Proposal No. **11-103**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

Submitter	Thomas L. Howell
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Proposal Subject	Alternative Male-specific Coliphage Meat Standard for Restricted Classification of Growing Areas Impacted by wastewater treatment plant outfall.
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Area @ .02 Bacteriological Standards
Text of Proposal/ Requested Action	G. Standard for the Restricted Classification of Growing Areas Affected by Point Sources and Used as a Shellstock Source for Shellstock Depuration. (4) Exception. If the Male-specific Coliphage indicator is used for supplemental process verification using an end-point meat standard of < 50PFU/100gm and existing fecal coliform testing requirements in Chapter XV .03 J. are used, then FC water quality monitoring is not required for the restricted classification of growing areas affected by point sources such as wastewater treatment plant outfall.
Public Health Significance	Under shellfish relay, water quality requirements are not needed for the restricted classification when a contaminant reduction study is conducted and a minimum time period of two weeks is used. For depuration, the restricted classification requires water quality monitoring and standards. The reason for these upper FC limits is that FC meat indicator does not adequately reflect the viral risk and/or viral depuration kinetics. Male-specific coliphage is a viral indicator organism to be used in growing areas impacted by point source sewage contamination. MSC demonstrates significant advantages over FC alone for both the assessment of viral contamination and assessment of viral depuration kinetics. Upper FC limits were put into the NSSP to prevent shellfish with higher levels of viruses from being depurated. Several studies clearly show that conventional depuration using FC for process validation is not adequate to protect public health with respect to virus contamination in growing areas with significant wastewater treatment plant and sewage impact. Studies have also shown that viral levels in shellfish impacted by sewage and partially treated sewage detected using MSC and molecular techniques are much lower in the summer months than the winter months. Additionally, the viral depuration rate is higher in the summer with process waters >18°C. Recent studies have also shown that MSC is an appropriate viral indicator to assess viral depuration. Therefore, seasonal viral depuration using male-specific coliphage as well as FC for process verification is a superior approach to taking water samples using FC in a growing area adjacent to wastewater treatment plant outfall. Combining the bacterial indicator of FC and the viral indicator MSC for mitigation strategies that use meat scores is far more direct and effective than water quality sampling in this context.

Cost Information	The Male-specific Coliphage (MSC) method is an inexpensive double-agar pour plate method that can be run in any state-certified microbiological laboratory. A refrigerated centrifuge capable of 9,000G is required which costs $10K to $12K (USD). Significant cost savings and a higher level of public health protection may be realized using strategies such as seasonal coliphage depuration process validated using MSC and seasonal coliphage relay using MSC in contaminant reduction studies than requiring water quality limits using FC.
Action by 2011 Task Force I	Recommend referral of Proposal 11-103 to the appropriate committee as determined by the Conference Chairman.
Action by 2011 General Assembly	Adopted recommendation of 2011 Task Force I on Proposal 11-103.
Action by FDA February 26, 2012	Concurred with Conference action on Proposal 11-103.
Action by 2013 Growing Area Classification Committee	Recommend referral of Proposal 11-103 to the appropriate committee as determined by the Conference Chairman. It was additionally recommended that a workgroup be formed to look at current MSC data and the science behind its potential use and applicability for use in the NSSP. The workgroup will organize a summit of outside experts, academia, and scientists to present current information and science on MSC. The group will meet at least quarterly and respond back to the Growing Area Classification Committee on its findings and recommendations. Recommended that the ISSC pursue funding to facilitate scheduling a summit to bring together experts to present the current science in the use of MSC.
Action by 2013 Task Force I	Recommended adoption of Growing Area Classification Committee action on Proposal 11-103.
Action by 2013 General Assembly	Adopted recommendation of 2013 Task Force I on Proposal 11-103.
Action by FDA May 5, 2014	Concurred with Conference action on Proposal 11-103.
Action by 2015 Growing Area Classification Committee	Recommended referral of Proposal 11-103 to appropriate committee as determined by the Conference Chair.
Action by 2015 Task Force I	Recommended adoption of Growing Area Classification Committee recommendation on Proposal 11-103.
Action by 2015 General Assembly	Adopted recommendation of Task Force I on Proposal 11-103.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 11-103.
Action by 2017 Growing Area Committee	Recommended adoption of Proposal 11-103 as amended. Add a new section as follows: Chapter XV. Depuration .03 Other Model Ordinance requirements K. Supplemental Requirements for Depuration using MSC Viral Controls for Shellstock Harvested from Conditionally Restricted Growing Areas Impacted by Wastewater System Discharge (WWSD).

	If the conditionally restricted growing area from which the shellstock is being depurated is impacted by wastewater treatment system discharge (generally that section of the conditionally restriced growing area located within the 300:1 to 1000:1 dilution lines), then supplemental requirements for depuration using MSC viral controls may be required. Depuration using MSC viral controls may be seasonally limited and may be species and depuration facility specific. Contaminant reduction studies as described in (1) below are recommended unless the SSCA and the Depuration Facility Operator have significant experience with the depuration process using MSC viral controls. (1) Male-specific coliphage may be used in addition to fecal coliform for species-specific, growing area-specific, and depuration system-specific contaminant reduction studies. These contaminant reduction studies should demonstrate that; (a) Predictable periods of time exist when male-specific coliphage levels are less than 1,000 PFU/100gm in shellfish meats, (b) Male-specific coliphage and fecal coliform can be consistently reduced below end-point requirements, and (c) Critical limits of season, process water temperature and salinity, and system design and operation limitations can be assessed and determined (d) Species-specific operating protocols may be developed from the contaminant reduction studies for each conditionally restricted growing area that includes; (i) Calendar dates when depuration shall be permitted, (ii) Water temperature and salinity limitations, (iii) Minimum processing time, (iv) Sampling requirements and release criteria, and (v) Operating Protocol. (2) All requirements of Chapter XV shall be followed, (3) A single 0-day MSC shellfish meat sample is required. (4) The MSC end-point requirement for depuration is 50 PFU/100gm. If the single 0-day sample exceeds 50 PFU/100gm, then triplicate samples are required prior to release of product. (5) The geometric mean of the triplicate samples used for product release must not exceed 50PFU/100gm and no single sample over 100 PFU/100gm. (6) Extended depuration may be permitted to achieve end-point requirements. (7) Evaluation of male-specific coliphage samples shall be performed in an NSSP conforming laboratory,
Action of 2017 Task Force I	Recommended adoption of Growing Area Classification Committee recommendation on Proposal 11-103.

Action by FDA February 7, 2018	Did not concur with Conference action on proposal 11-103
Action by ISSC Executive Board	Referred Proposal 11-103 to an appropriate committee as determined by the Conference Chair.

Proposal No. **13-107**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

Submitter	Robert Rheault
Affiliation	East Coast Shellfish Growers Association
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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.

Proposal No. **13-111**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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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	Concurred with Conference action on Proposal 13-111.

January 11, 2016	
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.

Proposal No. **13-114**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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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.

Proposal No. **13-116**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

Submitter	Florida Department of Agriculture and Consumer Services
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Proposal Subject	Shellfish Quarantine Guidance Document
Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.04 Marine Biotoxin Control Section IV. Guidance Documents Chapter II. Growing Areas .02 Guidance for Developing Marine Biotoxin Contingency Plans
Text of Proposal/ Requested Action	Model Ordinance Chapter IV. Shellstock Growing Areas @.04 Marine Biotoxin Control Section A. (4) describes agreements or memoranda of understanding between the Authority and individual shellfish harvesters or individual shellfish dealers, to allow harvesting during marine Biotoxin closures under specific, controlled conditions. The State of Florida has successfully implemented such an agreement to address Neurotoxic Shellfish Poisoning (NSP) for over a decade. This pilot project, developed in consultation with FDA, has resulted in zero cases of NSP in commercially harvested shellfish from Florida waters. NSP may affect any Gulf or South Atlantic state and therefore Florida wishes to provide ISSC member states with a proven quarantine protocol template for incorporation into the Model Ordinance Section IV. Guidance Documents. Guidance Documents Chapter II. Growing Areas .02 Guidance for Developing Marine Biotoxin Contingency Plans. Text of the proposed guidance is as follows: Example Protocol for Quarantine Harvest of Shellfish from Aquaculture Leases During *Karenia brevis* Closures: A. Closure of an entire shellfish growing area due to *Karenia brevis* shall be in accordance with Model Ordinance Chapter IV. @.04 C. (1). B. When a shellfish growing area is closed due to *Karenia brevis*, the Authority may allow harvest of shellfish from selected aquaculture leases within a specific zone by authorized harvesters and subsequent controlled quarantine at a certified shucker packer or shellstock shipper. This option would not be

available if any Authority collected water samples in the specific zone exceeded 200,000 cells per liter of *Karenia brevis*. Zone is defined as an Authority delineated geographic area within a Conditionally Approved or Approved classified shellfish growing area.

Controlled quarantine conditions:

The Authority will determine and plot the specific zones. Certified processors possessing a valid shellfish processing plant certification license must have written permission from the Authority to engage in this activity. To be eligible for participation in the quarantine program, the certified processor must:

(1) Provide the Authority with written and signed agreements the processor has with shellfish aquaculture leaseholders who would be supplying the shellfish and;
(2) Notate on their application letter which FDA-approved marine Biotoxin laboratory will be used to conduct the approved mouse bioassay and;
(3) Provide the Authority with the cooler capacity, physical address and current certification number of the facility to be used for controlled quarantine of shellfish. All quarantine coolers must be non-mobile, secure from unauthorized access and equipped with warning signs in a language readily understood by all employees.

Participation in each week's quarantine program is only possible for certified processors who:

(1) Have written permission on file with the Authority and are on an Authority-controlled document listing current approved quarantine program processors and;

(2) Possess emailed permission granted by the Authority the day before harvest for that one specific quarantine and;

(3) Propose harvesting a quantity of shellfish that meets the Authority established minimum number but does not exceed the maximum allowed number of shellfish of one specific species for that day.

Under no circumstances may any approved processor participate in any quarantine until they possess written (emailed) documentation sent by the Authority before each specific quarantine event.

- The authorization email sent by the Authority shall explicitly state the permissible species that may be harvested by that approved processor.
- The Authority will notify the appropriate law enforcement entity in charge of patrol of shellfish growing areas with a list of participants in that specific day's harvest.
- Persons harvesting a species not authorized for that day's harvest will be subject to seizure of that harvest by the Authority. In addition, the Authority will immediately seize and destroy product

which is improperly tagged, violates any National Shellfish Sanitation Program (NSSP) Model Ordinance regulations, state laws or is from non-authorized participants.

- Co-mingling of species is not allowed to make up an individual lot.

Violation of the terms of this protocol may result in the termination of the participant's future eligibility in the quarantine program, as determined by the Authority.

Prior to being considered for participation in any specific quarantine event, approved processors shall be contacted by the Authority and asked to provide the name of the species they plan to harvest and the quantity they plan on harvesting. Quantities shall be described as approximate total number by species in addition to total number of baskets, containers, bags, etc. with specific weights (if applicable) for those baskets, containers, bags, etc.

Eligible processors should be aware that daily implementation of this program is contingent on marine Biotoxin laboratory availability as well as Authority staffing considerations given staff time necessary to fulfill the requirements of the program.

Regulatory considerations on behalf of the Authority and staffing considerations on behalf of the marine Biotoxin lab necessitate an Authority developed maximum number of samples that could be potentially tested on any given week.

The Authority may implement a lottery, random rotation or similar procedure to ensure a fair distribution of testing opportunities among the eligible processors. It is suggested that the Authority develop this procedure with industry involvement.

Once specific permission is received from the Authority, the processor:

(2) May receive properly tagged shellfish from eligible aquaculturists only as indicated in the Authority's authorization email;

(3) Must upon receipt of shellfish, separate and maintain the shellfish into specific lots [A Lot is defined as shellfish of one species from no more than one day's harvest from a specific zone within a shellfish growing area];

(4) Must place shellfish under proper controls and quarantine; Proper controls and quarantine are defined by bold, clear, warning signage signaling the properly tagged and segregated shellfish within the processor's cooler are under quarantine and must not be moved until Authority permission is obtained pending outcome of laboratory testing. The signage should be such that it is clear to anyone entering the cooler (including facility employees and/or regulatory inspectors) that the affected shellfish are under quarantine. Wrapping of the entire lot with a single bright red or yellow ribbon or equivalent attached to the bold warning sign will

	further reinforce the warning message. (5) Must allow the Authority to take two (2) random samples [minimum of twenty (20) shellfish per each sample] from each lot and deliver to the approved laboratory for approved mouse bioassay; (6) Must hold all shellfish in quarantine at the approved processor's certified facility until receiving official written test result notice from the Authority via email or fax that the shellfish are cleared for sale; (7) Must either return shellfish to aquaculture lease(s) in the zone(s) from where harvested if any sample in a lot is 20 Mouse Units / 100 grams or greater or destroy the shellfish, both activities of which must be witnessed and documented by the Authority; (8) Must cease this activity if any Authority collected red tide cell counts in the specific zone exceeds 200,000 cells per liter of *Karenia brevis*; and (9) Must document all of the requirements listed above in the approved facility HACCP plan. C. If cell counts in all water samples fall to 5,000 cells/L or less Karenia brevis in the entire area, the Authority will collect shellfish meat samples for toxicity testing and the entire Shellfish Harvesting Area will be reopened if results of all samples are <20 MU/100g. I _____ (print name) have received a copy of this quarantine protocol and I agree to abide by all terms and conditions. I understand I am bound by the terms of this agreement during the period of time that I am processing shellfish from a shellfish growing area that is currently in the closed status due to Karenia brevis. _____ _____ Signed Date
13. Public Health Significance	Closures of shellfish growing areas due to Neurotoxic Shellfish Poisoning (NSP) may occur at any time in the Gulf of Mexico and to a lesser degree, the Atlantic coast. Well established procedures for detecting and responding to *Karenia brevis* blooms have safeguarded public health. Clear early warning signs, a cell count action level with a high factor of safety and established sampling networks provide excellent public health protection. A very real impact of *Karenia brevis* blooms is the resulting long-term closures of shellfish growing areas and severe economic impact to commercial shellfish operations. Florida addressed this issue after studying years of water quality samples and mouse bioassay results from shellfish growing areas. Hydrodynamic studies linked to water samples obtained from fixed stations over an extended period of time established clear patterns in distribution of *Karenia brevis*. Working in conjunction with harmful algal bloom researchers, shellfish growing area managers, FDA and industry, Florida developed a NSP quarantine protocol that has resulted in the retention of a shellfish industry in one of the most severely impacted HAB regions of the Gulf while protecting public health as required by the Model Ordinance. An enormous amount of data has been generated and reviewed during the years this protocol has been used. Repeated mouse bioassay testing on shellfish exposed to different levels of *Karenia brevis*

	has provided Florida with sufficient data to refine the protocol into a powerful management tool. Florida's experience pre-quarantine protocol was unfortunate, as several fledgling businesses failed due to repeated NSP closures. It was this economic damage that spurred the aforementioned collaborative effort between leading edge HAB researchers, shellfish growing area managers, FDA and industry. If adopted, shellfish producing states impacted by *Karenia brevis* could reference this protocol in the Guidance Document and use it to effectively manage NSP closures.
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 Task Force I	Recommended referral of Proposal 13-116 to an appropriate committee as determined by the Conference Chairman
Action by 2013 General Assembly	Adopted recommendation of 2013 Task Force I on Proposal 13-116.
Action by FDA May 5, 2014	Concurred with Conference action on Proposal 13-116.
Action by 2015 Biotoxin Committee	Recommended adoption of Proposal 13-116 with substitute language as follows: (4) The plan may include agreements or memoranda of understanding, between the Authority and individual shellfish harvesters or individual shellfish dealers, to allow harvesting in designated parts of a state growing area while other parts of the same ~~the~~ growing area are placed in the closed status. Such controlled harvesting shall be conducted with strict assurances of safety. In state growing areas or designated portions of state growing waters that are closed, the authority may allow for harvesting if an end product testing program is developed and~~, such as by batch release of shellfish lots only after~~ samples of each lot are tested and found to be below the action levels specified in Section C. The program must include at a minimum: i. Establishment of appropriate pre-harvest screening levels; ii. Establishment of appropriate screening and end product testing methods; iii. Establishment of appropriate laboratories/analysts to conduct screening and end product testing methods; iv. Establishment of representative sampling plan for both i. and ii. above; and v. Other controls as necessary to ensure that shellstock are not released prior to meeting all requirements of the program. Should the above amended proposal be adopted by the conference, then the Biotoxin Committee should develop a Guidance Document that includes guidance for development of end-product testing programs to address biotoxins in closed state waters.
Action by 2015 Task	Recommends adoption of Biotoxin Committee recommendation on Proposal 13-

Force I	116.
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 13-116.
Action by 2017 Task Force I	Recommended the Biotoxin Committee should develop a Guidance Document that includes guidance for development of end-product testing programs to address Biotoxins in closed State waters.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 13-116.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 13-116.

Proposal No. **15-109**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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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	The PCOX method was developed to provide a rapid, high throughput chemical

Significance	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.

Proposal No. **15-112**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting

a. ☒ Growing Area
b. ☐ Harvesting/Handling/Distribution
c. ☐ Administrative

Submitter	Executive Board
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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)

5. Approved Methods for Vibrio Enumeration

	Vibrio Indicator Type:	Application: PHP Sample Type: Shucked	Applicatio Reopenin
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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

Submitter	Executive Board
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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	Adopted recommendation of Task Force I on Proposal 15-114.

General Assembly	
Action by FDA January 11, 2016	Concurred with Conference action on Proposal 15-114.
Action by 2017 Laboratory Committee	Recommended referral of Proposal 15-114 to an appropriate committee as determined by the Conference Chair.
Action by 2017 Task Force I	Recommended adoption of Laboratory Committee recommendation on Proposal 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.

Proposal No. **17-100**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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Proposal Subject	Marina Definition
Specific NSSP Guide Reference	Section I Purposes and Definitions B. Definition of Terms (71) Marina
Text of Proposal/ Requested Action	**(71) Marina** means any water area with a structure (docks, basin, floating docks, etc.) which is: (a) Used for docking or otherwise mooring vessels to a dock or pier; and (b) Constructed to provide temporary or permanent docking space for more than ten boats.
Public Health Significance	There has been ever increasing pressure to include mooring areas which are not defined in the Model Ordinance into the Marina Proper; Section II- Chapter IV **@ .05 Marinas**. When the criteria were developed to deal with the classification of Marinas as defined, and the determination of a buffer zone in adjacent waters; mooring areas were purposely not included. It was left to the discretion of the SSCA to determine, classification criteria that could be different from the marina calculations depending on local circumstances and local knowledge. FDA is now interpreting anchors, chains and mooring blocks as "structures "and as such is requiring that mooring areas be treated as Marinas. Structure in the Marina definition means "(docks, basin, floating docks, etc.)" not anchors and chains. There are many different kinds of marinas, some essentially parking lots with no overnight occupancy and others that are destination mooring areas. Some states have outstanding boat pump out programs and large areas, if not the entire state, that are federal No Discharge Areas, in addition to local well enforced no discharge and occupancy regulations or by-laws. SSCAs should be allowed to assess the pollution impact of mooring areas based on actual circumstances and data not just an assumed risk.
Cost Information	NONE, Possible savings to SSCAs.
Action By 2017 Task Force I	Recommended referral of Proposal 17-100 to an appropriate committee as determined by the Conference Chair.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-100.
Action by FDA February 7, 2018	Concurred with Conference action on proposal 17-100 with comments. (See February 7, 2018 FDA response to ISSC Summary of Actions)

Proposal No. **17-103**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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.

Proposal No. **17-106**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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.

Proposal No. **17-108**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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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.

Proposal No. **17-110**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
2. Submitter	J. Michael Hickey Margaret Barette David Fyfe
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10. Proposal Subject	Reconditioning of Recalled Shellfish Implicated in a Norovirus Outbreak
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter II. Risk Assessment & Risk Management @.01 Outbreaks of Shellfish Related Illness.
12. Text of Proposal/ Requested Action	J. Molluscan shellfish product that is recalled as a result of an illness outbreak associated with *V.v.*, *V.p.*, or Norovirus may be reconditioned. 1. Validated reconditioning processes for *V.v.* and *V.p.* include subjecting product to validated PHPs or placing into approved, conditionally approved, conditionally restricted, or restricted growing areas for an appropriate period of time, not less than fourteen (14) days, with appropriate controls and documentation to be determined by the State Shellfish Control Authority (SSCA). 2. Product associated with a Norovirus outbreak may be reconditioned by returning the product, within three (3) days of the recall, to the growing area from which it was harvested for an appropriate period of time. The period of time shall not be less than twenty-one (21) days. The Authority shall ensure appropriate controls and provide documentation of the activity.
13. Public Health Significance	A twenty-one (21) day submergence period is consistent with the amount of time required at Section II. Chapter IV. A. (5) (b) (ii) and C. (2) (c) (iii), Shellstock Growing Areas.
14. Cost Information	No substantial increased cost to SSCAs and to the shellfish industry. would

	constitute a cost saving
Action By 2017 Task Force I	Recommends referral of Proposal 17-115 to an appropriate committee as determined by the Conference Chair.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-114.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-114.

Proposal No. **17-116**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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 frequency with 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.

Proposal No. **17-121**

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	☒ Growing Area ☐ Harvesting/Handling/Distribution ☐ Administrative

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Proposal Subject	Disposal of Human Sewage and Bodily Fluids
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	Chapter VIII. .02 Shellstock Harvesting and Handling D. Disposal of Human Sewage and Bodily Fluids~~from Vessels~~. (1) Human sewage and bodily fluids shall not be discharged overboard from ~~a~~any vehicle or vessel used in the harvesting of shellstock, or from vehicles or vessels which buy shellstock while the vehicles or vessels are in growing areas. (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 vehicle or vessel to contain human sewage and bodily fluids. (3) Portable toilets shall: (a) Be used only for the purpose intended; (b) Be secured while on board and located to prevent contamination of shellstock by spillage or leakage; (c) Be emptied only into a sewage disposal system; (d) Be cleaned before being returned to the vehicle or vessel~~boat~~; and (e) Not be cleaned in equipment used for washing or processing food. (4) Use of other receptacles for sewage disposal may be approved by the Authority if the receptacles are: (a) Constructed of impervious, cleanable materials and have tight fitting lids; (b) Indelibly labeled “Human Waste” in contrasting letters at least three (3) inches in height; and (c) Meet the requirements in Section D. (3). Chapter IX. .01 Conveyances Used to Transport Shellstock to the Original Dealer G. Disposal of Human Sewage and Bodily Fluids (1) Human sewage and bodily fluids shall not be discharged overboard from any vehicle or vessel used in the harvesting of shellstock, or from vehicles or vessels which buy shellstock while the vehicles or vessels are in growing areas. (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 vehicle or vessel to contain human sewage and bodily fluids. Portable toilets shall meet the requirements of VIII. .02. D. (3). Chapter IX. 02 Conveyances Used to Transport Shellstock from Dealer to Dealer C. Disposal of Human Sewage and Bodily Fluids (1) Human sewage and bodily fluids shall not be discharged overboard from any vehicle or vessel used in the harvesting of shellstock, or from vehicles or vessels which buy shellstock while the vehicles or vessels are in growing areas. (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 vehicle or vessel to contain human sewage and bodily fluids. Portable toilets shall meet the requirements of VIII. .02. D. (3).
Public Health Significance	During evaluations, harvesters and certified dealers buying trucks are observed within harvesting areas and aquaculture lease site areas. The vehicles are often there for hours while harvesting, husbandry, and purchasing activities are taking place. In many areas, there are no nearby toilet facilities to accommodate emergency (or non-emergency) needs for toilet facilities to accept human digestive waste or vomit, putting the area at risk of foodborne illness, e.g. norovirus, hepatitis A, etc. The requirement for marine sanitation devices should not only pertain to vessels in order to protect the public health.
Cost Information	~$5.00 for a five (5) gallon bucket with a lid.
Action By 2017 Task Force I	Recommended referral of Proposal 17-121 to an appropriate committee as determined by the Conference Chair.
Action by 2017 General Assembly	Adopted the recommendation of Task Force I on Proposal 17-121.
Action by FDA February 7, 2018	Concurred with Conference action on Proposal 17-121.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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9. Email	Melissa.Abbott@fda.hhs.gov
10. Proposal Subject	Determining Emergency Conditions
11. Specific NSSP Guide Reference	Section I. Purposes and Definitions Section II. Model Ordinance Chapter IV @.03 A.(1)
12. Text of Proposal/ Requested Action	Section I. Purposes and Definitions New Definition: B.(39) **Emergency Conditions** means potential or actual pollution conditions which were not specifically represented in the sanitary survey information used to establish the classification and support the status of a shellfish growing area. Emergency conditions include, but are not limited to, tropical storms, hurricanes, sewage spills, oil spills, poisonous or deleterious substance spills, excessive rainfall, and flooding events. Chapter IV @.03 A.(1): (1) Emergency Conditions. A growing area shall be placed in the closed status under Section @.03A. (5) when ~~pollution conditions exist which were not included in the database used to classify the area~~ emergency conditions exist. The Authority shall: (a) Develop a written emergency conditions protocol defining the thresholds and criteria used to determine if emergency conditions exist, including defining what conditions would trigger a growing area closure, and how to reopen a growing area once the emergency conditions no longer exist. The thresholds and criteria used to determine if emergency conditions exist, shall be based on the potential or actual pollution conditions which were not specifically represented in the sanitary survey information or database used to establish the classification and support the status of a shellfish growing area. These potential or actual pollution conditions may include, but are not limited to, tropical storms, hurricanes, sewage spills, oil spills, poisonous or deleterious substance spills, excessive rainfall, and flooding events; (b) Make a determination within 24 hours of a potential emergency condition event as to whether conditions exceed the established thresholds and criteria defined in the emergency conditions protocol and maintain a written record of the determination assessment; (c) Notify FDA and ISSC of the determination within 24 hours; (d) Once it is determined that an emergency condition exists, ~~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.~~ place the growing area in the closed status; (e) If a determination cannot be made within 24 hours, notify FDA and ISSC and immediately place the growing area in the closed status; (f) If the growing area is closed due to a precautionary closure and a determination is later made that the growing area did not experience emergency conditions based on the established protocol, the area may be immediately re-opened. The determination shall be documented in a written report and included in the sanitary survey for the area; and ~~(e)~~(g) If the growing area is closed due to emergency conditions, prior to re-opening, conduct an assessment of the growing area based on the established protocol and field observations and document the results in a written report to be included in the sanitary survey. Field observations include, but are not limited to, observations of actual or potential pollution sources made via shoreline survey, boat survey, sample collection, and/or analysis of sample results. The assessment shall include documentation of any new pollution sources and their effect on the growing area.
13. Public Health Significance	Current Model Ordinance language in Chapter IV states "If it is determined that an emergency condition or situation exists…", but does not specify the circumstances under which a determination must be made by the Authority. It will not be clear to a state Authority that pollution conditions exist which were not included in the data used to classify a growing area unless the Authority decides to check the data within the sanitary survey and perform an assessment in a situation which has the potential to meet emergency conditions. Not all Authorities do this in all situations that have the potential to meet "Emergency Conditions" under NSSP MO @.03 A.(1), such as excessive rainfall events with higher rainfall totals that what's recorded in the Authority's database. Additionally, the current language for "Emergency Conditions" does not clearly define "pollution conditions" or "the database used to classify the area". The "database" could be referring to the most recent 12 year sanitary survey or to all of the data ever collected for a growing area or to the most recent 30 water quality samples – it is not clear. In some instances, this has led to disagreements between FDA and state Authorities as to when a growing area needs to be closed due to emergency conditions, such as in the event of a tropical storm with rainfall levels or river stage levels which may or may not exceed the levels in the state's database. Since emergency conditions have the potential to significantly impact the water quality of a growing area and could lead to human fecal contamination, petroleum contamination, or poisonous or deleterious substance contamination in the area and possible shellfish-borne illnesses, it is important to clarify the definition of "Emergency Conditions".
14. Cost Information	Minimal Cost

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Conditionally Conforming Laboratory Status
11. 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)
12. 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;
13. Public Health	A technical Laboratory evaluation, as outlined in MO Chapter 1.@.03B.1.b.ii, is

Significance	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.
14. Cost Information	Cost of conducting SLV, MV or Proficiency Participation

Proposal 19-102 was moved to Task Force II as Proposal 19-239

Proposal 19-103 was moved to Task Force II as Proposal 19-240

Proposal 19-104 was moved to Task Force II as Proposal 19-241

Proposal No. 19-105

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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9. Email	Scott.Berbells@doh.wa.gov
10. Proposal Subject	Laboratory approval for sample analysis with no Model Ordinance defined method or action level
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter III. Laboratory @.01 Quality Assurance (A)
12. 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.
13. 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.
14. Cost Information	

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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9. Email	issc@issc.org
10. Proposal Subject	Delete Notification Requirement to Pollution Control Agencies
11. Specific NSSP Guide Reference	Section II Model Ordinance Chapter IV Shellstock Growing Areas @.01
12. Text of Proposal/ Requested Action	**@.01 Sanitary Survey** A. General. (1) The sanitary survey is the written evaluation report of all environmental factors, including actual and potential pollution sources, which have a bearing on water quality in a shellfish growing area. The sanitary survey shall include the data and results of: (a) A shoreline survey; (b) A survey of the microbiological quality of the water. In growing areas adjacent to waste water system discharge (WWSD)s the Authority may utilize male specific coliphage (MSC) results from analysis of shellfish meat samples and the analysis of the data will be included in the sanitary survey report; (c) An evaluation of the effect of any meteorological, hydrodynamic, and geographic characteristics on the growing area; and (d) A determination of the appropriate growing area classification. (2) The sanitary survey shall be periodically updated through the triennial reevaluation and the annual review in accordance with Section C. to assure that data are current and that conditions are unchanged. (3) The documentation supporting each sanitary survey shall be maintained by the Authority. For each growing area, the central file shall include all data, results, and analyses from: (a) The sanitary survey; (b) The triennial reevaluation; and (c) The annual review. ~~(4) Wherever possible, the Authority shall provide the necessary information to Federal, State, or local agencies which have the responsibility to minimize or eliminate pollution sources identified in the sanitary survey.~~ ~~(5)~~(4) The Authority shall maintain a current comprehensive, itemized list of all growing areas, including maps showing the boundaries and classification of each shellstock growing area.
13. Public Health Significance	This requirement does not have public health significance.

14. Cost Information	

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Determining shoreline survey area.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.01 Sanitary Survey D.(1) and (2)(a).
12. Text of Proposal/ Requested Action	(1) In the shoreline survey for each growing area, the Authority shall: (f) Conduct an in-field assessment of pollution sources which may include: (i) A drive-through survey; (ii) Observations made during sample collection; and/or (iii) Information from other sources. (2) The Authority shall assure that the shoreline survey meets the following minimum requirements: (a) The boundaries, ~~based on the area topography,~~ of each shoreline survey area are determined by an ~~in-field~~ investigation ~~which identifies only the properties with the potential to impact the shellfish waters~~ that shall include, but not limited to, all properties with the potential to impact the shellstock growing area based on area topography, as well as field observations, and other sources of information;
13. Public Health Significance	The minimum requirements of the shoreline survey include an investigation and evaluation of pollution sources by trained, qualified, personnel. The investigation must be accomplished through an in-field assessment where the surveyor identifies actual and potential sources of pollution that might influence water quality. Given the technology available today, there are mutltiple options for identifing properties with the potential to impact growing areas. The Authority can define the shoreline survey area boundry by using various data resources such as geoprapohic information such as on-line maps. Using the term “only” as it is used in the existing language is confusing and, if taken literally, limiting. Example: One property two miles from the growing contains a large wastewater treatment plant that has the potential to impact shellfish waters. Another property one- and one-half miles from the growing area between that growing area and the property with the wastewater treatment plant on it has no identifiable pollution sources on it so that it does not have potential to impact shellfish waters. If the shoreline survey area is defined as a single area that includes the property with the

	wastewater treatment plant, it will also include the property with no identifiable pollution sources on it. Thus, it will not be an area that has "only" the properties with potential to impact the shellfish waters in it.
14. Cost Information	No cost.

Proposal No. 19-108

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Aquaculture Seed Shellstock
11. Specific NSSP Guide Reference	Section II Model Ordinance, Chapter VI. Shellfish Aquaculture, Requirements of the Authority @.02
12. 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.
13. 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)
14. Cost Information	Proposal would not impact the enforcement costs for the authority and would simplify management for growers.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Offshore State Water classification requirements
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.02
12. Text of Proposal/ Requested Action	**@.02 Microbiological Standards** Note: The NSSP allows for a growing area to be classified using either a total or fecal coliform standard. The NSSP further allows the application of either standard to different water bodies within the State. The NSSP also allows for two (2) sample collection strategies for the application of the total or fecal coliform standard: adverse pollution condition and systematic random sampling. The 1992 Task Force II recommended that this portion of the Ordinance be codified in two (2) ways: a total coliform strategy and a fecal coliform strategy so that the State may choose sampling plans on a growing area basis. Within each strategy, provisions would appear for use of both systematic and adverse pollution condition sample collection. The Ordinance has been recodified in this manner. For maximum flexibility, an Authority may wish to adopt the use of both standards and both sampling strategies for each standard. This codification represents the fecal coliform standards. Additionally, the Authority may choose to use MSC sample data in conjunction with total or fecal coliform data to evaluate areas impacted by WWSD. A. General. Either the total coliform or fecal coliform standard shall be applied to a growing area. The Authority may utilize MSC data in conjunction with bacteriological data to evaluate WWSD impacts on shellfish growing areas. B. Water Sample Stations. The Authority shall assure that the number and location of sampling stations is adequate to effectively evaluate all pollution sources. C. Exceptions. (1) Except for growing areas classified as prohibited, in growing areas where there are pollution sources having an impact on the water quality, a minimum of thirty (30) samples, collected under various environmental conditions, shall be required to classify any growing area not previously classified under Section @.03. (2) Except for growing areas classified as prohibited or when the systematic random sampling standard is applied, in growing areas where there are no pollution sources having an impact on the water quality, a minimum of fifteen (15) samples shall be required to classify any growing area not previously classified under Section @.03.

	(3) Except for offshore state waters where a sanitary survey shows that there are no pollution sources that will impact the microbiological quality of the water. Offshore state waters are classified as approved.
13. Public Health Significance	State waters extend 9 miles off shore of the State of Florida. If a sanitary survey can show there are no pollution impacts (ie. Rivers, WWTPs discharges) to proposed areas for aquaculture the required 30 samples to classify should not be required.
14. Cost Information	This would reduce the cost and burden to state authorities having to sample waters that are far removed from any potential pollution sources.

Proposal No. 19-110

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Point source approved standard station locations.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.02 Microbiological Standards E.(3)(c).
12. Text of Proposal/ Requested Action	(c) 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.
13. 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.
14. Cost Information	No cost.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Allowing the use of the SRS method in areas impacted by point sources
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.02E; Chapter IV. Shellstock Growing Areas @.02F; Chapter IV. Shellstock Growing Areas @.02F(2)(b); Chapter IV Shellstock Growing Areas @.02G; and Chapter IV. Shellstock Growing Areas @.02H
12. Text of Proposal/ Requested Action	Chapter IV, @.02 E. Standard for the Approved Classification of Growing Areas ~~Affected by Point Sources~~ when Evaluated for Adverse Pollution Conditions. Chapter IV, @.02 F. Standard for the Approved Classification of Growing Areas ~~Affected by Nonpoint Sources~~ when Evaluated for Nonpoint Sources. (1) Exception. If the tidal stage increases the fecal coliform concentration, the authority shall use sample results collected during that tidal stage to classify the area. (2) Pollution Sources. Growing areas shall be~~:~~ ~~(a)~~ Impacted only by randomly occurring, intermittent events~~; and~~ ~~(b) Not impacted by discharges from sewage treatment facilities or combined sewer overflows.~~ Chapter IV, @.02 G. Standard for the Restricted Classification of Growing Areas ~~Affected by Point Sources~~ when Evaluated for Adverse Pollution Conditions and Used as a Shellstock Source for Shellstock Depuration. Chapter IV, @.02 H. Standard for the Restricted Classification of Growing Areas ~~Affected by Nonpoint Sources~~ when Evaluated for Nonpoint Sources and Used as a Shellstock Source for Shellstock Depuration
13. Public Health Significance	This proposed amendment to Chapter IV, @.02 updates the conditions under which

	the APC and SRS methods may be used. The proposal allows the use of the SRS method in areas impacted by discharges from sewage treatment facilities or combined sewage overflows where marine water stations have been placed to monitor nonpoint pollution. The intent of this proposal is to use the sampling methodology and statistical analysis most acceptable for the purpose of the marine water sampling station. If the station is placed to monitor nonpoint pollution, the SRS methodology should be used. If the station is placed to monitor adverse pollution conditions, the APC methodology should be used. In Washington state, marine water stations located in Conditionally Approved areas impacted by wastewater treatment plants are placed to monitor nonpoint pollution from the surrounding upland areas. The APC criterion is used to sample and evaluate data from these stations with the adverse condition defined as an upset at the treatment plant. Many wastewater treatment plants are high performing and upset conditions occur infrequently. The infrequency of the impact to the growing area does not allow for the intended use of the APC sampling strategy. Hydrographic studies and dilution analyses are more appropriate for the evaluation of the impact area around high performing wastewater treatment plants.
14. Cost Information	No impact

Proposal No. 19-112

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Nonpoint source approved standard station locations.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.02 Microbiological Standards F.(6)(b)(i).
12. 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;
13. 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.
14. Cost Information	No cost.

Proposal No. 19-113

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Authorizing unclassified areas and multiple classifications for single area.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.03 Growing Area Classification A.(2).
12. Text of Proposal/ Requested Action	(2) Classification of ~~All~~ Growing Areas. ~~All~~ Each growing ~~areas~~area which: (a) ~~Are~~ Is not subjected to a sanitary survey every twelve (12) years shall be classified as prohibited or, if unclassified, shall be treated as prohibited for NSSP purposes; or ~~(b) Have a sewage treatment plant outfall or other point source outfall of public health significance within or adjacent to the growing area shall have an area in the prohibited classification established adjacent to the outfall in accordance with Section E. Prohibited Classification; and~~ (b~~e~~) ~~Are~~ Is subjected to a sanitary survey shall be correctly classified based on the twelve (12) year sanitary survey, and its most recent triennial or annual reevaluation when available, as ~~only~~ one or more~~(1)~~ of the following: (i) Approved; (ii) Conditionally Approved; (iii) Restricted; (iv) Conditionally Restricted; and/or (v) Prohibited.
13. Public Health Significance	There is no reason to require that all growing areas be classified if the Authority is required to treat unclassified areas as prohibited areas. The current Section II. Chapter IV.@.03A.(2)(b) language is unnecessary. Requiring that each growing area be characterized by only one classification is not realistic and does not reflect common practice. There are many circumstances in which one growing area contains several classifications. Example: A 10 square mile growing area is generally classified as approved. However, there is a marina in it, so some waters associated with that marina are classified as prohibited and restricted. There is a business with a 5,000 gallon per day wastewater treatment system discharging along the shoreline so there is a prohibited zone adjacent to that point source. That circumstance literally represents violation of Chapter IV.@.03A.(2)(c) as that requirement now reads because there are multiple classifications within a single growing area.

14. Cost Information	No cost.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
2. Submitter	US Food & Drug Administration (FDA)
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9. Email	Melissa.Abbott@fda.hhs.gov
10. Proposal Subject	Emergency Conditions re-opening studies.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.03 Growing Area Classification A.(5)(c)(i).
12. Text of Proposal/ Requested Action	(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. When pathogens are of concern, ~~S~~studies establishing sufficient elapsed time shall document the interval necessary for reduction of ~~contaminant~~ coliform levels in the shellstock to pre-closure levels. ~~In addressing pathogen concerns, the~~ Such coliform studiesmay establish criteria for reopening based on coliform levels in the water. When poisonous or deleterious substances are the concern, studies 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; or
13. Public Health Significance	National Shellfish Sanitation Program Guide for the Control of Molluscan Shellfish, Section IV Guidance Documents, Chapter II Growing Areas, .08 Action Levels, Tolerances and Guidance Levels for Poisonous or Deleterious Substances in Seafood contains target levels for many poisonous or deleterious substances. Target levels for other substances can be established through risk evaluation. The 2010 Deepwater Horizon crisis provides an example of how emergency conditions involving poisonous or deleterious substances are addressed in practice. Levels of concern were established through risk evaluation then areas were re-opened based on determining that contaminant levels were below levels of concern rather than based on comparisons between pre and post closure levels.
14. Cost Information	Cost would potentially be reduced because studies to compare post closure levels of poisonous or deleterious substances to pre closure levels would no longer be required.

Proposal No. 19-115

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Emergency Conditions/closed status to reflect Chapter II use of harvest area
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification A. General (1) and (5)
12. Text of Proposal/ Requested Action	@.03 Growing Area Classification A. 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. (c) 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:
13. 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.
14. 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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. 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.
11. Specific NSSP Guide Reference	Section II, Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification A. (5) (b).
12. 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….
13. 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.
14. 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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. X Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Shellfish cleansing studies
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification. C. Conditional Classifications. (2) (c) (iii)
12. Text of Proposal/ Requested Action	(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. The study may establish criteria for reopening based on coliform levels in the water. If the conditional management plan is based on effects of non-point sources of pollution such as rain events and /or storm water runoff, an area can be reopened 48 hours after the water quality has met acceptable classification criteria as long as shellstock are actively feeding.
13. Public Health Significance	There are a number of problems related to the current M. O. language." There is no guidance or criteria in the Guide concerning what constitutes an adequate study. There are a number of study related questions: 1) How many shellfish samples of each species of shellfish and sampling stations (locations) are needed in a growing area; 2) Are studies required in every conditional area? 3) can information obtained in one growing area be applied to shellstock in another growing area? 4) The first sentence at (iii) refers "*to reducing pathogens...to acceptable levels*", what are acceptable levels of pathogens. The second sentence at (iii) refers to *reduction of coliform levels in shellstock to pre-closure levels*. Pre-closure levels in shellstock can be variable both temporally and spatially. Thus the concept of reducing coliforms to pre-closure levels is at best ambiguous. In order to obtain the required data, there is a sampling and laboratory burden. This requires time consuming shellstock sampling during open periods and again after pollution events over the year as well as increased laboratory effort to establish a data base. Shellfish samples require two lab days thus reducing lab capacity to handle water samples. In the 1980's and early 1990's Massachusetts and other states sampled shellstock one or two days after water in Conditionally Approved areas reached the criteria for an Approved classification to ensure that the shellstock was well below the then existing NSSP 230 FC market standard. Usually 150 FC or less was considered adequate to reopen because there was no actual coliform harvest standard and it made sense to only allow harvest well below the market standard. This reduction was accomplished within two days or less of the water quality returning to

	acceptable levels. This approach compared coliform levels in shellfish after water quality reached acceptable levels to an existing standard. When this policy was established, it was endorsed by the FDA Shellfish Specialist. \Shellstock can accumulate bacteria up to 100 times the level in the water. In theory shellstock in water at geometric mean of 10 FC per 100 ml could accumulate FC bacteria to a level of 1000 FC per 100 g. Thus opening an area at a level below the former 230 FC market standard would seem appropriate. Two day purging time is well established. Literature supports elimination of greater than 95% of FC bacteria from shellstock in less than 24 hours including NSSP workshop studies. Temperature is the most important factor affecting elimination of bacteria because it governs shellfish feeding activity. Naturally contaminated shellfish can eliminate fecal coliform levels in 48 hours to levels below most market standards over a range of environmental conditions (Perkins, et al, 1979). Other studies show that soft –shelled clams at MPN 10,000 FC /100 g reduced to values below 50 in 48 hours (Arcisz, et al, 1955) and oysters at MPN 39,000FC/1000g can purge to values below 50 in 48 hours.
14. Cost Information	Could produce significant savings to state shellfish classification programs.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
2. Submitter	US Food & Drug Administration (FDA)
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10. Proposal Subject	Conditional areas not based on predicting microbiological indicator levels.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas Section @.03 Growing Area Classification C.(1).
12. Text of Proposal/ Requested Action	(1) Survey Required. The sanitary survey meets the following criteria: (a) ~~The area will be in the open status of the conditional classification for a reasonable period of time.~~ The factors determining the~~is~~ period the growing area is in open status are known and ~~, are~~ predictable~~, and are not so complex as to preclude a reasonable management approach~~; (b) Each potential source of pollution that may adversely affect the growing area is evaluated; (c) When conditional management is based at least in part on predicted changes in microbiological water quality,~~M~~microbiological water quality correlates with environmental conditions or other factors affecting the distribution of pollutants into the growing area; and (d) For Authorities utilizing MSC meat sample data, when conditional management is based at least in part on predicted changes in MSC levels, those~~is~~ data correlate~~s~~ with environmental conditions or other factors affecting the distribution and persistence of viral contaminants into the growing area.
13. Public Health Significance	Not all conditional management is based on predicted changes in microbiological water quality. Conditional management can be based, for example, on the operation of a wastewater treatment system that has never failed. In such a circumstance, demonstrating correlation with environmental conditions or other factors may play no role. The plan can be based completely on other means of predicting the impact of plant failure. Conditional management can also be based on changes in marina occupancy. Similarly, the Authority may use MSC data in some way to support conditional management without demonstrating correlation between MSC levels in shellfish tissues and environmental conditions or other factors.
14. Cost Information	No cost.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Reduced marine water sampling in conditionally approved areas impacted by point sources
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification C3. Reevaluation of Conditional Classification(b)(ii)
12. Text of Proposal/ Requested Action	Section II Model Ordinance Chapter IV Shellstock Growing Area @.03 Growing Area Classification C3. Reevaluation of Conditional Classification (b) Water Sample Collection (ii) When the conditional management plan is based on the operation and performance of a WWSD (s); combined sewer overflows(s); or other point sources of pollution, monthly water samples are required when the growing area is in the open status of its conditional classification except when: (a) Hydrographic or dilution analysis has been completed to determine the impact of a performance failure; and (b) Communication requirements are documented and the WWSD operator provides immediate notification to the Shellfish Authority during a performance failure.
13. Public Health Significance	This proposed amendment to Chapter IV, @.03C3(b)(ii) updates the requirements related to the monthly sampling requirement in Conditionally Approved areas classified based on the operation and performance of a WWSD, combined sewer overflow, or other point source. The proposal allows the Shellfish Authority to reduce the number of marine water samples in the area from monthly to five or six times per year, based on the sampling methodology used, if additional studies and appropriate communication channels have been developed. Based on the high performance of many treatment plants, upset conditions occur infrequently and are not evaluated through the placement of permanent marine water sampling stations. Dye and drogue studies coupled with computer modelling are commonly used to determine the potential impact from a point source of pollution on the growing area and are used to calculate the dilution available throughout the area. In Washington state, all NPDES permits issued to wastewater treatment plants contain requirements for operators to provide immediate notification to the Shellfish Authority during upset conditions. Failure of the operator to respond in a

	timely fashion could result in a significant penalty. Upset conditions impacting Conditionally Approved shellfish growing areas in Washington State are infrequent; however, during each event the Shellfish Authority has been immediately informed. The high performance of current treatment plants, effective use of hydrographic and dilution analysis, and immediate communication during upset conditions provide more effective and efficient protection of public health in Conditionally Approved areas impacted by point sources. Upset conditions are infrequent and random which can make monthly sampling inefficient and ineffective at evaluating impacts from the point source.
14. Cost Information	The reduced sampling option would be a cost savings for the Shellfish Authority.

Proposal No. 19-120

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Classification of Federal Waters
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Growing Area Classification F.
12. Text of Proposal/ Requested Action	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, or marine biotoxin hazards) and involve commercial shellfish resources. Should FDA allow harvesting in Federal waters with known marine biotoxin hazards, the FDA will classify the harvest area in a manner equivalent to the requirements of Model Ordinance Chapter IV.
13. Public Health Significance	The FDA has taken the position that all Federal waters are approved unless closed. Currently shellfish harvesting is being allowed in areas with known marine biotoxin hazards. To address these hazards, harvesting restrictions are being required without the designation of appropriate harvesting classification. Currently the Model Ordinance does not include any restrictions for approved areas. Shellfish harvesting areas that have been closed are considered prohibited and harvesting for human consumpltion purposes ia not allowed. If the FDA wants to continue to allow harvesting in Federal waters with restrictions, appropriate classification should be designated.
14. Cost Information	

Proposal No. 19-121

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	*Karenia brevis*
11. Specific NSSP Guide Reference	Section II Model Ordinance Chapter IV. Shellstock Growing Areas @.04
12. Text of Proposal/ Requested Action	Chapter IV. Shellstock Growing Areas @.04 C. Closed Status of Growing Areas. A growing area, or portion(s) thereof as provided in Section A.(4), shall be placed in the closed status for the taking of shellstock when the Authority determines that the number of toxin-forming organisms in the growing waters and/or the level of biotoxin present in shellfish meats is sufficient to cause a health risk. The closed status shall be established based on the following criteria: (a) PSP - 80 µg saxitoxin equivalents/100 grams (b) NSP - 5,000 cells/L *(Karenia brevis)* or 20 MU/100 grams (0.8 mg brevetoxin-2 equivalents/kg) (c) AZP - 0.16 mg azaspiracid-1 (AZA-1) equivalents/kg (0.16 ppm) (d) DSP – 0.16 mg okadaic acid (OA) equivalents/kg (0.16 ppm) (e) ASP – 2 mg domoic acid/100 grams (20 ppm)
13. Public Health Significance	The 5,000 cell count standard applies to *Karenia brevis* only
14. Cost Information	

Proposal No. 19-122

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Use of "growing area" rather than "harvest area" in Patrol requirements language.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter VIII. Control of Shellfish Harvesting @.01 Control of Shellstock Growing Areas A.(2)(d), A.(3)(b), B.(2).
12. Text of Proposal/ Requested Action	A. General. (1) The Authority shall maintain an effective program to control shellstock growing areas and to assure that shellstock are harvested only: (a) From areas in an open status; and (b) With approval from areas classified as restricted, conditionally restricted, or prohibited, or in the closed status of the approved or conditionally approved classification. (2) This program shall include: (a) The patrol of growing areas; (b) The licensing of harvesters; (c) Enforceable legal penalties sufficient to encourage compliance; and (d) Appropriate identification of growing~~harvest~~ areas and/or portions of growing areas where shellstock harvest is not allowed. (3) At the time of issuance or renewal of a harvester's license or a dealer's certification, or an annual mail out to all licensed shellfish harvesters, the Authority shall provide each harvester or dealer with: (a) Information which explains the public health risk associated with illegal harvesting shellstock in areas classified as restricted, conditionally restricted, or prohibited or in the closed status; and (b) When requested, a current, comprehensive, itemized listing of all growing~~harvest~~ areas including their geographic boundaries and their classification. B. Patrol of Growing Areas. (1) The Authority shall assure that shellstock are harvested only as provided in this Chapter. (2) The Authority shall patrol growing~~harvest~~ areas classified as restricted, conditionally restricted, or prohibited, or conditionally approved and approved when in the closed status at sufficient intervals to deter illegal harvesting...
13. Public Health	The NSSP Guide for the Control of Molluscan Shellfish contains definitions for

Significance	"Harvest Area" and "Growing Area." "Growing Area" is the more appropriate term for the indicated locations.
14. Cost Information	No cost.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Marine Biotoxin Control - Public Health Reasons
11. Specific NSSP Guide Reference	Section III. Public Health Reasons and Explanations, Model Ordinance Chapter IV. Shellstock Growing Areas, @.04
12. 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 to 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	ASP is characterized by gastrointestinal disorders (vomiting, diarrhea, abdominal pain) and neurological problems

Course	(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.htm
Diarrhetic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0e.htm
Neurotoxic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0o.htm
Amnesic Shellfish Poisoning	http://www.fao.org/3/y5486e/y5486e0n.htm
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 whelk, snails, and, in some cases, even lobster and octopus), may become toxic in areas where the source algae are present.

References

Deeds, J.R., & Landsberg, J.H., Etheridge, S.M., Pitcher, G.C., Longan, S.W. (2008). Non-traditional vectors for paralytic shellfish poisoning. *Marine Drugs, 6(2)*, 308-348. Retrieved from https://doi.org/10.3390/md6020308.

Degrasse, S., & Rivera, V., Roach, J., White, K., Callahan, J., Couture, D., Simone K., Peredy, T., Poli, M. (2014). Paralytic shellfish toxins in clinical matrice extension of AOAC official method 2005.06 to human urine and serum and application to a 2007 case study in Maine. *Deep Sea Research Part II: Topical Studies in Oceanography*, 103, 368-375. Retrieved from https://doi.org/10.1016/j.dsr2.2012.08.001.

Food and Agriculture Organization of the United Nations. (2015) Codex Alimentar Standard for Live and Raw Bivalve Molluscs Codex Stan 292-2008. Retrie from http://www.fao.org/fao-who-codexalimentarius/codex-texts/all-standards/en/

Food and Agriculture Organization of the United Nations. (2004). FAO Food and Nutrition Papers, 80 - Marine Biotoxins. Retrieved from http://www.fao.org/3/y5486e/y5486e00.htm

Joint Sanitation Seminar on North Pacific Clams Juneau, A., Felsing, W. A. (Willi August)., United States. Public Health Service., Alaska. Dept. of Health an Welfare. (1966). *Proceedings of Joint Sanitation Seminar on North Pacific Clams*. Washington, D.C.: For sale by the Supt. of Docs., G.P.O.. Retrieve from https://babel.hathitrust.org/cgi/pt?id=pur1.32754081175147&view=1up&se

Klontz, K.C., & Abraham, A., Plakas, S., Dickey, R. (2009). Mussel-associated azaspiracid intoxication in the United States. *Annals of Internal Medicine*, 150(5), 361. Retrieved from https://www.researchgate.net/publication/24174858_Mussel-

Associated_Azaspiracid_Intoxication_in_the_United_States

Liston, J. (1994). Association of *Vibrionaceae*, natural toxins, and parasites with fe indicators, p. 215-216. In Hackney, C.R. and M.D. Pierson (eds.). *Environmental Indicators and Shellfish Safety*. Chapman and Hall, New Yo NY.

Lloyd, J.K., & Duchin, J., Borchert, J., Quintana, H.F., Robertson, A. (2013). Diarrh Shellfish Poisoning, Washington, USA, 2011. Emerging Infectious Diseases 19(8), 1314-1316. Retrieved from https://doi.org/10.3201/eid1908.121824.

Marsden I.D., & Contreras, A.M., MacKenzie, L., Munro, M.H.G. (2015). A comparison of the physiological responses, behaviour and biotransformatic of paralytic shellfish poisoning toxins in a surf-clam (*Paphies donacina*) a the green-lipped mussel (*Perna canaliculus*). *Marine and Freshwater Research*, 67, 1163-1174. Retrieved from http://www.publish.csiro.au/mf/MF14374

McCabe, R.M., & Hickey, B.M., Kudela, R.M., Lefebvre, K.A., Adams, N.G., Bill B.D., Gulland, F.M.D., Thomson, R.E., Cochlan, W.P., Trainer, V.L. (201 An unprecedented coastwide toxic algal bloom linked to anomalous ocean conditions. *Geophysical Research Letters*, 43(19), 10,366–10,376. Retriev from https://DOI.org/10.1002/2016GL070023.

Morris, P.D., & Campbell, D.S., Taylor, T.J., Freeman, J.I. (1991). Clinical and epidemiological features of neurotoxic shellfish poisoning in North Carolir *American Journal of Public Health*, 81(4), 471-474. Retrieved from: https://DOI.org/10.2105/ajph.81.4.471.

National Shellfish Sanitation Workshop., United States. Shellfish Sanitation Branch. (1964). *Proceedings - National Shellfish Sanitation Workshop.* [Washington]: U.S. Dept. of Health, Education, and Welfare, Public Health Service, Food and Drug Administration, Shellfish Sanitation Branch. Retrieved from https://catalog.hathitrust.org/Record/006685147

Perl, T.M., & Bedard, L., Kosatsky, T., Hockin, J.C., Todd, E.C.D., NcNutt, L.A., Remis, R.S. (1990). Amnesic shellfish poisoning: a new clinical syndrome due to domoic acid. In: Hynie, I., Todd, E.C.D., editors. Proceedings of a symposium, domoic acid toxicity. Canada Disease Weekly Report; Ottawa, Ontario. Pp. 7-8.

Prakash, A., & Medcof, J.C., Tennant, A.D. (1971). Paralytic shellfish poisoning i eastern Canada. Bulletin 177, Fisheries Research Board of Canada. Ottawa, Canada. Retrieved from http://dfo-mpo.gc.ca/library/1498.pdf.

Pulido, O.M. (2008). Domoic acid toxicologic pathology: a review. *Marine Drugs, 6(2)*, 180-219. Retrieved from https://doi.org/10.3390/md20080010.

Quayle, D.B. (1969). Paralytic shellfish poisoning in British Columbia. Bulletin 168, Fisheries Research Board of Canada. Ottawa, Canada.

Schwalm, D.J. (1973). The 1972 PSP outbreak in New England. FDA Report, Boston, MA. U.S. Food and Drug Administration, Washington, D.C.

Tong, M., & Smith, J.L., Richlen, M.L., Steidinger, K., Kulis, D., Fux, E., Anderson, D.M. (2014) Characterization and comparison of toxin-producing isolates of *Dinophysis acuminata* from New England and

Canada. *Journal of Phycology*, 51(1), 66-81. Retrieved from https://www.researchgate.net/publication/267340694_Characterization_and_comparison_of_toxin-producing_isolates_of_Dinophysis_acuminata_from_New_England_and_Canada .

Trainer, V.L., & Moore, L., Bill, B.D., Adams, N.G., Harrington, N., Borchert, J., da Silva, D.A.M., Eberhard, B.T.L. (2013). Diarrhetic shellfish toxins and other lipophilic toxins of human health concern in Washington State. *Marine Drugs*, 11, 1815–1835. Retrieved from https://doi.org/10.3390/md11061815.

Twiner, M.J., & Bottein Dechraoui, M.Y., Wang, Z., Mikulski, C.M., Henry, M.S., Pierce, R.H., Doucette, G.J. (2007). Extraction and analysis of lipophilic brevetoxins from the red tide dinoflagellate *Karenia brevis*. *Analytical Biochemistry*, 369(1), 128-135. Retrieved from https://DOI.org/10.1016/j.ab.2007.06.031.

Twiner, M.J., & Rehmann, N., Hess, P., Doucette G.J. (2008). Azaspiracid shellfish poisoning: a review on the chemistry, ecology, and toxicology with an emphasis on human health impacts. *Marine Drugs*, *6(2)*, 39-72. Retrieved from https://doi.org/10.3390/md6020039.

Uchida, H., & Watanabe, R., Matsushima, R., Oikawa, H., Nagai, S., Kamiyama, T., Baba, K., Miyazono, A., Kosada, Y., Kaga, S., Matsuyama, Y., Suzuki, T. (2018). Toxin profiles of okadaic acid analogues and other lipophilic toxins in Dinophysis from Japanese Coastal Waters. Toxins (Basel). 10(11), 457. Retrieved from https://doi.org/10.3390/toxins10110457 .

US Center for Disease Control. (1973). Shellfish poisoning - Florida. *Morbidity Mortality Weekly Report*, 22(48), 397-398. Retrieved from https://stacks.cdc.gov/view/cdc/1843

US Food and Drug Administration. (1997). Poisonous or Deleterious Substances Food. *Federal Register*, 42(190), 52814-52819.

US Food and Drug Administration. (2000). Guidance for Industry: Action Levels fo Poisonous or Deleterious Substances in Human Food and Animal Feed. Retrieved from https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-action-levels-poisonous-or-deleterio substances-human-food-and-animal-feed.

US Food and Drug Administration. (2011). Fish and Fishery Products Hazards and Controls Guidance 4th Edition. Retrieved from https://www.fda.gov/food/seafood-guidance-documents-regulatory-information/fish-and-fishery-products-hazards-and-controls-guidance-4th-edition

US Public Health Service (PHS). (1958). Proceedings: 1957 Conference on Shellfish Poison. U.S. PHS, Washington, D.C. 125 pages. Retrieved from https://babel.hathitrust.org/cgi/pt?id=uc1.31822005678131&view=1up&seq

Watkins, S.M., & Reich, A., Fleming, L.E., Hammond, R. (2008). Neurotoxic shellfish poisoning. *Marine Drugs*, *6(3)*, 431-455. Retrieved from: https://doi.org/10.3390/md6030431.

Wekell, J.C., & Hurst, J., Lefebvre, K.A. (2004). The origin of the regulatory limits f PSP and ASP toxins in shellfish. *Journal of Shellfish Research*, 23(3), 927-9 Retrieved from: https://www.researchgate.net/publication/285809374_The_origin_of_the_re atory_limits_for_PSP_and_ASP_toxins_in_shellfish

Wiese, M., & D'Agostino, P.M., Mihali, T.K., Moffitt, M.C., Neilan, B.A. (2010). Neurotoxic alkaloids: saxitoxin and its analogs. *Marine Drugs*, *8(7)*, 2185-2211. Retrieved from https://doi.org/10.3390/md8072185.

~~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 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.~~
13. 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.
14. Cost Information	None

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Marine Biotoxin Control – Guidance Document
11. Specific NSSP Guide Reference	Section IV Guidance Documents Chapter II. Growing Areas Chapter IV. Shellstock Growing Areas .02
12. 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 h 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 v 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 dangerous 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 fluctuations coastal phytoplankton populations due to the influence of meteorological and 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 bloom 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, Chap 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 of 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 defining 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, authority 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 phytoplankte 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 the 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 v 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, wh 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 exce~~
~~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 vary 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 exceed 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 are 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 gathe 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. Channels 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. T 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 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 promptly identify which shellfish produ 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 advar 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**~~

	1. Center for Disease Control (a). 1973. Shellfish Poisoning - Florida. *Morbid. Mortal. Weekly Rep*.22(48):397-398. 2. Center For Disease Control (b). 1973. Neurotoxic Shellfish Poisoning - Florida. *Morbid. Mortal.Weekly Rep*. 22(48):397-398. 3. Felsing, W.A., Jr. 1966. Proceedings of Joint Seminar on North Pacific Cla September 24-25,1965. U.S. Public Health Service, Washington, D.C. 4. Food and Drug Administration. 1977. Poisonous or Deleterious Substances Food. *FederalRegister* 42(190):52814-52819. 5. Food and Drug Administration. 1985. Action Levels For Poisonous or Deleterious Substances in Human Food and Animal Feed. U.S. Department of Health and Human Services, Public Health Service, Washington, D.C. 20204. 1 pages. 6. Gordon, K., M.D., *et al*. 1973. Shellfish Poisoning. *Morbid. Mortal. Weekly Rep*. 22, (48):397-398. 7. Liston, J. 1994. Association of *Vibrionaceae*, natural toxins, and parasites v fecal indicators, p.215-216. In Hackney, C.R. and M.D. Pierson (eds.), *Environmental Indicators and Shellfish Safety*. Chapman and Hall, New York, 8. Prakash, A., J.C. Medcof, and A. D. Tennant. 1971. Paralytic shellfish poisoning in easternCanada. Bulletin 177, Fisheries Research Board of Canada. Ottawa, Canada. 9. Quayle, D.B. 1969. Paralytic shellfish poisoning in British Columbia. Bulle 168, FisheriesResearch Board of Canada. Ottawa, Canada. 10. Schwalm, D.J. 1973. The 1972 PSP outbreak in New England. FDA Report Boston, MA. U.S.Food and Drug Administration, Washington, D.C. 11. U.S. Public Health Service (PHS). 1958. Proceedings: 1957 Conference on Shellfish Poison. U.S.PHS, Washington, D.C. 125 pages. 12. Wilt, D.S. (ed). 1974. Proceedings of Eighth National Shellfish Sanitation Workshop. January 16-18. New Orleans, LA. National Technical Information Services (PB8-6 236916/AS), U.S. Dept. of Commerce, Springfield, VA. 158 p
13. 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.
14. Cost Information	None

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☐ Growing Area b. ☒ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	*Karenia brevis* Guidance
11. Specific NSSP Guide Reference	Section IV Guidance Documents – Chapter II. Growing Areas
12. Text of Proposal/ Requested Action	**.02 Guidance for Developing Marine Biotoxin Plans** Introduction Shellfish are filter… There are a… There are five… Both *Alexandrium* and… The minimum concentration… The NSSP Model… In shellfish growing… 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, or when the cell counts for ~~members of the genus~~ *Karenia brevis* in the water column equal or exceed 5,000 cells per liter of water.
13. Public Health Significance	The 5,000 cell count standard applies to *Karenia brevis* only
14. Cost Information	

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting

1. a. ☒ Growing Area
   b. ☐ Harvesting/Handling/Distribution
   c. ☐ Administrative

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10. Proposal Subject	MPN-Real-Time PCR for Enumeration of Vibrio vulnificus in Oysters
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas .14 Approved NSSP Laboratory Tests.

12. Text of Proposal/ Requested Action

5. Approved Methods for Vibrio Enumeration

	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**	
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**	

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 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 the vvh gene for total *V. vulnificus* as described in Kinsey et al., 2015.
13. Public Health Significance	This MPN-real-time PCR method provides results in as little as 24 h from receipt of sample. The current NSSP methods for enumeration of *Vv* have limitations: the traditional MPN requires a minimum of 3 days and the SYBR Green PCR is only validated on an instrument platform which is no longer supported by the manufacturer. This method provides an additional option for laboratories to maintain the same level of testing as has been maintained in the program.
14. Cost Information	This method costs ~$100 per sample for laboratory consumables, supplies, and reagents. Most equipment needed for testing is standard microbiology equipment, but purchase of a heat block (~$400) and/or centrifuge (~$2,500) may be necessary. Purchase of a real-time PCR instrument will be required ($30,000-$45,000). Additional costs for a laboratory would vary based on their operational overhead and labor.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Modification of the MARBIONC Brevetoxin ELISA Standard Operating Procedures
11. Specific NSSP Guide Reference	Section IV. Guidance Documents Chapter II. Growing Areas. 14 Approved NSSP Laboratory Tests 4. Approved Limited Use Methods for Marine Biotoxin Testing
12. Text of Proposal/ Requested Action	In 2017, the ISSC approved the MARBIONC Brevetoxin ELISA as a Limited Use Method under the NSSP (Proposal 17-107). The Standard Operating Procedure (SOP) for the MARBIONC Brevetoxin ELISA submitted as a part of the supporting documents for Proposal 17-107 specifies that quantification of sample dilutions is restricted to those dilutions falling within the linear portion of the standard curve, which is specified as the range of concentrations that yield 20-70% inhibition in the assay. One of the QA/QC criterion in the SOP requires that the variation (%CV) of concentrations calculated from sample dilutions falling within this range must be <20%. This proposal is to modify the MARBIONC ELISA SOP to: a) narrow the range for quantifying sample dilutions to 30%-70%, b) update the QA/QC criteria to reflect this change, and c) make minor additions and corrections to the text of the SOP. The modified SOP with proposed changes is provided in Appendix A. Data and justification for the proposed changes are provided in Appendix B.
13. Public Health Significance	The approval of this ELISA as a Limited Use Method for testing to support the NSSP has enabled rapid testing for NSP, which has enhanced the protection of public health by enabling more frequent NSP testing. Revising the SOP and QA/QC criteria will help to minimize avoidable QA/QC failures while still controlling for errors and protecting public health.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting

1. a. ☒ Growing Area
   b. ☐ Harvesting/Handling/Distribution
   c. ☐ Administrative

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10. Proposal Subject	Laboratory Method for *Vibrio parahaemolyticus* and *Vibrio vulnificus* Enumeration and Detection Through MPN and Real-Time PCR
11. Specific NSSP Guide Reference	Section IV Guidance Documents Chapter II Growing Areas .14 Approved NSSP Laboratory Tests
12. Text of Proposal/ Requested Action	5. Approved Methods fir 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
MPN-Real Time PCR[9]	***Vibrio parahaemolyticus (V.p.)* and *Vibrio vulnificus (V.v.)***	X	X
Direct Plating Method[8]	***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 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.
13. 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.
14. 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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Micropipettor Verification
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, NSSP Laboratory Evaluation Checklists, 2. Shellfish Laboratory Evaluation Checklist for Mouse Bioassay (MBA) and Scotia Rapid Test for PSP.
12. Text of Proposal/ Requested Action	The requested action is to adopt the new text to be consistent across checklists for the NSSP MBS and Scotia Rapid Test (SRT) for PSP under Part III, Section 3.1, Screening by SRT item 3.1.7.
13. Public Health Significance	Quality Assurance and Standardization are integral to the validity of the NSSP laboratory. This includes verifying the measurement accuracy of pipetting instruments including micropipettors. There are no recognized references that state micropipettors must receive third party certifications. There is no indication as to what "Level" calibration should exist. The reference for this item is only **#2, Good Laboratory Practice**. Accuracy measurement assurance should be based on workload and use. Pipette calibration values on certificates obtained in a calibration laboratory (known as a controlled laboratory) do not accurately transfer to the NSSP laboratory and therefore do not provide assurance and defensibility. A pipette's measurement accuracy is influenced by its *physical uncertainty*, *environmental uncertainty* (i.e., temperature, vibration and humidity) and *operator use uncertainty*. These uncertainties will differ between laboratories. Pipette performance in the NSSP (non-controlled laboratories) is impacted by the temperature and viscosity of the fluid, the skill of the operator and choice of tip. Conducting in-house verifications for each operator, using a verified balance provides a better assessment of the actual measurement accuracy of what the pipet is delivering. When the uncertainty of measurement exceeds the stated laboratory established threshold, adjustments are made. As a component of a Laboratory's Quality Management System, the individual laboratory can institute legally defensible and measurement assurance practices appropriate for the laboratory's workload, testing and ambient conditions. Calibration Cost Information from one Pipet Manufacturer: 1. Calibration and Maintenance - Offers three "levels" of examination, with an

	assorted number of readings at 3 volumes, across different channel pipettors. Cost Range $30 - $225 per unit. 2. Calibration only (center channel only) - $30 - $180 if unit passed on the initial attempt. 3. Non-Operational pipette repair evaluation (no calibration and parts additional cost) starting at $28/unit.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. **X** Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Microbiology Laboratory Evaluation Checklist- Standards Thermometer
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, 1. NSSP Laboratory Evaluation Checklist for Microbiology
12. Text of Proposal/ Requested Action	The requested action is to adopt modified standards thermometer language to correct checklist inconsistencies in Section 1.4 Laboratory Equipment item 1.4.21.
13. Public Health Significance	All standards thermometers allowed for in section 1.4.23, not just mercury-in-glass thermometers, should be calibrated and traceable to NIST at the points of use.
14. Cost Information	Cost of calibration.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	NSSP Microbiology Laboratory Evaluation Checklist – Reagent Water Quality
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, 1. NSSP Laboratory Evaluation Checklist for Microbiology.
12. 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.
13. 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.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Microbiology Laboratory Evaluation Checklist - Working Thermometers
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, 1. NSSP Laboratory Evaluation Checklist for Microbiology
12. 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:
13. 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."
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Microbiology & PCR Laboratory Evaluation Checklists - Working Thermometers
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, NSSP Laboratory Evaluation Checklists
12. 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.
13. 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.* <u>Savings</u>: Calibration costs <u>per thermometer</u>: $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.
14. Cost Information	none

Proposal No. 19-134

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	J. Michael Hickey, Jeff Kennedy, Diane Regan
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10. Proposal Subject	Membrane Filtration Technique for Seawater using mEndo Agar LES 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 , NSSP Laboratory Evaluation Checklists, NSSP Laboratory Evaluation Checklist for Microbiology
12. Text of Proposal/ Requested Action	The Requested Action is to adopt the attached checklist for the Membrane Filtration Technique for Seawater using mEndo Agar LES and to append the NSSP Laboratory Evaluation Checklist for Microbiology found at the end of section .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists to include this checklist.
13. Public Health Significance	The NSSP does not have a checklist for Total Coliform analysis on UV Seawater using the NSSP approved method of Membrane Filtration with mEndo Agar LES. Checklists provide quality assurance and method support for laboratories and for Laboratory Evaluation Officers to standardize and evaluate laboratories which use approved methods in support of the NSSP. The attached checklist for this NSSP approved method provides such standardization, quality assurance and background documentation for method procedures. As a laboratory evaluation tool with critical and key codes identified it will be used for determination of laboratory conformance and compliance.
14. Cost Information	none

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
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10. Proposal Subject	Microbiology Laboratory Evaluation Checklist - Sterilization
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, 1. NSSP Laboratory Evaluation Checklist for Microbiology
12. Text of Proposal/ Requested Action	The requested action is to adopt the modified text of the NSSP microbiology checklist, section 1.6 Sterilization and Decontamination, item 1.6.3:
13. Public Health Significance	The laboratory's goal is to ensure high-quality data using accepted scientific practices. The denoted information acknowledges recommended best practices used in recognized scientific publications to develop a laboratory's Quality Assurance Program (QAP) for sterilization practices at a wider range of temperature. The sterilization temperature range and the verification of working thermometers are now acceptably referenced to support past and present practices in program laboratories. The current reference material is cited to foster confidence in accepting the changes to an elevated sterilization temperature range and strengthen confidence in the acceptability of past practices for checking accuracy of working temperature monitoring devices. Most references for media sterilization simply state "121ºC for no less than 15 minutes." ***Difco***, a leading media manufacturer, states "A temperature range of 121-124ºC for 15 minutes is an accepted standard condition for sterilizing up to one liter of culture medium. The definition of "autoclave at 121ºC for 15 minutes" refers to the temperature of the contents of the container being held at 121ºC for 15 minutes, not to the temperature and time at which the autoclave has been set." ***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.….For general sterilization tasks, the recommended autoclave temperature range is 121 to 124ºC (at 200 kPa/29 PSI), although higher temperatures (≥121ºC) are acceptable for decontaminating laboratory material." *Each lab's QAP **must validate** temperature, time and pressure parameters for successful sterilization for media, reagents, supplies and spores using a verified working temperature monitoring device.*

14. Cost Information	No Cost. Minor adjustment during regularly scheduled sterilizer preventative maintenance service.

Proposal No. 19-136

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	NSSP DSP 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
12. Text of Proposal/ Requested Action	The requested action is to adopt the laboratory evaluation checklist for Diarrhetic Shellfish Poisoning LC-MS/MS.
13. 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.
14. Cost Information	N/A

Proposal No. 19-137

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ **Growing** Area b. ☐ **Harvesting**/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Checklist for the Bacteriological Analysis of UV Treated Process Water Samples by Membrane Filtration (MF) using mEndo Agar LES
11. Specific NSSP Guide Reference	NSSP *Guide for the Control of Molluscan Shellfish*, 2017 Revision, "Guidance Documents", Chapter II. Growing Areas, .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists, 1. NSSP Laboratory Evaluation Checklists for Microbiology.
12. Text of Proposal/ Requested Action	Incorporate Sections 2.11 through 2.14 for the Bacteriological Analysis of UV Treated Process Water Samples by Membrane Filtration using mEndo Agar LES into the NSSP Laboratory Evaluation Checklist for Microbiology.
13. Public Health Significance	Incorporation of the mEndo Agar LES membrane filtration method into the Microbiology Checklist will provide the means of assessing the competence of the laboratory to perform the test method.
14. Cost Information	NA

Proposal No. 19-138

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	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
12. 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.
13. Public Health Significance	The proposed modifications are to improve consistency in current NSSP microbiology checklist language and account for technology improvements to laboratory equipment.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	NSSP Laboratory Evaluation Officers Team
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10. Proposal Subject	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
12. Text of Proposal/ Requested Action	The requested action is to adopt the modified text of NSSP microbiology checklist item 1.4.24 in the Laboratory Equipment section and 3.2.7 in the Preparation of Shellfish for Examination section and add an additional reference to item 3.2.7.
13. Public Health Significance	1.4.24: One of the most basic attributes of any thermometer is its accuracy, and because a thermometer is only as valuable as the temperature it measures, accuracy is of the utmost importance. Calibration defines the accuracy by quantifying and controlling uncertainties within the measurement process. The quality of data must be known and established beyond a reasonable doubt before it can be used logically in any application; thus, calibration is an integral part of the lab's Quality Assurance. When individuals record and maintain data, proof of calibration demonstrates that the measurements performed are consistent with the "true value." An intermediate check is an action that the user takes to verify that the measuring instrument continues to be suitable for its purpose. Currently, the NSSP requires laboratories to perform intermediate checks on incubator and water bath thermometers at the temperature at which they are used. This requirement does not include refrigerator or freezer thermometers; however, NSSP Microbiology checklist items 1.4.9 and 1.4.10 require laboratories to measure and record refrigerator temperature data. When properly performed, an ice point is recommended as a “fixed point” for calibration of liquid in glass thermometers as it provides a reliable reference temperature at 0 °C with an estimated measurement uncertainty of ± 0.002 °C for determining the thermometer’s accuracy at all calibration points. The reliability and high degree of accuracy achieved by performing a proper ice point is due to the ice-water mixture stabilizing at its own “triple point.” Due to the nature of an ice point, it is the most common calibration point used for intermediate checks. 3.2.7 and reference addition: This change corrects an oversight in the current checklist regarding the role of gloves when shucking.
14. Cost Information	N/A

Proposal No. 19-140

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	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
12. 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.
13. 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.
14. Cost Information	N/A

Proposal No. 19-141

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	US Food and Drug Administration (FDA)
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10. Proposal Subject	NSSP Receptor Binding Assay for Paralytic Shellfish Poisoning (PSP) 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
12. 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).
13. 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.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Add the use of a mechanical shaker to the water microbiology methods checklist in the sample preparation requirements section and include a reference.
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 SHELLFISH LABORATORY EVALUATION CHECKLIST PART II - SEAWATER SAMPLES 2.2 Bacteriological Examination of Seawater by the APHA MPN 2.2.3 Sample and dilutions of sample are shaken vigorously (25 times in a 12" arc in 7 seconds) before inoculation. 2.5 Bacteriological Examination of Seawater by the MA-1 Method 2.5.5 Sample and dilutions of sample are shaken vigorously (25 times in a 12" arc in 7 seconds) before inoculation. 2.9 Sample Analyses - MF using mTEC Agar 2.9.3 The sample is shaken vigorously (25 times in a 12″ arc in 7 seconds) before filtration.
12. Text of Proposal/ Requested Action	Adopt the text of update the shellfish laboratory evaluation microbiology checklist (attached) to include the use of a mechanical shaker for sample preparation and include a reference for the use in the checklist's lists of references.
13. Public Health Significance	This proposal does not have direct public health significance but directly impacts the health of laboratorians performing water microbiological testing by allowing the use of a mechanical shaker to reduce or alleviate repetitive motion injuries caused by hand shaking the water samples. Work related injuries in the laboratory due to poor ergonomics are increasing every year and are costly to the laboratory due to work related injury claims. FDA LEO's currently allow the use of this equipment but there is no mention of the use of the equipment, no guidance for use of the equipment nor any reference from a reliable source in the current microbiology checklist for allowing the use of a mechanical shaker for sample preparation purposes.
14. Cost Information	This proposal updates text in the NSSP Manual wherever found in the microbiology checklist if approved by the conference. Minimal costs will be incurred by the ISSC administration when the laboratory evaluation checklist development and updating occurs at the ISSC office as part of the biannual NSSP Manual update process.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	MARBIONC Brevetoxin (Neurotoxic Shellfish Poisoning; NSP) ELISA Method 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
12. Text of Proposal/ Requested Action	The requested action is to adopt the text of the attached checklist for the MARBIONC Brevetoxin ELISA method 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.
13. Public Health Significance	The MARBIONC Brevetoxin ELISA method was approved for limited use at the 2017 ISSC meeting. Currently, there is no checklist adopted by the ISSC for this method. The attached 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. The checklist documents the number of critical, key or other nonconformities and how overall laboratory status for the method is determined.
14. Cost Information	N/A

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. 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.
11. 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
12. 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.
13. 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.
14. 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.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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10. Proposal Subject	Guidance on cleansing studies
11. Specific NSSP Guide Reference	NSSP Section IV Chapter II .19 VI B.
12. 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.
13. Public Health Significance	This language will provide state shellfish Authorities with guidance regarding 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.
14. Cost Information	No additional cost. This is simply providing guidance for a requirement already in place.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. X Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	Leonora Porter - Spokesperson
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10. Proposal Subject	Micropipettor Verification
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, NSSP Laboratory Evaluation Checklists, 6. Shellfish Laboratory Evaluation Checklist for PCR Microbiology
12. Text of Proposal/ Requested Action	The requested action is to adopt the new text for the NSSP PCR Microbiology checklist, section 1.4 Laboratory Equipment item 1.4.24.
13. Public Health Significance	Quality Assurance and Standardization are integral to the validity of the NSSP laboratory. One QA component includes verifying the measurement accuracy of pipetting instruments including micropipettors. There are no recognized references that state micropipettors must receive third party certifications. There is no indication as to what "Level" calibration should exist. The reference for this item is only **#2, Good Laboratory Practice**. Accuracy measurement assurance should be based on workload and use, not calendar year. Pipette calibration values on certificates obtained in a calibration laboratory (known as a controlled laboratory) do not accurately transfer to the NSSP laboratory and therefore do not provide assurance and defensibility. A pipette's measurement accuracy is influenced by its *physical uncertainty*, *environmental uncertainty* (i.e., temperature, vibration and humidity) and *operator use uncertainty*. These uncertainties will differ between laboratories. Pipette performance in the NSSP (non-controlled laboratories) is impacted by the temperature and viscosity of the fluid, the skill of the operator and choice of tip. Conducting in-house verifications for each operator, using a verified balance provides a better assessment of the actual measurement accuracy of what the pipet is delivering. When the uncertainty of measurement exceeds the stated laboratory established threshold, adjustments are made. As a component of a Laboratory's Quality Management System, the individual laboratory can institute legally defensible and measurement assurance practices appropriate for the laboratory's workload, testing and ambient conditions. Savings: Calibration Cost Information from one Pipet Manufacturer: 1. Calibration and Maintenance - Offers three "levels" of examination, with an

	assorted number of readings at 3 volumes, across different channel pipettors. Cost Range $30 - $225 per unit. 2. Calibration only (center channel only) - $30 - $180 if unit passed on the initial attempt. Non-Operational pipette repair evaluation (no calibration and parts additional cost) starting at $28/unit.
14. Cost Information	N/A

Proposal No. 19-147

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	US Food & Drug Administration (FDA)
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9. Email	Melissa.Abbott@fda.hhs.gov
10. Proposal Subject	Relay contaminant reduction studies.
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter V. Shellstock Relaying Section @.02 Contaminant Reduction B. (2)
12. Text of Proposal/ Requested Action	(2) Contaminant levels of poisonous or deleterious substances in shellstock do not exceed FDA ~~tolerance~~action levels, tolerances and/or guidance levels and/or levels that are deemed safe through risk evaluation; or
13. Public Health Significance	Action levels, tolerances and/or guidance levels have not been established for all poisonous or deleterious substances. When there is concern about contamination of shellstock by a poisonous or deleterious substance and no action level, tolerance, or guidance level for that substance, regulators must evaluate risk and establish a level of concern. Suggested change from "tolerance" to "action levels, tolerances, and/or guidance levels" is made to make the language consistent with the title of National Shellfish Sanitation Program Guide for the Control of Molluscan Shellfish, Section IV Guidance Documents, Chapter II Growing Areas, .08 Action Levels, Tolerances and Guidance Levels for Poisonous or Deleterious Substances in Seafood.
14. Cost Information	Possible increased cost of unknown magnitude related to time necessary to conduct risk evaluations.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	ISSC Executive Office
3. Affiliation	Interstate Shellfish Sanitation Conference
4. Address Line 1	209 Dawson Road
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6. City, State, Zip	Columbia, SC 29223
7. Phone	(803) 788-7559
8. Fax	(803) 788-7576
9. Email	issc@issc.org
10. Proposal Subject	Correct language of MO to reflect current checklists
11. Specific NSSP Guide Reference	Section II Model Ordinance – Chapter I. Shellfish Sanitation Program for the Authority @.03 Evaluation of Shellfish Sanitation Program Elements B. Criteria for evaluation of shellfish sanitation program elements shall be as follows: 1. Laboratory
12. Text of Proposal/ Requested Action	Section II Model Ordinance – Chapter I. Shellfish Sanitation Program for the Authority @.03 Evaluation of Shellfish Sanitation Program Elements B. Criteria for evaluation of shellfish sanitation program elements shall be as follows: 1. Laboratory (a) Requirements for evaluation of shellfish laboratories shall include at a minimum: i. Records audit of laboratory operations both Quality Systems and Technical methods; ii. Direct observation of current laboratory operating conditions; and iii. Information collection from the Authority and other pertinent sources concerning laboratory operations. (b) Laboratory status is determined by the number and types of nonconformities found in the evaluation using NSSP standardized criteria contained in the FDA Shellfish Laboratory Evaluation Checklists found in Section IV Guidance Documents Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists. i. Quality System Evaluation. (a) This checklist includes a conforming and nonconforming status only. All nonconformities must be reconciled prior to scheduling an onsite evaluation of technical

methods in NSSP laboratories. As this part of the evaluation specifically refers to the Quality manual and SOPs and other documentation considered the basis for data defensibility, this documentation must be in order prior to further Laboratory Evaluation Officer (LEO) scheduling. The Quality Systems evaluation is performed as a desk audit and is in accordance with the checklist found in Section IV Chapter II.

ii. Technical Evaluation: Shellfish Laboratory will be technical.y evaluation and will be assigned the designation of conforms, provisionally conforms or non-confomance. The criteria used in determining the evaluation designations are included in the NSSP Shellfish Laboratory Evaluation Checklist designated for the specific type of laboratory evaluation being performed. (For more information see Section IV. Guidance Documents Chapter II. Growing Areas .15 Evaluation of Laboratories by State Shellfish Laboratory Evaluation Officers Including Laboratory Evaluation Checklists

~~(b) Conforms. In order to achieve or maintain conforming status under the NSSP, a laboratory must meet the following laboratory evaluation criteria:~~

~~(c) No critical nonconformities in the microbiological or marine biotoxin component under evaluation have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist; and~~

~~(d) (b) Not more than thirteen (13) key nonconformities in the microbiological component or six (6) in the marine biotoxin components have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist; and~~

~~(c) Not more than eighteen (18) critical, key, and other nonconformities in total in the microbiological component, twelve (12) critical, key and other nonconformities in total for the paralytic shellfish poisoning (PSP) and amnesic shellfish poisoning (ASP) components, or ten (10) critical, key and other~~

~~nonconformities in total for the neurotoxic shellfish poisoning (NSP) component have been identifiedusing the appropriate NSSP Shellfish Laboratory Evaluation Checklist. This number must not exceed the numerical limits established for either the critical or key criteria; and~~

~~(d) No repeat key nonconformities have been identified in the microbiological or marine biotoxin component under evaluation in consecutive evaluations using the appropriate NSSP Shellfish Laboratory Evaluation Checklist.~~

~~iii. Technical Evaluation: Provisionally Conforms. In order to be deemed provisionally conforming under the NSSP, a laboratory must meet the following laboratory evaluation criteria:~~

~~(a) Not more than three (3) critical nonconformities in the microbiological component, four (4) in the PSP and ASP components, or three (3) in the NSP component have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist; and~~

~~(b) Not more than thirteen (13) key nonconformities in the microbiological component or six (6) in the marine biotoxin component have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist; and~~

~~(c) Not more than eighteen (18) critical, key and other nonconformities in total in the microbiological component, or twelve (12) critical, key and other nonconformities in total in the PSP and ASP components or ten (10) critical, key and other nonconformities in total in the NSP component have been identified using the appropriate NSSP Shellfish Laboratory Evaluation umber must not exceed the numerical limits established for either the critical or key criteria; and~~

~~(d) Not more than one (1) repeat key nonconformity has been identified in the microbiological or marine biotoxin component under evaluation in consecutive evaluations using the appropriate NSSP Shellfish Laboratory Checklist.~~

~~iv. Technical Evaluation: Nonconformance. When~~

	~~a laboratory exceeds the following criteria, it will be determined to be in nonconformance:~~ (a) ~~More than three (3) critical nonconformities in the microbiological component or four (4) in the PSP and ASP components, or three (3) in the NSP component have been identified using the appropriate NSSP Shellfish Laboratory Checklist; or~~ ~~(b) More than thirteen (13) key nonconformities in the microbiological component or six (6) in the marine biotoxin component have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist;~~ ~~(c) More than eighteen (18) critical, key, and other nonconformities in total in the microbiological component, or more than twelve (12) critical, key and other nonconformities in total in the PSP and ASP components, or more than ten (10) critical, key, and other nonconformities in total in the NSP component have been identified using the appropriate NSSP Shellfish Laboratory Evaluation Checklist; or~~ ~~(d) One (1) or more repeat critical or two (2) or more repeat key nonconformities have been identified in consecutive evaluations in either the microbiological or marine biotoxin components using the appropriate NSSP Shellfish Laboratory Evaluation Checklist.~~
13. Public Health Significance	The goal of a laboratory evaluation is to monitor implementation of NSSP Quality Systems and Approved methods. Laboratory data is standardized as a result of this process and reciprocity of shellfish in the commercial market is protected and preserved through defensible practices and transparent requirements. As the laboratory program in the NSSP continues to develop and grow it is prudent to keep requirements in accessible documents with few deviations. Checklists are a cornerstone document for laboratories, referring to these documents ensures laboratories have access to requirements at all times. As laboratorians are the target audience, this is the most sensible place for the actual numbers of nonconformities to reside, and the reference to the checklists in the Model Ordinance ensures the checklists are part of the overarching document adopted by reference or into legislation. Multiple locations of numbers of permissible nonconformities only ensures updates will be missed. As existing structure is in place through the Lab Committee to handle checklists and edits therein, this seems the most reasonable solution.
14. Cost Information	No cost incurred by change. Practice is already in place.
15. Research Needs Information (Optional)	

a. Proposed specific research need/ problem to be addressed	none
b. Explain the relationship between proposed research need and program change recommended in the proposal	**There is no research need to implement proposal recommendation. This is a change requested to reflect language that exists in the MO. The language changes proposed have not been changed as new Checklists were introduced and the numbers of Critical key and other nonconformities are not constant. Therefore, it makes sense to refer to the checklist rather than continue to have to occasionally update arbitrary numbers in Chapter 1. This will save time and money in the future as more checklists are introduced. Checklists have a great deal of attention by the Lab Committee, in fact, they have a subcommittee dedicated entirely to their drafting or editing. Any questions would be answered here.**
c. Estimated cost	**none**
d. Proposed sources of funding	**N/A**
e. Time frame anticipated	**N/A**
For Research Guidance Committee Use Only	Relative priority rank in terms of resolving research need ☐ Immediate ☐ Required ☐ Valuable ☐ Important ☐ Other

Proposal No. 19-149

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	ISSC Executive Office
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7. Phone	(803) 788-7559
8. Fax	(803) 788-7576
9. Email	issc@issc.org
10. Proposal Subject	Biotoxin Guidance
11. Specific NSSP Guide Reference	Section II. Chapter IV Shellstock Growing Areas
12. Text of Proposal/ Requested Action	In conjunction with the adoption of Proposal 13-116 at the 2017 ISSC Biennial Meeting, the voting delegates recommended the Biotoxin Committee develop a guidance document to include guidance for end product testing programs in closed state waters. In addition to proposing guidance, the committee will be making recommendations to modify the monitoring requirements of Chapter IV @.04 Marine Biotoxin Control. These proposed changes are under development. The purpose of this proposal is to advise the ISSC membership that the Biotoxin Committee will be making recommendations to modify Chapter IV @.04 as part of their committee charge from Proposal 13-116
13. Public Health Significance	The proposed changes should clarify and simplify biotoxin monitoring.
14. Cost Information	

Proposal for Task Force Consideration at the ISSC 2017 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

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8. Fax	1-517-372-2006
9. Email	broman@neogen.com
10. Proposal Subject	Neogen's 'Reveal 2.0 for PSP' for detection of PSP
11. Specific NSSP Guide Reference	Section IV. Guidance Documents, Chapter II. Growing Areas, .11 Approved NSSP Laboratory Tests
12. 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.
13. 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
14. Cost Information	Approximately $20 per test. Reader based assay – approximate cost of reader is $2,700.00 USD.

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative
2. Submitter	Catalina Sea Ranch, LLC (CSR)
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9. Email	maria@catalinasearanch.com
10. Proposal Subject	Update the Protocol for the Landing of Shellfish from Federal Waters
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.03 Section IV. Guidance Documents Chapter II Growing Areas .06
12. Text of Proposal/ Requested Action	**Section II. Model Ordinance** **Chapter VI. Shellfish Aquaculture** **@.03 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 frequency with which NOAA will audit the aquaculture facility and vessels; (b) biotoxin 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. **Section IV. Guidance Documents** **Chapter II. Growing Areas** **.06 Protocol for the Landing of Shellfish from Federal Waters** Harvest of molluscan shellfish in Federal Waters not routinely monitored for toxins in shellfish ~~(such as the Federal waters on Georges Bank closed due to PSP risks)~~ may be authorized provided the Authority in the State of landing in cooperation with appropriate Federal agencies shall develop agreements or memoranda of understanding between the Authority and individual shellfish harvesters or individual shellfish dealers. The following guidance provides descriptions of the specific information to be included in the protocol. A. Harvest Permit Requirements ~~If harvesting from Federal waters closed due to toxins, t~~The Authority in the landing State will only allow the landing of shellfish from vessels in

possession of an appropriate Aquaculture Permit issued by NOAA or an Exempted Fishing Permit (EFP) issued by the National Marine Fisheries Service (NMFS) by vessels participating in the Federal Vessel Monitoring System (VMS). ~~The NMFS shall receive concurrence from the Authority in the State of landing. Vessels operating in open Federal waters will also need applicable permits.~~

~~Training~~

~~The Authority shall ensure that all shipboard persons conducting onboard testing have been trained by a U.S. FDA LEO (LEO) or an FDA marine biotoxin expert to conduct onboard toxin screening using an NSSP recognized method(s). Shipboard persons conducting onboard toxin testing must receive refresher training every three (3) years. A designee of the FDA LEO or FDA marine biotoxin expert may be appointed in writing to provide the training and/or refresher training.~~

B. Vessel Monitoring

The Authority shall monitor the harvesting location(s) of each landing vessel.

C. Identification of Shellfish

Prior to landing each vessel Captain or Mate shall provide the Authority with a Harvest Record, which may be electronic provided that it is made available to the authorized individual at dockside, for each harvesting trip identifying each lot of shellfish as follows:

1. Vessel name and Federal Fishing Permit number;
2. Name and telephone number of the vessel Captain and vessel owner;
3. Date(s) of harvest;
4. Number of lots and volume of catch per lot or number of containers per lot;
5. Location(s) of harvest (GPS coordinates or latitude/longitude coordinates in degrees:minutes:seconds);
6. Identification of each harvest lot, including cage tag numbers for surf clams and ocean quahogs, and container numbers or identification codes for other shellfish species;
7. Location (GPS coordinates or latitude/longitude coordinates in degrees:minutes:seconds) of each toxin screening sample;
8. Results of each toxin screening test; and
9. Destination(s) and purchaser(s) of each lot and amount of each lot to each destination

The Captain or Mate shall sign the Harvest Record. ~~The Harvest Record shall be checked by the individual authorized to sample the harvested shellfish. Failure to provide complete and accurate information will result in revocation or suspension of the NMFS EFP and rejection of the entire lot(s) of harvested shellfish. Four (4) copies of the Harvest Record~~

~~shall be prepared. One (1) copy shall remain with the vessel, one (1) copy shall be provided to the Authority in the State of landing, one (1) copy shall accompany the catch to the processing firm(s), and one (1) copy shall be retained by the laboratory authorized to conduct lot sample analyses.~~

Container Labeling:

Each container of shellfish shall be clearly labeled (indelible and legible) with the following NSSP required information at the time of harvest:

1. Surf clams and ocean quahogs existing NMFS tagging requirements.
2. All other molluscan shellfish (including Stimpson clams also known as Arctic surf clams) using durable, waterproof, Authority sanctioned prior to use tags:
   a. Vessel name;
   b. Type and quantity of shellfish;
   c. Date of harvest; and
   d. Harvest lot area defined by GPS coordinates or latitude/longitude coordinates in degrees:minutes:seconds.

D. ~~Pre-Harvest~~Shellfish Sampling

~~Prior to harvesting of molluscan shellfish, a minimum of five (5) screening samples shall be collected within each area of intended harvest (lot area) and tested for marine biotoxins that are likely to occur in accordance with an NSSP recognized method. Each screening sample shall be collected during a separate and distinct gear tow. Screening sample tows shall be conducted in a manner that evenly distributes the five (5) samples throughout the intended harvest area for each area of intended harvest (see Section H.). Only shipboard officials trained by an FDA LEO or FDA marine biotoxin expert (or their designee as expressly indicated in writing) in the use of the designated NSSP method may conduct these tests. Each of the five (5) samples must test negative for toxins (i.e., below half of the established criteria in Section II. Model Ordinance Chapter IV @04.C. (1)). A positive result from any one (1) sample shall render the lot area unacceptable for harvest. The harvest vessel Captain shall immediately report all positive screening test results, by telephone or email, to the Authority within the intended State of landing, the FDA Shellfish Specialist, and the processor. The FDA shall notify the NMFS. The NMFS shall notify permitted harvesters to advise them to cease fishing in the affected area(s). For each screening test, whether positive or negative, the remaining sample material (homogenate) shall be maintained under refrigeration for later use should the Authority in the State of landing request confirmatory testing using an NSSP recognized method.~~

Each commercial shellfish grower is required to submit at least one shellfish sample per week, per lot, to an FDA conforming laboratory for

testing of ASP and PSP during all harvest periods. Sample test results will be submitted to the Authority for review and data compilation.

Harvester representatives performing sample collection must receive initial training to ensure proper collection technique from the appropriate Authority. Sample collectors must receive refresher training every three (3) years.

Location of sampling stations:
The sampling station should be centrally located in each harvest lot.

Sampling Frequency:
Samplers are required to achieve a sampling frequency of at least once sample per week during the months of May through October, and at least one sample per month during the months of November through April. When either PSP toxins or domoic acid are detected in shellfish, the frequency of sampling will double to allow better characterization of the event.

If test results of any sample collected equal or exceed 50% of the established criteria in Section II. Model Ordinance Chapter IV@.04 C. (1) (e.g., 40 µg /100 g for PSP toxins), sampling will double for all harvesters. If test results of any samples collected equal or exceed 75% of the established criteria in Section II. Model Ordinance Chapter IV@.04 C. (1) then sampling will commence for each harvest and the harvest will be held until final test results indicate toxin levels below that established criteria in Section II. Model Ordinance Chapter IV@.04 C. (1).

If test results equal or exceed that established criteria in Section II. Model Ordinance Chapter IV@.04 C. (1) then the growing area will be placed in Closed Status pursuant to Section II. Model Ordinance Chapter IV@.04 C. (1).

Testing shall be according to NSSP recognized methods and shall be conducted by laboratories evaluated in accordance with NSSP guidelines. Private laboratories may be used if evaluated by an LEO in accordance with NSSP guidelines.

Sampling Methods:

Each screening sample shall be comprised of at least twelve (12) whole animals with the exception of mussels and “whole” or “roe-on” scallops. For mussels each sample shall be comprised of thirty (30) animals. For “whole” scallops each sample shall be comprised of twenty (20) scallop viscera and gonads. For “roe-on” scallops each sample shall be comprised of twenty (20) scallop gonads.

~~E. Submittal of Onboard Screening Homogenates and Test Results~~

~~F.~~

~~All screening results shall be recorded on the Harvest Record as stipulated in Section D. of this Protocol. Upon landing of the harvest vessel, the Harvest Record and screening homogenates shall be provided to the Authority or designee and the testing of those samples for toxins using an NSSP method by an NSSP conforming laboratory in the State of landing authorized to sample the harvested shellfish as described in Section G. of this Protocol.~~

~~Dockside Sampling~~

~~After dockside samples are collected by the Authority or designee, molluscan shellfish may be processed while awaiting toxin results. Each lot must be identified and segregated during storage while awaiting dockside sample test results. Under no circumstances will product be released from the processor prior to receiving satisfactory toxin results that demonstrate that toxin levels are below the established criteria in Section II. Model Ordinance Chapter IV @04.C.(1).~~

~~The dockside sampling protocol for molluscan shellfish shall be as follows:~~

~~For each lot of molluscan shellfish, a minimum of seven (7) composite samples, each comprised of at least twelve (12) whole animals, shall be taken at random by the individual authorized by the Authority to sample, with the following exceptions:~~

~~For each lot of mussels, a minimum of seven (7) composite samples, each comprised of at least thirty (30) whole animals, shall be taken at random by the individual authorized to sample.~~

~~For each lot of "whole" scallops, a minimum of seven (7) composite samples, each comprised of twenty (20) scallop viscera and gonads, shall be taken at random by the individual authorized to sample.~~

~~For each lot of "roe-on" scallops, a minimum of seven (7) composite samples, each comprised of twenty (20) scallop gonads, shall be taken at random by the individual authorized to sample.~~

~~Shellfish samples collected in accordance with G.1 shall be tested for the presence of toxins using an NSSP recognized method(s).~~

~~Laboratory test results for each lot of shellfish shall be forwarded to the Authority in the State in which the shellfish is being held prior to the product being released by the Authority in the State of landing, or if processed in another State, the Authority in the State of processing.~~

~~G.~~E. Holding and Lot Separation

A harvest lot is defined as all molluscan shellfish harvested during a single period of uninterrupted harvest activity within a geographic area not to exceed three (3) square miles. Once harvesting has ceased and the harvest vessel moves to another location, regardless of the distance, a new harvest lot will be established. Any harvest vessel containing more than one (1)lot shall clearly mark and segregate each lot while at sea,

during off loading, and during transportation to a processing facility. Prior to harvesting in Federal waters, each harvest vessel shall submit to the NMFS a written onboard lot segregation plan. The Authority in the intended State of landing and the FDA Shellfish Specialist must approve the proposed lot segregation plan.

~~H.~~F. Disposal of Shellfish

If test results of any harvest held based on D. Shellfish Sampling ~~one (1) of the seven (7) samples collected in accordance with G.1~~ equal or exceed the established criteria in Section II. Model Ordinance Chapter IV@.04 C. (1) (e.g., 80 µg /100 g for PSP toxins)(n=7, c=0), the entire lot must be discarded or destroyed at the cost of the harvester under the supervision of the Authority in accordance with State laws and regulations except when:

A lot of "whole" or "roe-on" scallops equals or exceeds the established criteria in Section II. Model Ordinance Chapter IV@.04C.(1), the adductor muscle may be shucked from the viscera and/or gonad and marketed. The remaining materials (viscera and/or gonad) must be discarded or destroyed under supervision of the Authority in accordance with State laws and regulations.

Dockside toxin testing shall be according to NSSP recognized methods and shall be conducted by laboratories evaluated in accordance with NSSP guidelines. Private laboratories may be used if evaluated by an LEO in accordance with NSSP guidelines.

~~I.~~G. Notification Prior to Unloading by Harvesters Under NMFS Permts

Prior to the issuance of an EFP, the harvester shall be responsible for notifying the Authority in the State of landing and in a manner approved by the Authority that molluscan shellfish is being harvested for delivery to the intended receiving processor.

Each vessel shall give at least twelve (12) hours' notice to the individual authorized to sample prior to unloading shellfish. Notice of less than twelve (12) hours may be approved by the authorized individual at his/her discretion. Authorities may appoint a designee in writing for sampling and sample transport to the NSSP certified testing laboratory in accordance with the practices and procedures used by the Authority under the NSSP. The procedures, as well as training and certification records, must be available for evaluation.

Shellfish from a Federal water harvest area(s) must be kept separate and not sold until so authorized by the Authority in the State of landing or, if processed in another State, the Authority in the State of processing.

	Failure to comply with the provisions of this Protocol will result in the suspension or revocation of the vessel's permits through the NMFS. ~~J.~~H. Unloading Schedule for Harvesters Under NMS Permits Unloading shall take place between 7:00 A.M. and 5:00 P.M. Monday through Friday, unless otherwise mutually agreed upon by the individual authorized to sample, the processing plant manager, the harvest vessel captain, and the Authority in the State of landing. ~~K. Access for Dockside Sampling~~ ~~L.~~ ~~Individuals authorized to sample shall be provided access to the catch of shellfish.~~ ~~M.~~I. Record Keeping Record keeping requirements shall be as follows: 1. The vessel shall maintain Harvest Records for at least one (1) year. 2. The processor(s) shall maintain Harvest Records for at least one (1) year or two (2) years if the product is frozen. 3. The Authority in the State of landing shall retain Harvest Records for at least two (2) years. ~~N.~~J. Early Warning/Alert System Toxin data acquired as a result of ~~onboard screening and dockside~~sample testing shall be transmitted to the FDA. These data~~, both screening and dockside,~~ shall be transmitted to the FDA by the NSSP certified laboratory conducting toxin testing of the sampled lot(s) within one (1) week of the completion of the toxin analyses. The data provided shall include the following: 1. Shellfish species; 2. Harvest location name and coordinates (GPS or latitude/longitude); 3. Harvest date; 4. Onboard screening test method, date, and results; and 5. Laboratory test date, test method, and test results for dockside samples. Results of all samples having unacceptable levels of toxins (e.g.,<80 µg/100 g for PSP toxins) shall immediately be reported to the Authority in the State of landing. If the results of any one (1) sample equal or exceed the established criteria in Chapter IV @.04(c)(1) the testing laboratory shall immediately notify the FDA Shellfish Specialist, the Authority, and the processor by telephone and email. The FDA shall notify the NMFS. The NMFS shall notify permitted harvesters to advise them to cease ~~fishing~~ harvesting in the affected area(s).
13. Public Health Significance	This proposal provides clarification to Chapter VI. @.03 by clarifying the type of testing requirements for aquaculture facilities. Additionally, the proposal modifies

	Section IV. Guidance Documents for the landing of shellfish in Federal Waters. These modifications would improve and simplify the protocols for landing shellfish in Federal Waters where a biotoxin concern exists.
14. Cost Information	

Proposal No. 19-152

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	Catalina Sea Ranch, LLC (CSR)
3. Affiliation	Catalina Sea Ranch, LLC (CSR)
4. Address Line 1	2303 S. Signal street, Berth 58
5. Address Line 2	
6. City, State, Zip	San Pedro, CA 90731
7. Phone	844-922-8254
8. Fax	
9. Email	maria@catalinasearanch.com
10. Proposal Subject	Update the Protocol for Marine Biotoxin Control
11. Specific NSSP Guide Reference	Section II. Model Ordinance Chapter IV. Shellstock Growing Areas @.04 B.
12. Text of Proposal/ Requested Action	**@.04 Marine Biotoxin Control** B. Marine Biotoxin Management Plan. In those areas that have been implicated in an illness outbreak or where toxin-producing phytoplankton are known to occur and the toxins are prone to accumulate in shellfish, and when appropriate at those times when marine biotoxins can be reasonably predicted to occur, representative samples of the water may be collected and shellfish shall be collected during harvest periods. The samples shall be collected from indicator stations at intervals determined by the Authority. Water samples may be assayed for the presence of toxin-producing phytoplankton and shellfish meat samples shall be assayed for the presence of toxins. **NOTE:** In situations in which the toxin of concern has an established cell count standard, such as *Karenia brevis*, water and shellfish samples would not be required. Management decisions could be made on either water or shellfish sampling results. (1) The Authority shall develop and adopt a marine biotoxin management plan for all marine and estuarine shellfish growing areas if there is a history of biotoxin closures related to PSP, ASP, NSP, DSP, or AZP; if toxin-producing phytoplankton are known to occur in the growing area; or a reasonable likelihood that biotoxin closures could occur. (2) For Federal waters harvesters, each company is considered an Authority and must develop and adopt their own plan. (~~2~~3) The plan shall… (~~3~~4) The Authority may… (~~4~~5) Except that the…

	(~~5~~6) The plan may… (~~6~~7) Prior to allowing…
13. Public Health Significance	This proposal would expand the definition of Authority to include harvesters in the definition of Authority.
14. Cost Information	

Proposal for Task Force Consideration at the ISSC 2019 Biennial Meeting	1. a. ☒ Growing Area b. ☐ Harvesting/Handling/Distribution c. ☐ Administrative

2. Submitter	ISSC Executive Office
3. Affiliation	Interstate Shellfish Sanitation Conference
4. Address Line 1	209-1 Dawson Road
5. Address Line 2	
6. City, State, Zip	Columbia, SC 29223
7. Phone	803-788-7559
8. Fax	803-788-7576
9. Email	issc@issc.org
10. Proposal Subject	Alternative Pre-harvest Screening
11. Specific NSSP Guide Reference	Section II Model Ordinance – Chapter IV. Shellstock Growing Area @.04 Marine Biotoxin Control B. Marine Biotoxin Management Plan (6)e
12. Text of Proposal/ Requested Action	(6) Prior to allowing the landing of shellfish harvested from Federal waters where routine monitoring of toxin levels is not conducted, in addition to following State requirements in the Model Ordinance, the State Authority in the landing State, in cooperation with appropriate Federal agencies, shall develop agreements or memoranda of understanding between the Authority and individual shellfish harvesters or individual shellfish dealers. The agreements or memoranda of understanding shall provide strict safety assurances. At a minimum agreements or memoranda of understanding shall include provisions for: (a) Harvest permit requirements; (b) Training for individuals conducting onboard toxicity screening using NSSP methods; (c) Vessel monitoring; (d) Identification of shellfish for each harvesting trip to include: (i) Vessel name and owner; (ii) Captain's name; (iii) Person conducting onboard screening tests; (iv) Port of departure name and date; (v) Port of landing name and date; (vi) Latitude and longitude coordinates of designated harvest area; (vii) Onboard screening test results; (viii)Volume and species of shellfish harvested; (ix) Intended processing facility name, address and certification number; and (x) Captain's signature and date; (e) Pre-harvested (onboard) sampling that includes a minimum of five (5) samples from the intended harvest area be tested for toxins that are likely to be present. Harvesting shall not be permitted if any of the pre-harvested samples contain toxin levels in excess of half of the established criteria listed in Chapter IV@.04(c)(1)As an alternative to pre-harvest (on-board) screening samples, end product (dockside) testing samples alone may be used. Should alternative be chosen, the minimum number of seven (7) dockside samples as stated in section (g) below must be expanded to ten

	(10). (e.g., 44 µg/l00 g when using a quantitative test or a positive at a limit of detection of 40 µg/100 g for the qualitative screening test for PSP toxins); (f) Submittal of onboard screening homogenates and test results to the Authority in the State of landing; (g) The collection of a minimum of seven (7) dockside samples by the Authority or designee and the testing of those samples for toxins using a NSSP method by a NSSP conforming laboratory; the Authority may require more samples based on the size of the vessel and the volume of shellfish harvested; (h) Holding and providing separation until dockside samples verify that toxin levels are below the established criteria (e.g., 80 µg/100 g for PSP toxins); (i) Disposal of shellfish when dockside test results meet or exceed the established criteria in Chapter IV@.04C.(1) (e.g., 80 µg /100 g for PSP toxins); (j) Notification prior to unloading; (k) Unloading schedule; (l) Access for Dockside Sampling; (m) Record Keeping; and (n) Early Warning/Alert System.
13. Public Health Significance	The ISSC Executive Board adopted the proposed language as an interim measure to address concerns with the Abraxis PSP Shipboard ELISA Kit. See attached report.
14. Cost Information	

Follow-up Actions by Eurofin Abraxis

Rich Quashne
Eurofins Abraxis
124 Railroad Drive
Warminster, PA 18974

RE: Updated product insert/instructions for Abraxis PSP Shipboard ELISA

In reference to Section IV Guidance Documents, Chapter II Growing Areas, .14 Approved NSSP Laboratory Tests, 4. Approved Limited Use Methods for Marine Biotoxin Testing, Eurofin Abraxis requests the following action:

The product insert/kit instructions for the Abraxis PSP Shipboard ELISA should be updated. The proposed updated kit instructions do not change how the assay is performed. Nor does it require a change to the language in the NSSP method table. Rather the updates provide clarity and additional information specifically in the kit instructions that come with the kit on two critical steps to aid the user in more consistently achieving the required quality control (QC) measures.

It was found that some users were experiencing challenges in meeting the required QC (i.e., $r^2 > 0.99$) for data to be used in support of making NSSP decisions. There were two critical steps in the assay instructions that if not followed contributed significantly to a decreased dynamic range of the assay and difficulty in passing QC. The first step is allowing the kit reagents to come to room temperature prior to use. Current instructions do not specify how to bring the reagents to room temperature. The proposed update states that all reagents must be removed from the kit packaging and left at room temperature for two hours prior to use. The second critical step relates to the swirling of reagents in the plate during the incubation. The update states that plate swirling must be achieved using a rotating shaker, or equivalent, to achieve the degree of shaking needed to ensure antigen-antibody contact for binding. Adding these specific instructions will improve user experience, consistency, and ability to successfully perform the kit, yielding the required QC for optimal kit performance.

Supporting documents, including the proposed updates to the product insert/kit instructions, are attached.

Paralytic shellfish poisoning (PSP) toxins are a family of neurotoxins produced by certain species of dinoflagellates primarily belonging to the Genus *Alexandrium* and *Pyrodinium*. These toxins may be accumulated by filter-feeding bivalves, thereby posing a risk to human health. Shellfish growing areas in state waters are monitored for marine biotoxins and the monitoring information is used to place growing areas in the closed or open status accordingly. For remote, offshore areas such as federal waters, routine monitoring programs are not feasible. Instead, onboard screening and dockside testing (also referred to as pre-harvest screening and lot testing) is the marine biotoxin control strategy used. This strategy requires an NSSP method for pre-harvest screening to determine when/where it is safe to harvest in a given intended harvest area. This level of testing serves as the first level of protection for public health but also economic insurance for the harvester who must also submit samples for dockside or lot testing upon landing. Currently, the Abraxis PSP Shipboard ELISA is the only Approved Limited Use Method for onboard screening for PSP toxins. This method has been used successfully in this capacity for ~10 years. Recent expansion of the kit to additional users identified that more detailed instructions were needed for new users to successfully perform the kit.

The cost of the testing supplies are:
Saxitoxins (PSP) Shipboard (ISSC 09-107), ELISA kit, 96 tests, $590
Saxitoxins (PSP) Shipboard (ISSC 09-107), accessory pack, 20 test, $165

Abraxis PSP Ship Board Data Analysis Worksheet (Quantitative)

Vessel		Kit Lot #	
Date		Kit Expiration	
Time		Technician	

r^2=_____ (>0.990?)

Well	ID (ug/100g)	Abs.	Average Abs.	B/B_0, Result	Well	ID	Abs.	Average Abs.	B/B_0, Result
A1	0				A3				
						(Sam 3)			
B1	0		B_0=		B3				
				Abs 1.0 – 2.0					
C1	20				C3				B/B_0, Result
						(Sam 4)			
D1	20		B_1=		D3				
				B_1/B_0					
E1	40				E3				B/B_0, Result
						(Sam 5)			
F1	40		B_2=		F3				
				B_2/B_0					
G1	80				G3				B/B_0, Result
						(Sam 6)			
H1	80		B_3=		H3				
				B_3/B_0					
A2	120				**Key**:	Average = $\frac{\text{Absorbance 1 + Absorbance 2}}{2}$			
					How to label samples: VVSSDDMMYYT Where VV=MD/SW, SS=Station number, DD=day (01-31), MM=month (01-12), YY= (10) T= Q for quahog, C for surfclam				
B2	120		B_4=						
				B_4/B_0	Station Log:	Station #	Latitude	Longitude	
C2	Control								
D2	Control					01			
				B/B_0, Result 48-72µg/100g					
E2						02			
	(Sam 1)								
F2						03			
				B/B_0, Result					
G2						04			
	(Sam 2)								
H2						05			
				B/B_0, Result					

I. Evaluation

Evaluation of ELISA results may be performed using a spreadsheet macro available from Eurofins Abraxis or other commercial ELISA evaluation programs using Log/Logit. For manual evaluation, calculate the mean absorbance value for each of the standards. Calculate the $\%B/B_0$ for each standard by dividing the mean absorbance value for each standard by the Zero Standard (Standard 0) mean absorbance. Construct a standard curve by plotting the $\%B/B_0$ for each standard on a vertical linear (y) axis versus the corresponding Saxitoxin concentration on horizontal logarithmic (x) axis on graph paper. $\%B/B_0$ for the control and samples will then yield levels in µg/100 g of Saxitoxin by interpolation using the standard curve.

The concentrations of the samples are determined using the standard curve run with each test. Samples showing lower concentrations of Saxitoxin than standard 2 (40 µg/100 g) are considered as negative. Samples showing a higher concentration than standard 2 (40 µg/100 g) are considered positive.

As with any analytical technique (HPLC, LC/MS, etc.), positive samples requiring action should be confirmed by an alternative method.

Importance of Saxitoxin Determination

Saxitoxin, known as "paralytic shellfish poison" (PSP), is one of the toxins produced by several marine dinoflagellates and freshwater cyanobacteria. Contamination of shellfish with saxitoxin has been associated with harmful algal blooms throughout the world.

In humans, PSP causes dose-dependent perioral numbness or tingling sensations and progressive muscular paralysis, which can result in death through respiratory arrest. The maximum guidance level established by the EU and FDA is 80 µg per 100 g of fresh, frozen, or tinned shellfish.

The PSP Shipboard ELISA kit allows for the determination of 42 samples in duplicate determination. The assay can be performed in about 1 hour.

Performance Data

Test reproducibility: Coefficients of variation (CVs) for standards: <10%, CVs for samples: <15%.

Selectivity: This ELISA recognizes Saxitoxin and other PSP toxins to varying degrees:

Cross-reactivities:

Saxitoxin (STX)	100% (per definition)
Decarbamoyl STX	29%
GTX 2 & 3	23%
GTX-5B	23%
Lyngbyatoxin	13%
Sulfo GTX 1 & 2	2.0%
Decarbamoyl GTX 2 & 3	1.4%
Neosaxitoxin	1.3%
Decarbamoyl Neo STX	0.6%
GTX 1 & 4	<0.2%

Cross-reactivities with other classes of algal toxins have not been observed.

General Limited Warranty: Eurofins Abraxis warrants the products manufactured by the Company, against defects and workmanship when used in accordance with the applicable instructions for a period not to extend beyond the product's printed expiration date. **Eurofins Abraxis makes no other warranty, expressed or implied. There is no warranty of merchantability or fitness for a particular purpose**

For ordering or technical assistance contact:

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R092519

PSP Shipboard ELISA Kit, Microtiter Plate

Enzyme-Linked Immunosorbent Assay for the Determination of Saxitoxin (PSP) in Shellfish Samples

Product No. 52255SB

1. General Description

The PSP Shipboard ELISA Kit is an immunoassay for the quantitative and sensitive detection of Saxitoxin. Saxitoxin is one of the toxins associated with paralytic shellfish poisoning (PSP). This test is suitable for the quantitative and/or qualitative detection of Saxitoxin in shellfish. A sample preparation is required (see Sample Preparation, Section E). Positive samples should be confirmed by HPLC, LC/MS, or other conventional methods as appropriate.

2. Safety Instructions

The standard solutions in the test kit contain small amounts of Saxitoxin. In addition, the color solution contains tetramethylbenzidine and the stop solution contains diluted sulfuric acid. Avoid contact of stop solution with skin and mucous membranes. If these reagents come in contact with the skin, wash with water.

3. Storage and Stability

The PSP Shipboard ELISA Kit should to be stored in the refrigerator (4–8°C). The solutions must be allowed to reach room temperature (20-25°C) before use (see Test Preparation, Section F). Reagents may be used until the expiration date on the box.

4. Test Principle

The test is a direct competitive ELISA based on the recognition of Saxitoxin by specific antibodies. Saxitoxin, when present in a sample, and a saxitoxin-enzyme conjugate compete for the binding sites of rabbit anti-saxitoxin antibodies in solution. The saxitoxin antibodies are then bound by a second antibody (anti-rabbit) immobilized on the microtiter plate. After a washing step and addition of the color solution, a color signal is produced. The intensity of the blue color is inversely proportional to the concentration of the Saxitoxin present in the sample. The color reaction is stopped after a specified time and the color is evaluated using an ELISA plate reader. The concentrations of the samples are determined by interpolation using the standard curve constructed with each run.

5. Limitations of the PSP Shipboard ELISA Kit, Possible Test Interference

Numerous organic and inorganic compounds commonly found in samples have been tested and found not to interfere with this test. However, due to the high variability of compounds that might be found in samples, test interferences caused by matrix effects can not be completely excluded.

Samples containing methanol must be diluted to a concentration < 20% methanol to avoid matrix effects.

Mistakes in handling the test can also cause errors. Possible sources for such errors include: Inadequate storage conditions of the test kit, incorrect pipetting sequence or inaccurate volumes of the reagents, too long or too short incubation times during the immune and/or color reaction, extreme temperatures during the test performance (lower than 10°C or higher than 30°C), or exposure to direct or indirect sunlight during the color reaction.

The PSP Shipboard ELISA Kit provides screening results. As with any analytical technique (HPLC, LC/MS, etc.), positive samples requiring action should be confirmed by an alternative method.

Working Instructions

A. Materials Provided

1. Microtiter plate coated with a second antibody (anti-rabbit)
2. Standards (5): 0, 20, 40, 80, 120 µg/100 g, 1 mL each
3. Control at 60 µg/100 g, 1 mL
4. Reagent 1 (Saxitoxin-HRP Conjugate Solution), 6 mL
5. Reagent 2 [Antibody Solution (rabbit anti-Saxitoxin)], 6 mL
6. Wash Buffer (5X) Concentrate, 100 mL, must be diluted before use, see Test Preparation (Section F)
7. Color Solution (TMB), 12 mL
8. Stop Solution, 12 mL

B. PSP Shipboard Accessory Pack Materials PN 530009 (available separately)

1. Diluent in dilution vials with blue stickers, 20, with labels (Dilution 1)
2. Diluent in dilution vials with red stickers, 20, with labels (Dilution 2)
3. 4 mL glass vials with caps, 20, with labels (Sample Extract)
4. Pipette tips, 1 rack of 96, 10-200 µL
5. Plastic transfer pipettes, 20
6. Microtiter plate frame with strip of blank wells (for zeroing reader)
7. Adhesive plate covers, 3
8. Simplified qualitative procedure/flow chart, data sheets (5), graph papers (5)

C. Additional Materials (not provided with the test kit)

1. Fixed volume 50 and100 µL micro-pipettes with disposable plastic tips
2. Deionized or distilled water
3. Squeeze wash bottle with 500 mL capacity (for diluted 1X Wash Buffer, see Test Preparation, Section F)
4. Fisherbrand™ 3D Platform Rotator (Fisher catalog # 88-861-04540) for plate mixing
5. Strip/tube combo reader or microtiter plate reader (wavelength 450 nm)
6. Timer
7. Absorbent paper towels
8. Materials for sample preparation:
   a. Shucking knife
   b. Strainer (#10 sieve)
   c. Plastic tablecloth (to protect work area)
   d. Deionized or distilled water (for rinsing samples prior to homogenization)
   e. Immersion blender or appropriate grinder
   f. 600 mL plastic beaker (VWR 83008-810)
   g. Permanent marker
   h. Conical tubes with caps (VWR # 21008-169)
   i. 125 mL plastic container with lid (VWR # 89202-838) or Ziploc bag
   j. 25 mL disposable plastic pipettes (VWR # 89130-900) with Pipetting device (VWR # 53502-244) or 25 mL graduated cylinder (VWR # 83008-874 or 83008-870)
   k. Paint filters or coffee filters
9. Reagents for sample preparation:
   a. Isopropyl alcohol/white vinegar extraction solution – Combine 5 parts rubbing alcohol (70% isopropyl alcohol) and 2 parts white vinegar (5% acetic acid). Mix thoroughly. Store in a tightly capped container at room temperature.
   b. 10% bleach solution (for cleaning equipment between samples)

D. Sample Collection and Storage

1. Fill out all necessary collection data.
2. Harvest shellfish as follows:
   Note: A minimum of 150 g of meat for each sample should be processed
   a. Blue mussels – 30 mussels per sample
   b. Littleneck clams – 1.5" size – 20 clams per sample
   c. Butter clams - > 3" size – 5 per sample; < 2" size – at least 12 per sample
   d. Surf clams – > 3" size - at least 12 per sample
   e. Other shellfish – at least 20 per sample
3. Verify all data sheets have been completed after sampling.
4. Place shellfish into a plastic Ziploc bag with the data sheet.
5. For on-site or testing within 2 days, store shellfish in refrigerator (2-8°C). For storage greater than 2 days, samples must be homogenized and stored frozen until extraction.

E. Shellfish Sample Preparation, Extraction, and Dilutions

Note: Thoroughly clean the immersion blender and beaker with the 10% bleach solution between samples to prevent contamination.

1. Thoroughly rinse the outside of the shellfish with deionized or distilled water to remove any sand or mud.
2. Open the shellfish with the shucking knife by cutting the adductor muscles. Remove the desired tissue and place in the strainer.
3. Rinse tissue with fresh water to remove any grit or shell fragments. Drain thoroughly (about 5 minutes).
4. Transfer the sample to a 600 mL beaker and puree with immersion blender for 1 minute or until the entire sample is homogenized.
5. Using a 25 mL disposable plastic pipette and pipetting device, transfer 10 mL of the homogenized sample to an appropriately labeled 50 mL conical tube. Transfer the remaining sample to an appropriately labeled plastic container, cap tightly, and freeze.
6. Using a clean pipette or graduated cylinder, add 10 mL of the isopropyl alcohol/white vinegar extraction solution to the 10 mL of sample in the conical tube (1:1 ratio). Cap tightly and shake vigorously for 30 seconds.

Note: If the pipette or graduated cylinder comes in contact with any sample, obtain a new pipette or thoroughly clean the graduated cylinder with the 10% bleach solution before using for additional samples to avoid contamination.

7. Filter the sample extract through the paint/coffee filter into a clean, appropriately labeled plastic container or measuring cup. This extract can then be diluted and tested immediately, stored refrigerated (2-8°C) up to 2 days, or frozen for long-term storage.

Note: The following steps use the materials contained in the PSP Shipboard Accessory Pack.

8. Using a disposable plastic transfer pipette, transfer about 1 mL of the filtered extract to an appropriately labeled 4 mL glass vial.
9. Add 100 µL of the extract to an appropriately labeled blue stickered dilution vial (Dilution 1). Cap and shake well.
10. Add 100 µL of Dilution 1 (from step 9) to an appropriately labeled red stickered dilution vial (Dilution 2). Cap and shake well. Analyze Dilution 2 as the sample (see Assay Procedure, Section H, Step 1)

F. Test Preparation

Micro-pipetting equipment and disposable pipette tips for pipetting the standards, samples, Reagent 1, Reagent 2, color, and stop solutions are necessary. Use only the reagents and standards from one package lot in one test, as they have been adjusted in combination.

1. **Remove the foil bag containing the microtiter plate and all reagents from the kit box. Remove all reagent bottles and vials from the protective foam. Allow the microtiter plate and reagents to sit at room temperature (20-25°C) for at least 2 hours before use.**
2. Remove the number of microtiter plate strips required from the foil bag. The remaining strips are stored in the foil bag and zip-locked closed. After analysis, store the remaining kit in the refrigerator (2-8°C).
3. The standard solutions, Reagent 1, Reagent 2, color, and stop solutions are ready to use and do not require any further dilutions.
4. Dilute the 5X Wash Buffer Concentrate at a ratio of 1:5. Empty the entire contents of the 5X Wash Buffer Concentrate into the squeeze wash bottle and fill to the neck with deionized or distilled water.
5. The Stop Solution must be handled with care as it contains diluted H_2SO_4.

G. Working Scheme

The microtiter plate consists of 12 strips of 8 wells, which can be used individually for the test. The standards must be run with each test. Never use the values of standards which have been determined in a test performed previously.

Std 0-Std 4: Standards
0, 20, 40, 80, 120 µg/100 g

Cont.: Control - 60 µg/100 g

Sam 1, Sam 2, etc.: Samples

	1	2	3	4	5	6	7	8	9	10	11	12
A	Std 0	Std 4	etc.									
B	Std 0	Std 4	etc.									
C	Std 1	Contr.										
D	Std 1	Contr.										
E	Std 2	Sam 1										
F	Std 2	Sam 1										
G	Std 3	Sam 2										
H	Std 3	Sam 2										

H. Assay Procedure

1. Add **50 µL of the standards, control, or sample extract (Dilution 2)** into the wells of the test strips using a fixed volume 50 µL micro-pipette according to the working scheme above. We recommend using duplicate or triplicate wells for each standard, control, and sample.
2. Add **50 µL of Reagent 1** to the individual wells successively using a fixed volume 50 µL micro-pipette.
3. Add **50 µL of Reagent 2** to the individual wells successively using a fixed volume 50 µL micro-pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a circular motion on the benchtop for 30 to 60 seconds. Be careful not to spill the contents.
4. **Place the strip holder on the Fisherbrand™ 3D Platform Rotator and mix at 80 RPM for 30 minutes at room temperature. P**rotect from direct or indirect sunlight.
5. Remove the covering, decant the contents of the wells into an appropriate waste container. Blot the inverted plate on a stack of absorbent paper towels. Wash the strips **four times** using the **diluted 1X wash buffer**. ***Blot the inverted plate after each wash step*** on a stack of paper towels. For each washing step, flood the wells with the diluted 1X wash buffer using a squeeze wash bottle. After the last wash/blot, check the wells for any remaining buffer in the wells, and if necessary, remove by additional blotting.
6. Add **100 µL of color solution** to the wells successively using a fixed volume 100 µL micro-pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a circular motion on the benchtop for 30 to 60 seconds. Be careful not to spill the contents.
7. **Place the strip holder on the Fisherbrand™ 3D Platform Rotator and mix at 80 RPM for 30 minutes at room temperature.** Protect from direct or indirect sunlight.
8. Add **100 µL of stop solution** to the wells in the same sequence as for the color solution using a fixed volume 100 µL micro-pipette.
9. Read the absorbance at 450 nm using a microplate or manual well strip ELISA photometer within 15 minutes after the addition of the stop solution.

Eurofins Abraxis Shipboard Kit Proposal for Method Clarification

Scope:

The Saxitoxins (PSP) Shipboard ELISA Kit, product number 52255SB, is an Approved Limited Use Method for onboard screening of shellfish toxicity in the National Shellfish Sanitation Program (NSSP).[1] In an effort to reduce some reported user challenges, particularly when using the kit in a laboratory setting as opposed to onboard, in meeting quality control (QC) requirements for the Saxitoxins (PSP) Shipboard ELISA kit and support the use of data in making NSSP decisions, Eurofins Abraxis identified two critical steps in the assay procedure that contribute significantly to overall assay binding, as expressed by the absorbance value of Standard 0 (B0). As overall binding increases, the dynamic range of the assay improves differentiation of absorbance values for standards, controls and samples. This enhanced dynamic range supports the ability to achieve QC requirements of the method.

These two critical steps were in the original instructions, yet they were identified as the sources of inconsistency and error among certain users:

1. Allowing kits/reagent to come to room temperature prior to analysis
2. Plate mixing after the addition of reagent

Experiments were performed to identify clearer and more specific instructions that could be included in the product insert to aid users in achieving consistent, successful results.

Experiment 1 – Kit Reagent Temperature

Kits were removed from refrigerated storage and runs were completed at various times after removal.

Amount of Time Out of Refrigerator (minutes)	Standard 0 Absorbance
Experiment 1-No Shaking During Incubation	
0	0.502
30	0.855
60	1.11
Experiment 2-With Shaking During Incubation	
15	1.63
90	1.89

The longer the kit reagents were allowed to come to room temperature before use, the more the binding improved, especially when the incubation was performed with a shaker.

[1] NSSP 2017. NSSP Guide for the Control of Molluscan Shellfish: 2017 Revision. Section IV Guidance Documents, Chapter II Growing Areas, .14 Approved NSSP Laboratory Tests.

Eurofins Abraxis Shipboard Kit Proposal for Method Clarification

Experiment 2- Plate Mixing/Shaking During Incubation Steps

Runs were performed side by side, with one plate placed on a rotating shaker (Fisherbrand™ 3D Platform Rotator Catalog No.88-861-04540) at 80 RPM for the duration of both incubation steps within the assay instructions, while the other plate was left stationary on the bench.

		No Shaking	Shaking
Run 1	B0 Abs	1.265	1.57
	R^2	0.9997	1
	QC	67.1	53.7
Run 2	B0 Abs	1.174	1.433
	R^2	0.9924	0.9993
	QC	57.1	63.1

Active shaking through use of a rotating shaker during the incubation steps improved binding as measured by B0 absorbance.

Conclusions:

Allowing kits to come to room temperature prior to use and adequately shaking plates after reagent addition are two important factors that are directly related to the overall binding. These steps, as currently written in the Saxitoxins (PSP) Shipboard ELISA kit protocol leave some room for interpretation by individual users. It is concluded that user ability to achieve QC requirements might be improved by expanding the current instructions to be more specific for these steps. By specifying the length of time that kits should be removed from refrigerated storage prior to use in the test kit instructions, the risk of running the kit with cold reagents can be reduced. This will help to ensure better binding and improved dynamic range for the assay. Likewise, specifying the use of a rotating mixer at a specific RPM for mixing during incubations also removes the human variables of how long and vigorously users mix plates prior to incubation, leading to greater operator to operator and run to run consistency.