SMR 1550 - 1

WORKSHOP ON THE USE OF RECEPTOR BINDING ASSAY (RBA)

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Co-organized by the International Atomic Energy Agency (I.A.E.A.)

METHOD VALIDATION FOR PSP TOXINS
Canadian Situation

James F. LAWRENCE
Bureau of Chemical Safety, Health Canada, Banting Research Centre
PL 2203G2, Ottawa, Ontario, K1A OL2, Canada

These are preliminary lecture notes, intended only for distribution to participants.

METHOD VALIDATION FOR PSP TOXINS
Canadian Situation

James F. Lawrence
Bureau of Chemical Safety
Health Canada
Banting Research Centre
PL 2203G2
Ottawa, Ontario
Canada K1A 0L2

Health Canada   Santé Canada

Health Canada is responsible for <u>setting regulations</u>, standards and guidelines for marine toxins in fish and shellfish

The Canadian Food Inspection Agency (CFIA) has the responsability to <u>enforce Health Canada regulations</u>

Health   Santé
Canada   Canada

Health Canada needs to assist the CFIA by developing and validating analytical methods to enforce our regulations

Health Canada's research on marine toxins:

- ❖ **Simple validated methods for PSP and ASP**

- ❖ **DSP toxins: pectenotoxins and DTX toxins**

- ❖ **Spirolides**

- ❖ **Others: ciguatoxins, azaspiracids, gymnodimine**

- ❖ **Freshwater toxins: microcystins, anatoxins**

Health   Santé
Canada   Canada

PRESENT SITUATION WITH PSP TOXINS IN CANADA

❖ **In Canada, the AOAC Mouse Bioassay is written in our regulations as the method to use for enforcing the PSP maximum acceptable concentration of 80μg/100g saxitoxin equivalents**

❖ **The CFIA have been using this method for more than 30 years**

❖ **Presently, about 30,000-50,000 mice are used per year for 10,000-15,000 analyses for PSP in shellfish**

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Canada   Canada

HEALTH CANADA IS WORKING TOWARDS REMOVING REFERENCE TO THE AOAC MOUSE BIOASSAY IN OUR REGULATIONS

This requires : 📁 **Development of a validated / collaboratively studied alternative method**

📄 **A legal change to our regulations**

📄 **Implementing new testing methods / technology into the CFA inspection laboratories**

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DEVELOPMENT OF VALIDATED / COLLABORATIVELY STUDIED ALTERNATIVES

Over the past 15 years, Health Canada has foscussed its research efforts on HPLC methods for marine toxins

➢ Successfully completed an AOAC collaboratively studied method for domoic acid (ASP)

➢ Developed alternative HPLC methods for domoic acid, okadaic acid, microcystins and anatoxin

➢ Last year completed and submitted to AOAC an HPLC collaborative study for PSP toxins in shellfish that can be used for screening and quantitation

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IMPLEMENTING NEW ANALYTICAL METHODS IN ROUTINE TESTING LABORATORIES

- **Introduce new methods slowly – some resistance to new technology**

- **In parallel with current mouse bioassay testing**

- **In one lab only at first**

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RAPID SCREENING VERSUS QUANTITATIVE METHODS

- Both are important and have their place
  - Cost
  - Reliability / Accuracy
- False positives / negatives / interferences
  - Speed
- Effective use of methods, combinations

Health Canada   Santé Canada

Comparison of collaborative study with mouse bioassay and mist alert test results

MATRIX	HPLC ANALYSIS		MOUSE BIOASSAY	MIST ALERT
	Total PSP (µg/100)	Total PSP as STX Equivalent (µg/100)	(µg/100)	+ / -
Mussel	634	310	380	+
Scallop	338	185	180	+
Clams	364	156	120	+
Oyster	Nd		<42	-
Mussel	Nd		<42	-
Clams	104	65	55	+
Clams	Nd		<42	-
Mussel	256	36	<42	+
Oyster	74	29	<42	+
Oyster	140	54	<42	+
Mussel	93	64	76	+
Mussel	372	149	190	+
Mussel	53	21	<42	+
Mussel	592	91	73	+

PRESENT SITUATION WITH THE AOAC COLABORATIVE STUDY APPROVAL PROCESS

❖ **AOAC International validation of collaborative studies is of highest quality and their official methods are used by regulatory agencies around the world including Canada**

❖ **Two years ago, AOAC Int. changed its policy on collaborative study approval.**

❖ **AOAC now requires a minimum of $35,000 US to review and officially approve completed collaborative studies. For an additional fee, they will organize and conduct collaborative studies**

❖ **Laboratories / organizations must consider this cost before new collaborative studies are initiated**

❖ **AOAC Int. general referees will assist collaborative study leaders in finding financial support**

Health Santé
Canada Canada