

Marine biotoxin sampling: preconcentration and passive sampling; Environmental data to support aquaculture decision-making



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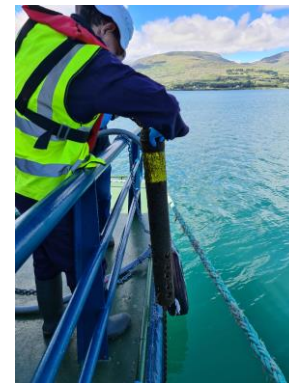
Halpin, NMCI, MTU - Cork  
**Mike Griew,**

STREAM Conference – Waterford 21<sup>st</sup> June 2023



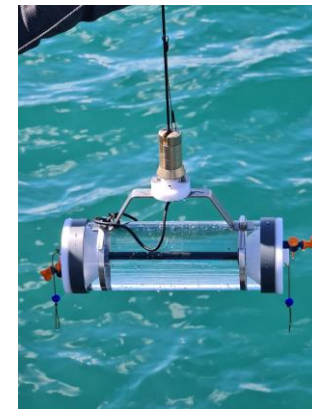
# Introduction

- Overview of MTU contribution to STREAM
- Aquaculture, Algae and Marine Biotoxins
- Polymer Resin Absorbent beads, aka ‘Solid Phase’, biotoxin trap principles – live demo.
- SPATT monitoring.
- STREAM ‘Smart’ pumps.



## Work Package 5

- Multiparameter sonde deployment and water quality sampling program at Castletownbere.
- SPATT (Solid Phase Adsorption Toxin Tracking) bag deployment - Castletownbere.
- Profile toxin level profiling (SPATT bags and vertical hauls) using LC-MS
- ‘SMART’ pump marine toxin preconcentration system development.
- Environmental data gathering system development: precipitation monitoring, water column data backhaul.



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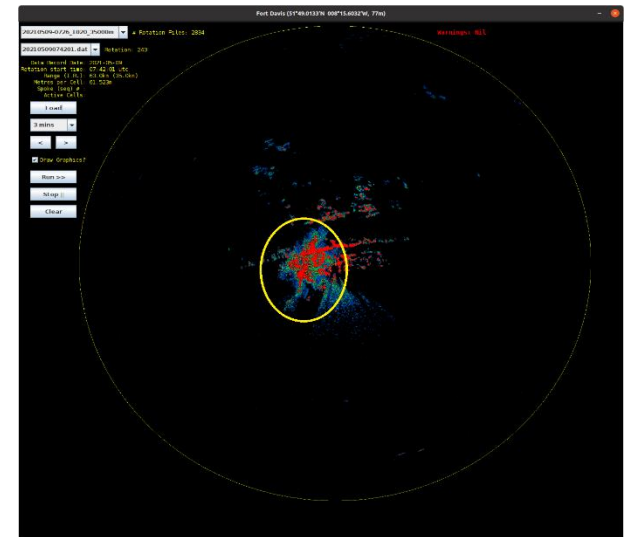
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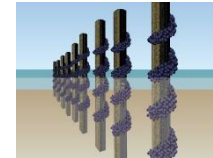
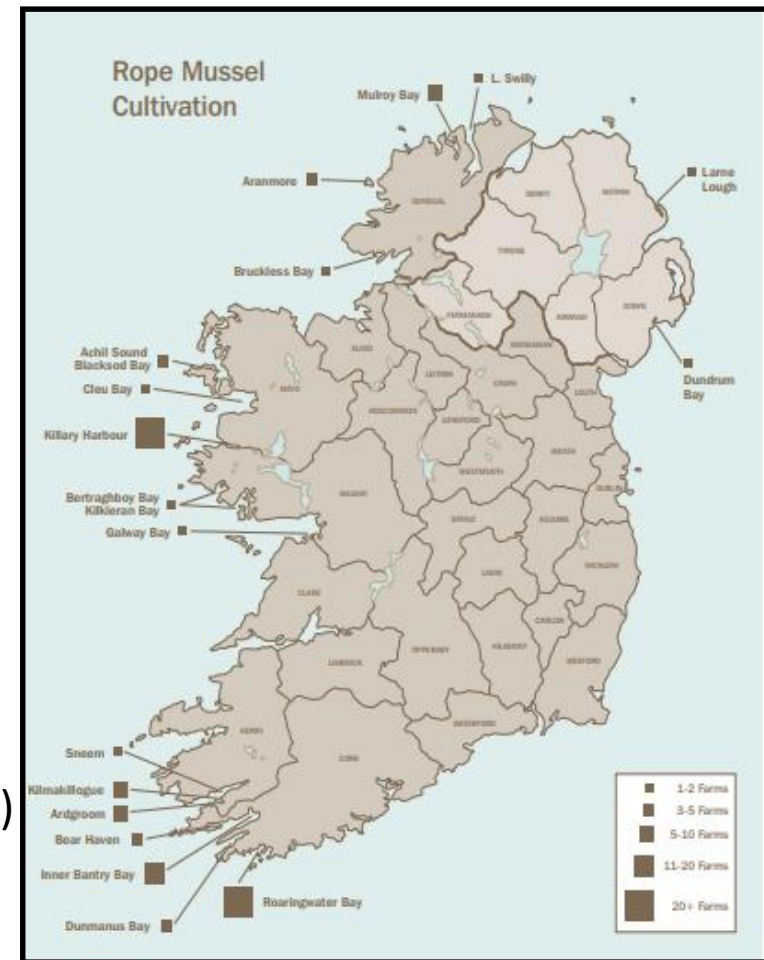
# Irish Aquaculture

The total market value of Irish Seafood in 2016 was €943 Million, of which €563 Million (60%) was due to exportation of produce (Bord Bia, 2017)

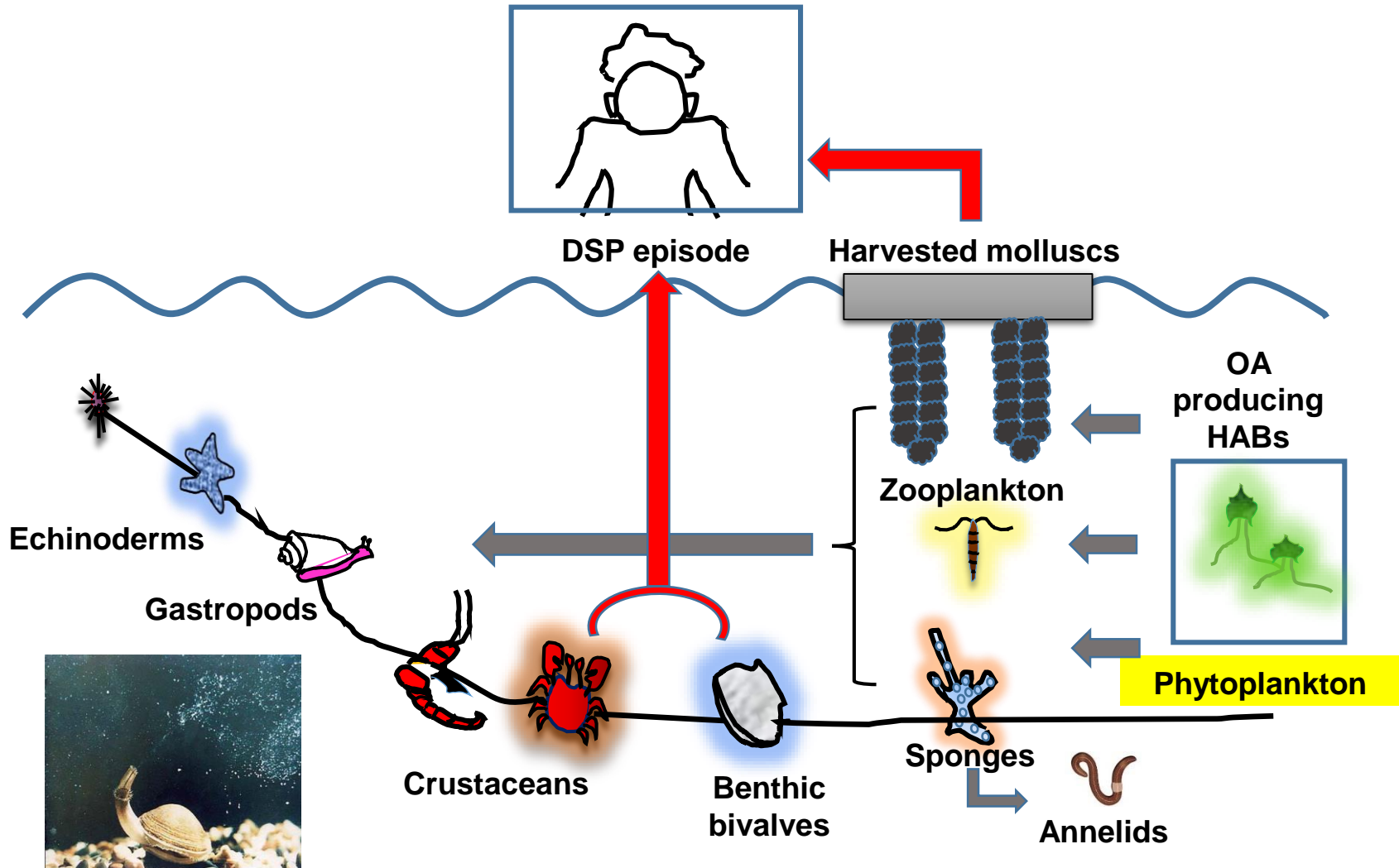
As of 2017, Ireland had an 8% share in mussel production – scientific name *Mytilus edulis* (Bord Bia, 2017).

Mussel industry is split into two main production techniques:

1. Bottom mussel (naturally growing on the seabed and harvested by specialised dredging equipment)
2. Rope mussels (cultivated on rope structures in an aquaculture environment) (Bord Iascaigh Mhara, 2006)



# Bioaccumulation of marine toxins



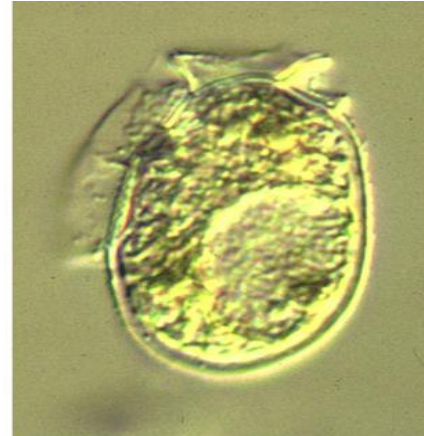
# Algal Blooms



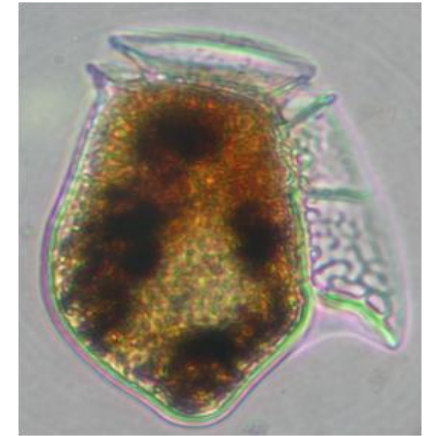
*Prorocentrum lima*



*D. fortii*



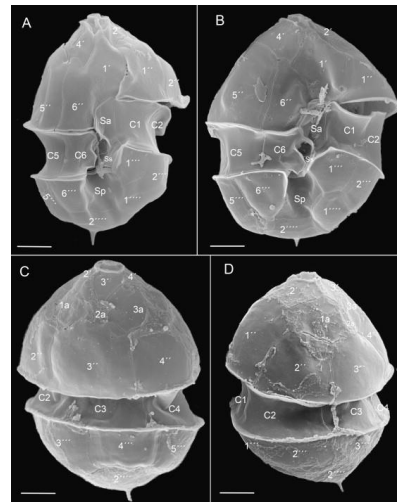
*D. acuminata*



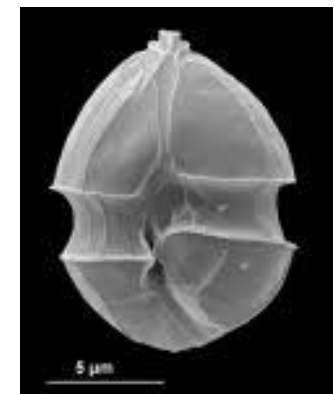
*D. acuta*

## Marine Algae Effects:

- **Positive**
- **Negative**



AZA producer: *Azadinium* spp.



*Amphidoma languida*

# Harmful Algal Blooms (HABs)

- Algae populations can increase rapidly: “Red Tides”
- Blooms are *not always* visible
- Phytoplankton generally proliferate in Summer when the water is calm and warmer.



# Marine Toxin Syndromes

- Five major toxic syndromes:
  - Diarrhetic Shellfish Poisoning (DSP)
  - Azaspiracid Shellfish Poisoning (AZP)
  - Amnesic Shellfish Poisoning (ASP)
  - Paralytic Shellfish Poisoning (PSP) **Saxitoxin**
  - Neurotoxic Shellfish Poisoning (NSP) **Brevetoxins**



# Marine toxins

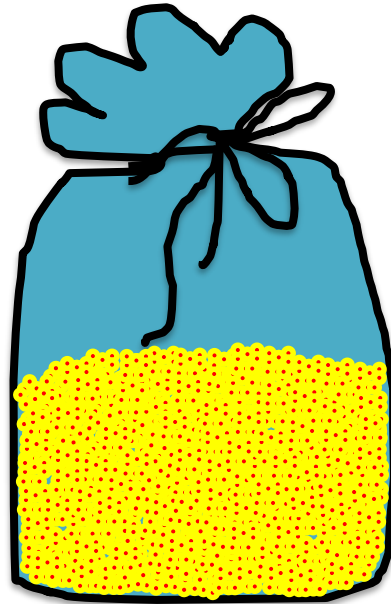
	Toxin group	Syndrome
Hydrophilic toxins	Domoic acid	ASP
	Saxitoxins	PSP
	Tetrodotoxin	TTX poisoning
	Palytoxin	Palytoxin poisoning

	Toxin group	Syndrome
Lipophilic toxins	Okadaic acid	DSP
	Dinophysistoxins	
	Pectenotoxins	
	Saxitoxin	PSP
	Azaspiracids	AZP
	Cyclic imines	Not known

# Early Warning: Solid Phase Adsorption Toxin Tracking (SPATT)

- *In-situ* adsorption sampling can warn when harmful algal bloom imminent.

'SPATT' bag



Permeable Bag



Polymer resin beads ('solid phase'):  
e.g. DIAION HP20, SEPABEADS SP700 or  
Amberlite.

[Solid phase adsorption toxin tracking \(SPATT\): a new monitoring tool that simulates the biotoxin contamination of filter feeding bivalves.](#)

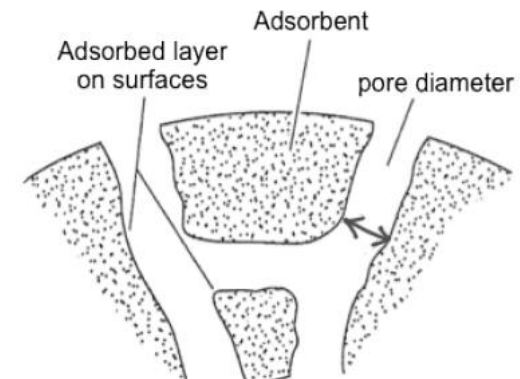
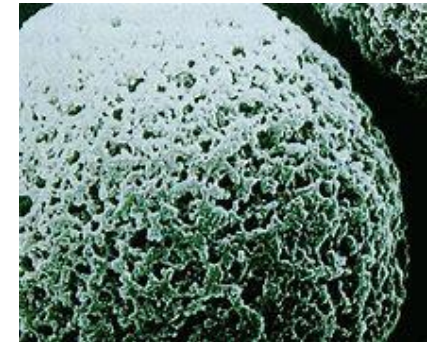
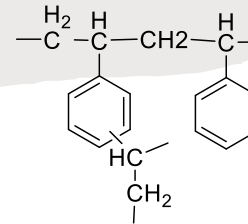
MacKenzie L, Beuzenberg V, Holland P, McNabb P, Selwood A.

Toxicon. 2004 Dec 15;44(8):901-18.



## Structure and physical characteristics

- What are resin adsorbents?
- How do they work?
- How are they used to concentrate biotoxins?





# Polymer Resin Adsorbents

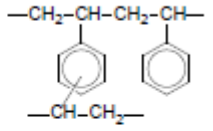
## HP20 Datasheet...

- What are resin adsorbents?
- How do they work?
- How are they used to concentrate biotoxins?

Product Data Sheet No.01-10-A-0103

### DIAION™ HP20

DIAION™ HP20 is based on a unique rigid polystyrene/divinylbenzene matrix. A controlled pore size distribution and large surface area offer excellent resolution and the capacity for a wide range of molecules, from small peptides and oligonucleotides up to large proteins.

Grade Name	DIAION™ HP20	
Bead Form	Spherical, porous	
Matrix	Polystyrene/divinylbenzene	
Chemical Structure		
Shipping Density*	g/L	690
Water Content	%	55 - 65
Particle Size Distribution thr. 250 µm	%	10 max.
Effective Size	mm	0.25 min.
Uniformity Coefficient	-	1.6 max.
Particle Density*	g/mL	1.01
Specific Surface Area*	m <sup>2</sup> /g	590
Pore Volume*	mL/g	1.3
Pore Radius*	Å	290

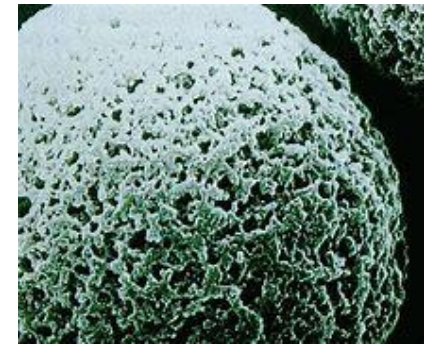
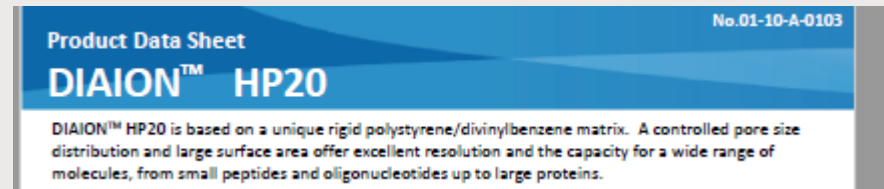
\* Made - measurement value is based on IUPAC and international data.



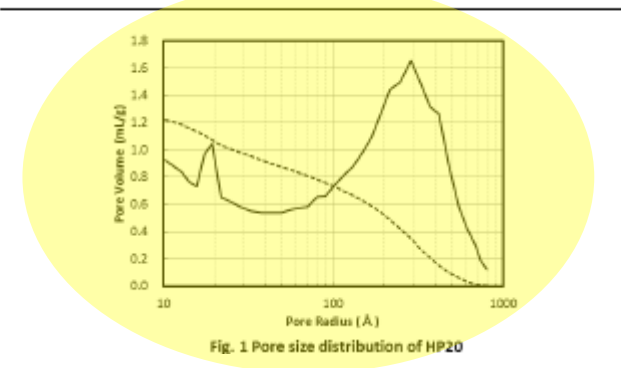
# Polymer Resin Adsorbents

## HP20 Datasheet...

- What are resin adsorbents?
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- How are they used to concentrate biotoxins?



Pore size distribution



# Polymer Resin Adsorbents

- What are resin adsorbents?
- How do they work?
- How are they used to detect biotoxins?

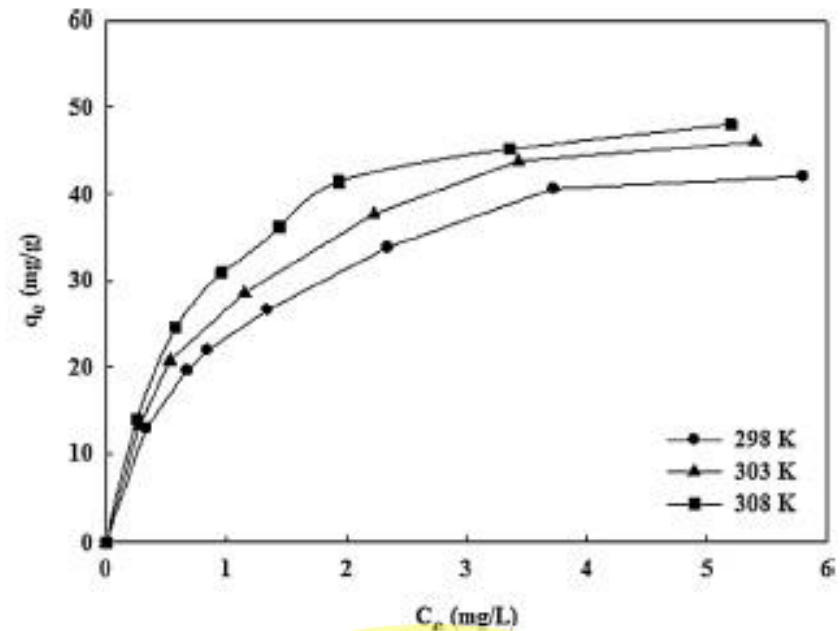
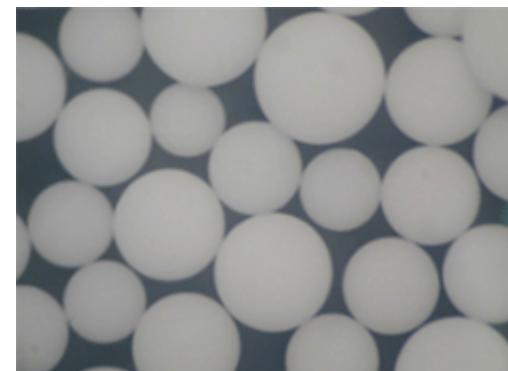
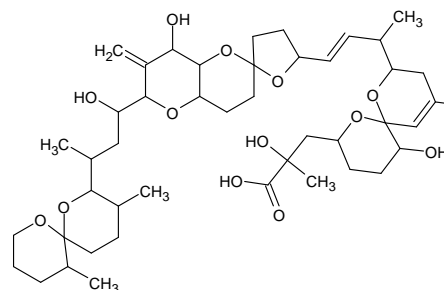
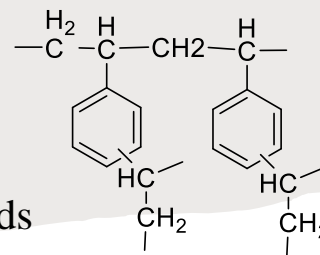


Fig. 2. Adsorption isotherm for paclitaxel onto HP-20 at different temperatures.

# Polymer Resin Adsorbents

## Amberlite XAD-7HP and SEPABEADS SP700

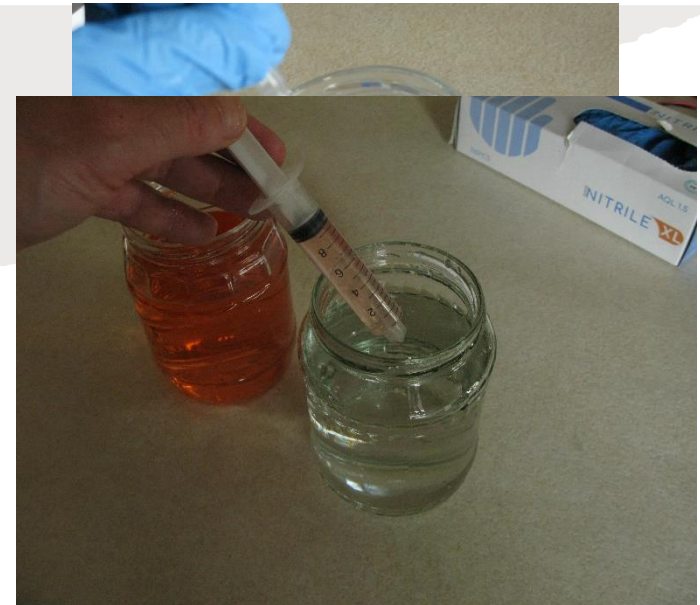
- **XAD-7 Lipophilic shellfish toxin sorbent**
- **SP700 for pharmaceutical applications, compounds up to 60,000 MW (molecular weight).**
- Mixed-mode phases (cross-polarity range of compounds, simultaneous preconcentration).



# Polymer Resin Adsorbents

## Practical session AFTER LUNCH

1. Bulk Adsorption (SPATT bag, 'IAV')
2. Large volume, fast flow  
preconcentration ('Smart Pump', '7-  
Day-Rig' technique)

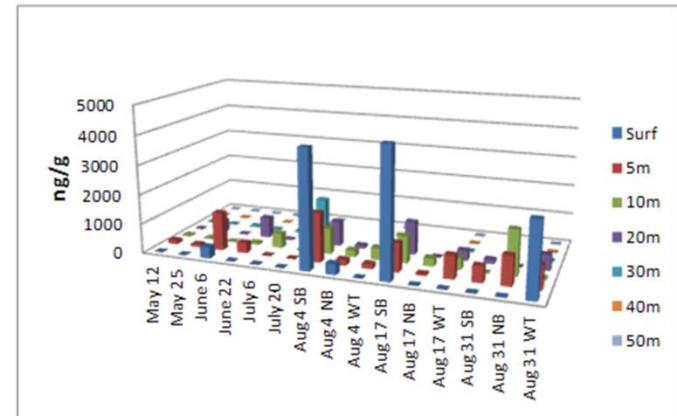


# Passive sampling method (SPATT)

## Spatial and temporal analysis of marine biotoxins



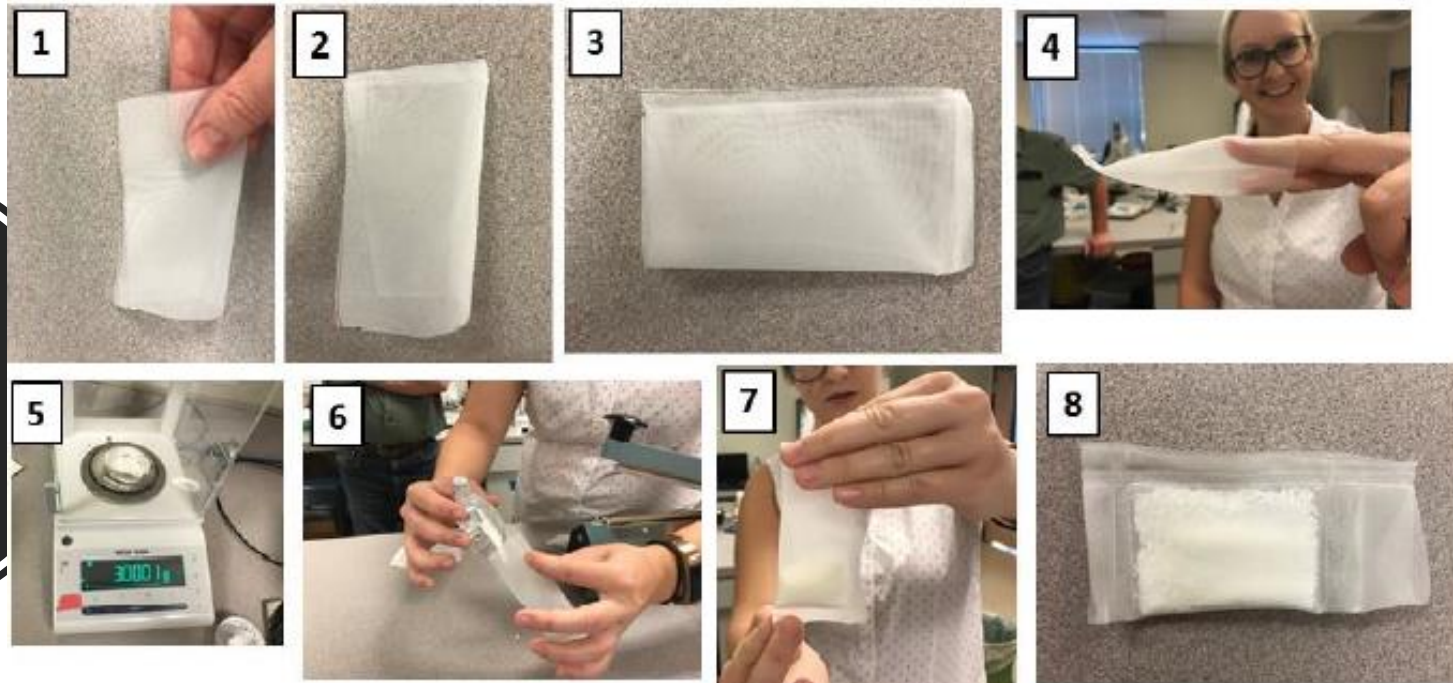
Lough Hyne Marine Nature Reserve, West Cork, Ireland



The distribution of Okadaic acid (DSP) at 7 depths over 4 months

Based on method originally devised by MacKenzie *et al.* 2004

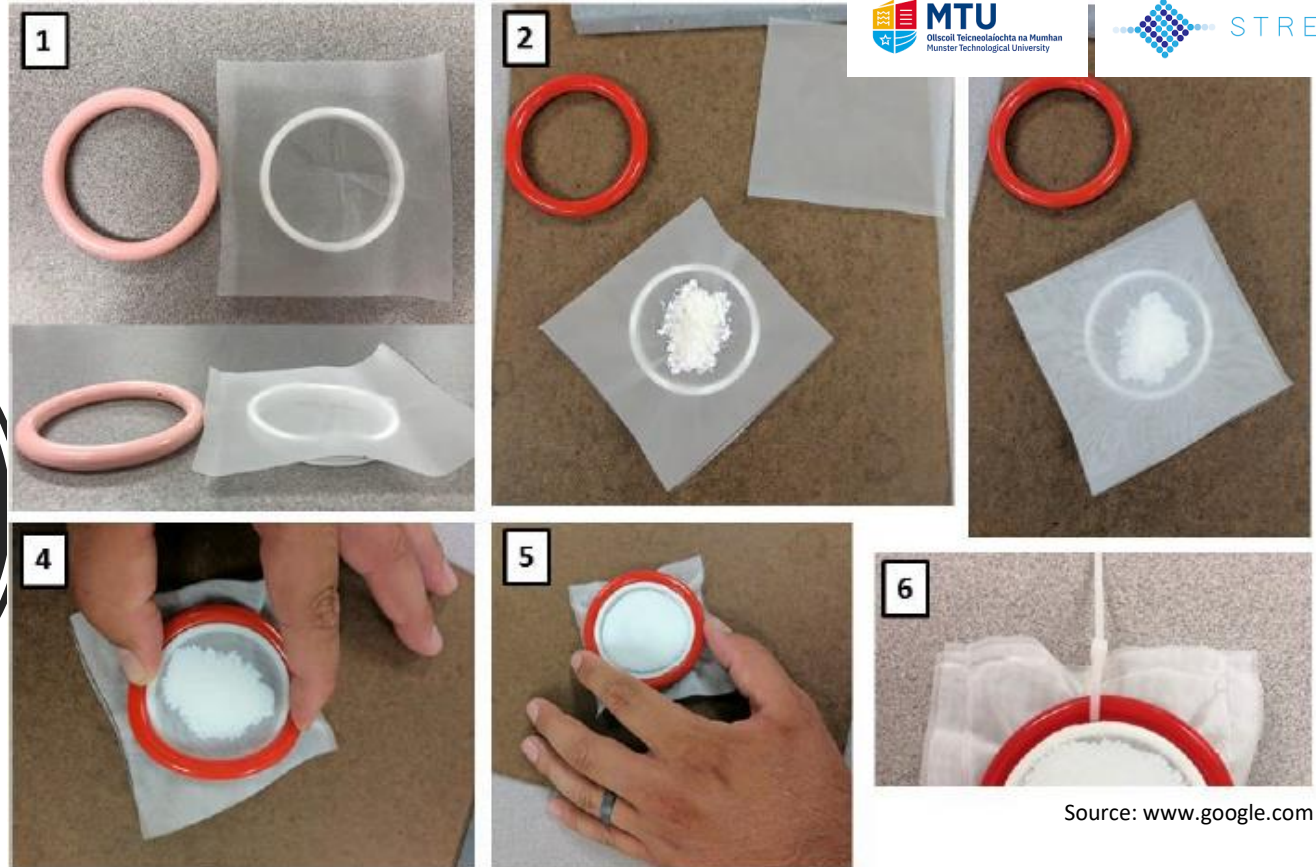
SPATT  
preparation-  
Tea bag  
design



Source: [www.google.com](http://www.google.com)

Bag's dimension : 4.5" X 4.5"  
Made from 95  $\mu\text{m}$  polyester mesh

**SPATT**  
 preparation  
 using  
 embroidery  
 ring



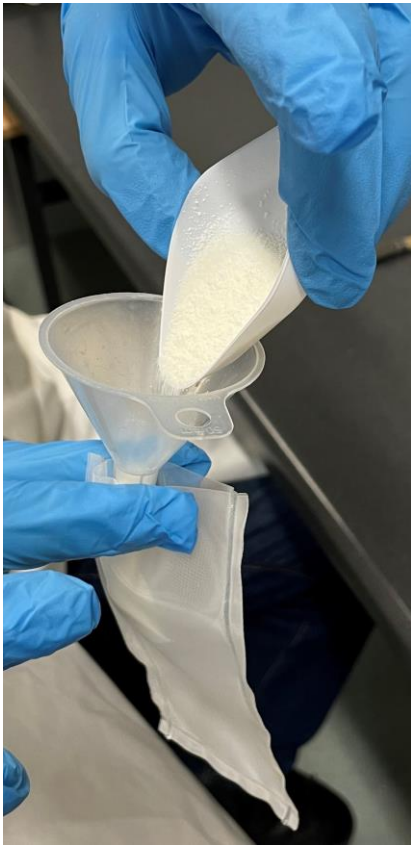
Source: [www.google.com](http://www.google.com)

To form a thin layer of resin Rundberget *et al.*\*, placed the resin between two layers of nylon mesh that was clamped tightly into a frame.

**\*T. Rundberget, et al, *Toxicol 50* (2007) 960–970**



# Redesigned SPATT bags



# Previous Project\_Active toxin sampling

## Bio-harvesting of algal toxins

Sampling

Extraction

Clean-up

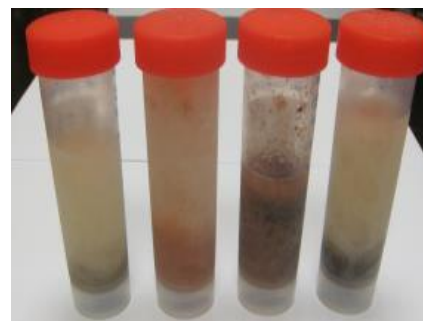
Detection  
LC-MS

Chromato-  
graphy

Ion-  
suppression

Validation

Data  
processing



# 'Traditional' method - collect bulk samples of phytoplankton

Pumping water at specified depths into a phytoplankton dual net

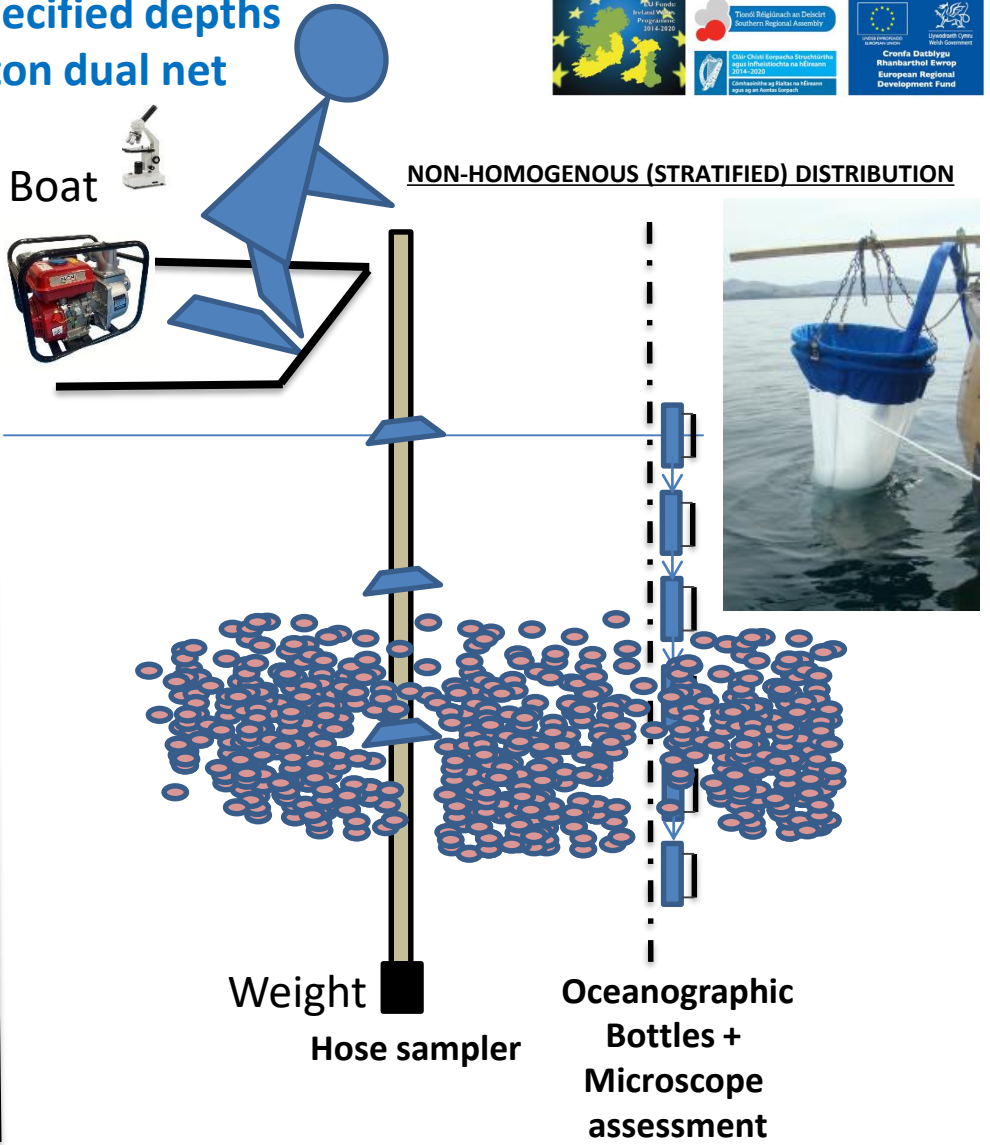
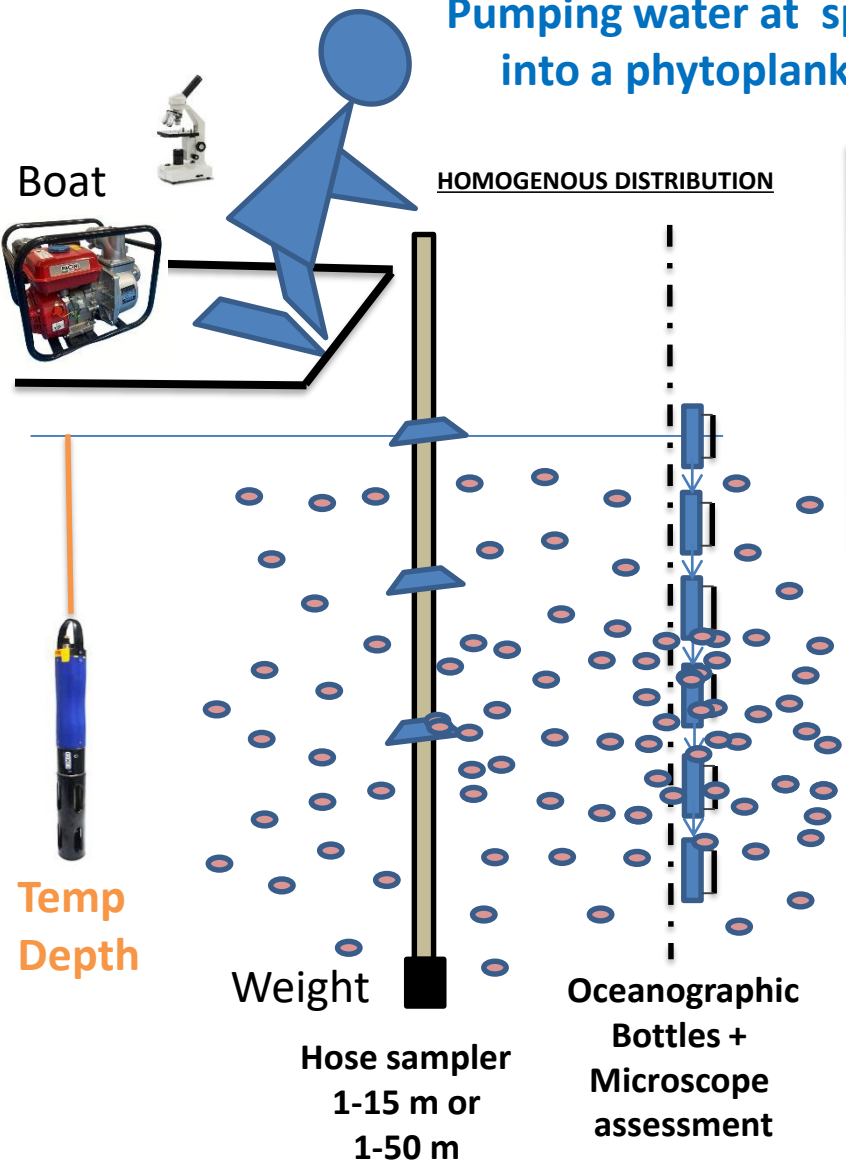
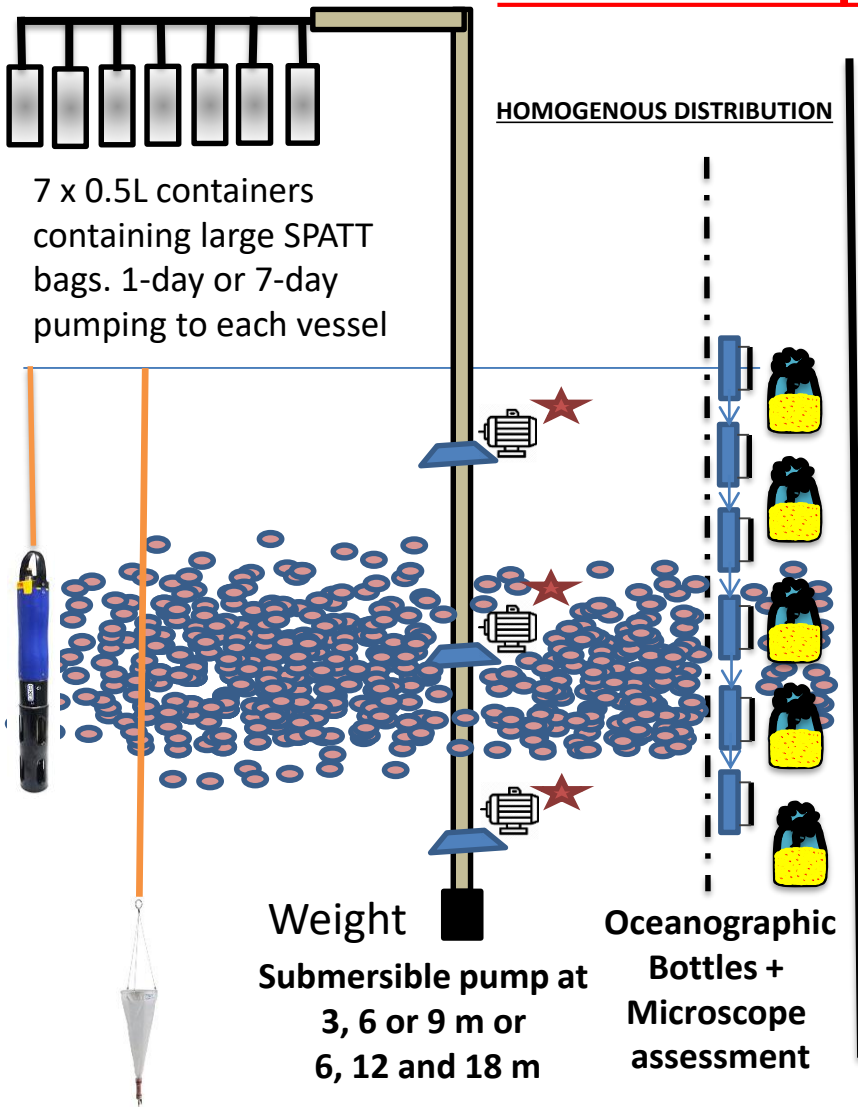


Figure: On-board Pump + Hose (dropped to various depths)

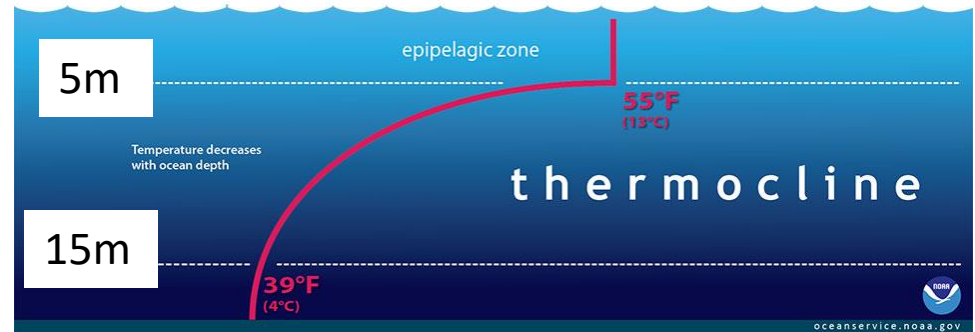
# 'Modular Bulk Sampling' method on BIM barge (Castletownbere) to collect bulk phytoplankton – submersible pumps with sensors

## BIM Barge



### 1. 7-day Rig: Pumping water at 3 specified depths

Elie Fux et al. 2010 Toxicon "Production of diarrhetic shellfish poisoning toxins and pectenotoxins at depths within and below the euphotic zone" – Section 2.3.1 - Water samples were obtained every 3 h at the maximum chlorophyll layer located just above the thermocline using the profiler's peristaltic pump and filtered over a 200  $\mu\text{m}$  and a 20  $\mu\text{m}$  mesh. The depth of the layer varied from 22 to 34 m depending on the time of the day (SW coast of Ireland).

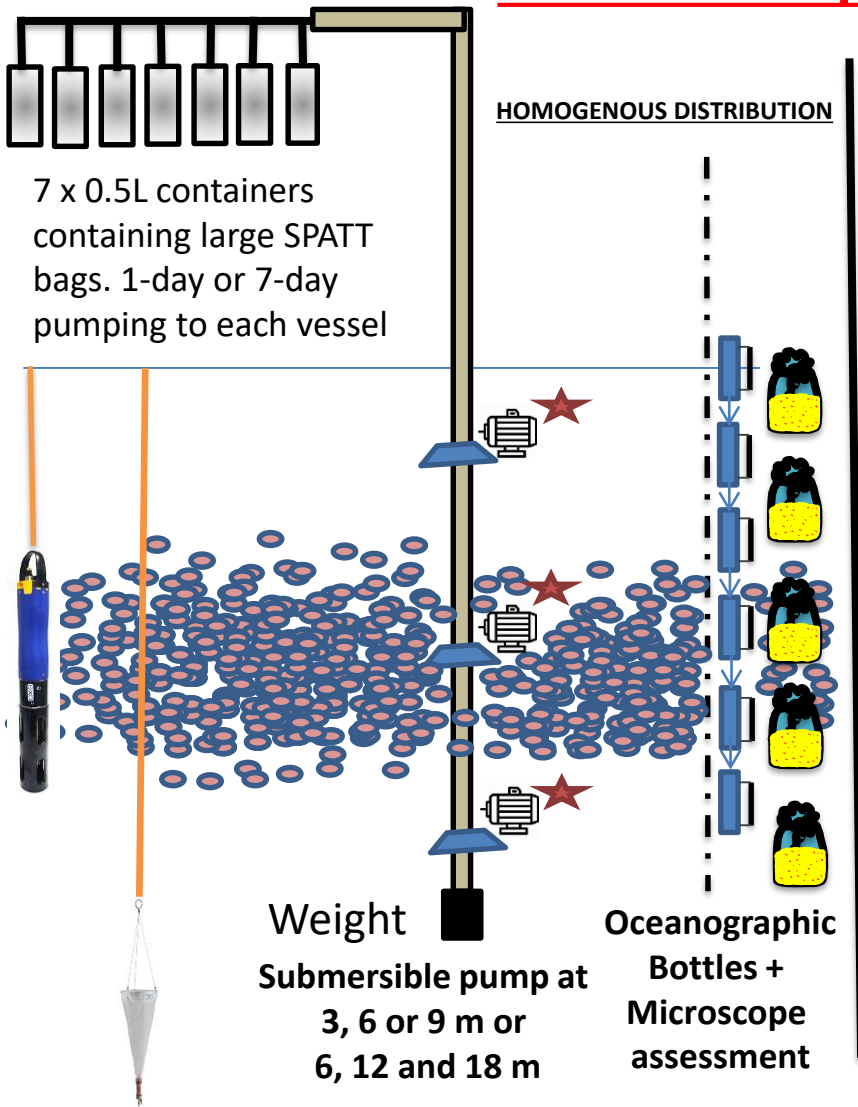


2. SPATT bags – 3 g of adsorbent each
3. Water samples collected for LC-MS analysis
4. Phytoplankton vertical hauls
  - Once every two weeks



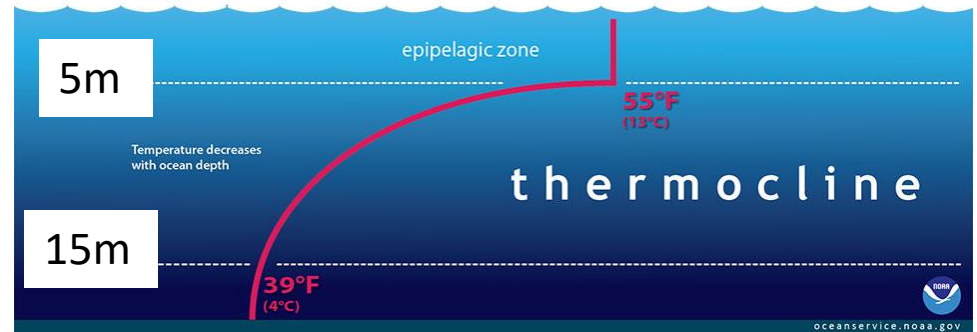
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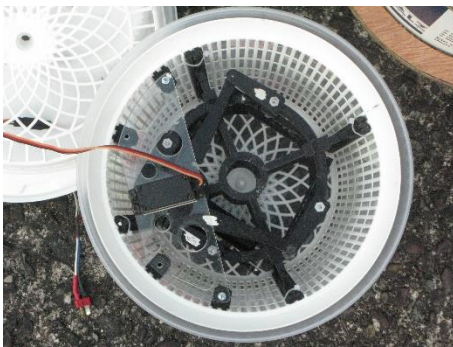
# Modular Bulk Sampling: '7-Day-Rig'

1. Sample from three depths simultaneously (e.g., 5,10,15m)
2. Seven independent collection capsules
3. Temporal sampling distribution or different phase types.
4. Preconcentration of water-borne toxin (accumulator tank, sampling water drawn from between coarse and fine plankton net, differential feed and sampling pump rates).
5. In-line self-cleaning (purging) filter.
6. Remote control and monitoring with industry standard techniques (SCADA via ModBus and GSM communication).
7. Portable: 24V DC (potentially battery operated).



# Bulk Sampling: 'I.A.V.'

1. Sample large water volume brought to the surface over several hours.
2. Sample with up to four different phases simultaneously.
3. Apply agitation strategy to optimize adsorption.
4. Portable, battery operated.



# Bulk Sampling: Submersible Pump Sample Concentrator

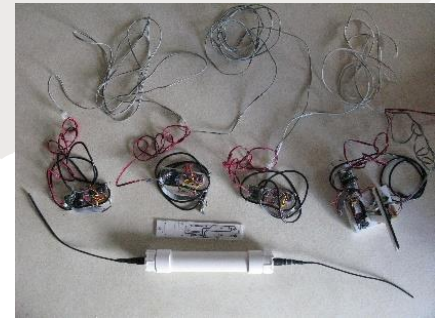
1. **High flow - large water volume sampled in-situ.,**
2. **Large quantity of phase, sample captured when algal bloom event suspected.**
3. **Inexpensive, commonly available components (off-the-shelf AC pump).**





# REMINDER:

1. **SEAMOTE Water Column Sensors**  
(live data to MQTT broker).
2. **HP20 Adsorbency demonstration!**



# THANK YOU FOR YOUR ATTENTION!



STREAM is Part-Funded by the ERDF via the Ireland  
Wales Programme.