For thermometers there are several factors that influence temperature readings; therefore, controlling thermometer accuracy will impact thermometer standardization across NSSP laboratories. A thermometer's accuracy is a product of its *manufacturing uncertainty*, *measurement uncertainty* and *environmental uncertainty* which all must be considered and evaluated by the purchaser. Only thermometers that are manufactured accurately and are found *fit for purpose* for the NSSP laboratory should be purchased. Some Liquid-in-Glass thermometers are manufactured with accuracies (> 0.2ºC) that are greater than the water bath temperature limit of ±0.2°C; these thermometers should not be purchased for the NSSP laboratory. As stated in Reference #4, NIST Monograph 150 "the accuracy attainable is principally limited by the characteristics of the thermometer itself." Therefore, a working thermometer's accuracy should be assessed prior to purchase. Calibration is performed post purchase. *Calibration quantifies <u>only</u> the temperature* ***measurement uncertainty*** *at the single temperature point assessed.* Calibration without also considering the ***manufacturing uncertainties*** of the thermometer is inaccurate: generating a false security for accuracy. Calibration values are only accurate at the environmental conditions found within the calibration laboratory; when total immersion thermometers are immersed to the test temperature being measured with the emergent stem at ambient temperature. In the NSSP laboratory, the emergent stem <u>is not</u> at ambient temperature. This creates ***environmental uncertainty*** which invalidates the calibration certificate and requires experience and knowledge in generating an accurate stem correction. An inaccurate stem correction compounds the degree of error in the final temperature

	reading. The current NSSP practice of calibrating an inappropriate thermometer against the undefined calibration standard (NIST, ASTM, Primary, Secondary, etc) and then using this thermometer incorrectly in the laboratory environment negates any assurance received by having a calibration certificate. This practice would not be legally defensible. NSSP Quality Assurance and Standardization would be better served to establish manufacturing accuracy requirements that only allow for the use of appropriate working thermometers. *These working thermometers will then be verified against a calibrated standards thermometer, that is traceable to NIST in section 1.4.24.* Savings: Calibration costs per thermometer: $125 for the first point and $60 for each additional point. Most lab are locked into local calibration facilities, within driving distance of their labs, if their thermometers are mercury. Postal hazard restrictions prohibit mercury thermometers being shipped in the mail.
Cost Information	none
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-133 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-133.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-133.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-133.
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 19-133 as amended.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-133.

Submitter	US Food and Drug Administration (FDA)
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Proposal Subject	NSSP DSP Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt the laboratory evaluation checklist for Diarrhetic Shellfish Poisoning LC-MS/MS.
Public Health Significance	The Diarrhetic Shellfish Poisoning (DSP) LC-MS/MS checklist will provide the means of assessing the competence of the laboratory to perform the test method.
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-136 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-136.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-136.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-136.
Action by 2021 Laboratory Committee	Recommended adoption of Proposal 19-136 as amended with Interim Approval by the Executive Board
Action by 2021 ISSC Executive Board	Granted Interim Approval in effect until the Conference convenes at the 2023 ISSC Biennial Meeting.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-136.

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Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt the modified text of four (4) NSSP microbiology checklist items in the Laboratory Equipment and Sterilization and Decontamination sections; said NSSP checklist items are 1.4.5, 1.4.21, 1.6.10, and 1.6.11.
Public Health Significance	The proposed modifications are to improve consistency in current NSSP microbiology checklist language and account for technology improvements to laboratory equipment.
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-138 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-138.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-138.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-138.
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 19-138 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-138.

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Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt the modified text of the attached checklist for Bacteriological Examination of Soft-shelled Clams and American Oysters for Male Specific Coliphage (MSC), starting at section 3.10.
Public Health Significance	The proposed modifications are to provide clarification to bench analysts and LEOs for consistent performance and evaluation of the method for the NSSP.
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-140 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-140.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-140.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-140.
Action by 2022 Laboratory Committee	Recommended adoption of Proposal 19-140 as amended with Interim Approval by the Executive Board
Action by 2022 ISSC Executive Board	Granted Interim Approval in effect until the Conference convenes at the 2023 ISSC Biennial Meeting.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-140.

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Proposal Subject	NSSP Receptor Binding Assay for Paralytic Shellfish Poisoning (PSP) Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to adopt the laboratory evaluation checklist for the Receptor Binding Assay for Paralytic Shellfish Poisoning (PSP).
Public Health Significance	The Receptor Binding Assay for Paralytic Shellfish Poisoning (PSP) checklist will provide the means of assessing the competence of the laboratory to perform the test method.
Cost Information	N/A
Action by 2019 Laboratory Committee	Recommended referral of Proposal 19-141 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force I	Recommended the adoption of Laboratory Committee recommendation on Proposal 19-141.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-141.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-141.
Action by 2022 Laboratory Committee	Recommended adoption of Proposal 19-141 as amended with Interim Approval by the Executive Board
Action by 2022 ISSC Executive Board	Granted Interim Approval in effect until the Conference convenes at the 2023 ISSC Biennial Meeting.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-141.

Proposal No. 19-144

Submitter	Thomas Howell
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Proposal Subject	Guidance for Assessing the Viral Impact from Waste Water Treatment Plant Outfall on Adjacent Growing Areas using the Male-specific Coliphage Method on Effluent Samples.
Specific NSSP Guide Reference	Section IV Guidance Documents - Chapter II. Growing Areas - .19 Classification of the Shellfish Growing Waters Adjacent to Waste Water Treatment Plants
Text of Proposal/ Requested Action	The requested action is that an ISSC committee be formed to draft guidance language describing how to best use MSC effluent sampling techniques to assess the viral impact on adjacent growing areas. This proposed action is the result of recent collaborative work funded by New Hampshire Sea Grant. The PI's and project participants on this project included University of New Hampshire Sea Grant, Connecticut Sea Grant, Spinney Creek Shellfish, Connecticut Department of Agriculture, New Hampshire Department of Environmental Services, US Food and Drug Administration Center for Food Safety and Applied Nutrition, and US Food and Drug Administration Gulf Coast Seafood Laboratory. An optimized method to determine MSC in effluent samples, both pre-treatment (disinfection) and final effluent has been submitted to the Lab Committee for approval. Two years of field studies were recently completed which looked closely at 2 plants in CT and 4 plants in NH. Results of these field studies were reported at the 2019 NESSA meeting in Plymouth MA. By taking effluent samples from WTP's two to three times per week over an extended period, a database can be assembled including Geomean and P95 values in a strategy consistent with NSSP practices. Plotting the effluent time-series data can be used to identify times when plant performance is degraded by predictable, challenging, conditions whether they are operational or environmental. By informing dye study work with WWTF effluent analysis, much more informed decisions can be made with respect to classification of adjacent growing waters. Simply multiplying the P95 results from final effluent statistical analysis by the dilution line in question, an upper level of MSC concentration MSC in the growing waters can be estimated. An interpretation matrix for final effluent MSC time-series analysis to interpret results in a relative way is proposed.
Public Health Significance	The Public Health Significance of this proposal is substantial. Dye studies alone are protective of public health using the 1000:1 dilution line for classification purposes. However, MSC assessment of effluent samples gives a much more informed picture of how appropriate the 1000:1 line is in a particular situation. If an under-designed, problematic WWTP is not adequately deactivating viruses, a higher dilution may be required. This is an important consideration when dealing with a WWTP that does not perform to typical standards of secondary treatment with effective disinfection. However, the study has shown that many modern and advanced WWTPs can be reliably operated at sufficient performance levels to justify the 300:1 dilution line for the establishment of a prohibited classification

	around the WWTP outfall. As time continues and WWTPs are upgraded, this method and technique may permit increased utility of the growing area between the 300:1 and 1000:1 dilution line. In conclusion, public health can be informed and optimized while maximum commercial utilization of growing areas can be achieved.
Cost Information	The MSC method for WWTP effluent samples is inexpensive and easy to perform. Costs become more significant when one considers the personnel and travel time needed to sample the WWTP's. The state control agency can optimize this work by focusing field work during the winter months when the WWTP are likely more challenged and personnel resources are more available.
Action by 2019 Task Force I	Recommended referral of Proposal 19-144 to an appropriate committee as determined by the Conference Chairman.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-144.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-144.
Action by 2023 Male Specific Coliphage Committee	Recommended referral of Proposal 19-144 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Male Specific Coliphage Committee recommendation for Proposal 19-144.

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Proposal Subject	Guidance on cleansing studies
Specific NSSP Guide Reference	NSSP Section IV Chapter II .19 VI B.
Text of Proposal/ Requested Action	B. Guidance for a Conditional Area Management Plan The management plan for a growing area in the conditionally approved or conditionally restricted classification must meet certain minimum requirements to ensure that the safety of the shellfish for human consumption is maintained. The use and success of the conditional classification depends upon a thorough and accurate management plan. Therefore, it is important that all aspects of the management plan be fully considered and implemented. The minimum requirements to be addressed are: (1) An understanding of and an agreement to the conditions of the management plan by the one (1) or more Authorities involved, other local, State and Federal agencies which may be involved, the affected shellfish industry, and the persons responsible for the operation of any treatment plants or other discharges that may be involved; (2) A written management plan for the growing area being placed in the conditional classification, which includes a general description of the growing area with a map showing the area's boundaries, and which addresses all items in C. through H. (3) A sanitary survey that shows the growing area will be in the open status of its conditional classification for reasonable periods of time. The survey must provide a description of the factors determining the growing area's suitability for being classified conditionally approved or conditionally restricted, and the supporting information and data. (4) A description of the predictable pollution event or events that are being managed and the performance standards established for each pollution source contributing to the pollution event including: (a) For a wastewater treatment facility, the performance standard should be based on: (i) Peak effluent flow (ii) Bacteriological quality of the effluent (iii) Physical and chemical quality of the effluent (iv) Bypasses from the treatment plant or its collection system (v) Design, construction, and maintenance to minimize mechanical failure or overloading (i.e., the reliability of the treatment system and collection system components)

(vi) Provisions for verifying and monitoring efficiency of the wastewater treatment plant and the feedback system for addressing inadequate treatment.

(vii) Identification of conditions that lead to WWTP failure, a lapse in WWTP treatment leading to untreated or partially treated sewage discharge, and closure of the conditionally approved area.

(b) For meteorological or hydrological events, the performance standard should be based on:

(i) Identification of the specific meteorological and/or hydrologic event that will cause the growing area to be placed in the closed status;

(ii) Discussion and data analyses concluding that effects on water quality from these specific meteorological and/or hydrologic events are predictable, and that the data are sufficient to establish meaningful performance standards or criteria for the establishment and implementation of a management plan for the growing area placed in the conditional classification; and

(iii) The predicted number of times, based on historical findings, that the pollution event will occur within one (1) year.

(c) For seasonal events, such as marina operation, seasonal rainfall, and waterfowl migration, the performance standard should be based on:

(i) Identification of the seasonal event that will cause the growing area to be placed in the closed status, including its estimated duration; and

(ii) Discussion and data concluding that the seasonal event is predictable, and that the data are sufficient to establish meaningful performance standards or criteria for the establishment and implementation of a management plan for a growing area placed in the conditional classification;

(5) A description of the plan for monitoring water quality including numbers and frequency;

(6) A description of how the closed status for the conditional classification will be implemented, which must include:

(a) A clear statement that when the performance standards are not met, the growing area will immediately be placed in the closed status;

(b) A requirement to notify the Authority or Authorities that the management plan performance standards have not been met, including:

(i) The name of the agency or other party responsible for notifying the Authority;

(ii) The anticipated response time between the performance standards not being met and notification of the Authority; and

(iii) The procedures for prompt notification including contingencies such as night, weekend and absences of key personnel;

(c) A description of the implementation and enforcement, including:

(a) The response time between the notification to the Authority of the failure to meet performance standards and activation of the legal closure of the growing area by the Authority;

(b) The procedures and methods to be used to notify the shellfish industry; and

(c) The procedures and methods to be used to notify the patrol agency (enforcement agency) including:

- The name of the responsible patrol agency;
- The anticipated response time between the Authority's legal closure of the growing area and notification of closure to the patrol agency; and
- A description of the patrol agencies anticipated activities to enforce the closed status.

(7) A description of the criteria that must be met prior to reopening a growing area in the closed status, including the need to determine that:

(a) The performance standards established in the management plan are again fully met;

(b) The flushing time for pollution dissipation is adequate;

(c) A time interval has elapsed which is sufficient to permit reduction of human pathogens as measured by the coliform indicator group in the shellstock; . Studies shall be conducted to document the time interval necessary for the reduction of coliform levels in the shellstock to pre-closure levels. The Authority shall develop and implement a study design that includes:

(i) The utilization of NSSP-conforming laboratories and NSSP-approved methods to analyze coliform in shellstock and water.

(ii) Establishing a pre-closure coliform baseline in shellstock for each species under consideration in the conditional area management plan.

(iii) If re-opening is to be based on coliform levels in the water, identify and describe an association between coliform levels in shellstock for each species under consideration in the conditional area management plan and coliform levels in growing area water.

(iv) Defining conditions under the conditional area management plan which considers various factors including water temperature, salinity, seasonality,

	and other environmental conditions that may affect the pumping activity of each species of shellstock under consideration. (i)(v) A study design and data analysis approach providing statistical reliability. At a minimum, this should include consideration of: • variability of measurements of indicator levels in replicate samples • the likelihood or probability that a significant difference in indicator levels will be identified based on the sample outcomes if a substantial difference exists between the populations being sampled. Irrespective of the type of study design, these considerations apply and should be used to ensure that the number of samples collected is adequate. The number of samples needed increases with increasing variability of the measurements. When there is a substantial difference between indicator levels in the populations being sampled, the study should have at least an 80% probability of identifying this as such. (ii)(vi) Determining the time interval for post-closure coliform levels in shellstock and water to return to the pre-closure established baseline. (d) When utilizing MSC in shellstock in growing areas subjected to suspected human sewage to reopen a closed growing area, studies (utilizing the same format as (c) above) establishing sufficient elapsed time shall document the interval necessary for reduction of viral levels in the shellstock. The utilization of NSSP-conforming laboratories and NSSP-approved methods to analyze MSC in shellstock. Analytical shellstock sample results shall not exceed a level of 50 MSC per 100 grams or pre-determined levels established by the Authority based on studies conducted on regional species under regional conditions. These studies may establish criteria for reopening based on viral levels in the shellfish meats or the area must be in the closed status until the event is over and twenty-one (21) days have passed; (d)(e) Where necessary, the bacteriological quality of the water must be verified; and (e)(f) Shellstock feeding activity is sufficient to achieve reduction of pathogens to levels present prior to the pollution event. (8) A commitment to a reevaluation of the management plan at least annually using, at a minimum, the reevaluation requirements in the NSSP Model Ordinance.
Public Health	This language will provide state shellfish Authorities with guidance regarding

Significance	establishing the elapsed time to reopen closed conditional management areas and assure that shellstock are not adulterated. The public health significance of the proposed guidance for statistical reliability of studies used to establish an elapsed time to reopen is evident by considering an example of the effect of application of these criteria. While several different types of study designs are suitable to identify a minimum elapsed time for pathogen reduction, a common approach is to compare mean log concentrations of fecal indicators in a group of samples collected pre-closure, and representative of baseline, to that in a group of samples collected at the candidate elapsed time post-closure. For this type of study, a two-sample one-sided t-test is typically applied to test the null hypothesis that mean log concentrations are equal. If the test statistic is statistically significant (i.e., $p<0.05$), the null hypothesis is rejected; otherwise, mean concentrations are considered equivalent and the candidate elapsed time sufficient for pathogen reduction. To satisfy the proposed criteria of statistical reliability the sample size of the study will need to be large enough to achieve, based on expected variability of sample measurements about mean levels, an 80% probability of rejecting the null hypothesis when a minimally consequential difference in means exists. This determination of the sample size is made based on what is called the power function of the test statistic. Explicit formula and/or software to calculate sample sizes based on power functions are widely available for most commonly used hypothesis tests and test statistics. Using such calculations, it can be determined that, when the expected standard deviation of log sample measurements about mean levels is 0.5 logs, the example study design requires 13 samples per group to achieve 80% power (probability) to reject the null hypothesis when a true difference in means of 0.5 logs exists. Consequently, when a difference in means of 0.5 logs is considered consequential, a study of this type with fewer than 13 samples per group would not be considered sufficiently reliable. With an expected standard deviation of 0.5 logs, a sample size of 3 per group would have only a 27% probability of rejecting the null hypothesis when a consequential difference in means of 0.5 logs exists and an 80% probability of rejecting the null hypothesis would be achieved only when the true difference in means is equal to or greater than 1.25 logs.
Cost Information	No additional cost. This is simply providing guidance for a requirement already in place.
Action by 2019 Task Force I	Recommended referral of Proposal 19-145 to an appropriate committee as determined by the Conference Chairperson with the following instructions to develop guidance for cleansing studies and to assess scenarios where water quality sampling could be used in place of cleansing studies.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-145.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-145.
Action by 2023 Cleansing Study Committee	Recommended: 1) The committee Recommended adoption of the following Guidance. **Guidance on Studies Used in the Reopening of an Area Temporarily Placed in the Closed Status Due to an Emergency Condition, a Discharge of Raw Sewage, or when Conditional Area Management Plan (CAMP) Performance Standards are not Met** *Note: Similar contaminant reduction studies associated with shellstock relaying and*

validation studies associated with the depuration process are not covered in this guidance document. Instead, each has their own specific requirements which are covered in Chapter V. and Chapter XV., as well as Guidance Documents Chapter II .10 and .19, respectively.

A. When Are Studies Required?

Per Chapter IV. @.03 A.(5)(d) and C.(2)(c), studies are required for reopening a closed area to establish the environmental conditions and time required for pathogens (as measured by microbiological indicators) in shellstock and water to return to acceptable levels following the impact from an emergency condition, discharge of raw sewage, or when conditional area management plan (CAMP) performance standards are not met. Listed below is a summary of scenarios for reopening options:

1) Scenarios where studies are required to reopen once the emergency situation or condition has returned to normal, or CAMP performance standards are fully met, and sufficient time has elapsed to allow the shellstock to reduce pathogens and for the growing area water quality to return to acceptable levels:

(a) Chapter IV. @.03A.(5)(d):

- Reopening due to closures resulting from an emergency condition or situation when pathogens are of concern (other than raw untreated sewage discharged from a sewage collection system or WWSD), studies establishing sufficient elapsed time shall document the interval necessary for reduction of coliform levels in the shellstock to pre-closure levels. Such coliform studies may establish criteria for reopening based on coliform levels in the water.

- Reopening due to emergency closures caused by the occurrence of raw untreated sewage discharged from a sewage collection system or WWSD, when the closure duration is less than 21 days or when analytical shellstock samples are utilized for comparison to the levels established in the Chapter IV. @.02 E. (4). The authority may use studies to establish pre-determined male-specific coliphage (MSC) levels in shellfish samples that are conducted no sooner than seven (7) days after contamination has ceased and from representative locations in each growing area potentially impacted.

(b) Chapter IV. @.03 C.(2)(c)(iii):
For management plans based on WWSD function or pollution sources other than WWSD criteria that reliably predict when an area that was placed in the closed status because of failure to comply with its conditional management plan can be returned to the open status.

- Reopening due to closures impacted by pathogens (other than raw untreated sewage discharged from a sewage collection system or WWSD) from a failure to comply with its conditional management plan, studies establishing sufficient elapsed time shall document the interval necessary for reduction of coliform levels in the shellstock to pre-closure levels. These studies may establish criteria for reopening based on coliform levels in the water.

- Reopening due to temporary closures impacted by sewage from a failure to comply with the conditional management plan based on the WWSD performance standards, studies may be conducted to establish sufficient

	elapsed time and shall document the interval necessary for reduction of viral levels in the shellstock. These studies may establish pre-determined levels based on regional species under regional conditions. These studies may establish criteria for reopening based on viral levels in shellfish meats. *2) Scenarios where sampling is required to reopen when a study is not conducted, include:* (a) Chapter IV. @.03A.(5)(d): • Reopening due to emergency closures of harvest areas caused by the occurrence of raw untreated sewage discharged from a sewage collection system or WWSD, when the closure duration is intended to be less than 21 days, the analytical sample results shall not exceed the levels established in Chapter IV. @.02 E. (4). • Reopening due to emergency closures of harvest areas when poisonous or deleterious substances are the concern, sampling shall establish that poisonous or deleterious substances in shellstock do not exceed FDA action levels, tolerances, guidance levels, and levels that are deemed unsafe through risk evaluation. (b) Chapter IV. @.03C.(2)(c)(iii): • Reopening due to temporary closures impacted by sewage from a failure to comply with the conditional management plan based on the WWSD performance standards, analytical sample results shall not exceed the MSC level established in Chapter IV. @.02 E. (4). • Water quality sampling can be used to reopen an area following temporary closures resulting from a failure to comply with conditional management plan performance standards based on the effects of non-point sources of pollution such as rain events and/or stormwater runoff. *3) Scenarios where no studies or sampling are required to reopen, include:* (a) Chapter IV. @.03A.(5)(d)(ii) and C.(2)(c)(iii): • Reopening due to the temporary closure from a discharge of raw untreated sewage or exceedance of management plan performance standards relating to WWTP function. If no studies or analytical samples are collected and compared to the levels established in Chapter IV. @.02 E. (4), the area must be in the closed status until the event is over and twenty-one (21) days have passed. 2) proposal be referred back to an appropriate committee as determined by the conference chair to allow for further development of additional sections of the Guidance Document. 3) expanding the charge of the committee to include reviewing Model Ordinance language relating to cleansing studies for reopening.
Action by 2023 Task Force I	Recommends adoption of the Cleansing Study Committee recommendation for Proposal 19-145.

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Proposal Subject	Neogen's 'Reveal 2.0 for PSP' for detection of PSP
Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas, .11 Approved NSSP Laboratory Tests
Text of Proposal/ Requested Action	The intention is for this method to be an Approved Limited Use Method for Biotoxin testing for PSP toxins under the NSSP (for mussels and oysters) and that it should appear in Section IV (Guidance Documents), Table 4 (Approved Limited Use Methods for Biotoxin Testing). Full SLV validation data is provided for mussels and oysters.
Public Health Significance	PSP is a serious intoxication which still occurs in the USA and elsewhere. The USFDA and the European Union (EU) have established action levels for PSP toxins at 800 ppb (800 µg/kg) STX equivalents in shellfish. PCOX, has been accepted as a quantitative reference method in the USA and some other countries, although Pre-COX is also accepted by regulatory agencies in other areas of the world such as the UK, various EU countries, AU and NZ. Shellfish need to be more easily screened for toxins that cause paralytic shellfish poisoning (PSP), and they need to be screened closer to growing/harvesting areas to better protect public health. A reliable and simple screening tool for end product testing (EPT) by industry, for community-based and remote surveillance, and for screening out negative samples from the regulatory sample stream. Implementation of these approaches would broaden the food safety net and reduce outbreaks of PSP intoxication. Neogen is the only antibody-based test to detect both the STX and NEO parts of the PSP family of toxins at similar levels. No other antibody-based rapid test for PSP can detect NEO to any significant degree. Other ISSC approved "rapid" methods for PSP screening are largely limited to laboratory settings because of complexity which limits their use in EPT and community-based and remote surveillance of shellfish resources. The only ISSC-approved LFA rapid method, the Scotia LFI, has had many issues with reliability that have limited its applicability in screening for PSP, and concerns about the stability of the method have also been published [1,2,3,4,5]. The Neogen Reveal 2.0 for PSP is an excellent candidate for rapid screening of shellfish for PSP toxins in both laboratory and field situations, and is an extension of a platform used by Neogen for many reliable rapid tests in the meat, dairy and food sectors, many of which are approved for use by FDA, USFDA and/or EPA. The test has undergone SLV and ILV evaluations [5,6]and has been shown to be an accurate and reliable candidate for approval for use in the NSSP. [1] Cefas 2006

	[2] Turner et al. 2015 [3] Harrison et al. 2016 [4] Dorantes-Aranda et al. 2017a [5] Jawaid et al. 2015 [6] Dorantes-Aranda et al. 2017b
Cost Information	Approximately $20 per test. Reader based assay – approximate cost of reader is $2,700.00 USD.
Action by 2019 Laboratory	Recommended referral of Proposal 19-150 to an appropriate committee as determined by the Conference Chair.
Action by 2019 Task Force	Recommended adoption of Laboratory Committee recommendation on Proposal 19-150.
Action by 2019 General Assembly	Adopted recommendation of Task Force I on Proposal 19-150.
Action by FDA February 21, 2020	Concurred with Conference action on Proposal 19-150.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 19-150 to an appropriate committee as determined by the Conference Chairperson
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation for Proposal 19-150.

Proposal No. **23-100**

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Proposal Subject	Mooring Area Definition Change
Specific NSSP Guide Reference	Section I Purposes & Definitions, B. 79.
Text of Proposal/ Requested Action	**(79) Mooring Area** means any water area that is used to provide temporary or permanent anchorage for more than twenty (20) boats with marine sanitation devices. Mooring areas do not include any structures for docking boats.
Public Health Significance	The proposed Mooring Area definition change adds clarification that only vessels which have marine sanitation devices onboard are to be included in the count of boats in a mooring area. Inclusion of only vessels with marine sanitation devices is consistent with the risk evaluation of illicit discharge of human waste in shellfish growing area. It is logistically difficult for human waste to be discharged from a vessel that does not have a marine sanitation device onboard. The risk of fecal coliform contamination of a growing area from persons on vessels such as dinghies, daysailers, and small open boats that do not have marine sanitation devices onboard is no different than the risk presented by swimmers, shoreline walkers or any other person in or adjacent to the growing area. Shellfish Sanitation Control Authorities have engaged in numerous regulatory and educational programs to prevent illicit discharge of human waste into shellfish growing areas from vessels. Inclusion of the proposed clarifying language does not weaken those efforts.
Cost Information	No cost would be associated with this proposal. Clarifying the definition of a mooring area may also ease Authorites' administrative, patrol and fieldwork burdens with no impact on risk.
Action by 2023 Task Force I	Recommends adoption of Proposal 23-100 as submitted.

Proposal No. **23-101**

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Proposal Subject	Definition of scallops
Specific NSSP Guide Reference	Section I. Definitions B. Definition of Terms. Section III. Intorduction
Text of Proposal/ Requested Action	Section I. Definitions B. Definition of Terms. (115) Shellfish means all species of: (a) Oysters, clams or mussels, whether: (i) Shucked or in the shell; (ii) Raw, including post-harvest processed; (iii) Frozen or unfrozen; (iv) Whole or in part; and (b)Scallops in any form, except when the final product form is the adductor muscle only, attached or unattached to the shell. Section III. Introduction ….The purpose of the NSSP is to promote and improve the sanitation of shellfish (oysters, clams, mussels and scallops in any form, except when the final product form is the adductor muscle only, attached or unattached to the shell) moving in interstate commerce through Federal/State cooperation and uniformity of State shellfish programs.
Public Health Significance	The current definition of scallops excludes the adductor muscle only. However, there is a value added market for scallop adductor muscles that remain attached to the ventral shell. This proposal seeks to allow scallop adductor muscles to be exempt from the NSSP attached or unattached from the ventral shell.
Cost Information	There is no cost associated with this change.
Action by 2023 Task Force I	Recommends adoption of Proposal 23-101 as submitted.

Proposal No. **23-102**

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Proposal Subject	Seed sourced from Prohibited areas
Specific NSSP Guide Reference	Section I Purposes & Definitions Definitions B. Definition of Terms. Section II Model Ordinance, Chapter IV. Shellstock Growing Areas E. Prohibited Classification. Section IV Guidance Documents, Chapter II. Growing Areas Growing Area Classifications Section IV Guidance Documents, Chapter II. Growing Areas .19 Classification of Shellfish Growing Waters Adjacent to Waste Water Treatment Plants I. Introduction IV. Prohibited Classification A. Definition C. Allowable Uses of Shellfish from a Prohibited Growing Area D. Model Ordinance Requirements for Depletion and Gathering of Seed H. Public Health Significance
Text of Proposal/ Requested Action	Section I Purposes & Definitions Definitions B. Definition of Terms. (96) **Prohibited** means a classification used to identify a growing area where the harvest of shellstock for any purpose, except depletion, gathering of seed or nursery culture for aquaculture or resource enhancement, is not permitted. (113) **Seed** means shellstock which is less than market size and complies with the criteria in NSSP Model Ordinance Chapter VI. Shellfish Aquaculture @.02 Seed Shellstock where necessary. Section II Model Ordinance, Chapter IV. Shellstock Growing Areas E. Prohibited Classification. (1) Exception. The prohibited classification is not required for harvest waters within or adjacent to marinas. The Authority, however, may use the prohibited classification for these waters. (2) General. The Authority shall: (a) Not permit the harvest of shellstock from any area classified as prohibited, except for the gathering of seed or nursery culture for aquaculture or resource enhancement or the depletion of the areas classified as prohibited; and (b) Ensure that shellstock removed from any growing area classified as prohibited is effectively excluded from human consumption unless it ~~is~~

~~seed to be cultured as outlined in the~~ complies with the criteria in NSSP Model Ordinance Chapter VI. Shellfish Aquaculture @.02 Seed Shellstock.

Section IV Guidance Documents, Chapter II. Growing Areas
Growing Area Classifications
A growing area is placed in the prohibited classification when the sanitary survey or marine biotoxin surveillance program indicates that fecal material, pathogenic microorganisms, poisonous or deleterious substances, marine biotoxin, or radionuclides may reach the harvest area in excessive concentrations. The NSSP Model Ordinance also requires that a growing area for which there is no sanitary survey be placed in the prohibited classification as a precautionary measure. Taking shellstock from a prohibited area for any human food purpose is not allowed except for the gathering of seed or nursery culture for aquaculture or resource enhancement or the depletion of the areas classified as prohibited.

Section IV Guidance Documents, Chapter II. Growing Areas
.19 Classification of Shellfish Growing Waters Adjacent to Waste Water Treatment Plants
I. Introduction
(1) Prohibited – A classification used to identify a growing area where the harvest of shellstock for any purpose, except depletion or gathering of seed or nursery culture for aquaculture or resource enhancement, is not permitted.

IV. Prohibited Classification
A. Definition
A classification used to identify a growing area where the harvest of shellstock for any purpose, except depletion or gathering of seed or nursery culture for aquaculture or resource enhancement, is not permitted.
C. Allowable Uses of Shellfish from a Prohibited Growing Area
(1) Depletion
Depletion means the removal, under the direct control of the Authority, of shellstock from a growing area classified as prohibited.
(2) Seed
Seed means shellstock which is less than market size and complies with the criteria in NSSP Model Ordinance Chapter VI. Shellfish Aquaculture @.02 Seed Shellstock where necessary.
D. Model Ordinance Requirements for Depletion and Gathering of Seed
(1) Chapter IV. Shellstock Growing Areas
@.03 Growing Area Classification
E. Prohibited Classification
(1) Exception…
(2) General. The Authority shall:
(a) Not permit the harvest of shellstock from any area classified as prohibited, except for the harvest of shellstock for the gathering of seed or nursery culture for aquaculture or resource enhancement or the depletion of the areas classified as prohibited; and
H. Public Health Significance
The positive relationship between disease and consuming contaminated shellfish has been clearly established. Prevention of consumption of contaminated shellfish is the primary objective of the NSSP. The prohibited area classification is the most restrictive growing area classification and is used for areas subject to gross pollution. The use of

	this classification is also required for all growing areas immediately adjacent to a wastewater treatment plant and where the shellfish authority has not performed a sanitary survey. The harvesting of shellstock is not allowed for any human food use except for the gathering of seed or nursery culture for aquaculture or resource enhancement. For additional information concerning the classification of growing waters and the sanitary survey, see the NSSP Model Ordinance. Depletion and Gathering of Seed (Chapter IV @.03 E. Prohibited Classification (2) (a) & (b) and Chapter VI .03 Seed Shellstock A. & B.)
Public Health Significance	The NSSP MO prohibits any harvest from areas classified as Prohibited except for depletion and gathering of seed or nursery culture for aquaculture. The allowance for seed harvest from Prohibited areas for aquaculture purposes is coupled with a requirement for the Authority to define maximum seed sizes (Chapter VI. Shellfish Aquaculture @.02) that enable a minimum of 120 days of grow out before harvest and Control of Harvest requirements (Chapter VIII. Control of Shellfish Harvesting @.01). These requirements ensure safe harvest of seed coming from areas classified as Prohibited and should be extended to natural resource enhancement efforts. There are occasionally plentiful wild seed resources in Prohobited areas that can be safely transplanted to Approved areas for grow out and later harvest. Because of the existing maximum seed size regulation there is no risk of seed being harvested before 120 days. Allowing for the inclusion of harvest of seed from Prohibited areas for wild resource enhancement would not only increase resource utilization, but it would also deter illegal harvest by removing resources before they are market size.
Cost Information	There is no cost associated with this change.
Action by 2023 Task Force I	Recommends adoption of Proposal 23-102 as submitted.

Proposal No. **23-103**

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Proposal Subject	Illness Outbreak – Growing Area Closure
Specific NSSP Guide Reference	Section II Model Ordinance, Ch. II. Risk Assessment and Risk Management @.01 .01 Outbreaks of Shellfish-Related Illness G(2)
Text of Proposal/ Requested Action	G. When the growing area is determined the problem, the Authority shall: (1) Place the growing area in the closed status until: (a) The Authority verifies that the area is properly classified by conducting a review of the growing area to include: i. current data, in compliance with the NSSP Model Ordinance; ii. A field review of existing pollution sources; iii. A review of actual and potential intermittent pollution sources, such as vessel waste discharge and wastewater discharge from treatment plant collection systems. If a previously unknown pollution source can be corrected, the closure period shall be extended to allow for natural depuration following correction of the pollution source; and iv. Examination of water quality subsequent to the illness outbreak. (b) It has been determined that the event which caused the contamination no longer exists and sufficient time has elapsed for natural depuration; (2) Keep the area closed until at least ~~for a minimum of~~ 21 days have passed from ~~after~~ the last date of harvest of~~on~~ the implicated shellstock if the illness is consistent with viral etiology; and (3) Develop a written report summarizing the findings of the investigation and actions taken.
Public Health Significance	This proposal alters the language relating to when the 21 day timeline starts for closures due to viral etiology. The new language means that if a growing area is closed due to a viral illness outbreak, the 21 day viral cleansing timeline starts on the last day of harvest of implicated shellstock and the area must remain closed until 21 days following the last harvest date. This is different from the previous language where the area remained closed for 21 days from the first day a viral outbreak was identified. The existing requirement has resulted in growing area closures months after the shellstock was harvested and the risk is no longer present, as viral outbreaks are often identified many months after consumption. There is usually a delay in illness reporting. Requiring a full 21 day closure later than the implicated harvest dates, sometimes weeks or even months later, does not offer additional protections to the consuming public specific to the related outbreak. Section G (1) addresses the need for a closure for investigation related to the outbreak and G (1)(b) addresses the source of contamination and time for natural depuration prior to reopening the growing area. If the source of contamination continues, the Authority has the ability to keep the area closed until the criteria of G(1)(b) is met.
Cost Information	N/A
Action by 2023 Task Force I	Recommends adoption of Proposal 23-103 as submitted.

Proposal No. **23-104**

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Proposal Subject	Vibrio illness reporting- time frame for action to close shellfish growing areas
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter II. Risk Assessment and Risk Management @02A@.02 Shellfish Related Illnesses Associated with Vibrio parahaemolyticus (V.p.)
Text of Proposal/ Requested Action	A. When the investigation outlined in Section @.01 A. (6) indicates the illness(es) are associated with the naturally occurring pathogen Vibrio parahaemolyticus (V.p.), the Authority shall determine the number of laboratory confirmed cases epidemiologically associated with the implicated area. States will not be expected to close growing areas based on V.p. cases that are reported more than ~~sixty~~ thirty ~~(60)~~ (30) days when environmental parameters have changed, or monitoring indicates the V.p. risk is reduced. Actions taken by the Authority will be based on the number of cases and the span of time as follows.
Public Health Significance	According to the *Control of Communicable Diseases Manual 20th Edition*, the incubation period for Cholera and other vibrioses is a few hours to 5 days, usually 2-3 days. Section IV Guidance documents – Chapter II. Growing areas specifically states," The generally accepted minimum time period for elimination of microbial contaminants from shellstock is fourteen (14) days when environmental conditions are suitable for natural cleansing." Most states have requirements that communicable disease be reported to the state epidemiologist or health departments within set time frames- some as short as 24 hours. Closing a growing area beyond 30 days from the harvest date, due to inadequate reporting time frames, does not protect public health because after 30 days the molluscan shellfish will have had time to purge. In Section II Model Ordinance -Chapter II Risk Assessment and Risk Management @01 I(1) Molluscan shellfish that has been recalled because of an illness or outbreak is allowed to be reconditioned through placement into shellfish growing areas in the open status for a time frame not less than 14 days.
Cost Information	None
Action by 2023 Task Force I	Recommends adoption of Proposal 23-104 as submitted.

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10. Proposal Subject	Request to rescind the *Vibrio vulnificus* enzyme immunoassay (EIA) method
11. Specific NSSP Guide Reference	Section IV. Chapter II.14
12. Text of Proposal/ Requested Action	**Approved Methods for Vibrio Enumeration**

	Vibrio Type:	Application: PHP Sample Type: Shucked	Application: Reopening
~~**EIA**~~[1]	~~***Vibrio vulnificus (V.v.)***~~	~~X~~	
MPN[2]	***Vibrio vulnificus (V.v.)***	X	
SYBR Green 1 QPCR-MPN[5]	***Vibrio vulnificus (V.v.)***	X	
MPN[3]	***Vibrio parahaemolyticus (V.p.)***	X	
PCR[4]	***Vibrio parahaemolyticus (V.p.)***	X	
MPN-Real Time PCR[6]	***tdh+ and trh+ Vibrio parahaemolyticus (V.p.)***	X	X
MPN-Real Time PCR[7]	***Vibrio parahaemolyticus (V.p.)***	X	X
Direct Plating Method[8]	***Vibrio parahaemolyticus (V.p.)***		X
MPN-Real Time PCR[9]	***Vibrio vulnificus (V.v.)***	X	

Foot

	~~1 EIA procedure of Tamplin, et al, as described in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, 1992.~~ [2] MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, followed by confirmation using biochemical analyses or by the DNA -alkaline phosphatase e for vvhA as described by Wright et al., or a method that a State can demonstrate is equivalent.
13. Public Health Significance	The method for detection of Vibrio vulnificus (Vv) by the enzyme immunoassay (EIA) method should no longer be included in the NSSP. There are no laboratories using this method in support of the Program. The antibody required for the test method is not produced and has not been for many years, indicating it is unlikely to be produced again in the future. There are multiple alternative methods in the Program for the detection and confirmation of Vv isolates. Additionally, the ISSC Constitution, Bylaws, and Procedures states in Procedure XV, 8. that a method is subject to recantation when reagents are no longer available. As such, there should be no impact to the Program and the protection of public health and the table indicating approved methods for vibrio enumeration, validated and approved under the NSSP, will reflect the available choices of analyses.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-105 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-105.

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10. Proposal Subject	Request to rescind the *Vibrio vulnificus* SYBR Green real-time PCR method
11. Specific NSSP Guide Reference	Section IV. Chapter II.14 Approved Methods for Vibrio Enumeration
12. Text of Proposal/ Requested Action	[Section IV. Chapter II.14]

Approved Methods for Vibrio Enumeration

	Vibrio Type:	**Application: PHP Sample Type: Shucked**	**Application: Reopening**
EIA[1]	***Vibrio vulnificus (V.v.)***	X	
MPN[2]	***Vibrio vulnificus (V.v.)***	X	
~~**SYBR Green 1 QPCR MPN[5]**~~	~~***Vibrio vulnificus (V.v.)***~~	~~X~~	
MPN[3]	***Vibrio parahaemolyticus (V.p.)***	X	
PCR[4]	***Vibrio parahaemolyticus (V.p.)***	X	
MPN-Real Time PCR[6]	***tdh+ and trh+ Vibrio parahaemolyticus (V.p.)***	X	X
MPN-Real Time PCR[7]	***Vibrio parahaemolyticus (V.p.)***	X	X
Direct Plating Method[8]	***Vibrio parahaemolyticus (V.p.)***		X
MPN-Real Time PCR[9]	***Vibrio vulnificus (V.v.)***	X	

	[4]MPN method in Chapter 9 of the FDA Bacteriological Analytical Manual, 7th Edition, May 2004 revision, and as described in the "Direct Plating Procedure for the Enumeration of Total and Pathogenic Vibrio parahaemolyticus in Oyster Meats" developed by FDA, Gulf Coast Seafood Laboratory, or a method that a State can demonstrate is equivalent. [5]~~Vibrio vulnificus, ISSC Summary of Actions 2009. Proposal 09-113, Page 123~~. [65]MPN-Real Time PCR Method for the tdh and trh Genes for Total V. parahaemolyticus as described in Kinsey et al., 2015. ISSC 2015 Summary of Actions Proposal 15-111, Page 397. [Modifications to the Microbiology PCR Checklist] 3.2.3 The PCR forward and reverse primers used target. For Total and Pathogenic Vp Real-time PCR Method tdh_269-20: 6FAM-5'-TGACATCCTACATGACTGTG-3'-MGBNFQ trh_133-23: TET-5'-AGAAATACAACAATCAAAACTGA-3'-MGBNFQ tlh_1043: TEXAS RED-5'- CGCTCGCGTTCACGAAACCGT -3'-BHQ2 IAC_109: CY5-5'- TCTCATGCGTCTCCCTGGTGAATGTG -3'- BHQ2 trh_20F: 5'-TTGCTTTCAGTTTGCTATTGGCT-3' trh_292R: 5'-TGTTTACCGTCATATAGGCGCTT-3' tdh_89F: 5'-TCCCTTTTCCTGCCCCC-3' tdh_321R: 5'-CGCTGCCATTGTATAGTCTTTATC-3' tlh_884F: 5'-ACTCAACACAAGAAGAGATCGACAA-3' tlh_1091R: 5'-GATGAGCGGTTGATGTCCAAA-3' IAC_46F: 5'-GACATCGATATGGGTGCCG-3' IAC_186R: 5'-CGAGACGATGCAGCCATTC-3' ~~For Vv Real-time PCR Method~~ ~~vvhF 5'-TGTTTATGGTGAGAACGGTGACA-3'~~ ~~vvhR 5'-TTCTTTATCTAGGCCCCAAACTTG-3'~~
13. Public Health Significance	The specific instrumentation (Cepheid SmartCycler) required for the Vv Real-time PCR Method using SYBR Green for detection of Vibrio vulnificus (Vv) should no longer be included in the NSSP. There are no laboratories using this method in support of the Program. The instrumentation required for the test method is not produced and is no longer supported by the manufacturer, indicating a lack of ability to perform required maintenance and calibration to ensure integrity of results. There are multiple alternative methods in the Program for the detection and confirmation of Vv, including a Real-Time PCR Method. Additionally, the ISSC Constitution, Bylaws, and Procedures states in Procedure XV, 8. that a method is subject to recantation when equipment is no longer available. As such, there should be no impact to the Program and the protection of public health and the table indicating Approved Methods for Vibrio Enumeration will reflect the available choices of analyses.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-106 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-106.

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10. Proposal Subject	Data evaluation when the nonpoint sources impacting a growing area are not from a human sewage source.
11. Specific NSSP Guide Reference	Section II. Model Ordinance; Chapter IV Growing Areas; Section @.02 Microbiological Standards F.1.
12. Text of Proposal/ Requested Action	F. Standard for the Approved Classification of Growing Areas when Evaluated for Nonpoint Sources. (1) Exception. (a) If the tidal stage increases the fecal coliform concentration, the authority shall use sample results collected during that tidal stage to classify the area. (b) If the Authority has documentation supporting that the nonpoint sources impacting the growing area are not from a human sewage origin they may exclude up to two outlier datapoints from the dataset being evaluated. (2) Pollution Sources. Growing areas shall be impacted only by randomly occurring, intermittent events. (3) Water Quality. The bacteriological quality of every station in the growing area shall meet the fecal coliform standard in Section E. (2) or Section F. (4). (4) Fecal Coliform Standard for Systematic Random Sampling. The fecal coliform median (or geometric mean MPN or MF (mTEC) of the water sample results shall not exceed fourteen (14) per 100 ml and the estimated 90th percentile shall not exceed an MPN or MF (mTEC) of: (a) 43 MPN per 100 ml for a five-tube decimal dilution test; (b) 49 MPN per 100 ml for a three-tube decimal dilution test; or (c) 31 CFU per 100 ml for a MF (mTEC) test. (5) Estimated 90th Percentile. The estimated 90th percentile shall be calculated by: (a) Calculating the arithmetic mean and standard deviation of the sample result logarithms (base 10); (b) Multiplying the standard deviation in (a) by 1.28; (c) Adding the product from (b) to the arithmetic mean; (d) Taking the antilog (base 10) of the results in (c) to get the estimated 90th percentile; and (e) The MPN values that signify the upper or lower range of sensitivity of the MPN tests in the 90th percentile calculation shall be increased or decreased by one significant number. (6) Required Sample Collection.

	(a) Adverse Pollution Condition Standard. The Authority shall collect samples in the same intensity and frequency as described in Section E. (3) for application of the standard under Section E. (2). (b) Systematic Random Sampling Standard. The requirement for systematic random sample collection shall be met when: (i) Sample station locations are adequate to produce the data to effectively evaluate all nonpoint sources of pollution; (ii) Sample collection is scheduled sufficiently far in advance to support random collection with respect to environmental conditions. Compliance requires that, prior to implementation, the schedule for random sampling shall be documented in the master file for the growing area, and if conditions at the time of scheduled sample collection are believed to be hazardous to the safety of the individuals assigned to collect samples, sample collection shall be rescheduled at a later date as soon as practical; (iii) A minimum of six (6) random samples shall be collected annually from each sample station in the growing area; (iv) A minimum of two (2) random samples shall be collected annually from each sample station in the growing area while in the inactive status. The sample collection frequency of six (6) random samples per station per year specified under @.02 F. (6)(b) (iii) must resume at least six (6) months before an area is reactivated; and (v) A minimum of the thirty (30) most recent randomly collected samples from each sample station shall be used to calculate the median or geometric mean and 90th percentile to determine compliance with this standard. (c) Transition from Adverse Pollution Condition Standard to Systematic Random Sampling Standard. If the Authority: (i) Does not have thirty (30) recent randomly collected sample results from each station, then the previous fifteen (15) samples collected under adverse pollution conditions may be used with the most recent random samples to meet the minimum thirty (30) sample requirement for a transition period not to exceed three (3) years; and (ii) Uses the transition period described in (i), as additional random samples are collected; the random samples shall replace chronologically the samples collected under adverse pollution conditions (e.g. sample 31 replaces sample 1).
13. Public Health Significance	It is recognized that on occasion water quality may be impacted by non-human sources such as birds. Scientific literature also indicates that the presence of human enteric pathogens in wild birds is overestimated with the use of the coliform indicator (Smith et al. 2021) https://doi.org/10.1111%2Fbrv.12581 If a few aberrant samples can be reliably attributed to birds it is likely that the closure of the harvest area is an unwarranted response.
14. Cost Information	
15. Research Needs Information (Optional)	
a. Proposed specific	At this time we do not have an estimate of the correlation of human enteric pathogens with coliforms in wild bird waste. Our growing area classification has been entirely built on the correlation between pathogens and coliforms in

research need/ problem to be addressed	wastewater. Using the coliform standard to close harvest areas impacted by birds assumes the relationship is similar, when scientific literature indicates that the risk is being overestimated. Research is needed to describe the persistence of bird-sourced pathogens in the marine environment, and how long these pathogens persist in the shellfish if they are taken up by filter feeding bivalves
b. Explain the relationship between proposed research need and program change recommended in the proposal	Research to elucidate the relationship between human enteric pathogens and coliforms will help define the risk of illness associated with consumption of shellfish that may have been impacted by birds. Studies evaluating how these pathogens survive in the marine environment will further inform this relationship. Studies evaluating the purge rates of these pathogens will help growers devise management approaches to ensure potentially impacted product is held away for contaminated sites and is safe for consumption.
c. Estimated cost	Unknown
Action by 2023 Task Force I	Recommends referral of Proposal 23-107 as amended to an appropriate committee as determined by the Conference Chair. F. Standard for the Approved Classification of Growing Areas when Evaluated for Nonpoint Sources. (7) Exception. (a) If the tidal stage increases the fecal coliform concentration, the authority shall use sample results collected during that tidal stage to classify the area. ~~(b) If the Authority has documentation supporting that the nonpoint sources impacting the growing area are not from a human sewage origin they may exclude up to two outlier datapoints from the dataset being evaluated.~~ (8) Pollution Sources. Growing areas shall be impacted only by randomly occurring, intermittent events. (9) Water Quality. The bacteriological quality of every station in the growing area shall meet the fecal coliform standard in Section E. (2) or Section F. (4). (10) Fecal Coliform Standard for Systematic Random Sampling. The fecal coliform median (or geometric mean MPN or MF (mTEC) of the water sample results shall not exceed fourteen (14) per 100 ml and the estimated 90th percentile shall not exceed an MPN or MF (mTEC) of: (a) 43 MPN per 100 ml for a five-tube decimal dilution test; (b) 49 MPN per 100 ml for a three-tube decimal dilution test; or (c) 31 CFU per 100 ml for a MF (mTEC) test. (11) Estimated 90th Percentile. The estimated 90th percentile shall be calculated by: (a) Calculating the arithmetic mean and standard deviation of the sample result logarithms (base 10); (b) Multiplying the standard deviation in (a) by 1.28; (c) Adding the product from (b) to the arithmetic mean; (d) Taking the antilog (base 10) of the results in (c) to get the estimated 90th percentile; and (e) The MPN values that signify the upper or lower range of sensitivity of the MPN tests in the 90th percentile calculation shall be increased or decreased by one significant number. Required Sample Collection. (a) Adverse Pollution Condition Standard. The Authority shall collect samples in the same intensity and frequency as described in Section E. (3) for application of the standard under Section E. (2). (d) Systematic Random Sampling Standard. The requirement for

systematic random sample collection shall be met when:

(i) Sample station locations are adequate to produce the data to effectively evaluate all nonpoint sources of pollution;

(ii) Sample collection is scheduled sufficiently far in advance to support random collection with respect to environmental conditions. Compliance requires that, prior to implementation, the schedule for random sampling shall be documented in the master file for the growing area, and if conditions at the time of scheduled sample collection are believed to be hazardous to the safety of the individuals assigned to collect samples, sample collection shall be rescheduled at a later date as soon as practical;

(iii) A minimum of six (6) random samples shall be collected annually from each sample station in the growing area;

(iv) A minimum of two (2) random samples shall be collected annually from each sample station in the growing area while in the inactive status. The sample collection frequency of six (6) random samples per station per year specified under @.02 F. (6)(b) (iii) must resume at least six (6) months before an area is reactivated; and

(v) A minimum of the thirty (30) most recent randomly collected samples from each sample station shall be used to calculate the median or geometric mean and 90th percentile to determine compliance with this standard.

(e) Transition from Adverse Pollution Condition Standard to Systematic Random Sampling Standard. If the Authority:

(i) Does not have thirty (30) recent randomly collected sample results from each station, then the previous fifteen (15) samples collected under adverse pollution conditions may be used with the most recent random samples to meet the minimum thirty (30) sample requirement for a transition period not to exceed three (3) years; and

(ii) Uses the transition period described in (i), as additional random samples are collected; the random samples shall replace chronologically the samples collected under adverse pollution conditions (e.g. sample 31 replaces sample 1).

Proposal No. **23-108**

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Proposal Subject	Clarification of standards for reopening following WWTP sewage spill.
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @. 03 A. (5) (d)(ii)
Text of Proposal/ Requested Action	(ii) For emergency closures of harvest areas caused by the occurrence of raw untreated sewage discharged from a large community sewage collection system or WWSD, the analytical sample results shall not exceed ~~the~~ MSC levels established in Chapter IV @.02 E (4) or pre-determined levels established by the Authority based on studies conducted on regional species under regional conditions from shellfish samples collected no sooner than seven (7) days after contamination has ceased and from representative locations in each growing area potentially impacted or until the event is over and twenty-one (21) days have passed;
Public Health Significance	Chapt. IV @. 03 A. (5) (d)(ii) describes the how MSC can be utilized for reopening a growing area prior to 21 days in the case of a raw, untreated sewage spill closure. It is understood that MSC testing is the only acceptable method for reopening from raw sewage spills earlier than the mandated 21 day closure period. Including a reference to bacteriological data in this context is confusing and misleading since E. (4) is the regulation addressing the MSC standard., and utilizing MSC is the focus of (d) (ii).
Cost Information	None
Action by 2023 Task Force I	Recommends adoption of Proposal 23-108 as submitted.

Proposal No. **23-109**

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Proposal Subject	Growing area reopening criteria
Specific NSSP Guide Reference	Chapter IV. @.03 A.(5)(d) Chapter IV. @.03 C.(2)(c)
Text of Proposal/ Requested Action	Chapter IV. @.03 A.(5)(d): (d) Reopened Status. A growing area temporarily placed in the closed status as provided in (b) above, shall be returned to the open status only when: (i) The emergency situation or condition has returned to normal and sufficient time has elapsed to allow the shellstock to reduce pathogens or poisonous or deleterious substances that may be present in the shellstock to acceptable levels. (ii) When pathogens are of concern, and the area is not impacted by human sewage, studies establishing sufficient elapsed time shall document the interval necessary for reduction of coliform levels in the shellstock to pre-closure levels. Such coliform studies may establish criteria for reopening based on coliform levels in the water. (iii) When poisonous or deleterious substances are the concern, sampling shall establish that poisonous or deleterious substances in shellstock do not exceed FDA action levels, tolerances and/or guidance levels and/or levels that are deemed safe through risk evaluation. (v) For emergency closures of harvest areas caused by the occurrence of raw untreated sewage or partially treated sewage discharged from a ~~large community~~ sewage collection system or WWSD: a. The male-specific coliphage (MSC) analytical sample results in shellfish shall not exceed the levels established in Chapter IV @.02 E.(4) or b. ~~p~~Pre-determined MSC levels in shellfish established by the Authority based on studies conducted on regional species under regional conditions from shellfish samples collected no sooner than seven (7) days after contamination has ceased and from representative locations in each growing area potentially impacted or c. ~~until~~ Until the event is over, and twenty-one (21) days have passed. (vi) The requirements for biotoxins or conditional area management plans as established in Section @.04 and Section @.03, respectively, are met. (ivi) Supporting information is documented by a written record in the central file. Chapter IV. @.03 C.(2)(c): (c) For management plans based on WWSD function or pollution sources other than WWSD criteria that reliably predict when an area that was placed in the closed status because of failure to comply with its conditional management plan can be returned to the open status. The minimum reopening criteria for conditional management plans are: (i) Performance standards of the plan are fully met;

	(ii) Sufficient time has elapsed to allow the water quality in the growing area to return to acceptable levels; (iii) Sufficient time has elapsed to allow the shellstock to reduce pathogens that might be present to acceptable levels. ~~Studies establishing sufficient elapsed time shall document the interval necessary for reduction of coliform levels in the shellstock to pre-closure levels.~~ (iv) Shellstock feeding activity is sufficient to achieve ~~microbial~~ pathogen reduction. (v) If (i-iv) are met and if the conditional management plan closure performance standard(s) is(are) based on the effects of non-point sources of pollution such as rain events and/or storm water runoff, an area may be reopened when the water quality meets classification criteria without a shellstock cleansing study;. (vi) For conditionally managed areas based on WWSD performance standards,the Authority may utilize MSC levels in shellstock to establish that sufficient time has elapsed to allow water quality and shellstock to return to acceptable levels in growing areas adjacent to WWSD: a. Analytical shellstock tissue sample results shall not exceed the MSC levels established in Chapter IV @.02 E.(4) or b. Pre-determined MSC shellstock tissue levels established by the Authority based on studies conducted on regional species under regional conditions. These studies may establish criteria for reopening based on viral levels in the shellfish meats; or c. The area shall be in the closed status until the event is over and twenty-one (21) days have passed.
Public Health Significance	The NSSP MO requires certain criteria are met in order to reopen a growing area closed due to an emergency closure or based on the performance standards of a conditional management plan. There has been some confusion regarding the present reopening criteria language. This proposed language is intended to clarify the requirements for reopening criteria.
Cost Information	Not applicable.
Action by 2023 Task Force I	Recommends adoption of Proposal 23-109 as submitted.

Proposal No. **23-110**

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Proposal Subject	Marina classification
Specific NSSP Guide Reference	Section II Model Ordinance, Ch. IV Shellstock Growing Areas @.05 Marinas
Text of Proposal/ Requested Action	A. Marina Proper. The area within any marina which is in or adjacent to a shellstock growing area shall be classified as conditionally approved, restricted, conditionally restricted or prohibited. (1) Prior to the Authority establishing a classification of conditionally approved, restricted, or conditionally restricted in the marina proper, a pollution assessment supporting the classification will be conducted by the authority. (2) The assignment of a prohibited classification within the marina proper does not require a pollution assessment by the Authority.
Public Health Significance	Proper classification of shellfish havesting areas is critical to preventing shellfish related foodborne illnesses. The restricted classification is a key component of the proper classification of harvesting areas, this proposal is adding the restricted classification to the section governing the marina proper. The restricted classification should be an option in a marina proper with a pollution assessment justification by the Authority. A conditional classification management plan would only be needed if there is fluctuation in marina operation necessitating periodic and predictable closures of the growing area.
Cost Information	N/A
Action by 2023 Task Force I	Recommends no action on Proposal 23-110. Rationale: Restricted is not an appropriate classification for use in or adjacent to Marinas.

Proposal No. **23-111**

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Proposal Subject	Relay timeframe
Specific NSSP Guide Reference	Section II Model Ordinance, Ch. V Shellshock Relaying @.02 Contaminant Reduction C(3)
Text of Proposal/ Requested Action	(1) The Authority may waive the requirements for a contaminant reduction study if: (1) Only microbial contaminants need to be reduced; and (2) The shellstock are relayed from a conditionally approved, restricted, or conditionally restricted area meeting the bacteriological water quality for restricted areas used for shellstock depuration per Chapter IV. @.02 G. and Chapter IV. @.02 H.; and (3) The treatment period exceeds ~~sixty~~fourteen (~~60~~14) days. D. The time period shall be at least fourteen (14) consecutive days when environmental conditions are suitable for shellfish feeding and cleansing unless shorter time periods are demonstrated to be adequate
Public Health Significance	The change to 14 days is consistent with the literature available and already cited in the NSSP. The Guidance documents already have established 14 days as the ideal acceptable time for elimination of microbial contaminants. 60 days is not in any literature nor in any other already voted on sections of the NSSP for relaying. 21 days is the agreed upon value for harvesting waters adulterated with raw sewage, which is likely the worst-case scenario, relay from areas only impacted by microbial contamination should surely be less than those contaminated by raw sewage.
Cost Information	N/A
Action by 2023 Task Force I	Recommends referral of Proposal 23-111 to an appropriate committee as determined by the Conference Chairperson.

Proposal No. **23-112**

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Proposal Subject	Disposal of Human Sewage and Vomitus
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter VIII. Control of Shellfish Harvesting Requirements for Harvesters .02 Shellstock Harvesting and Handling. Section II. Model Ordinance Chapter IX. Transportation Requirements for Harvesters .01 Conveyances Used to Transport Shellstock to the Original Dealer and .02 Conveyances Used to Transport Shellstock from Dealer to Dealer
Text of Proposal/ Requested Action	Section II. Model Ordinance Chapter VIII. Control of Shellfish Harvesting Requirements for Harvesters .02 Shellstock Harvesting and Handling. (2) Disposal of Human Sewage and ~~Bodily Fluids~~ Vomitus. (1) Human sewage and ~~bodily fluids~~ vomitus shall not be discharged overboard from any vehicle or vessel used in the harvesting of shellstock. (2) As required by the Authority, in consultation with FDA, an approved marine sanitation device (MSD), portable toilet or other sewage disposal receptacle shall be provided on the vessel or available for the vehicle operator's use for the purpose of containing human sewage and ~~bodily fluids~~ vomitus. Section II. Model Ordinance Chapter IX. Transportation Requirements for Harvesters .01 Conveyances Used to Transport Shellstock to the Original Dealer (3) Disposal of Human Sewage and ~~Bodily Fluids~~ Vomitus (4) Human sewage and ~~bodily fluids~~vomitus shall not be discharged overboard from any vehicle or vessel which buys shellstock while the vehicles or vessels are in growing areas. (5) As required by the Authority, in consultation with FDA, an approved MSD, portable toilet or other sewage disposal receptacle shall be provided on the vessel or available for the vehicle operator's use for the purpose of containing human sewage and ~~bodily fluids~~vomitus. Portable toilets shall meet the requirements of VIII. .02. D. (3). Section II. Model Ordinance Chapter IX. Transportation Requirements for Harvesters .02 Conveyances Used to Transport Shellstock from Dealer to Dealer C. Disposal of Human Sewage and ~~Bodily Fluids~~Vomitus (1) Human sewage and ~~bodily fluids~~vomitus shall not be discharged overboard from any vessel used in the harvesting of shellstock, or from vessels which buy shellstock while the vessels are in growing areas.

	(2) As required by the Authority, in consultation with FDA, an approved MSD, portable toilet or other sewage disposal receptacle shall be provided on the vessel to contain human sewage and ~~bodily fluids~~vomitus. Portable toilets shall meet the requirements of VIII. .02. D. (3).
Public Health Significance	It is recognized that human digestive waste or vomit can put a shellfish growing area at risk of foodborne illness, e.g. norovirus, hepatitis A, etc. The current language references "bodily fluids" which is too broad a term for the recognized risks which include human digestive waste and vomitus. "Bodily fluids" can be interpreted to include liquids such as tears and sweat. This proposal attempts to limit the requirement to the recognized dangers of human digestive waste and vomitus.
Cost Information	There is no cost associated with this change.
Action by 2023 Task Force I	Recommends adoption of Proposal 23-112 as submitted.

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10. Proposal Subject	Redesigned Section IV. Guidance Table of Contents
11. Specific NSSP Guide Reference	Section IV. Guidance
12. Text of Proposal/ Requested Action	**Section IV. Guidance Documents** Chapter I. ~~General~~ Shellfish Sanitation Program @.01 Administration .01 Evaluation Standards .02 Procedures for Initiating a New State Program Under the National Shellfish Sanitation Program ~~.02~~. 03 Shellfish Plant Inspection Standardization Procedures NSSP Standardized Shellfish Processing Plant Inspection Form .04 Voluntary National Shellfish Regulatory Program Standards ~~.18~~.05 Decision Tree - Shellfish from Non-MOU Countries @.02 Dealer Certification ~~.03~~.01 Dealer Certification and the Interstate Certified Shellfish Shippers List (ICSSL) @.03 Evaluation of State Shellfish Sanitation Program Elements Chapter II. ~~Growing Areas~~ Risk Assessment and Risk Management @.01 Outbreaks of Shellfish-Related Illness .01 Guidance for Investigating an Illness Outbreak and Conducting Recall ~~.03~~.02 Guidance for Harvest Area Closure and Recall Notification ~~.02~~.03 Guidance for a Time-Temperature Evaluation of a Shellfish Implicated Outbreak ~~.03~~.04 Determining the Size of Closed Area as a Result of Illnesses ~~.04~~.05 Determining the Harvesting Periods Associated with Implicated Product for Identifying Shellfish to be Included in the Recall ~~.05~~. 06 Determining the Scope of Implicated Product for Conducting a Recall

@.03 Annual Assessment of *Vibrio vulnificus* and *Vibrio parahaemolyticus* Illnesses and Shellfish Production
.~~07~~.01 Production Reporting Guidance
@.04 Presence of Human Pathogens in Shellfish Meats
.~~06~~.01 *Vibrio cholerae*
@.06 *Vibrio vulnificus* Control Plan
.~~03~~.01 Guidance for Demonstrating the Effectiveness of Time to Temperature Reduction Criteria for *Vibrio vulnificus* and *Vibrio parahaemolyticus* (see below)
@.07 *Vibrio parahaemolyticus* Control Plan
.~~06~~.01 *Vibrio parahaemolyticus* (*V.p.*) Control Plan Guidance
.~~03~~.02 Guidance for Demonstrating the Effectiveness of Time to Temperature Reduction Criteria for *Vibrio vulnificus* and *Vibrio parahaemolyticus*

Chapter III. ~~Harvesting, Handling, Processing, and Distribution~~Laboratory
@.01 Quality Assurance
.~~15~~.01 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
@.02 Methods
.~~14~~.01 Approved NSSP Laboratory Tests
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Chapter IV. ~~Naturally Occurring Pathogens~~ Growing Areas
@.01 Sanitary Survey
.~~07~~.01 Sanitary Survey and the Classification of Growing Waters
@.02 Microbiological Standards
.01 Total Coliform Standards
.~~11~~.02 Systematic Random Sampling Monitoring Strategy
@.03 Growing Area Classification
.~~09~~.01 Management Plans for Growing Areas in the Conditional Classification
.~~16~~.02 Protocol for Reviewing Classification of Areas Implicated by Pathogens in Shellfish Meat Samples
.~~19~~ .03 Classification of Shellfish Growing Waters Adjacent to Waste Water Treatment Plants
.~~08~~. 04 Action Levels, Tolerances and Guidance Levels for Poisonous or Deleterious Substances in Seafood
@.04 Marine Biotoxin Control
.~~02~~.01 Guidance for Developing Marine Biotoxin Plans
@.05 Marinas
.01 Guidance TBD
@.06 Mooring Areas
.01 Guidance TBD

	Chapter V. ~~Illness Outbreaks and Recall Guidance~~ Shellstock Relaying ~~.10~~.01 Shellstock Relay Chapter VI. Shellfish Aquaculture .01 Guidance TBD Chapter VII. Wet Storage in Approved and Conditionally Approved Growing Areas .05.01 Protocol for Addressing Positive Coliform Sample in an Artificial Wet Storage Water Body Chapter VIII. Control of Shellfish Harvesting @.01 Control of Shellstock Growing Areas ~~.12~~.01 Growing Area Patrol and Enforcement ~~.13~~.02 Control of Shellfish Harvesting @.02 Shellstock Time to Temperature Controls ~~.08~~.01 Icing, Cold Water Dips and Ice Slurries for Cooling Shellstock Shellstock Harvesting and Handling See *Shellstock Tagging* (Chp. X. below) Chapter IX. Transportation See *Time and Temperature Controls* (Chp. XI-XIV below) Chapters X. General Requirements for Dealers .01-.03 Shellstock Identification, Shucked Shellfish Labeling, Shipping Documents and Records .04 Shellstock Tagging Chapter XI., XII., XIII., and XIV. – Shellfish Processing and Handling .01 Shellfish Industry Equipment Construction Guide ~~.06~~.02 Guidance for Reinstating a Previously Infected Employee ~~.07~~.03 Time and Temperature Controls Chapter XV. Depuration ~~.17~~.01 Calculating the Ninetieth (90th) Percentile for End-Product Depurated Shellfish Chapter XVI. Processes and Procedures for Pathogen Reduction ~~.02~~.01 Post- Harvest Processing (PHP) Validation/Verification Guidance for *Vibrio vulnificus (V.v.)* and *Vibrio parahaemolyticus (V.p.)* ~~.04~~.02 Method for Validation and Verification of a Two (2) or Three (3) Log Reduction of *Vibrio parahaemolyticus (V.p.)* in Oysters ~~.05~~.03 Template for Submission of Post-Harvest Process Validation Studies

	.09.04 Irradiation Pre-labeling Guidance Chapter XVII. Federal Waters .06.01 Federal Waters Guidance (DRAFT)
13. Public Health Significance	The proposed organizational redesign of the NSSP Guide for the Control of Molluscan Shellfish, Section IV. Guidance and associated Table to Contents will allow the guide to be more in line with the MO and therefore, make it easier to reference. In addition, the FDA has conducted a review and suggested update of the growing area guidance section. The idea is to use this suggested updated Table of Contents to suggest the establishment of a growing area guidance review committee where FDA can provide what we have put together and then have the ISSC input.
14. Cost Information	N/A
Action by 2023 Task Force I	Recommends adoption of Proposal 23-113 as submitted.

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Proposal Subject	Domoic Acid (Amnesic Shellfish Poisoning) HPLC Method Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to edit the text of the attached checklist for the HPLC method for detecting domoic acid and to append the checklist to the list of NSSP Laboratory Evaluation Checklists at the end of .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists.
Public Health Significance	The attached checklist provides the quality assurance and method requirements that laboratory evaluation officers will use to evaluate laboratories implementing the HPLC method for domoic acid to support the NSSP. The checklist documents the number of critical, key or other nonconformities and how overall laboratory status for the method is determined.
Cost Information	None.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 23-114 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-114.

Proposal No. 23-115

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Proposal Subject	Paralytic Shellfish Poisoning (PSP HPLC-PCOX) HPLC Method Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to edit the text of the attached checklist for the HPLC method for detecting domoic acid and to append the checklist to the list of NSSP Laboratory Evaluation Checklists at the end of .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists.
Public Health Significance	The attached checklist provides the quality assurance and method requirements that laboratory evaluation officers will use to evaluate laboratories implementing the HPLC method for domoic acid to support the NSSP. The checklist documents the number of critical, key or other nonconformities and how overall laboratory status for the method is determined.
Cost Information	None.
Action by 2023 Laboratory Committee	Recommended referral of Proposal 23-115 to an appropriate committee as determined by the Conference Chairperson.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-115.

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10. Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist Sample Diluent
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
12. Text of Proposal/ Requested Action	The requested action is to remove NSSP checklist item 3.2.13 - Specific edits in accompanying document. The current NSSP Microbiology Checklist has two duplicate items in 1.7.14 and 3.2.13 *Sterile phosphate buffered dilution water is used as the sample diluent.* This could result in a laboratory erroneously receiving two (2) Other cited nonconformities during an evaluation. By removing checklist item 3.2.13 it will ensure a laboratory is properly cited once in Microbiology Checklist Part I if they are not using an appropriate sample diluent for any method included in the Microbiology Checklist.
13. Public Health Significance	The proposed modifications are to improve consistency in the current NSSP Microbiology evaluation standard.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-116 as amended.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-116.

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10. Proposal Subject	Modifications to NSSP Quality Systems Evaluation Checklist
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
12. Text of Proposal/ Requested Action	The requested action is to adopt modified text in accompanying document.
13. Public Health Significance	The proposed modifications are to improve the current NSSP quality systems evaluation standard and remove redundant language.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-117 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-117.

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10. Proposal Subject	Part I Modifications to NSSP Microbiology Laboratory Evaluation Checklist
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists; References – NSSP Laboratory Evaluation Checklists 1. NSSP Laboratory Evaluation Checklist for Microbiology (link)
12. Text of Proposal/ Requested Action	The requested action is to adopt modified text of eleven (11) NSSP microbiology checklist items and remove one item in Part I; said NSSP checklist items are 1.4.8, 1.4.21, 1.4.22, 1.4.23, 1.6.4, 1.6.5, 1.6.6, 1.6.7, 1.6.21, 1.6.22, 1.7.2, 1.7.9. Specific text is in accompanying document.
13. Public Health Significance	The proposed modifications are to improve consistency in the current NSSP microbiology evaluation standard and account for technology improvements to laboratory equipment.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-118 as amended.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-118.

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10. Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist Productivity Controls
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
12. Text of Proposal/ Requested Action	The requested action is to remove NSSP checklist items 2.2.2, 2.3.3, 2.5.4, 2.9.2, 2.12.8, 3.3.2, 3.4.2, 3.8.12 and modify checklist item 1.7.13 to include the intent of items removed. Specific edits are reflected in supporting documentation. The current NSSP Microbiology Checklist includes multiple items related to the culture media productivity testing requirement. This could result in several Critical nonconformities being cited during an evaluation and deem a laboratory nonconforming unnecessarily. By removing checklist items 2.2.2, 2.3.3, 2.5.4, 2.9.2, 2.12.8, 3.3.2, 3.4.2, 3.8.12, it will ensure a laboratory is appropriately cited once in Microbiology Checklist Part I if they are not adequately performing media productivity testing across all media types. Once checklist items are removed, editorial renumbering of the checklist will be required to maintain orderliness.
13. Public Health Significance	The proposed modifications are to improve consistency in the current NSSP Microbiology evaluation standard.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-119 as amended.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-119.

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Proposal Subject	Modification of MARBIONC Brevetoxin (Neurotoxic Shellfish Poisoning, NSP) ELISA Method Laboratory Evaluation Checklist
Specific NSSP Guide Reference	Section IV Guidance Documents Chapter II Growing Areas .15 Evaluation of Laboratories by state Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists
Text of Proposal/ Requested Action	The requested action is to modify the current checklist to correct errors and make clarifications regarding specific quality assuarance parameters. (See attached.)
Public Health Significance	Brevetoxins produced by K. brevis are toxic to humans. Filter-feeding bivalves accumulate brevetoxins during blooms, and ingestion of contaminated shellfish can cause NSP in humans. The MARBIONC Brevetoxin ELISA method was approved for limited use at the 2017 ISSC meeting. The attached revised checklist provides the quality assurance and method requirements that laboratory evaluation officers will use to evaluate laboratories implementing the MARBIONC Brevetoxin ELISA method to support the NSSP.
Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-120 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-120.

Proposal No. **23-121**

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Proposal Subject	Mooring Area Guidance Document Request
Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II Growing Areas
Text of Proposal/ Requested Action	The requested action is to have the ISSC refer to an appropriate committee a charge to develop a guidance document for mooring areas.
Public Health Significance	Mooring areas were incorporated into the 2019 Guide to for the Control of Molluscan Shellfish without a related guidance document. State shellfish authorities would benefit from guidance on how to complete mooring area assessments and classifications.
Cost Information	No cost would be associated with this proposal.
Action by 2023 Task Force I	Recommend adoption of Proposal 23-121 as submitted.

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10. Proposal Subject	Addition of Vv MPN real-time PCR to Microbiology PCR Checklist
11. Specific NSSP Guide Reference	Section IV Guidance Documents - Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists; References – NSSP Laboratory Evaluation Checklists 6. Shellfish Laboratory Evaluation Checklist for PCR Microbiology (link)
12. Text of Proposal/ Requested Action	3.2.3 The PCR forward and reverse primers used target. For Total and Pathogenic Vp Real-time PCR Method tdh_269-20: 6FAM-5'-TGACATCCTACATGACTGTG-3'-MGBNFQ trh_133-23: TET-5'-AGAAATACAACAATCAAAACTGA-3'-MGBNFQ tlh_1043: TEXAS RED-5'- CGCTCGCGTTCACGAAACCGT -3'-BHQ2 IAC_109: CY5-5'- TCTCATGCGTCTCCCTGGTGAATGTG -3'- BHQ2 trh_20F: 5'-TTGCTTTCAGTTTGCTATTGGCT-3' trh_292R: 5'-TGTTTACCGTCATATAGGCGCTT-3' tdh_89F: 5'-TCCCTTTTCCTGCCCCC-3' tdh_321R: 5'-CGCTGCCATTGTATAGTCTTTATC-3' tlh_884F: 5'-ACTCAACACAAGAAGAGATCGACAA-3' tlh_1091R: 5'-GATGAGCGGTTGATGTCCAAA-3' IAC_46F: 5'-GACATCGATATGGGTGCCG-3' IAC_186R: 5'-CGAGACGATGCAGCCATTC-3' For Vv Real-time PCR Method (SYBR) vvhF 5'-TGTTTATGGTGAGAACGGTGACA-3' vvhR 5'-TTCTTTATCTAGGCCCCAAACTTG-3' For Vv Real-time PCR Method vvhF: 5'-TGTTTATGGTGAGAACGGTGACA -3' vvhR: 5'-TTCTTTATCTAGGCCCCAAACTTG-3' vvh_Probe: Cy5-5'-CCGTTAACCGAACCACCCGCAA-3'-IAbRQ IAC_46F: 5'-GACATCGATATGGGTGCCG-3' IAC_186R: 5'-CGAGACGATGCAGCCATTC-3' IAC_Probe: JOE-5'-TCTCATGCGTCTCCCTGGTGAATGTG-3'-IABkFQ
13. Public Health Significance	The current laboratory evaluation checklist for PCR methods does not include the details of the MPN-real-time PCR method for *V. vulnificus* adopted as an approved NSSP method at the 2019 Conference Biennial Meeting. The proposed modifications of this checklist will provide Laboratory Evaluation Officers an appropriate and standardized tool by which to evaluate laboratories implementing this method.
14. Cost Information	N/A
Action by 2023 Laboratory Committee	Recommended adoption of Proposal 23-122 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-122.

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10. Proposal Subject	Guidance for calculating the 90th percentile for end-product depurated shellfish
11. Specific NSSP Guide Reference	Section IV Guidance Documents; Chapter II Growing Areas; Section .17 Calculating the 90th percentile for end-product depurated shellfish
12. Text of Proposal/ Requested Action	Process verification in depuration is performed continuously to ensure that the microbial contaminant load is being effectively reduced. Two (2) indices of performance, the geometric mean and the ninetieth (90th) percentile have been developed to describe the effectiveness of the depuration process. Critical limits for these parameters have been established empirically by shellfish species. For soft clams (*Mya arenaria*), a geometric mean of fifty (50) and a ninetieth (90th) percentile of 130 have been set. For hard clams, oysters, manila clams and mussels, a geometric mean of twenty (20) and a ninetieth (90th) percentile of seventy (70) have been adopted. Geometric means and ninetieth (90th) percentiles are determined daily or as end-product results become available from the analysis of the most recent ten (10) consecutive harvest lots per species, per restricted harvest area used. If the critical limits for either the geometric mean and/or the ninetieth (90th) percentile are exceeded, the process is considered to be unverified; and, additional sampling requirements must be instituted to ensure effective process control. End-product depurated shellfish samples are analyzed using two (2) different methods of recovery, a pour plate procedure and a single dilution MPN test. Calculation of the ninetieth (90th) percentile for these samples is complicated by the fact that fecal coliforms recovered by the MPN and ETCP methods follow different statistical distributions. To accommodate these differences and maintain a high likelihood for detecting an unacceptable amount of process variability without having to change or alter the formula used requires the use of nonparametric or "distribution free statistics." Using "distribution free statistics," the position of the ninetieth (90th) percentile for end-product depurated shellfish samples is calculated by arraying the fecal coliform count data in ascending order and applying the formula (n + 1)P/100. As an example of the use of this formula, the Model Ordinance requires that the ninetieth (90th) percentile of the fecal coliform analytical data be calculated from the most recent ten (10) consecutive harvest lots for each shellfish species depurated from each restricted harvest area. Fecal coliform count data, whether from the ETCP or MPN procedure for these ten (10) lots must be arrayed from the smallest to the largest value using the arithmetic (not logarithmically transformed) count data. Applying the formula, n would be greater than or equal

	to ten (10) for the ten (10) most recent consecutive harvest lots required by the Model Ordinance. P, the percentile of interest would be ninety (90). Using the minimum sample set of n=10, ~~Multiplying~~ multiplying the formula out gives the position of the ninetieth (90th) percentile in the arrayed data. Performing these calculations, 10 + 1 = 11, 11 x 90 = 990/100 = 9.9. Thus, the ninetieth (90th) percentile for end-product depurated shellfish data when n=10 is the value of the 9.9th sample in the ten (10) sample array. Using ~~the~~ ten (10) samples ~~as required by the Model Ordinance~~, the ninetieth (90th) percentile for end- product depurated shellfish samples would always be the value of the 9.9th sample in the ascending array of the arithmetic count data. To calculate this value from the arrayed data, interpolation between samples nine (9) and ten (10) is necessary. This is best illustrated using several samples. Example 1… Example 2… Example 3… In cases where more than ten samples have been analyzed in the most recent ten (10) consecutive harvest lots for each species depurated or for each harvest area used, the geometric mean and estimated 90th percentiles may be calculated using the methodologies below in examples 4 and 5. Example 4 (attached) Example 5 (attached)
13. Public Health Significance	Incorrectly calculating the 90th percentile can lead to erroneous decisions that could affect public health. For instance, both the California Dept of Public Health and the FDA mis-calculated the 90th percentile for a data set in which n=36. They insisted, based on the examples given in the NSSP Guide, that the 90th percentile was **always found between the 2 largest numbers** in the data set, even when n is large, which is incorrect.
14. Cost Information	This clarification to the NSSP Guide, with additional examples, will make it easier to correctly calculate this depuration performance index and should reduce confusion and disagreements, which could save time and money.
Action by 2023 Task Force I	Recommended referral of Proposal 23-123 to an appropriate committee as determined by the Conference Chairperson.

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10. Proposal Subject	Updated Marina and Mooring Area Guidance
11. Specific NSSP Guide Reference	Section IV. Guidance (Mooring Area)
12. Text of Proposal/ Requested Action	MARINA and MOORING AREA GUIDANCE - **DRAFT** The following guidance is provided to ensure the uniform application of the National Shellfish Sanitation Program (NSSP) Model Ordinance (MO) criteria, as adopted by the Interstate Shellfish Sanitation Conference (ISSC), for the evaluation and classification of shellfish growing waters in and around docks, marinas, and boat mooring areas. BACKGROUND A marina policy was developed at the ISSC conference held in August of 1986. It was recognized that a marina is a potential pollution source in a shellfish growing area, and that a closure zone is required to prevent the harvest of shellfish for human consumption in and around occupied marinas and mooring areas. The purpose of the policy was to establish a uniform national approach to marina and mooring area closures. At the July 1988 ISSC conference, approval was given to incorporate the marina policy into the definition and growing area classification sections of the NSSP MO. The 1989 "*Evaluation of Marinas by State Shellfish Sanitation Control Officials*", better known as the **1989 Marina Guideline,** was released in order to further clarify the new marina policy adopted into the 1990 NSSP Manual of Operations Part I Sanitation of Shellfish Growing Areas. The 1989 Marina Guideline was originally intended for the U.S. Food and Drug Administration (FDA) and State Shellfish Control Authorities (Authority) to use as guidance when classifying growing areas in and around marina facilities. The 1989 Marina Guideline has been used in all the FDA growing area training courses since inception as a reference on implementation of the NSSP MO marina criteria. As a result of actions taken at the 2019 biennial conference, "marina" and "mooring area" were separated into two (2) definitions (NSSP MO Section I. B.). In addition, the NSSP MO Section II. Chapter IV. @.06 was created to allow for mooring areas to be classified as conditionally

approved and conditionally restricted in the open status if a detailed pollution assessment is conducted at the frequencies required by the NSSP MO Section II. Chapter IV. @.01 A. (2.), C., and D. indicating a significant reduced risk from pollution sources and if there is a Conditional Area Management Plan (CAMP) in place with sufficient controls to protect human health.

The justification for this change suggests that there may be a different level of human health risk associated with how a mooring area, as a pollution source, may be managed compared to a marina. Boats are considered a potential pollution source due to the capability to discharge human sewage into a growing area. As technology has improved and the management of mooring areas have evolved with the implementation of the Federal No Discharge Zone (NDZ) program and availability of boat waste pump out boats and facilities, there is the potential, with enough oversight and management controls in place, to limit the capacity for overnight occupancy and sewage discharge from boats in a mooring area compared to a marina.

This updated marina and mooring area guidance document is intended to serve as guidance for the FDA when evaluating state growing area classification programs and as guidance for authorities regarding the classification and management of marinas and mooring areas in accordance with the NSSP MO requirements.

GUIDANCE

This guidance will provide clarification for the pollution assessment, classification, dilution calculation, and conditional area classification management of marinas and mooring areas, in and adjacent to, shellfish growing areas.

Boats congregated into a marina or mooring area are operated and inherently occupied by people at some time and therefore, have the potential to discharge human sewage and graywater into associated shellfish growing areas. As a result, every public or private watercraft, barge, houseboat, or boat, that has the potential to produce an overboard discharge from a marine toilet or discharge graywater, should be considered a potential pollution source in the evaluation of shellfish growing areas.

Since marine toilets may provide only limited or no treatment, human sewage discharges from boats may contain bacteria and viruses attributed to human sewage and graywater. For this reason, discharges of graywater and marine toilets represent a greater public health risk than other discharges of sanitary waste, and since these discharges can be sporadic, it may represent a greater public health risk than the FC sources typically detected by routine bacteriological monitoring.

Since many marina facilities and mooring areas are in or adjacent to shellfish growing areas, and waste discharges are not uniformly distributed in the water column, detection of low levels of coliforms from waste discharges by current pollution monitoring methods may not provide sufficient information to properly classify the waters in or adjacent to a marina or mooring area. Therefore, each marina and mooring area pollution assessment, dilution analysis, classification, and closure zone should be considered on a site-by-site basis, given the potential significant public health risk combined with the unique characteristics of each site.

As a result, a classification other than approved or restricted is required for the area within a marina or mooring area. This requirement is based on the public health requisite that waters receiving sporadic waste discharges from marine toilets or discharge of graywater are not suitable for the direct harvest of shellfish destined for human consumption or for relay or depuration. A pollution assessment and dilution determination must be used for classifying and making status determinations for marinas and mooring areas and adjacent shellfish growing areas.

MARINAS

Per the 2019 Revision of the NSSP MO Section I. B.:

Definition: Marina - *any water area with a structure (docks, basin, floating docks, etc.) which is used for docking and constructed to provide temporary or permanent docking space for more than ten (10) boats.*

MARINA PROPER

Per the NSSP MO Section II. Chapter IV. @.05 A, the marina proper shall be classified as: conditionally approved, conditionally restricted, or prohibited. A *pollution assessment* shall also be conducted in order to support the conditionally approved or conditionally restricted classification. The FDA's interpretation is that the marina pollution assessment is not intended to allow direct harvesting in the marina proper while more than 10 boats are present, but to document the seasonality and the presence of boats for the development of a Conditional Area Management Plan (CAMP) and to assess the marina proper as a pollution source, gather information for the dilution analysis, and provide documentation in the sanitary survey.

If more than 10 boats are not present during certain seasons (as in some geographical areas) the marina proper may be reclassified or changed to the open status if already classified as conditionally approved or conditionally restricted to permit harvest. During such periods the Authority must document that the area meets the specific NSSP MO criteria for the classification allowing harvest in the CAMP.

ADJACENT WATERS

Per the NSSP MO Section II. Chapter IV. @.05 B., waters adjacent to a marina proper may be impacted by pollution associated with the marina. Therefore, when more than 10 boats are present, a dilution analysis shall be used to determine if there is any impact to the adjacent growing area waters. The dilution analysis shall be based on the volume of water in the vicinity of the marina proper.

If the dilution analysis predicts a theoretical fecal coliform (FC) loading greater than (>) 14 FC/100 ml, the waters adjacent to the marina shall be classified as: conditionally approved, restricted, conditionally restricted, or prohibited. If the dilution analysis predicts a theoretical FC loading less than (<) 14 FC/100 ml, the waters adjacent to the marina may be classified as: approved or conditionally approved.

In reference to NSSP MO Section II. Chapter IV. @.05 B. (3), the dilution analysis around a marina proper shall incorporate the following factors. The recommendations provided represent guidance for how the authority may meet the intent of each requirement:

(a) **Slip occupancy rate for the marina**:

This is the quantity of waste potentially originating in a marina and depends on the number of people who are present in the marina. The fewer boats that are found to be occupied, the smaller the expected impact from the marina proper. The NSSP MO provides for establishing an occupancy rate for each marina. The slip occupancy rate of the marina should be documented by actual observation of marina operations during the time of highest usage such as weekends or holidays. Document the overall number of boats in a marina proper and the number of boats being occupied as well as the number of people on each boat. Document the number of slips in the marina proper.

(b) **An actual or assumed rate of boats which will discharge untreated waste**:

Document the number of boats with a marine sanitation device (MSD) type used (i.e., MSD Type I, II, or III) in the marina. If the authority uses an assumed rate of discharge, that rate should be supported by data gathered during the pollution assessment of the marina.

(c) An occupancy per boat (number of persons per boat):

If the authority chooses not to determine a specific occupancy per boat rate by investigation, the authority shall assume a minimum occupancy rate of two (2) persons per boat (NSSP MO Section II. Chapter IV. @.05 B. (6)).

Document the number of boats with liveaboard capability as well as the number of people on liveaboard boats in the marina. This inventory should be taken during the expected high usage times such as weekends and holidays. The inventory should have continuity so that changes in population during high occupancy times will be documented. Regional differences in boat usage, and the percent of high usage, will vary.

(d) A fecal coliform discharge rate of 2 x 10^9 for the theoretical fecal coliform contribution per person per day.

(e) Assume that the wastes are completely mixed in the volume of water in and around the marina.

(f) **Documentation, verification and enforcement of Federal No Discharge Zones and locally well enforced no discharge and occupancy by-laws and regulations**:

Provide documentation of the NDZ: enforcement records, vessel inspection records, marina use agreements, available educational material, and graywater regulations. Document in the management plan how vessels are inspected to ensure that boats equipped with an MSD that is not properly sealed to prevent discharge of sewage into the water is documented and enforced. Document Memorandums of Understanding or Agreements with local towns, municipalities, and patrol enforcement agencies defining each agency's responsibility in administering and enforcing the NDZ.

(g) Availability and documented use of pump out boats or facilities:

Document the availability and number of pump out facilities and boats available to the marina. Document use and maintenance records, operation procedures, ease of use, hours of operation, pump out log, previous spills, and the individual responsible for pump out operations. The pump out log should include: date, boat name and length, approximate number of gallons pumped, and initials of the operator.

Document enforcement records and boat inspection records. Document the procedures used if there is a waste spill. Document the frequency of when inspections are conducted to ensure pump-out stations are properly maintained and compliant with Clean Vessel Act (CVA) grant requirements. The records of inspections must be maintained and available for review.

MOORING AREAS

Per the 2019 Revision of the NSSP MO Section I. B.:

Definition: Mooring Areas - *any water area that is used to provide temporary or permanent anchorage for more than twenty (20) boats. Mooring areas do not include any structures for docking boats.*

MOORING AREA PROPER

Per the NSSP MO Section II. Chapter IV. @.06, a designated mooring area, where there is anchoring or mooring of boats, which is in or adjacent to a shellstock growing area shall be classified as: conditionally approved, restricted, conditionally restricted, or prohibited.

Prior to the Authority establishing a classification of conditionally approved, conditionally restricted, or restricted in the mooring area proper, a *pollution assessment* supporting the classification will need to be conducted by the authority. The NSSP MO provides flexibility so that if the *pollution assessment* determines that the mooring area has controls in place and is not considered a pollution source and it is thoroughly documented in the CAMP, the area may be classified as conditionally approved or conditionally restricted and placed in the open status with boats present.

The following factors shall be considered and documented when conducting a *pollution assessment* to determine the classification of the mooring area and adjacent waters in accordance with the NSSP MO requirements.

POLLUTION ASSESSMENT

The NSSP MO Section II. Chapter IV. @.06 A. (1) requires that a *pollution assessment* supporting the classification of mooring areas be conducted by the authority. In accordance with the 1986 ISSC Marina Policy and the 1989 Marina Guidance, the basis for occupancy and discharge rates should reflect worst case conditions and the inventory should be taken during the expected high usage times such as weekends and holidays.

The *pollution assessment* shall include the following factors according to the NSSP MO Section II. Chapter IV. @.06 A. (1). The recommendations provided for each factor represents suggested guidance for how the authority may meet the intent of each required component of the *pollution assessment*:

(a) **Boat Type and Usage**:

- Documentation of the boat type and usage should be considered from a public health perspective and the risk of

the potential for overboard discharge from both treated and untreated sewage as well as graywater.

- Document the type and size of boats in the mooring or anchorage area such as cabin cruiser, houseboat, cuddy cabin, runabout, commercial fishing vessel, skiff, daysailer, etc.
- Document the number of boats in each type and size category.
- Document the usage of boats such as overnight, weekend, day use, as well as commercial, or recreational.
- The boat type and usage information may be used in a mooring area management strategy to separate out boats that might pose more of a human health risk into a different conditionally managed area using separate performance standards.

(b) **Density of Boats**:

- Document the geographic location of the mooring area and include a map defining the mooring area boundaries.
- If boats are geographically managed by type and use, document this management strategy using a map that defines the mooring area management areas.
- Document the density of boats as the number of boats per a unit of area (For example: 100 boats per 1 sq. mile).
- Each individual mooring or anchorage area in a growing area should be accounted for and evaluated and where multiple mooring areas are present in a growing area, the authority should evaluate the impact of those individual mooring areas on the growing area from a holistic or cumulative impact. As an example, using best human health protection management practices, it may not be appropriate to separate a single group of multiple mooring boats (greater than 20 boats) into numerous separate mooring areas of 20 or less boats.

(c) **Accessibility to boats which could reduce likelihood of overnight occupancy**:

- In reference to the term “parking lot” mooring area, such as a location where boats are temporarily moored for short periods of time, but not occupied overnight, document the factors which could reduce or increase the likelihood of overnight occupancy in the mooring area proper.
- Provide a detailed justification explaining how accessibility to boats in the mooring area increases or decreases the likelihood of overnight occupancy. This may include how

the access of the boats in the mooring area are managed and how accessible boats are to overnight occupancy.

- Document the municipal mooring area regulation(s), town charter(s), municipal regulation(s), and records documenting enforcement of said regulation(s) and charter(s) that limits or mandates no overnight occupancy.
- Document how boat owners access their vessels, such as through launch service (hours of operation), personal dinghy, etc.
- Provide and maintain records from the municipal or state enforcement agencies when overnight occupancy regulations are enforced or violated.

(d) **Occupancy Rates**:

- Document the number of mooring balls/buoys and the number of boats allowed on each.
- Document the overall number of boats in a mooring area and the number of boats being occupied as well as the number of people on each boat. If the mooring area is considered a "parking lot", such as a location where boats are temporarily moored for short periods of time but not occupied overnight, provide documentation to that effect, including justification for use.
- Document any transient mooring areas and their boat capacity.

(e) **Seasonal Use Pattern**:

- Document if there is a seasonal boat use pattern.
- Document what the seasonal boat use pattern is including the seasonal dates as to when more than 20 boats are present in the mooring area.

(f) **An actual or assumed rate of boats which will discharge untreated waste**:

- Conduct and document an onsite assessment of the mooring area and document the type and number of boats that have the potential for discharging treated or untreated sewage including graywater.
- Document boats with marine heads and include the number and location of boats with each type of MSD (Type: I, II, or III).

(g) **Documentation, verification, and enforcement of Federal No Discharge Zones (NDZ), and locally well enforced no discharge and occupancy regulations or by-laws**:

- Provide documentation of the NDZ: enforcement records, boat inspection records, mooring area use agreements, available educational material, graywater discharge regulations, and occupancy records during high-use times.
- Document how boats equipped with a MSD, not properly sealed to prevent discharge of sewage into the water, are inspected.
- Provide any Memoranda of Understandings or Agreements with local towns, municipalities, and patrol enforcement agencies. Define each agency's responsibility in administering and enforcing the NDZ; including references to the statue, regulation, or charter that confers authority to enforce the NDZ.
- Document the CAMP communication requirements (contact tree) in case an emergency closure is warranted.

(h) **Availability and documented use of shore-based pump out facilities and pump out boats**:

- Document the availability and number of pump out facilities and pump out boats available to the boats in the mooring area proper.
- Document pump out practices, pump out procedures, educational information, and employee/operator training.
- Document the use and maintenance records, operation procedures, ease of use, hours of operation, pump out log, previous spills, and who is responsible for the pump out operations. The pump out log should include date, boat name and length, approximate number of gallons pumped, and initials of the operator.
- Document enforcement records and boat inspection records.
- Document the procedures if there is a waste spill.
- Document the frequency as to when inspections are conducted to ensure pump-out stations are properly maintained and compliant with Clean Vessel Act grant requirements; with records of past inspections maintained and available for review.

The NDZ is only one factor to consider when conducting a *pollution assessment* to classify a growing area with a mooring area(s) as conditionally approved or conditionally restricted in the open status with boats present. The FDA does not consider the NDZ designation to be a standalone *pollution assessment*, control mechanism, or justification for classifying a mooring area(s) as conditionally approved or conditionally

restricted in the open status. As stated in the NSSP MO language, documentation, verification, and enforcement of the NDZ and locally well enforced no discharge and occupancy regulations or by-laws will be necessary for the *pollution assessment* and for review during FDA growing area program evaluations.

In addition, Section 312 of the Clean Water Act (CWA) contains the principal framework for domestically regulating sewage discharges from boats and is implemented jointly by the U.S. Environmental Protection Agency (EPA) and the U.S. Coast Guard (USCG). Sewage, treated or untreated, is prohibited in an NDZ. The NSSP utilizes the CWA definition of sewage.

Definition: Sewage - human body wastes and the waste from toilets and other receptacles intended to receive or retain body wastes.

Graywater is not defined as "sewage" and is not prohibited under the NDZ requirements. Graywater may contain high levels of human bacteria and viruses and poses a significant human health risk when present and this should also be considered in the *pollution assessment*.

CONDITIONAL AREA MANAGEMENT PLAN (CAMP) FOR THE MOORING AREA PROPER CLASSIFIED AS CONDITIONALLY APPROVED OR CONDITIONALLY RESTRICTED IN THE OPEN STATUS

Per the NSSP MO Section II. Chapter IV. @.06 A. (1), a *pollution assessment* of the mooring area proper is required to determine if the mooring area can be classified as conditionally approved or conditionally restricted. Per the NSSP MO Section II. Chapter IV. @.06 A. (2), after the mooring area proper pollution assessment determines that the mooring area proper is not a pollution source and it is documented in the CAMP, the growing area may be placed in the open status.

The CAMP for each mooring area placed in a conditional classification is based on the information gathered during the *pollution assessment*. The CAMP will establish a strict set of criteria or performance standards, which must be met for the growing area to remain in the open status. Failure to meet the criteria or performance standards automatically places the growing area in the closed status, with immediate notice to the CAMP participants, affected industry, and the public.

Performance Standards for a Mooring Area CAMP should include:

- Establishment of a Memorandum of Understanding and/or an agreement to the conditions of the CAMP by the one (1) or more authorities involved including: mooring area management organizations, local municipalities, other local, State and Federal agencies, enforcement, harbor master, or other organizations which

may be involved in the management and enforcement of the mooring area proper, pump out operations, and NDZ management and enforcement.

- A written CAMP for the mooring area(s) and associated growing area being placed in the conditional classification, which includes a description of the mooring area(s) with a map showing the mooring area(s) boundaries.

- A sanitary survey that shows the growing area will be in the open status of its conditional classification and provide a description of the factors determining the growing area's suitability for being classified conditionally approved or conditionally restricted with supporting information and data.

- A description of the *pollution assessment* for the mooring area documenting how the reduction of an illicit human sewage (treated or untreated) and graywater discharge will be prevented and what management strategies are in place including, documenting boat types and uses, inspection of boat MSDs, documentation of pump out boats and facilities, NDZ regulations, education, management, and enforcement.

- A description of the plan for monitoring water quality including what will be sampled and the location of sample stations on a map, numbers of sample stations, and frequency monitored.

- A description of how the closed status for the conditional classification will be implemented which must include:
    - A clear statement indicating when the performance standards are not met, the growing area will immediately be placed in the closed status;
    - A requirement to notify the authority or authorities that management plan performance standards have not been met, including:
        - The name of the agency or other party responsible for notifying the authority;
        - The anticipated response time between the performance standards not being met and notification of the authority; and
        - The procedures for prompt notification including contingencies such as night, weekend, and absences of key personnel;
    - A description of implementation and enforcement, including:

- The response time between the notification to the authority of the failure to meet performance standards and activation of the legal closure of the growing area by the authority;
- The procedures and methods to be used to notify the shellfish industry; and
- The procedures and methods to be used to notify the patrol agency (enforcement agency) including:
- The name of the responsible patrol agency;
- The anticipated response time between the Aathority's legal closure of the growing area and notification of closure to the patrol agency; and
- A description of the patrol agencies anticipated activities to enforce the closed status of an area.

- A description of the criteria that must be met prior to reopening a mooring area or growing area in the closed status, including the need to determine that:
  - The performance standards established in the management plan are again compliant;
  - The flushing time for pollution dissipation is adequate;
  - A time interval has elapsed which is sufficient to permit reduction of human pathogens as measured by the coliform indicator group in the shellstock;
  - Where necessary, the bacteriological quality of the water must be verified; and
  - Shellstock feeding activity is sufficient to achieve reduction of pathogens to levels present prior to the pollution event.

- A commitment to a reevaluation of the management plan, at least annually, using the reevaluation requirements in the NSSP MO, or other regulations/rules required as necessary.
- A designation in the CAMP whether the shellstock may be harvested for relaying or depuration in a conditionally approved (closed status) or whether the harvested shellstock are to be relayed or depurated in a conditionally restricted area (open status).

ADJACENT WATERS

Per the NSSP MO Section II. Chapter IV. @.06 B., waters adjacent to a mooring area proper may be impacted by pollution associated as a result. Based on the pollution assessment conducted in NSSP MO Section II. Chapter IV. @.06 A., if the authority determines that the mooring area proper is a pollution source, a dilution analysis shall be used to determine if there is any impact to the adjacent waters. The dilution analysis shall be based on the volume of water in the vicinity of the mooring area proper.

If the dilution analysis predicts a theoretical FC loading greater than (>) 14 FC/100 ml, the waters adjacent to the mooring area shall be classified as: conditionally approved, restricted, conditionally restricted, or prohibited. It the dilution analysis predicts a theoretical FC loading less than (<) 14 FC/100 ml, the waters adjacent to the marina may be classified as: approved or conditionally approved.

The dilution analysis shall include the following factors according to the NSSP MO Section II. Chapter IV. @.06 B. The recommendations provided, represents guidance for how the authority may meet the intent of each requirement:

(a) **An occupancy rate for the mooring area**:

Consider that the quantity of waste potentially originating in a mooring area depends on the number of people who are present in the mooring area. The fewer boats that are found to be occupied, the smaller the expected impact from the mooring area. The occupancy rate of the mooring area should be documented by actual observation of mooring area operations during the time of highest usage such as weekends or holidays. Document the overall number of boats in a mooring area and the number of boats being occupied as well as the number of people on each boat. Document the number of mooring balls and buoys in the mooring area.

(b) **An actual of assumed rate of boats which will discharge untreated waste**:

Document the number of boats with installed toilets and document the MSD type used (MSD Type I, II, or III) in the mooring area having the capability to discharge to the environment. If the authority uses an assumed rate of discharge, that rate should be supported by data gathered during the pollution assessment of the mooring area.

(c) An occupancy per boat (i.e., number of persons per boat):

If the authority chooses not to determine a specific occupancy per boat rate by investigation in specific areas or sites, the authority shall assume a minimum occupancy rate of two (2) persons per boat (NSSP MO Section II. Chapter IV. @.06 B. (6)).

Document the number of people on liveaboard boats in the mooring area. This inventory should be taken during the expected high usage times such as weekends and holidays. The inventory should have continuity so that changes in population during high occupancy times can be documented. Regional differences exist regarding boat usage; therefore, the percent of high usage will vary.

(d) A fecal coliform discharge rate of 2 x 10^9 for the theoretical fecal coliform contribution per person per day.

(e) Assume that the wastes are completely mixed in the volume of water in and around the marina.
Document the average depth of the area based on bathymetry charts and the volume of dilution water needed if complete mixing is assumed.

DILUTION ANALYSIS

The NSSP MO Section II. Chapter IV. @.05 and @.06 states that a dilution analysis will be used for making classification and closure determinations for waters adjacent to each marina proper and mooring area proper (if a pollution assessment determines the mooring area may be a pollution source). The information collected from a pollution assessment will help in determining the potential pollution impact and classification and size of the classification area or closure zone.

This dilution analysis requirement is based on the public health requisite that waters receiving waste discharges from marine toilets from marinas and mooring areas are not suitable for the direct harvest of shellfish destined for human consumption.

The intentional or unintentional direct discharge of treated or untreated human sewage and graywater discharge from a boat into a marina or mooring area is considered a point source and a high human health risk and therefore, pursuant to the NSSP MO Section II. Chapter IV. @.03 E. (5) (a), "An area classified as prohibited shall be established adjacent to each sewage treatment plant outfall or any other point source outfall of public health significance."

The estimated per capita discharge of fecal coliforms, coupled with the estimated population in the marina or mooring area, can be used to determine the classification and estimate a closure zone. Closures for existing or proposed marinas and mooring areas should be developed assuming two (2) persons per boat, and a 2 x 10^9 fecal coliform (FC) contribution per person per day, unless actual persons per boat or occupancy and discharge rates are documented by surveys conducted for individual marinas or mooring areas on a case-by-case basis. The authority should assume 100% boat slip and mooring ball occupancy unless the actual occupancy rate is documented through observation or credibly estimated. This documentation shall be maintained as specified by the NSSP MO, Chapter I, for reevaluation of sanitary survey information.

Similarly, any expansion, modification, or change to the operation of a marina or mooring area will necessitate the reevaluation of the marina or mooring area occupancy rate.

In determining the above loading rates, a minimum factor should be considered to provide protection against intentional or unintentional waste discharges from boats in the marina or mooring area.

The theoretical waste discharge based on the occupancy and discharge rate, will be completely mixed in and around the marina or mooring area. The marina or mooring area closure zone shall be calculated to reduce the assumed bacterial load to 14 FC/100 ml, in the volume of water in the vicinity of the marina or mooring area. If the results of hydrographic studies are used, the estimated fecal coliform contribution can be distributed throughout the volume of water calculated to flow by the site in 24 hours.

Dilution hydrographic studies may be used to determine the water volume available for dilution and limits of travel of discharges from a marina. The area to be closed shall provide sufficient water volume for calculations to show that theoretical discharges from the marina or mooring area are diluted to 14 FC/100 ml of water. In situations where there are no hydrographic studies, the closed or prohibited area is to be established on a volumetric basis as though the wastes are completely mixed and uniformly distributed in and around the marina or mooring area. The closed area volume is typically based on average water depth and shall be sufficient to dilute the assumed waste load to a value of 14 FC/100 ml.

EXAMPLE CALCULATIONS

The following examples show how various factors are to be considered in closure area determinations around marinas or mooring areas:

CASE 1: No Documentation of Occupancy or Discharge Rates	
Number of Boat Slips	50
Number of People	2 x 50 =100
Number of Fecal Coliforms (FC)	$100 \times 2 \times 10^9 = 200 \times 10^9$
Dilution Volume Required	200×10^9 FC (14 FC/100 mL) x (1000 mL/liter) Volume = 1.4×10^9 liters (5.0×10^7 cu ft)
Average Depth in Vicinity of Marina	3 meters (10ft)
Closed Area Required	1.4×10^9 liters (3 meters) x (1000 liters/cubic meter) A = 4.7×10^5 square meters (5.0×10^6 sq ft)

Radius of Half Circle Prohibited/Closed Area	$\sqrt{2/\pi\ (4.7\ xx\ 10^5)}$ R = 550 meters (1800 ft)

CASE 2: Boat Slip Occupancy, Population, Holding Tanks and Pumpout Facilities Documented	
Number of Boat Slips	50
Slip Occupancy- Holiday Weekends	40 (80%)
Boats with No Holding Tanks*	16 (16/40 = 40%)
Average People per Boat	1.5
Number of People	1.5 x .40 x .80 x 50 =24
Number of Fecal Coliform (FC)	$24 \times 2 \times 10^9 = 48 \times 10^9$
Dilution Volume Required	48×10^9 FC (14 FC/100 mL) x (1000 mL/liter) $V = 3.4 \times 10^8$ liters (1.2×10^7 cu ft)
Average Depth in Vicinity of Marina	3 meters (10ft)
Closed Area Required	3.4×10^8 liters (3 meters) x (1000 liters/cubic meter) $A = 1.1 \times 10^5$ square meters (1.2×10^6 sq ft)
Radius of Half Circle Closed Area	$\sqrt{2/\pi\ (1.1\ xx\ 10^5)}$ R = 265 meters (870ft)

* Assumes pumpout facilities are consistently used, increase percentage if otherwise

REFERENCES

1. Interstate Shellfish Sanitation Conference Marina Policy. August 1986.
2. Evaluation of Marinas by State Shellfish Sanitation Control Officials. Guideline 1.0. June 1989.
3. National Shellfish Sanitation Program Manual of Operations, Part I. 1988 revision.
4. Department of Health and Human Services NE Technical Unit. 1986. Hydrographic Studies of the Great Salt Pond, Block Island, Rhode Island.
5. Geldreich, Edwin, et al. Bacteria in the Feces of 295 (March). 1962. The distribution of Coliform Bacteria in the Feces of Warm-Blooded Animals. JWPCF 34(3),
6. U.S. Environmental Protection Agency (EPA), Region IV. 1985. Coastal Marina Assessment Handbook.

	7. U.S. Department of Health and Human Services, Northeast Technical Services Unit. 1983. Hydrographic Studies of the Kiawah River, South Carolina. 8. Title 33 Code of Federal Regulations, Section 159.7 https://www.govregs.com/regulations/expand/title33_chapterI_part159_subpartA_section159.7#title33_chapterI_part159_subpartA_section159.1 9. National Shellfish Sanitation Program (NSSP), Model Ordinance (MO). 2019 Revision 10. U.S. Environmental Protection Agency (EPA), Office of Waste Management. 2011. Graywater Discharges from Vessels. U.S. Environmental Protection Agency (EPA), Federal No Discharge Zone (NDZ) Link: https://www.epa.gov/vessels-marinas-and-ports/vessel-sewage-no-discharge-zones
13. Public Health Significance	The 2019 NSSP MO included new language separating out marinas and mooring areas. The adopted language does not have descriptive details as to how the new mooring area language will be evaluated by the FDA. Given that marinas and mooring areas may be considered a potential pollution source and high risk if mooring areas are not assessed correctly, the proposed updated marina and mooring area guidance is presented to help provide the guidance on how to meet those new requirements.
14. Cost Information	N/A
Action by 2023 Task Force I	Recommends referral of Proposal 23-124 to an appropriate committee as determined by the Conference Chairperson.

2. Submitter	ISSC Laboratory Committee
3. Affiliation	
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6. City, State, Zip	Richmond, VA 23227
7. Phone	(804) 330-6380
8. Fax	
9. Email	issc@issc.org
10. Proposal Subject	Guidance for Laboratory Method Matrix Extensions
11. Specific NSSP Guide Reference	PROCEDURE XV. PROCEDURE FOR THE APPROVAL OF ANALYTICAL METHODS FOR THE NSSP and Section IV Guidance Documents – Chapter II. Growing Areas
12. Text of Proposal/ Requested Action	PROCEDURE XV. PROCEDURE FOR THE APPROVAL OF ANALYTICAL METHODS FOR THE NSSP 10. ~~For methods already adopted into the NSSP, consideration of expanding a method to a new molluscan shellfish species is accomplished using the "ISSC Method Application Format for Biotoxin Methods Matrix Extension" and the "ISSC Method Application Format for Microbiology Methods Matrix Extension." The simplified, reduced approach to method validation for expanding an NSSP method to new molluscan shellfish species is visually represented in the "Matrix Extension Guidelines" schematic.~~ For methods already adopted into the NSSP, additional work must be done in order to expand the use of that method to a new molluscan shellfish matrix. To determine if a Matrix Extension is needed, please refer to the guidance provided in the NSSP Guide for the Control of Molluscan Shellfish, Section IV. Guidance Documents, Chapter II. Growing Areas .21 - Guidance for Laboratory Method Matrix Extensions. If a matrix extension is needed, the necessary information, studies, and data to be provided to the Laboratory Committee for consideration are summarized on the "ISSC Method Application Format for Biotoxin Methods Matrix Extension" and the "ISSC Method Application Format for Microbiology Methods Matrix Extension" documents available on the Laboratory tab of the ISSC website. This simplified, reduced approach to method validation for expanding an NSSP method to a new molluscan shellfish matrix is visually represented in the "Matrix Extension Guidelines" schematic, also available on the ISSC website. Section IV Guidance Documents – Chapter II. Growing Areas .20 Quantitative Analytical Method Verification This guidance is provided to aid laboratories verifying the performance of an NSSP Approved Method or Approved Limited Use Method of analysis being transferred from the originating laboratory/submitter to the implementing

laboratory before being placed in service by the implementing laboratory. When a laboratory implements an NSSP method for the first time, the method performance must be verified in that laboratory. In addition, when a laboratory expands an existing method to a new shellfish matrix, method performance may need to be verified. Guidance outlined in .21 should be followed to determine if the new shellfish matrix is in the same matrix category as matrices previously implemented in the laboratory. If so, the method does not need to be verified. However, if the new shellfish matrix is in a different matrix category, then the method performance must be verified.The following performance criteria are to be verified: recovery, measurement uncertainty, precision (repeatability and intermediate precision), linear range, limit of detection (LOD), limit of quantitation (LOQ), and comparability.

Section IV Guidance Documents – Chapter II. Growing Areas (new section .21)

.21 Laboratory Method Matrix Extensions

Validating Use of an Analytical Method With A New Shellfish Matrix

Analytical methods employed in the National Shellfish Sanitation Program (NSSP) are validated for their intended use before being adopted. Since differing characteristics of various molluscan shellfish matrices may impact the performance of certain methods, each validation is specific only to the shellfish species or matrices that were included in the validation studies.

In order to expand the use of any method already adopted into the NSSP for use with other molluscan shellfish matrices, additional validation studies need to be done. Based on proximate composition data (i.e. the amount of protein, fat, and carbohydrates in each species), as well as a review of existing empirical data where methods have been tested using multiple species, the Matrix Category Table below was developed to help determine if a Matrix Extension study is needed.

If a new shellfish species of interest is in the same matrix category (i.e. vertical column of the table) as an already validated species, then the method should not require further validation. For example, if a method has already been validated for use with the Eastern Oyster (*Crassostrea virginica*), and the new species of interest is the Pacific Oyster (*Crassostrea gigas*), then a matrix extension study is not necessary.

If a new species of interest is in a different matrix category from all previously validated species, then a Matrix Extension validation study should be conducted and data submitted to the ISSC for review following the process outlined in the ISSC Constitution, Bylaws, and Procedures, Procedure XV (10.). For example, if a method has already been validated for use with the Eastern Oyster (*Crassostrea virginica*) and the Soft Shell Clam (*Mya arenaria*), and the new species of interest is the Atlantic Surf Clam (*Spisula solidissima*), then a matrix extension study is needed.

If the new species of interest is not found in the Matrix Category Table, a request to add the new species should be submitted to the ISSC Executive Office.

The following information should be included in the request: common and scientific name of species, rationale for inclusion, and any available data for categorization (e.g, proximate composition, empirical data on use).

Regardless of the categorization of the species of interest, certain analytical methods require more species-specific data. The results of these studies will supersede the groupings described in the table below if significant matrix effects are identified.

1. For methods utilizing liquid chromatography, analyses shall be conducted to ensure sufficient separation of target analyte from sample matrix peaks through analysis of peak resolution utilizing retention times (e.g., AOAC[1]). Chromatograms supporting the analyses with labels noting peaks of interest as well as matrix peaks shall accompany the data package.

2. For methods utilizing mass spectrometry, comparison of neat and matrix-fortified standards shall be conducted to assess matrix effects on ionization.

1	2	3	4	5	6	7	8
Oysters	***Hard Clams***	***Non-US Hard Clams***	***Geoducks****	***Soft Clams***	***Mussels***	***Estuarine Mussels (non-***	***Scallops*** **
Eastern Oyster (*Crassostrea virginica*)	Atlantic Surfclam (*Spisula solidissima*)	Wedge Shell Clam (*Donax cuneatus*)	Pacific Geoduck Clam (*Panopea generosa*; formerly *P. abrupta*)	Softshell Clam (*Mya arenaria*)	Blue Mussel (*Mytilus edulis*)	Asian Green Mussel (*Perna viridis*)	Sea Scallop (*Placopecten magellanicus*)
Edible Oyster (*Ostrea edulis*)	Ocean Quahog (*Arctica islandica*)	Asiatic Hard Clam (*Meretrix meretrix*)	Atlantic Geoduck Clam (*Panopea bitruncata*)		Mediterranean Mussel (*Mytilus galloprovincialis*)		Rock Scallop (*Crassodoma gigantea*)
Olympia Oyster (*Ostrea lurida*)	Northern Quahog (*Mercenaria mercenaria*)				California Mussel (*Mytilus californianus*)		Bay Scallop (*Argopecten irradians*)
Pacific Oyster (*Crassostrea gigas*)	Southern Quahog (*Mercenaria campechiensis*)				Chilean Mussel (*Mytilus chelensis*)		Peruvian Scallop (*Argopecten purpuratus*)
	Northern Razor Clam (*Siliqua patula*)				Korean Mussel (*Mytilus coruscus*)		
	Pacific Littleneck Clam (*Protothaca staminea*)						
	Butter Clam (*Saxidomus gigantea*)						
	Sunray Venus Clam (*Macrocallista nimbosa*)						
	Japanese Littleneck Clam (*Venerupis philippinarum*)						

*Geoducks are generally analyzed as whole animals for microbiological methods and gutballs only for biotoxin methods. If a different form of the animal is to be processed (i.e., gutball for micro method or whole animal for biotoxin method), it should be considered a separate matrix.

**Scallops can be analyzed as whole animal or muscle excluded. These different forms of the animal should be considered a separate matrix. Methods for muscle only will not be considered as the product is not within the NSSP.

1 Association of Official Analytical Chemists. "AOAC Guidelines for Single Laboratory Validation of Chemical Methods for Dietary Supplements and Botanicals". Arlington, VA. 2002.

13. Public Health Significance	To ensure accurate reporting of analytical results within the NSSP, methods must be demonstrated to be fit-for-purpose. The program has recognized the potential interference from different shellfish types. This proposal is intended to provide additional detail on the conditions under which a matrix extension validation study is needed compared to when a method verification study is required.
14. Cost Information	Dependent upon the level of validation/verification needed.
Action by 2022 Executive Board	Granted Interim Approval in effect until the Conference convenes at the 2023 ISSC Biennial Meeting.
Action by 2023 Laboratory ommittee	Recommended adoption of Proposal 23-125 as submitted.
Action by 2023 Task Force I	Recommends adoption of the Laboratory Committee recommendation on Proposal 23-125.