

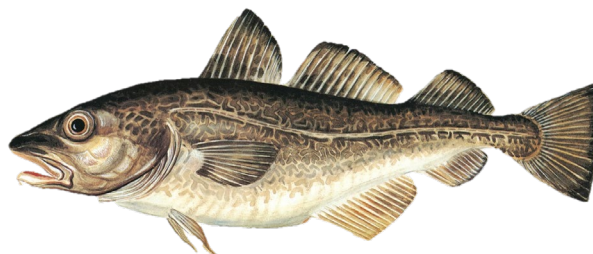
# Extracting microplastics from different environmental matrixes

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## Different sample types require different processing methods



### SAMPLING

non-reduced bulk sample

volume reduced sample

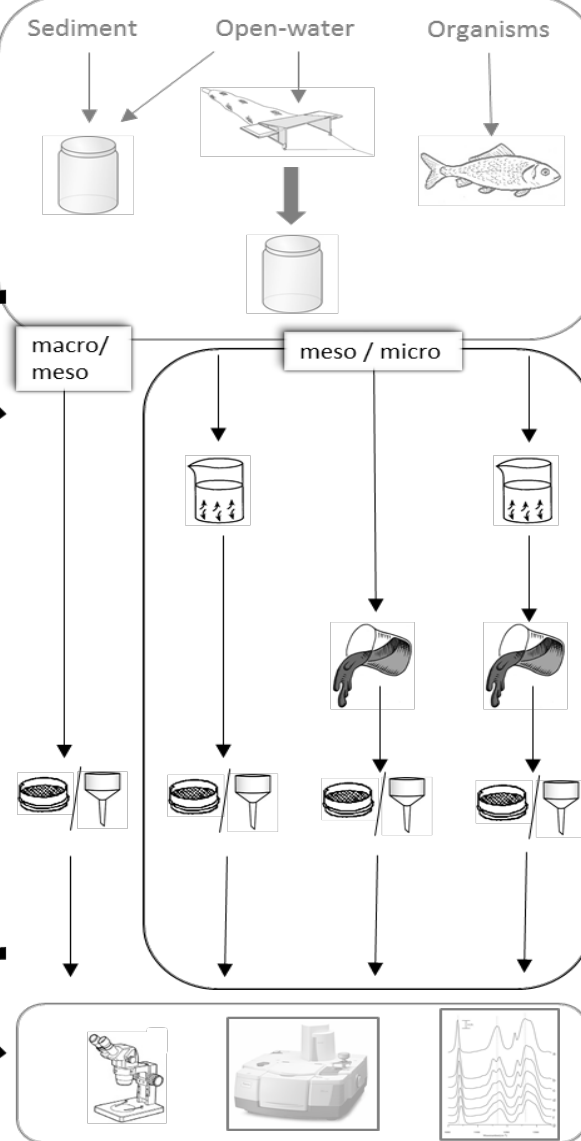
### SAMPLE PROCESSING

density separation

biological/chemical digestion

sieving/filtering

### PARTICLE ANALYSIS



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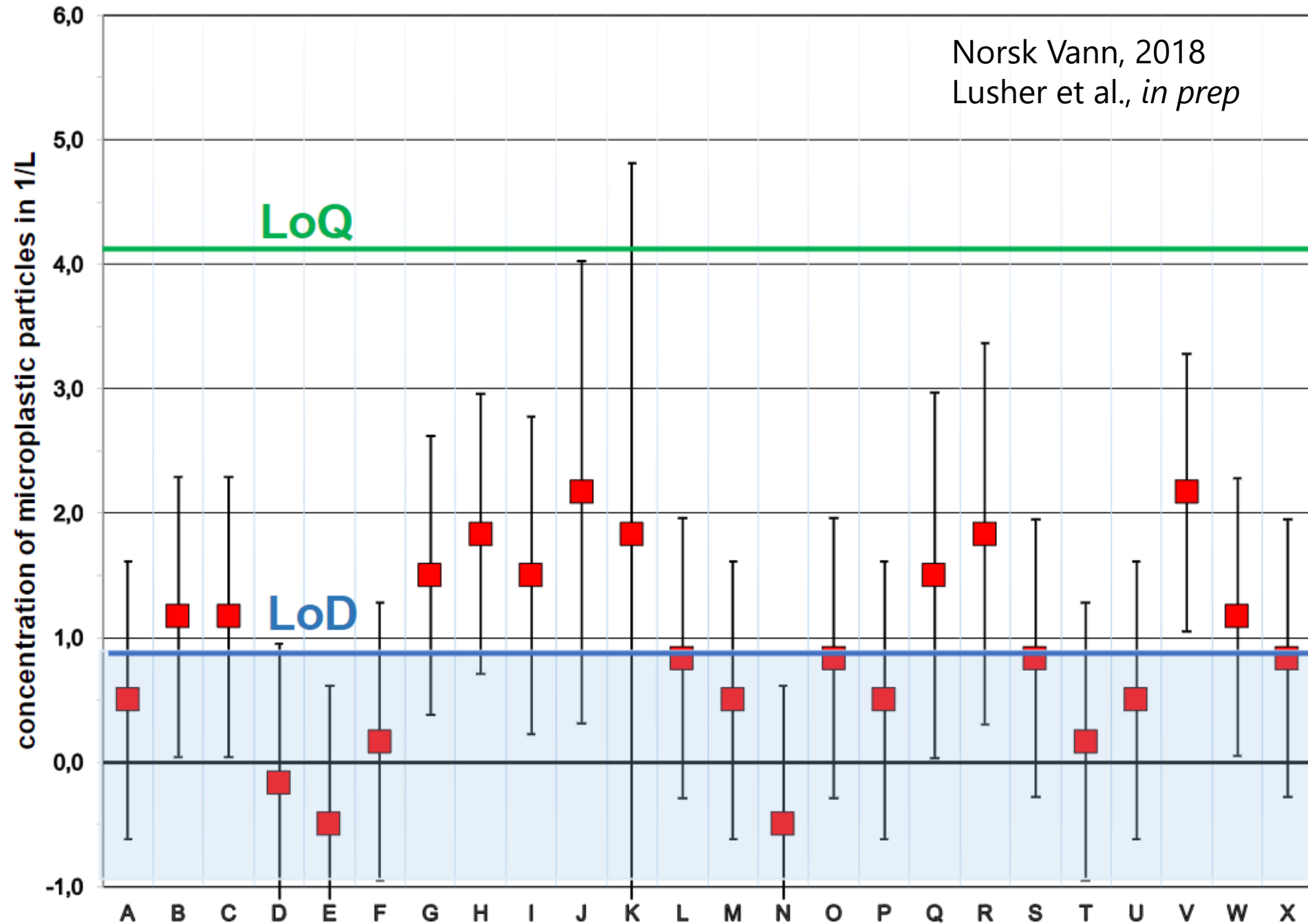
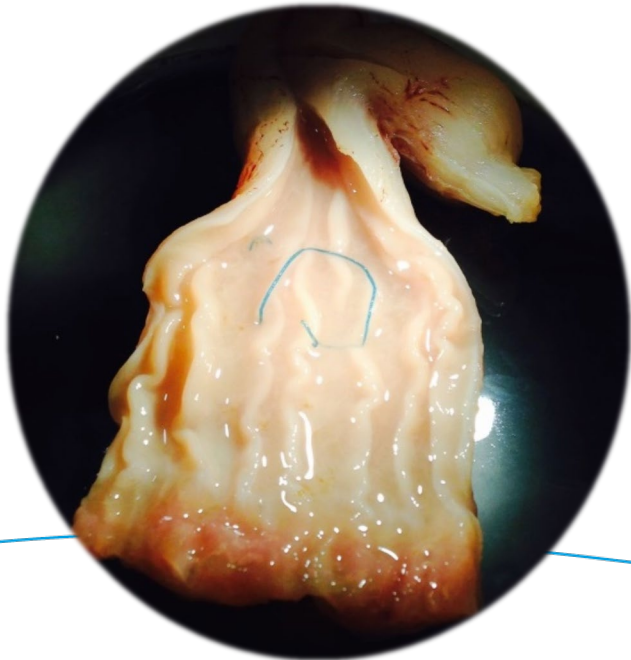
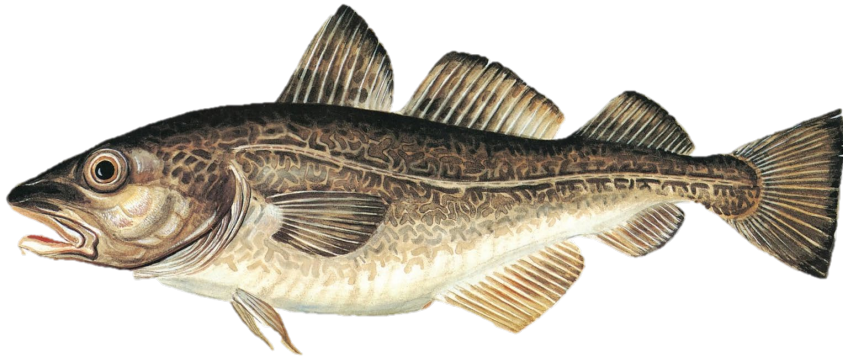


Figure 5: Measured microplastic concentrations in the raw water of the waterworks A-X, and 67% confidence intervals.

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## Biota: Dissection

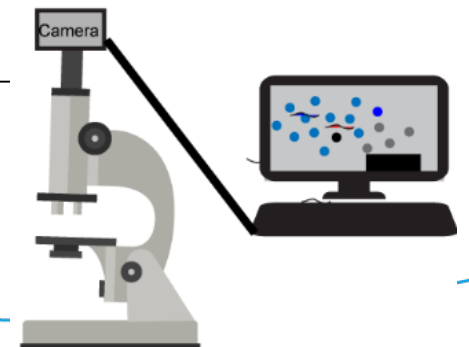
Simple visual sorting on a microscope

Investigation of stomachs and intestines is relevant for microplastic >0.5 mm in size

Must support with FT-IR or similar

Monitor for airborne fibers

Remember contamination control throughout!





## High plankton content

Too much plankton to just filter

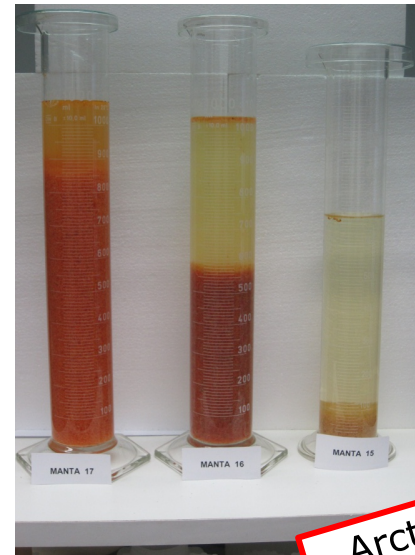
Can use gravity and density separation

Leave to stand

Pour off the supernatant, filtering it

Remaining material can be processed further

*Isobe et al., ILC study, MPs in seawater. Under review, MPB*



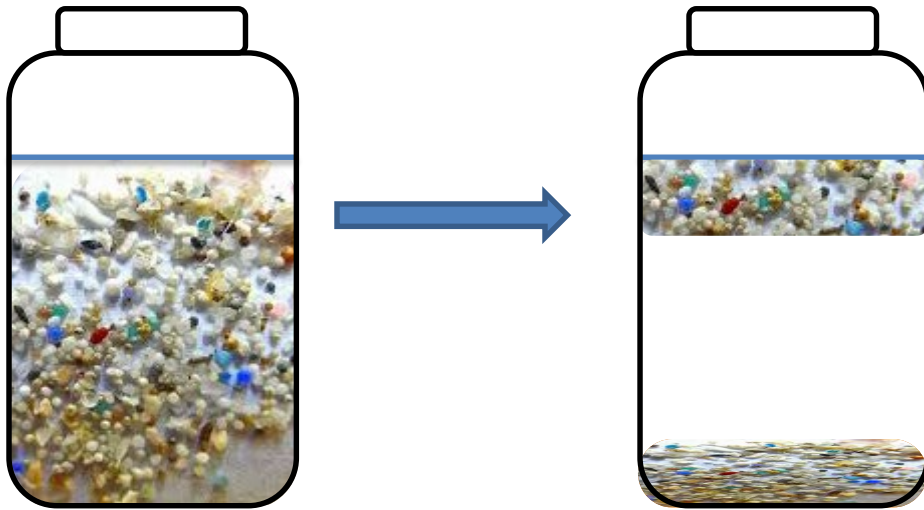
Arctic = highly  
productive  
LOTS OF  
PLANKTON



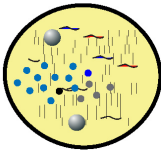
SCIENTIFIC REPORTS

*Lusher et al., 2015, Micropalstics in Arctic polar waters*

## Density separation

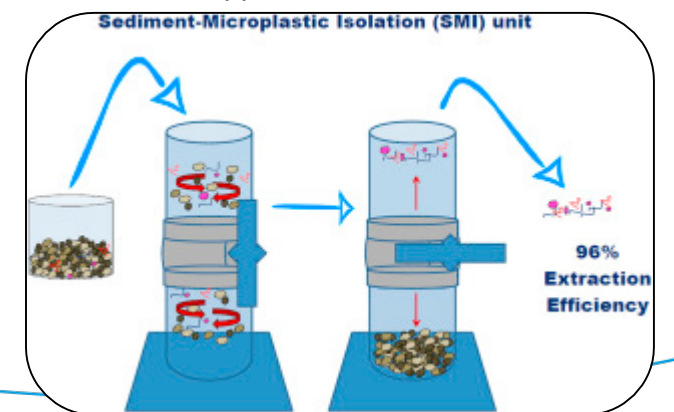


GF/D filter (2.7µm)



Salt	Density (g cm <sup>-3</sup> )
Sodium Chloride (NaCl)	1.2
Sodium Polytungstate (PST)	1.4
Sodium Iodide (NaI)	1.6-1.7
Zinc Chloride (ZnCl <sub>2</sub> )	1.6-1.7

Coppock et al. 2017



## Acid digestion

Strong mineral acid e.g.  $\text{HNO}_3$ ,  $\text{H}_2\text{SO}_4$

Strong oxidising agents:

- Peroxydisulfate
- Hydrofluoric acid
- Hydrochloric acid

Lower strength acids e.g.  $\text{HCl}$

Based on recommendation of Lusher et al., (2017), GESAMP advised against this method

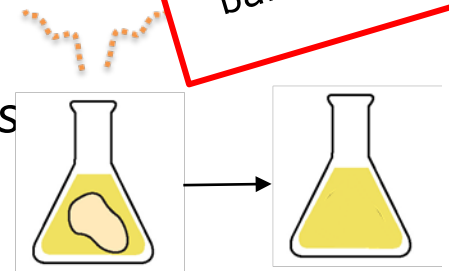
## Oxidative Digestion

Fenton's reagent:

30%  $\text{H}_2\text{O}_2$  with  $\text{Fe}^{2+}$  catalyst

Keeps temperature at optimum

Ice bath to prevent overheating ( $<40^\circ\text{C}$ )

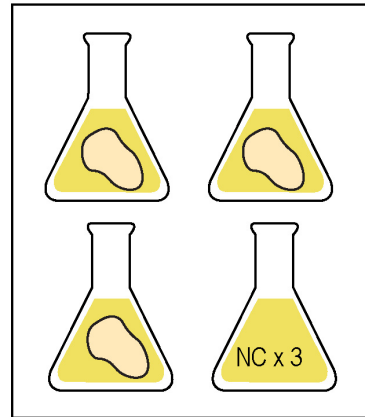


*Hurley et al. 2018, Validation of a Method for Extracting Microplastics from Complex, Organic-Rich, Environmental Matrices. ES&T*

## Alkaline digestion

Optimized protocol

Can be neutralized using HCl



10% KOH



145 rpm



60°C



24 h

NC: Negative Control x 3

Bråte et al. 2018, *Mytilus spp. as sentinels for microplastic pollution*, *Enviro. Poll.*

## Enzymatic Digestion

Enzymes break down lipids, proteins, cellulose, chitin etc.

Different sequences for use may be required

e.g., Basic Enzymatic Purification Protocol – BEPP, (Löder et al., 2017)

Time-consuming and costly

Chitinase

Cellulase

Protease

Protein



Peptides



a. PP + Protocol 1b  
 $H_2O_2$  (70°C)



b. PET + Protocol 3b  
10 M NaOH



c. PC + Protocol 3b  
10 M NaOH



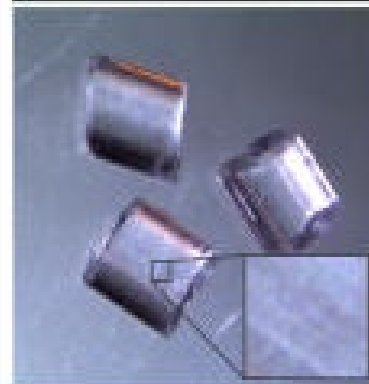
d. PS + Protocol 1a  
 $H_2O_2$  (60°C)



e. PS + Protocol 1b  
 $H_2O_2$  (70°C)



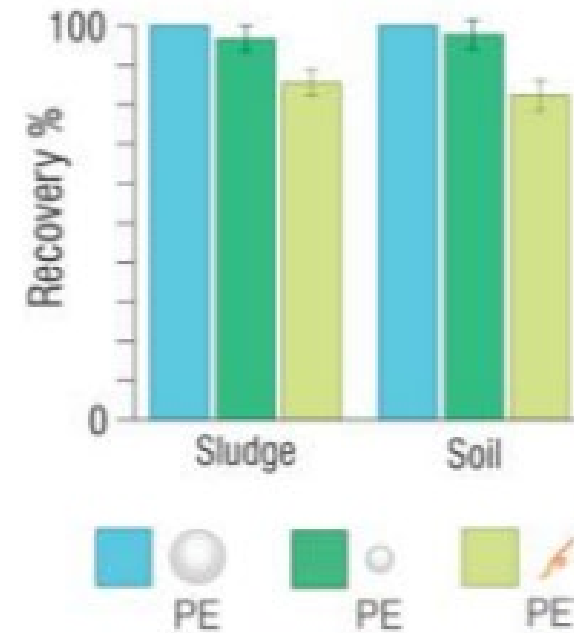
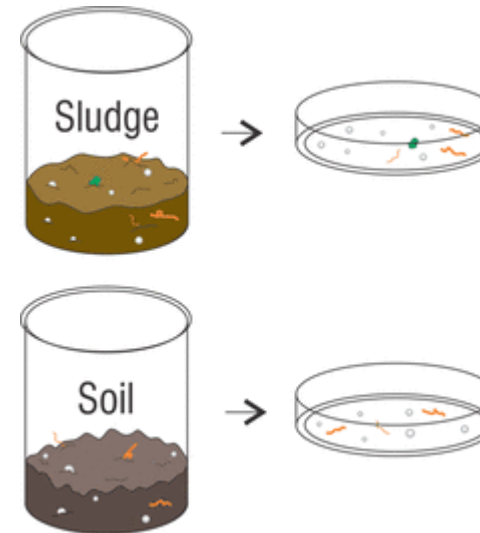
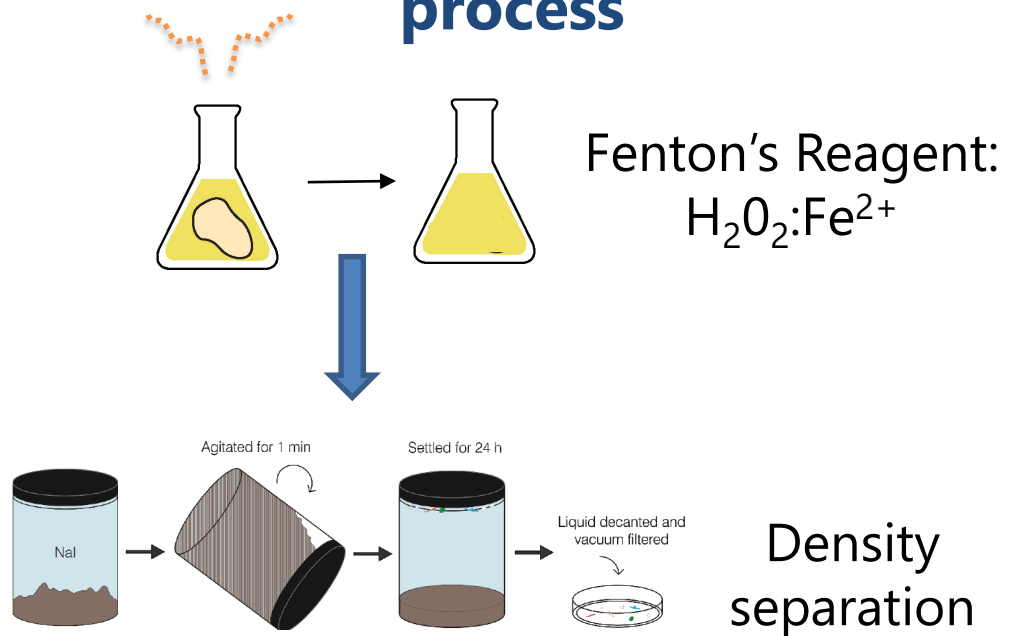
f. PS + Protocol 2  
Fenton's reagent



Strong acids denature proteins and hydrolyze  
chemical bonds

Hurley et al. 2018, Validation of a Method for Extracting Microplastics from  
Complex, Organic-Rich, Environmental Matrices. ES&T

## Some samples require a two step process





## Contamination controls

Processing steps		
Sampling	1	Sampling methods
	2	Sample size
	3	Sample processing and storage
Contamination mitigation	4	Laboratory preparation
	5	Clean air conditions
	6	Negative control
Sample purification and handing	7	Positive control
	8	Target control
	9	Sample treatment
Chemical analysis	10	Polymer identification and reporting

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- There are many different methods available
- Research question and sample type will influence choice
- Look at optimised and recommended methods
- **BUT ALWAYS TEST WITH KNOWN PLASTICS FIRST**
  - Check recovery rates
  - Check particles for deformations



# Thank you!

Natalie Welden



Analytical  
Methods



CRITICAL REVIEW

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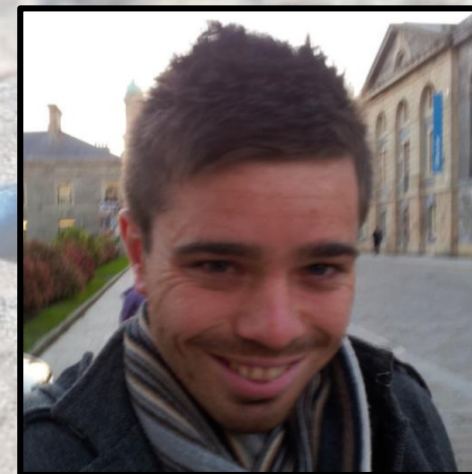


Cite this: *Anal. Methods*, 2017, 9, 1346

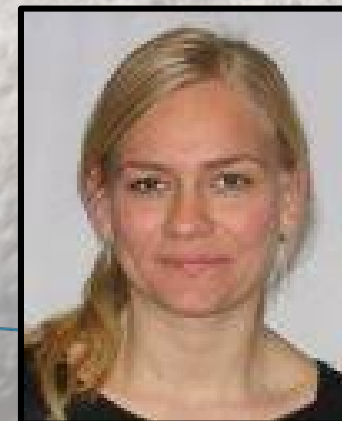
**Sampling, isolating and identifying microplastics ingested by fish and invertebrates†**

A. L. Lusher,<sup>\*a</sup> N. A. Welden,<sup>b</sup> P. Sobral<sup>c</sup> and M. Cole<sup>d</sup>

Matt Cole



**NIVA: Inger Lise N. Bråte,  
Rachel Hurley, Nina  
Buenaventura, Emilie  
Kallenbach et al.**



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**GUIDELINES FOR THE MONITORING  
AND ASSESSMENT OF PLASTIC  
LITTER IN THE OCEAN**

