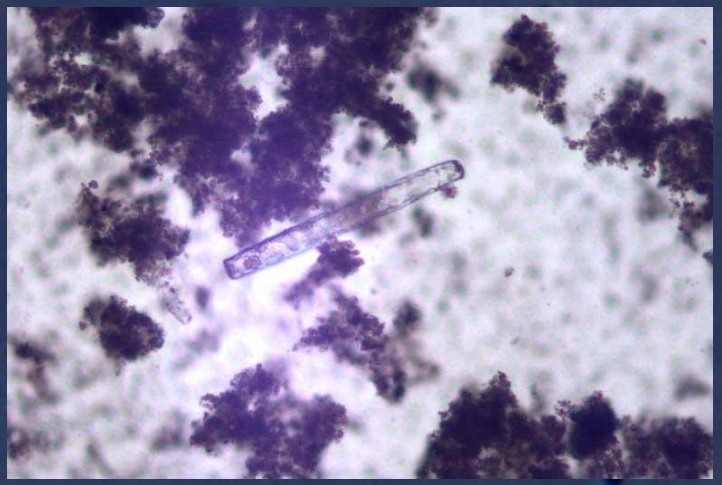
IMPROVING THE MICROSCOPIC PARTICULATE ANALYSIS



Peter Wallis

Hyperion Research Ltd., Medicine Hat, AB

Why is this important?

In the post-Walkerton era, Canadian regulations in several Provinces require that all wells used as potable sources must be tested for surface water intrusion.

Under Canadian regulations in some provinces an investigation of every well used as a potable water source must be carried out by a qualified hydrogeologist

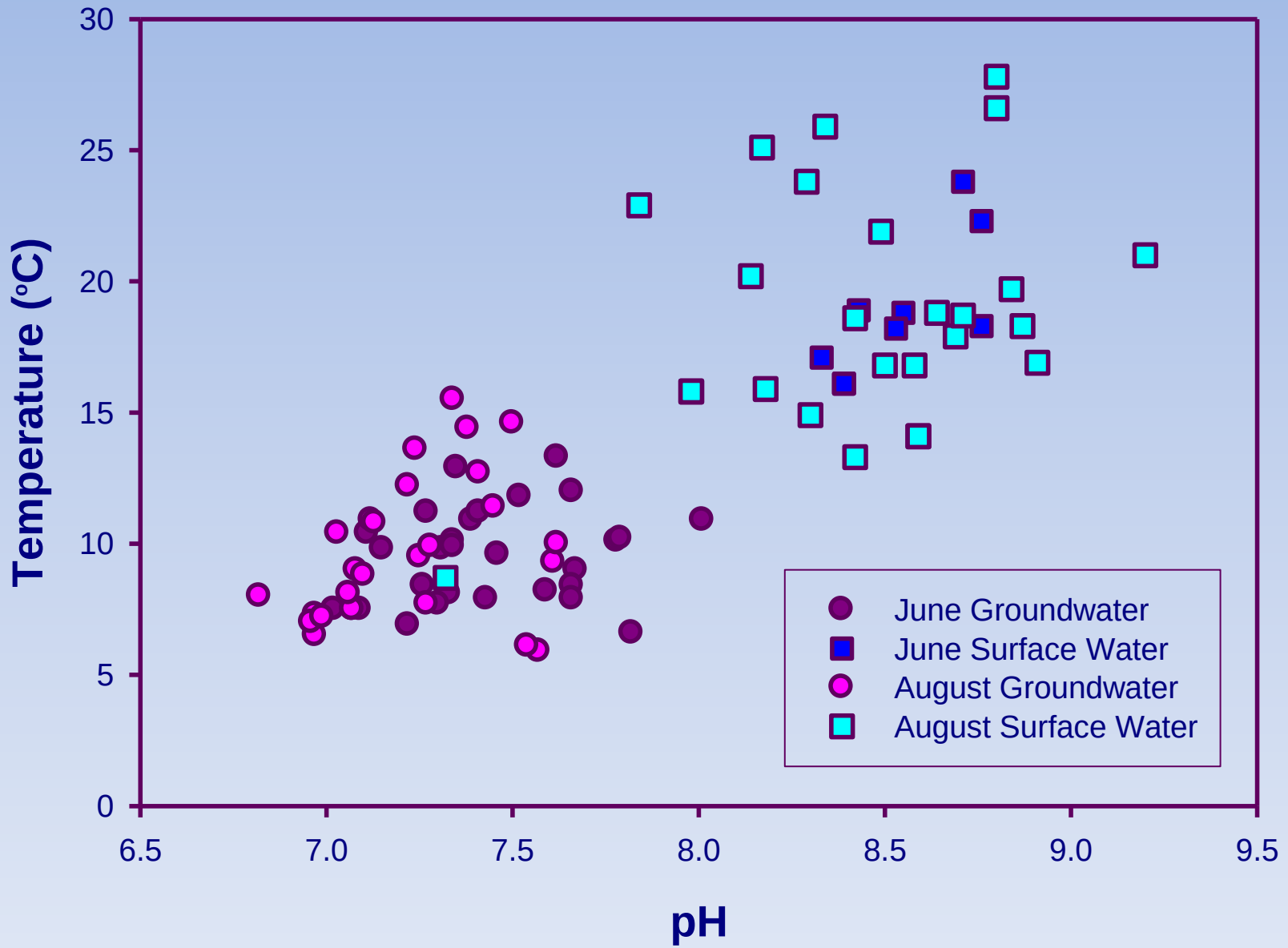
Phase I – Initial screening for risk factors such as distance to surface water

Phase II – Hydrogeological investigation including chemistry, well logs etc.

Phase III - Microscopic Particulate Analysis as specified by (EPA Consensus Method 910/9-92-029).

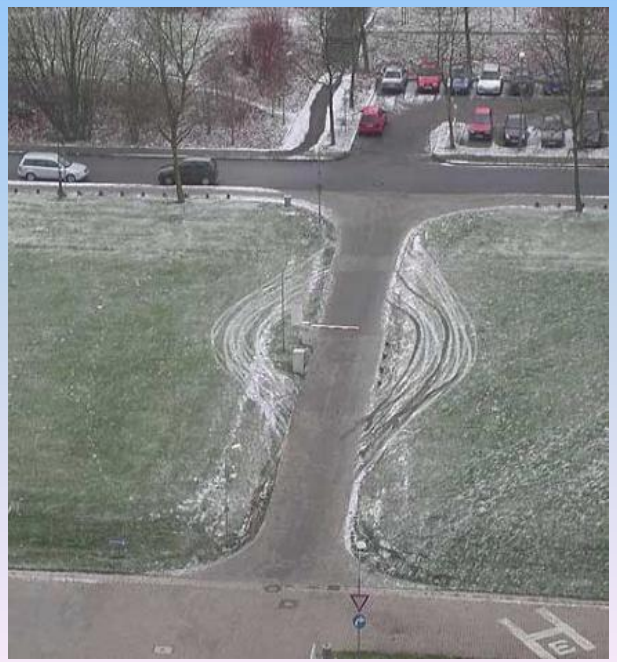
Nice cap!

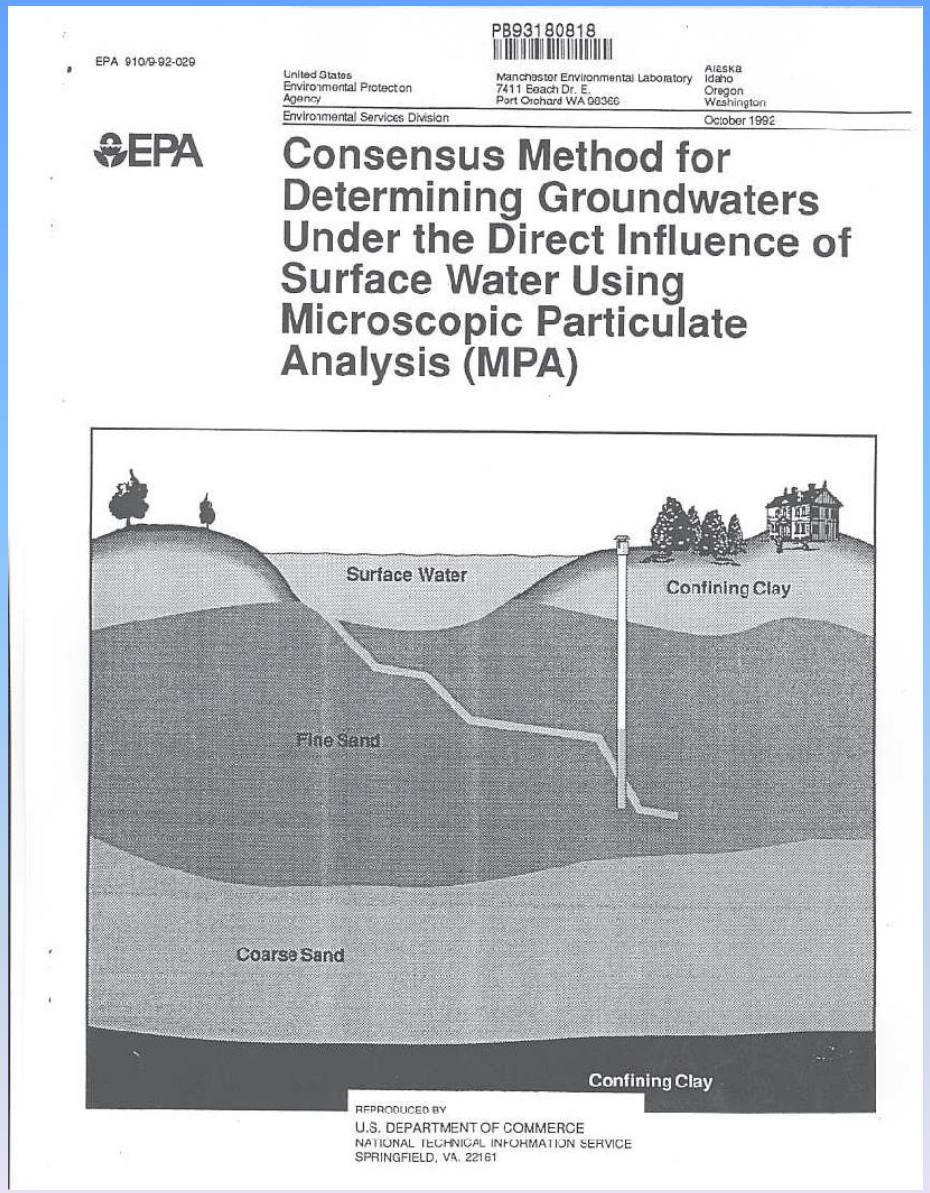
Saskatchewan Parks Well Water



Unfortunately, the Consensus MPA method has several drawbacks including:

- **Old filter technology (string wound cartridges)**
- **Inefficient recovery of particles**
- **Large filtration volume means extended field time**
- **Lack of quality control protocol**





PB93180818

EPA 910/9-92-029

United States Environmental Protection Agency
Environmental Services Division

Manchester Environmental Laboratory
7411 Beach Dr. E.
Port Orchard WA 98366

Alaska
Idaho
Oregon
Washington

October 1992

EPA

Consensus Method for Determining Groundwaters Under the Direct Influence of Surface Water Using Microscopic Particulate Analysis (MPA)

REPRODUCED BY
U.S. DEPARTMENT OF COMMERCE
NATIONAL TECHNICAL INFORMATION SERVICE
SPRINGFIELD, VA. 22161

Problems can be overcome!

The Consensus method was based on the original string-wound cartridge method for *Giardia* and *Cryptosporidium.*

This method has been superseded by Method 1623

Method 1623 specifies:

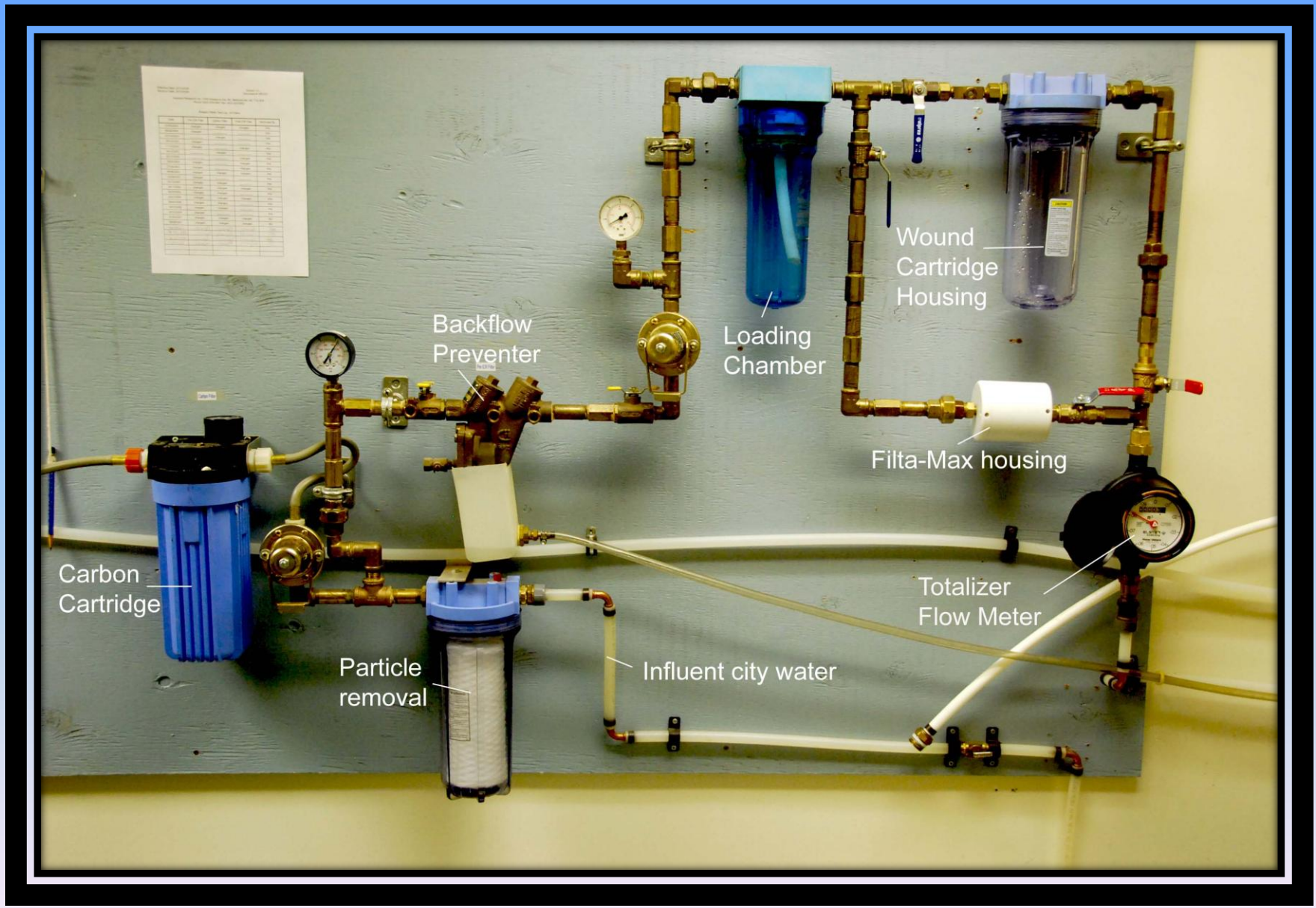
- much better filters which can be eluted more efficiently
- immunomagnetic separation to recover cysts and oocysts
- a rigorous quality control protocol

Additional benefits include:

- Much lower filtration volumes saving time in the field
- Smaller pellets for examination
- Ability to work at much higher pressures (Filta-Max)

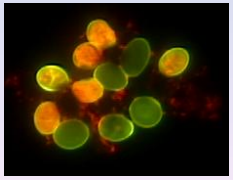
Why not modify the existing protocol to Method 1623?

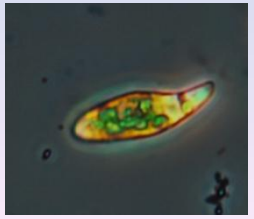
Lab testing apparatus



Recovery of *Giardia* cysts, *Cryptosporidium* oocysts and *Euglena gracilis* cells by the Consensus method in a complex matrix containing clay, precipitated iron and background minerals

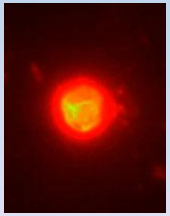
	Giardia Cysts			*Cryptosporidium* oocysts			*Euglena gracilis* cells		
Tube	Spike	Number Recovered	% Recovery	Spike	Number Recovered	% Recovery	Spike	Number Recovered	% Recovery
1	500	48	9.6	15,000	54	0.4	0	0	0
2	3000	216	7.2	10,000	36	0.4	2000	27	1.3
3	10,000	780	7.8	9000	24	0.3	4000	35	0.9
4	4500	264	5.9	8000	36	0.5	6000	347	5.8
5	1500	60	4.0	7000	12	0.2	8000	93	1.2
6	4000	156	3.9	6000	24	0.4	10,000	640	6.4
7	2500	144	5.8	5000	36	0.7	12,000	707	5.9
8	6000	240	4.0	4000	36	0.9	14,000	253	1.8
9	0	0	0	3000	24	0.8	16,000	1520	9.5
10	3500	228	6.5	2000	24	1.2	18,000	160	0.9
11	5000	420	8.4	1000	0	0	20,000	2307	11.5
12	2000	168	8.4	0	0	0	25,000	173	0.7
Mean			6.5			0.5			4.2
RSD			30.5			69.1			92.8





Recoveries of *Giardia* cysts, *Cryptosporidium* oocysts, *Euglena* sp. and *Sphaerocystis* cells from spiked Filta-Max cartridges pre-loaded with groundwater matrix

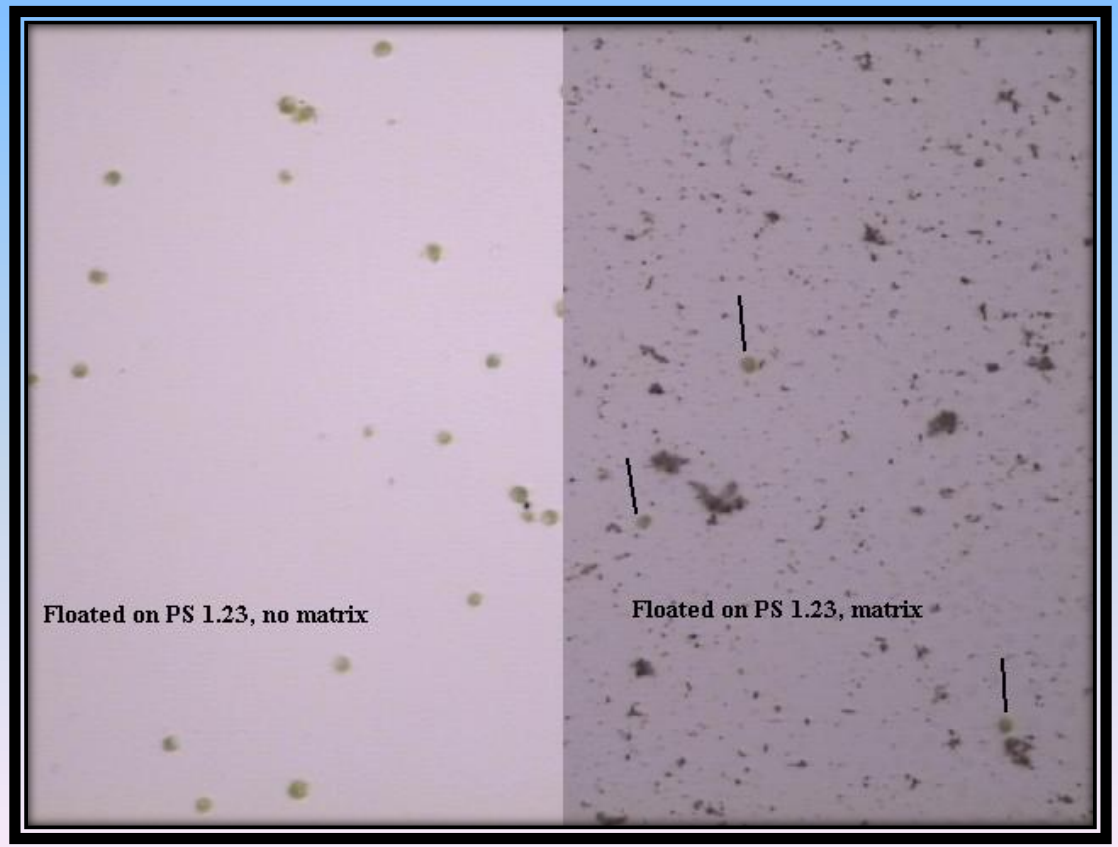
	Giardia Cysts		*Cryptosporidium* oocysts		*Euglena* sp. cells		*Sphaerocystis* sp. cells	
	Spike	Recovery %	Spike	Recovery %	Spike	Recovery %	Spike	Recovery %
	1000	38.6	10000	86.9	1000	28.0	250000	1.5
	8000	47.0	8000	94.0	8000	48.1	200000	1.8
	7000	44.3	7000	94.9	6000	35.0	150000	1.4
	6000	33.0	6000	105.5	4000	23.3	100000	3.5
	5000	23.2	5000	64.6	2000	46.7	50000	2.3
Mean (%)		37.2		89.2		36.2		2.1
RSD (%)		25.6		17.1		30.4		40.9



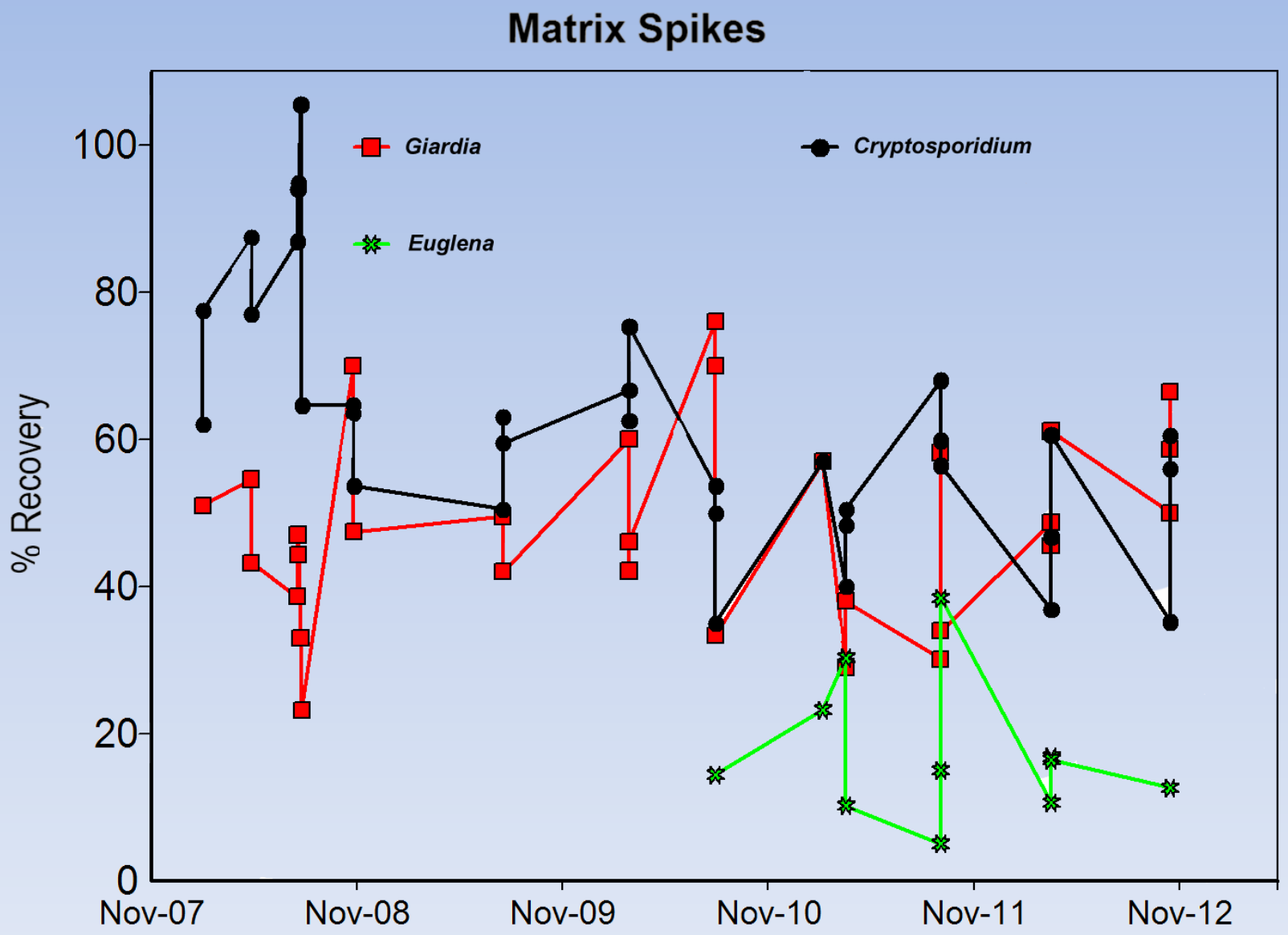
Consensus Method vs Modified Method 1623

	Giardia Cysts		*Cryptosporidium* oocysts		*Euglena* sp. cells		*Sphaerocystis* sp. cells	
	Consensus	Modified 1623	Consensus	Modified 1623	Consensus	Modified 1623	Consensus	Modified 1623
n	12	5	12	5	12	5	4	5
Mean %	6.5	37.2	0.5	89.2	4.2	36.2	0	2.1
RSD (%)	30.5	25.6	69.1	17.1	92.8	30.4	ND	40.9

A problem common to both methods is the need to reduce background when the pellet volume is too high



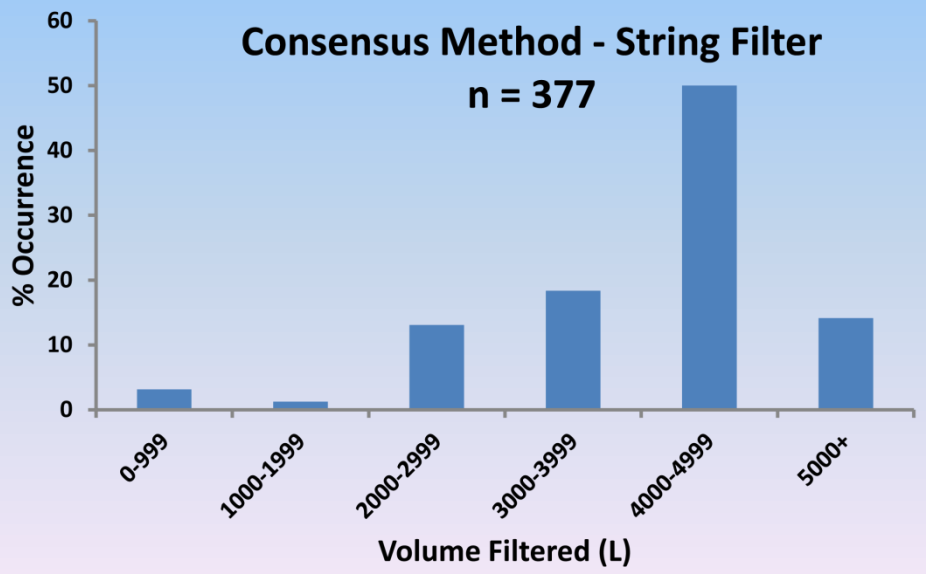
EPA Performance Evaluation Spike Testing: Hyperion Research Ltd. 2009 - 2012

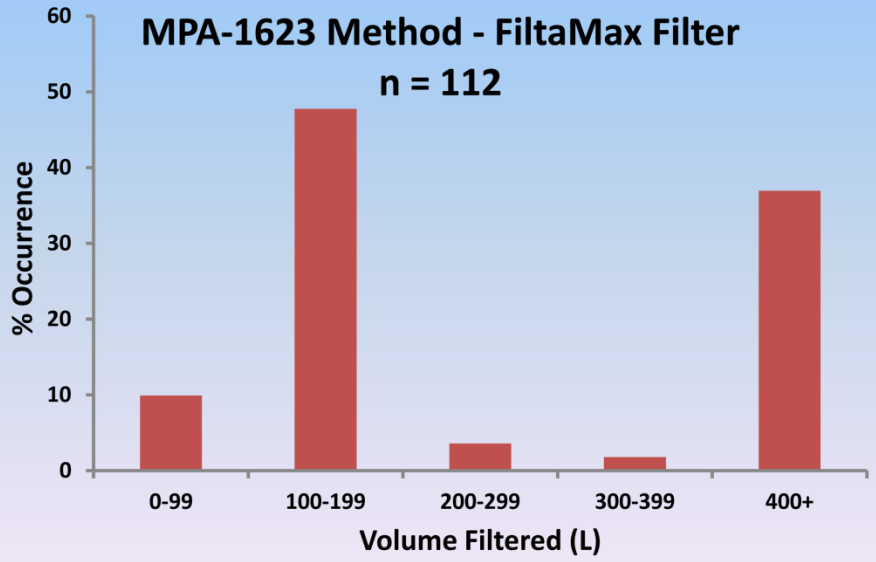


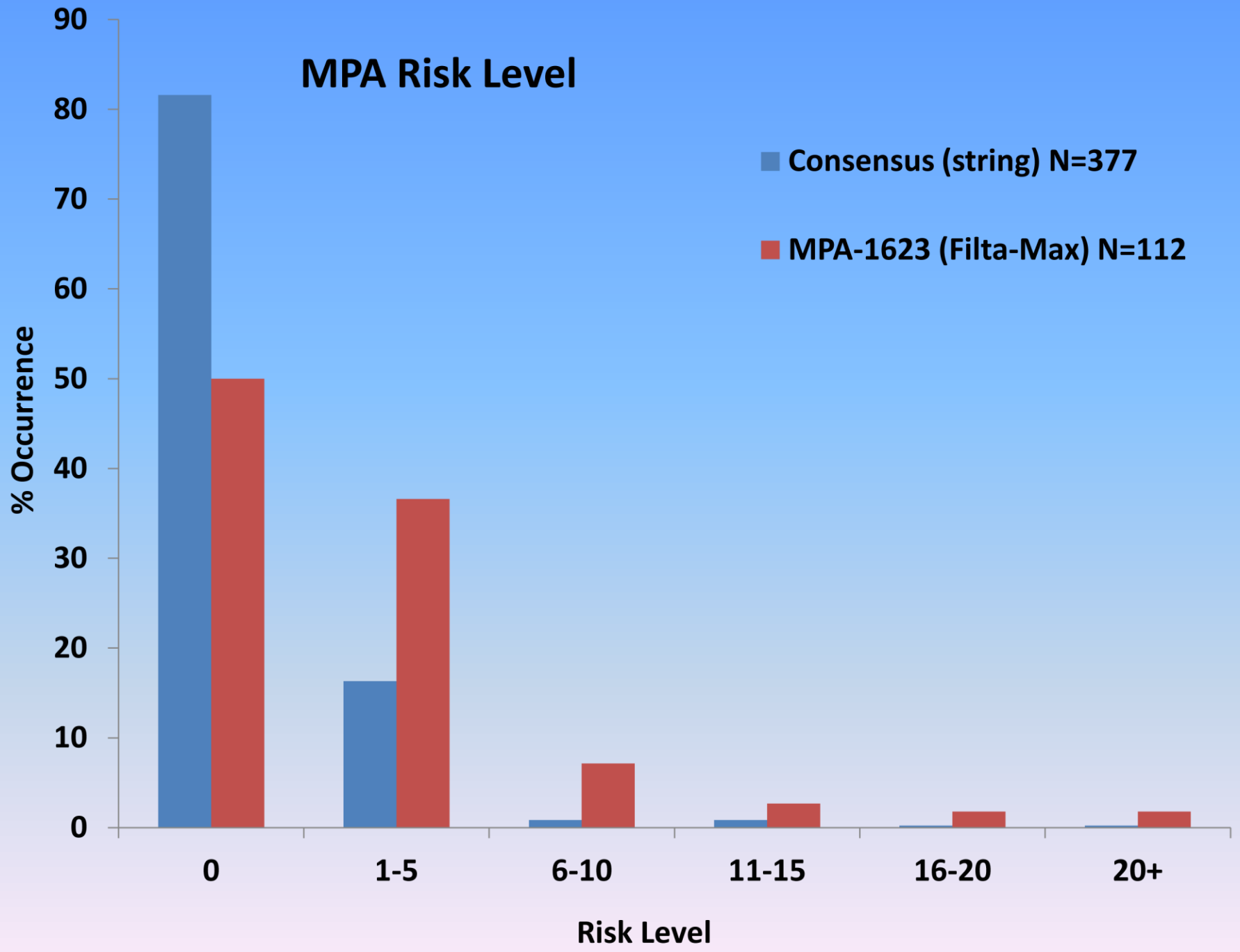
Field Testing the Modified Method 1623 – MPA Method

	Waterhen	Jeannette	Floten	First Mustus	Kimball	Matheson	Mistohay	Pierce
Other Algae Cells	53	89	9	107	152	50	46	84
Plant Debris	0	115	0	24	0	150	38	0
Pollen	2111	4545	2184	5804	3998	17700	726	2238
Calculated Risk Factor	9	11	4	12	12	11	10	9

Typical Volumes Filtered for Each Method






MPA Risk Level
Consensus (string) N=377
MPA-1623 (Filta-Max) N=112
% Occurrence
90
80
70
60
50
40
30
20
10
0
0
1-5
6-10
11-15
16-20
20+
Risk Level

Distribution (%) of relative risk factors in 377 Consensus Method samples

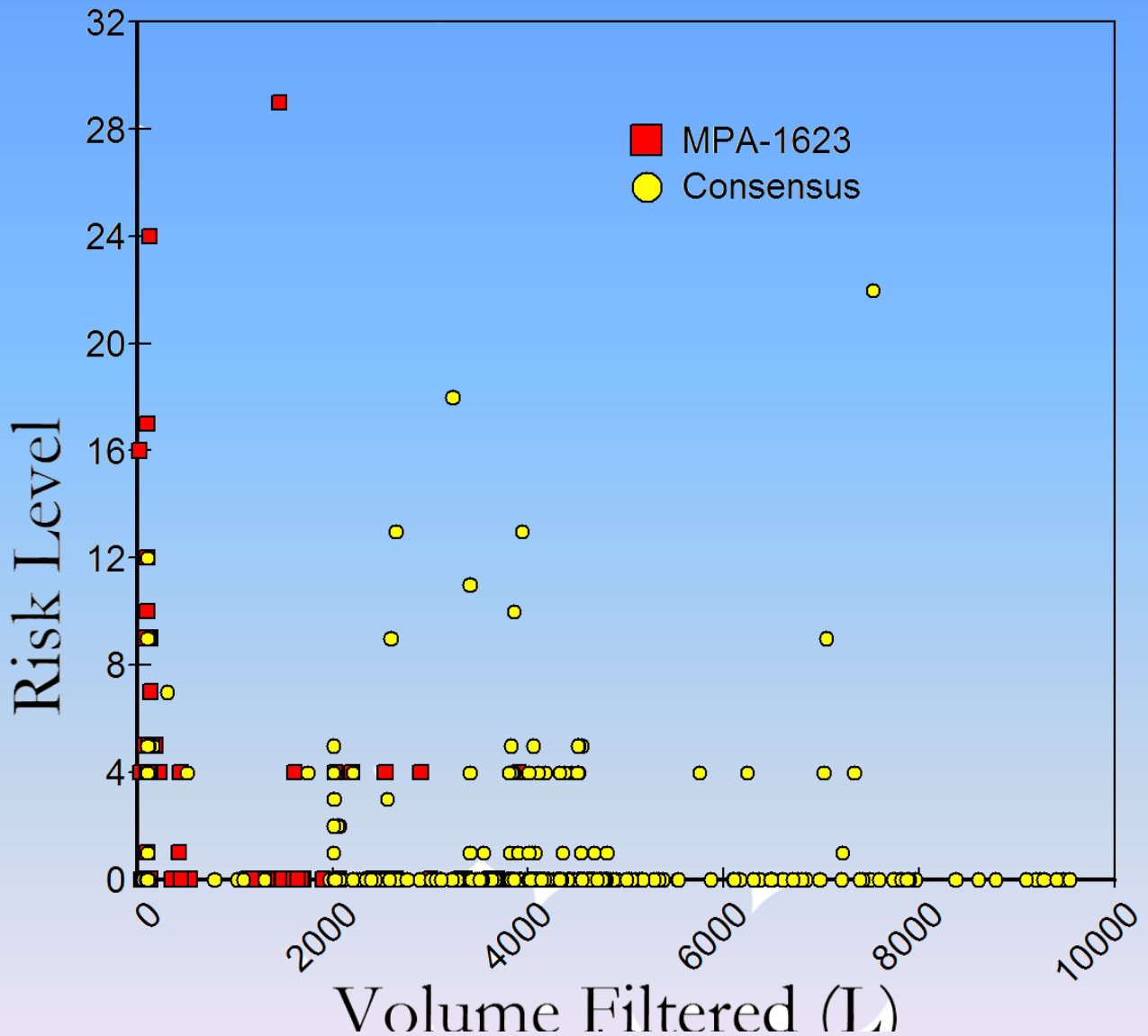
	Extra Heavy	Heavy	Medium	Rare	Not Significant
Diatoms	0.3	0	0.3	0.5	98.9
Other Algae	0.3	0.3	1.3	11.9	86.2
Insect	0.3	0	0	0.8	98.9
Rotifer	0.3	0.3	0.5	8.8	90.2
Plant	0.5	1.3	4.8	39.9	53.5

Distribution (%) of relative risk factors in 112 MPA-1623 samples

	Extra Heavy	Heavy	Medium	Rare	Not Significant
Diatoms	0	0	0	0	100
Other Algae	1.8	1.8	8	36.3	52.2
Insect	0	0	0	0.9	99.1
Rotifer	0	0.9	0.9	2.7	95.6
Plant	0.9	0.9	8	30.1	60.2

Other Algae are 3X more likely to be detected by MPA-1623 analysis

Risk Levels do not Increase with Higher Filtration Volumes



Another problem: recalibrating risk factors

TABLE 1. Numerical range of each primary bio-indicator (particulate) counted per 100 gallons water.

Indicators of surface water[1]	EH[3]	H	M	R	NS
Giardia[2]	>30	16-30	6-15	1-5	<1
Coccidia[2]	>30	16-30	6-15	1-5	<1
Diatoms[4]	>150	41-149	11-40	1-10	<1
Other Algae[4]	>300	96-299	21-95	1-20	<1
Insects/Larvae	>100	31-99	16-30	1-15	<1
Rotifers	>150	61-149	21-60	1-20	<1
Plant Debris[4]	>200	71-200	26-70	1-25	<1

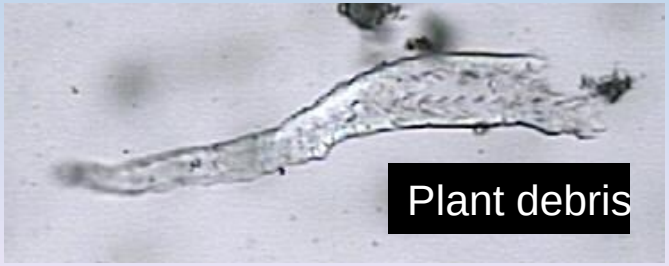
1. According to EPA "Guidance Manual for Compliance with the Filtration and Disinfection Requirements for Public Water Systems Using Surface Water Sources", March, 1991 ed.
2. If Giardia cysts or coccidia are found in any sample, irrespective of volume, score as above.
3. Key= EH -extremely heavy M -moderate NS -not significant
   H -heavy R -rare
4. Chlorophyll containing

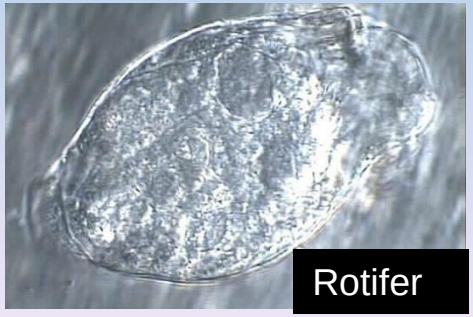
TABLE 2. Relative surface water risk factors associated with scoring of primary bio-indicators (particulate) present during MPA of subsurface water sources.

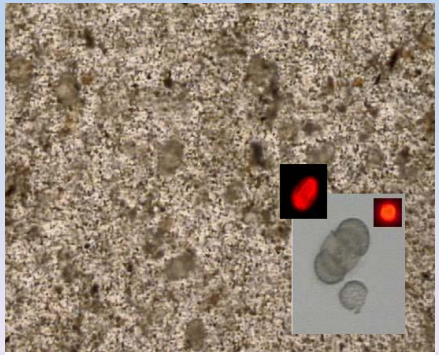
Indicators of surface water[1]	Relative Risk Factor[3]				
	EH[2]	H	M	R	NS
Giardia	40	30	25	20	0
Coccidia	35	30	25	20	0
Diatoms	16	13	11	6	0
Other Algae	14	12	9	4	0
Insects/Larvae	9	7	5	3	0
Rotifers	4	3	2	1	0
Plant Debris	3	2	1	0	0

1. According to EPA "Guidance Manual for Compliance with the Filtration and Disinfection Requirements for Public Water Systems Using Surface Water Sources", March 1991 ed.
2. Refer to Table 1 for range of indicators counted per 100 gallons.

   Key= EH -extremely heavy M -moderate NS -not significant
   H -heavy R -rare
3. Risk of surface water contamination:
   ≥20 - high risk
   10-19 - moderate risk
   ≤9 - low risk



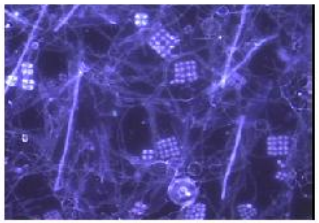


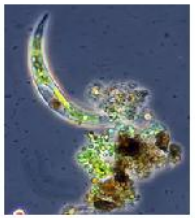


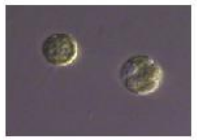
Pollen in groundwater

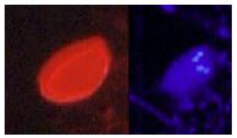
UNDER THE MICROSCOPE

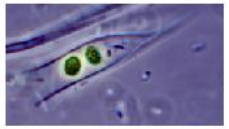
Primary Particulates

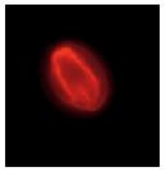


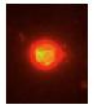


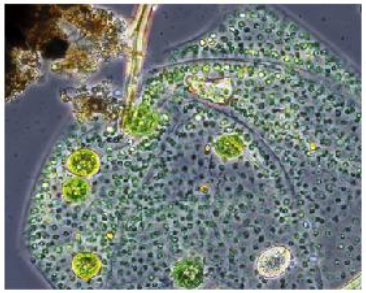


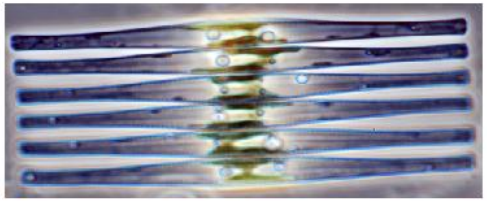


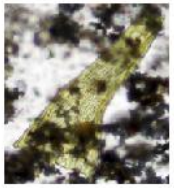




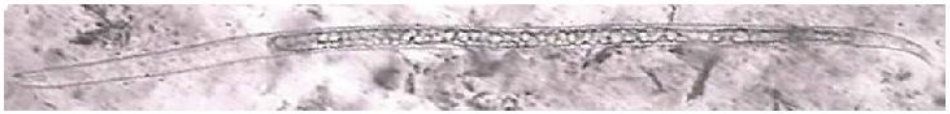


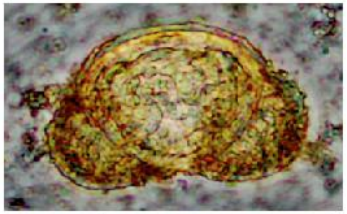


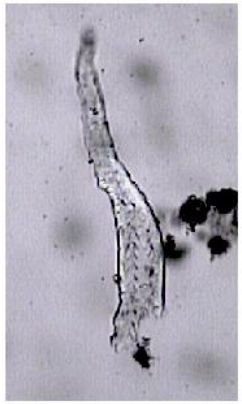


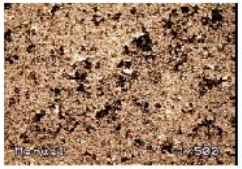


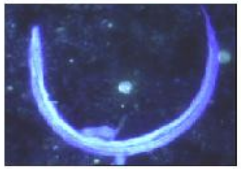
Secondary Particulates

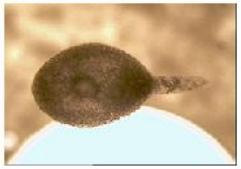


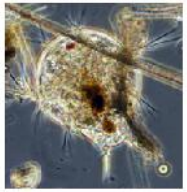


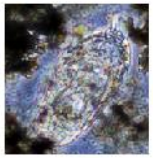


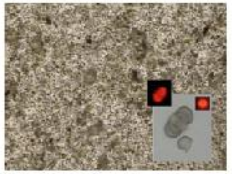


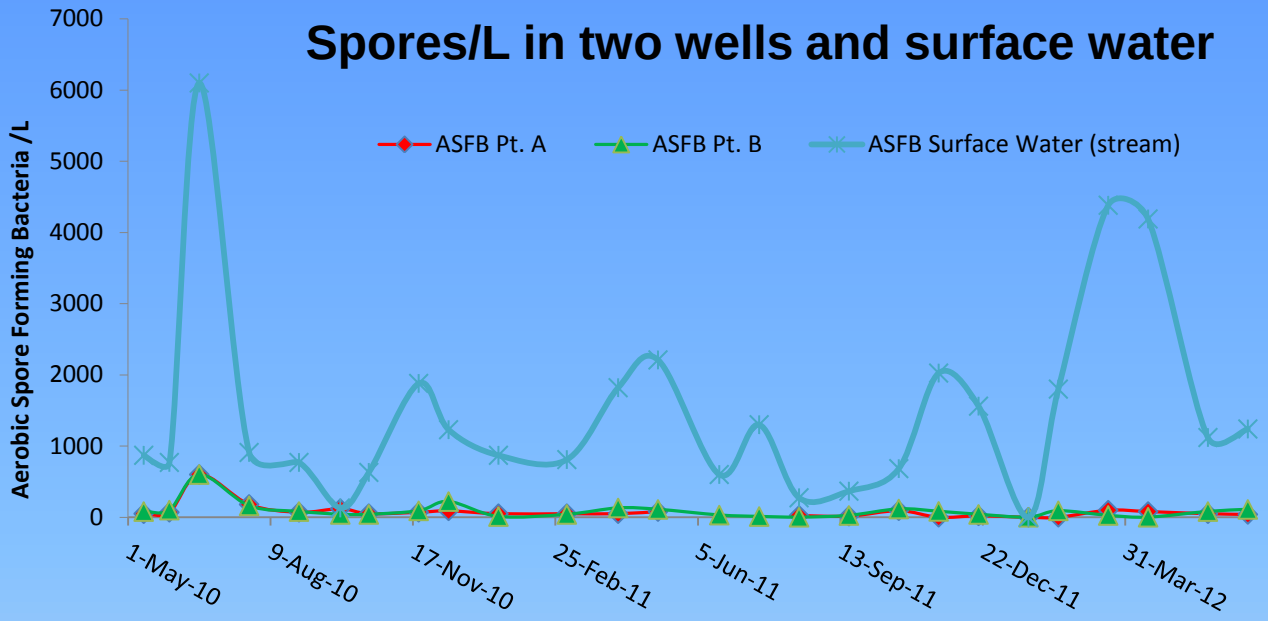


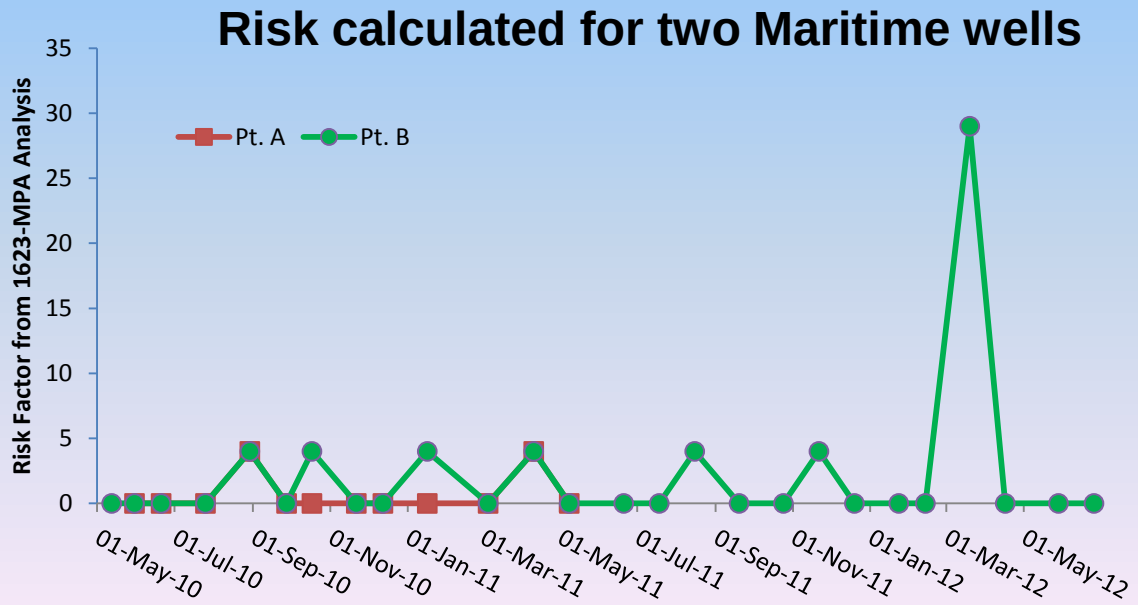






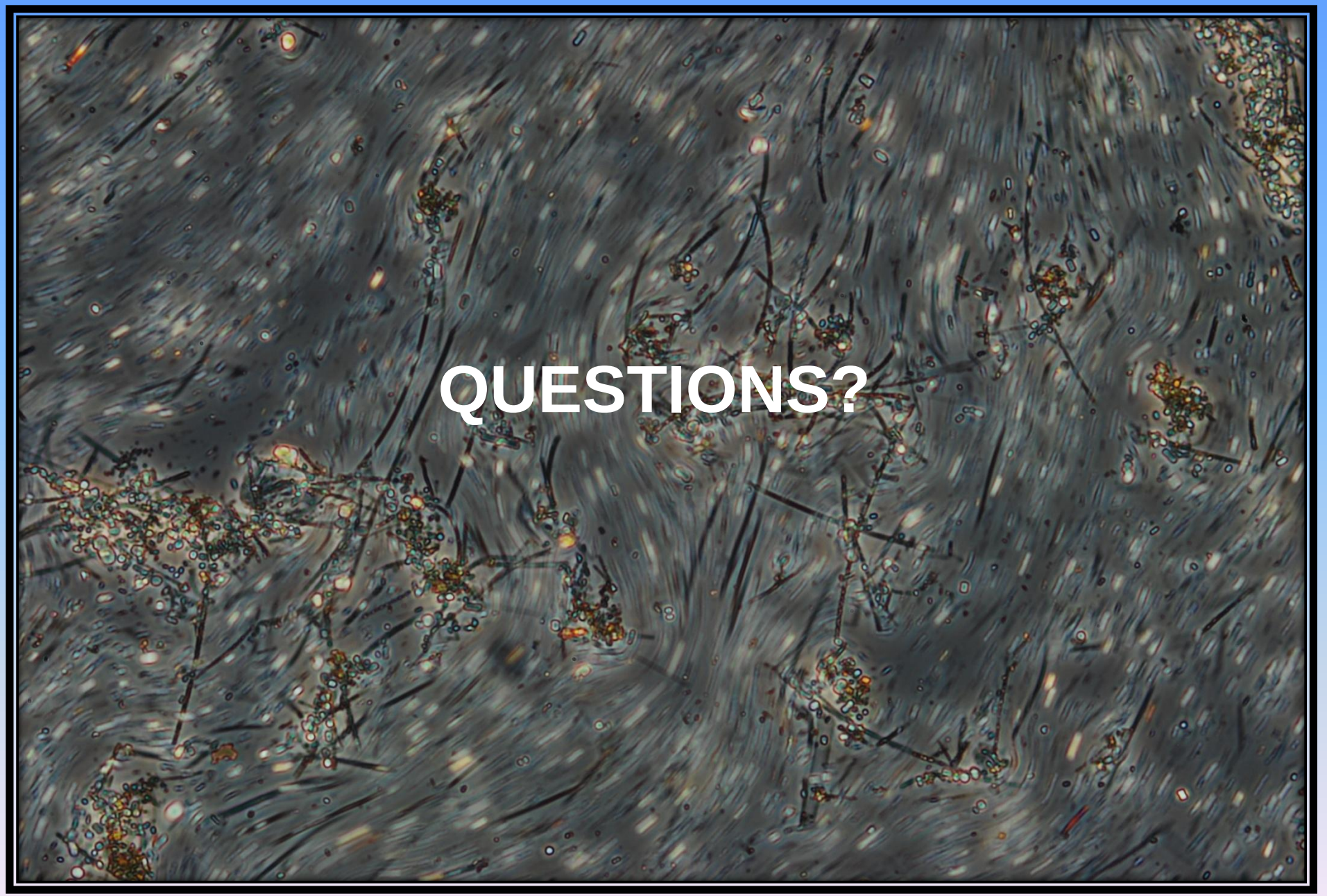



Spores/L in two wells and surface water
Aerobic Spore Forming Bacteria /L
7000
6000
5000
4000
3000
2000
1000
0
ASFB Pt. A
ASFB Pt. B
ASFB Surface Water (stream)
1-May-10
9-Aug-10
17-Nov-10
25-Feb-11
5-Jun-11
13-Sep-11
22-Dec-11
31-Mar-12


Risk calculated for two Maritime wells
Risk Factor from 1623-MPA Analysis
35
30
25
20
15
10
5
0
Pt. A
Pt. B
01-May-10
01-Jul-10
01-Sep-10
01-Nov-10
01-Jan-11
01-Mar-11
01-May-11
01-Jul-11
01-Sep-11
01-Nov-11
01-Jan-12
01-Mar-12
01-May-12

Conclusions

- **The Microscopic Particulate Analysis is a useful tool in determining whether groundwater is under the influence of surface water**
- **Recovery of particulate matter is inefficient with string-wound filters**
- **Method 1623 offers much better detection of the primary pathogens *Giardia* and *Cryptosporidium* and the pellet can be used for surface water organism detection**
- **Higher efficiency of particulate recovery means lower volumes need to be filtered resulting in reduced field sampling costs**
- **A quality control procedure is needed and already exists for Method 1623**
- **Higher recovery efficiency means that the risk tables need to be recalibrated**
- **High sediment background is still a problem**
- **Aerobic spore forming bacteria are an inexpensive addition to the MPA**
- **Nearby surface water should be sampled along with well water**


QUESTIONS?