IMS/ATP Rapid Method for the Determination of *E. coli* Concentrations in Recreational Waters

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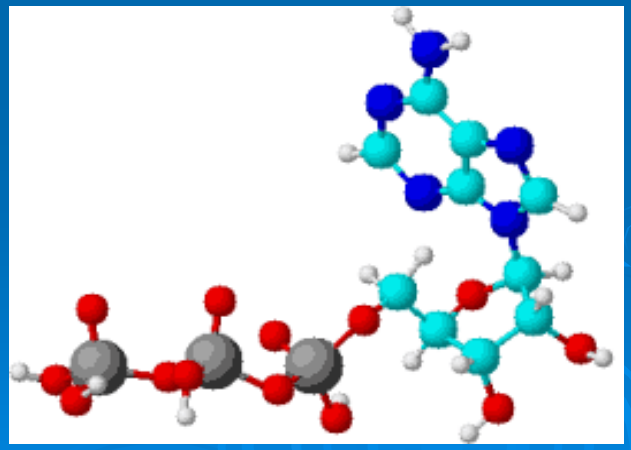
IMS/ATP Definition

- Immunomagnetic separation (IMS)
  - Method of capturing microorganisms
  - Uses antibody-coated paramagnetic beads which bind to antigens present on the surface of cells

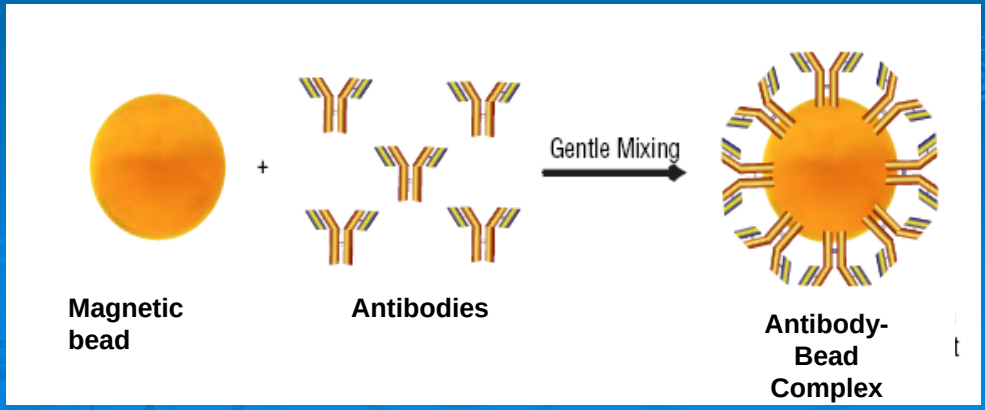
- Adenosine triphosphate (ATP)
  - ATP is present in all cells
  - ATP transports chemical energy within cells



IMS/ATP Rapid Method

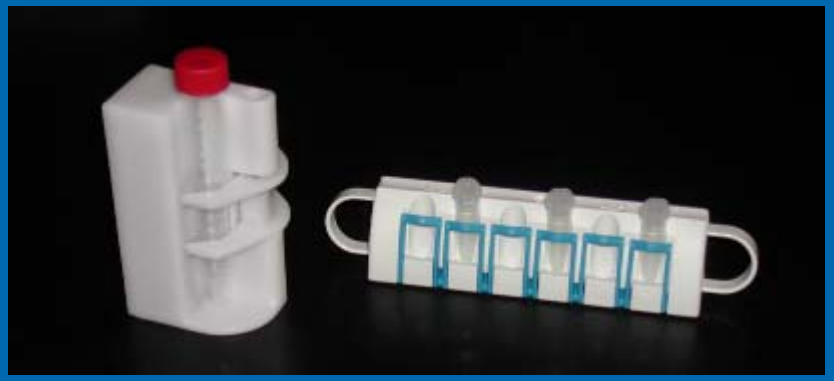
- Concentrate bacteria by filtration, if needed

- Add magnetic beads that are coated with antibodies



IMS/ATP Rapid Method

- Separate bacteria from the sample by IMS
- Rupture bacterial cells
- Measure ATP using luciferin-luciferase
- Results in Relative Light Units (RLUs)



IMS/ATP Rapid Method

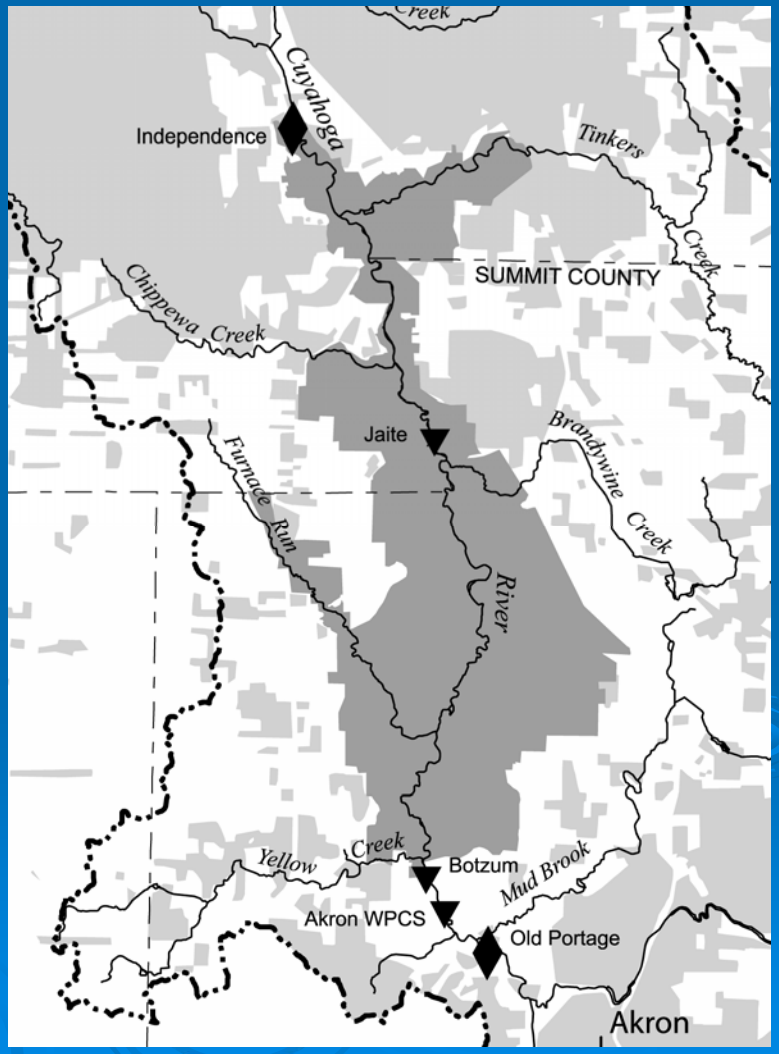
- Time to obtain results = ~1 hour
- Measures viable cells
- Supplies cost per sample = ~$15-20
- Equipment costs = ~$4,000 - 6,000
- Field portable

IMS/ATP Research

- Method developed by researchers at University of Michigan (Lee and Deininger; *Luminescence,2004*)
- IMS/ATP methods were developed for *E. coli* and enterococci
- Limited field studies:
  - University of Michigan – Lake Michigan and Huron River
  - USGS – Cuyahoga Valley National Park, Ohio
  - USGS – Lake Erie beaches, Ohio
  - SCCWRP – Rapid methods comparison study, California

Field Testing of IMS/ATP Method

- Cuyahoga River, Cuyahoga Valley National Park
  - In cooperation with the National Park Service
  - Project duration - 2004-2006
  - Results will be published this year



Approach

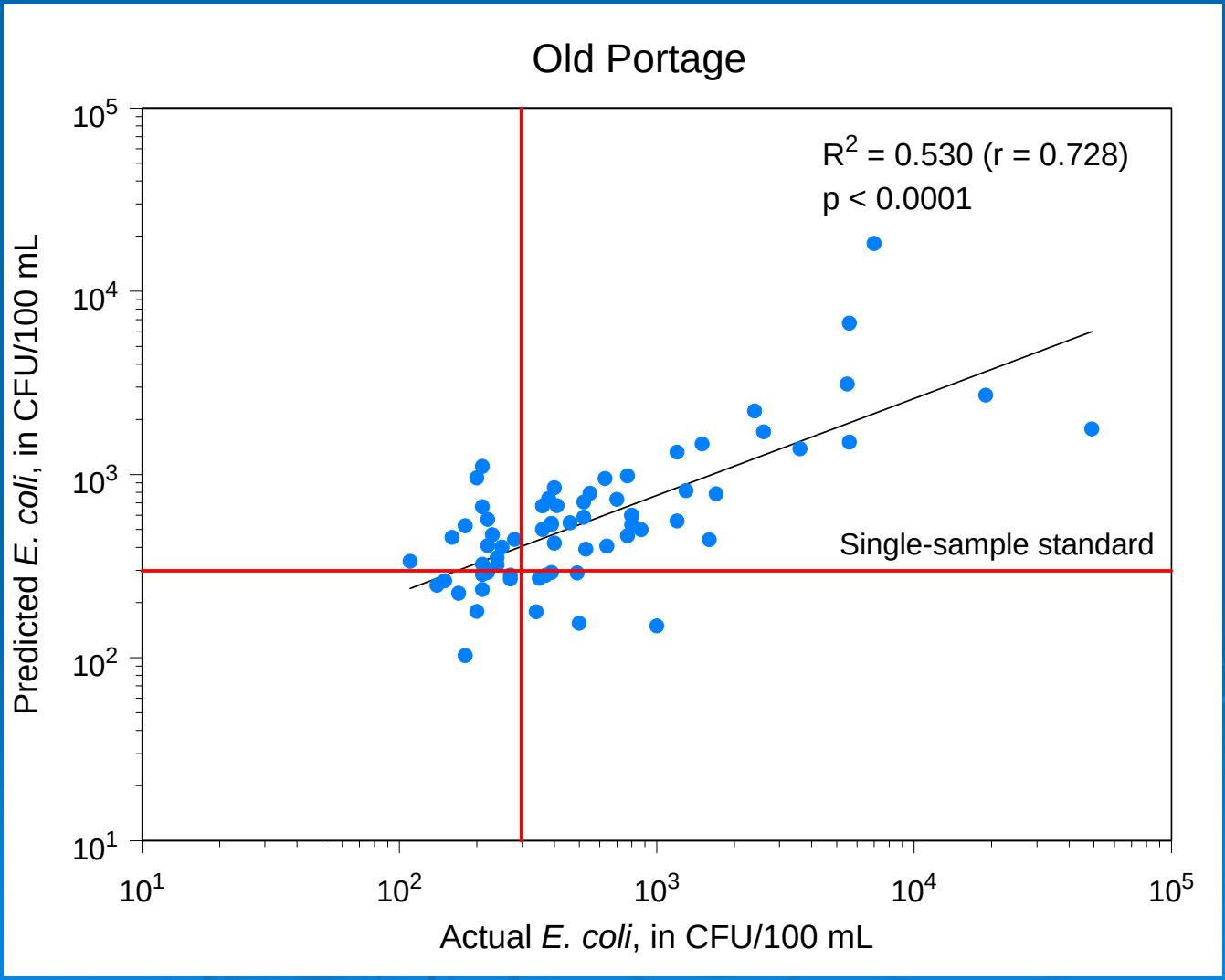
- Samples collected 7 days per week from May-August in 2004; 5 days per week in 2005
- Ancillary data collected: turbidity, rainfall, and streamflow
- Statistical analyses:
  - Correlations
  - Simple-linear regression
  - Multiple-linear regression

MLR results for *E. coli*

2004-2005

Model variables:

- log RLUs (direct analysis)
- log turbidity
- Rainfall yesterday



MLR results for *E. coli*

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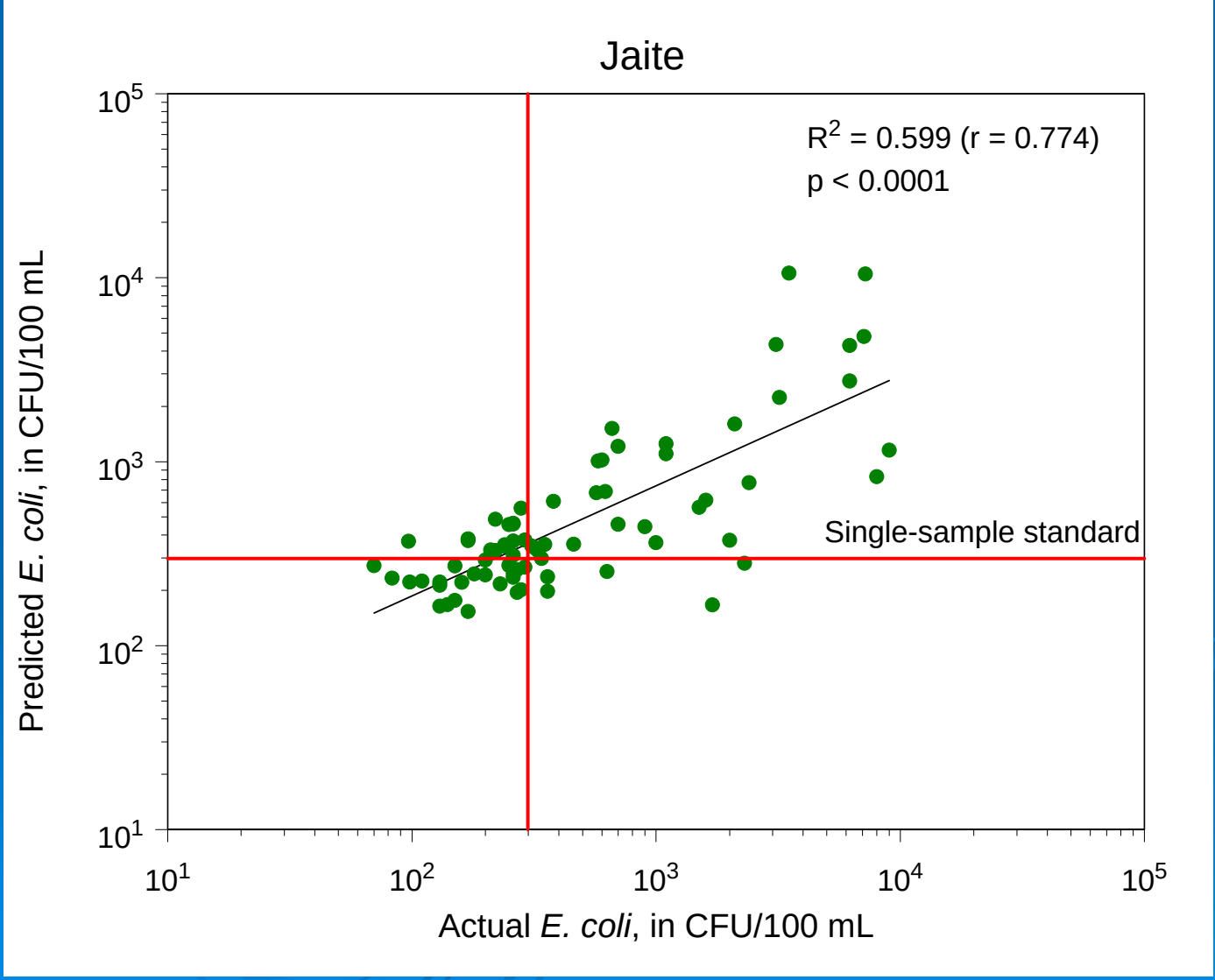


MLR versus yesterday's *E. coli*

	Correct Responses	False Positive	False Negative
Old Portage			
MLR	69.1% (47)	20.6% (14)	10.3% (7)
Yesterday's count	82.7% (48)	10.3% (6)	6.9% (4)
Jaite			
MLR	72.0% (54)	20.0% (15)	8.0% (6)
Yesterday's count	69.0% (49)	12.7% (9)	18.3% (13)

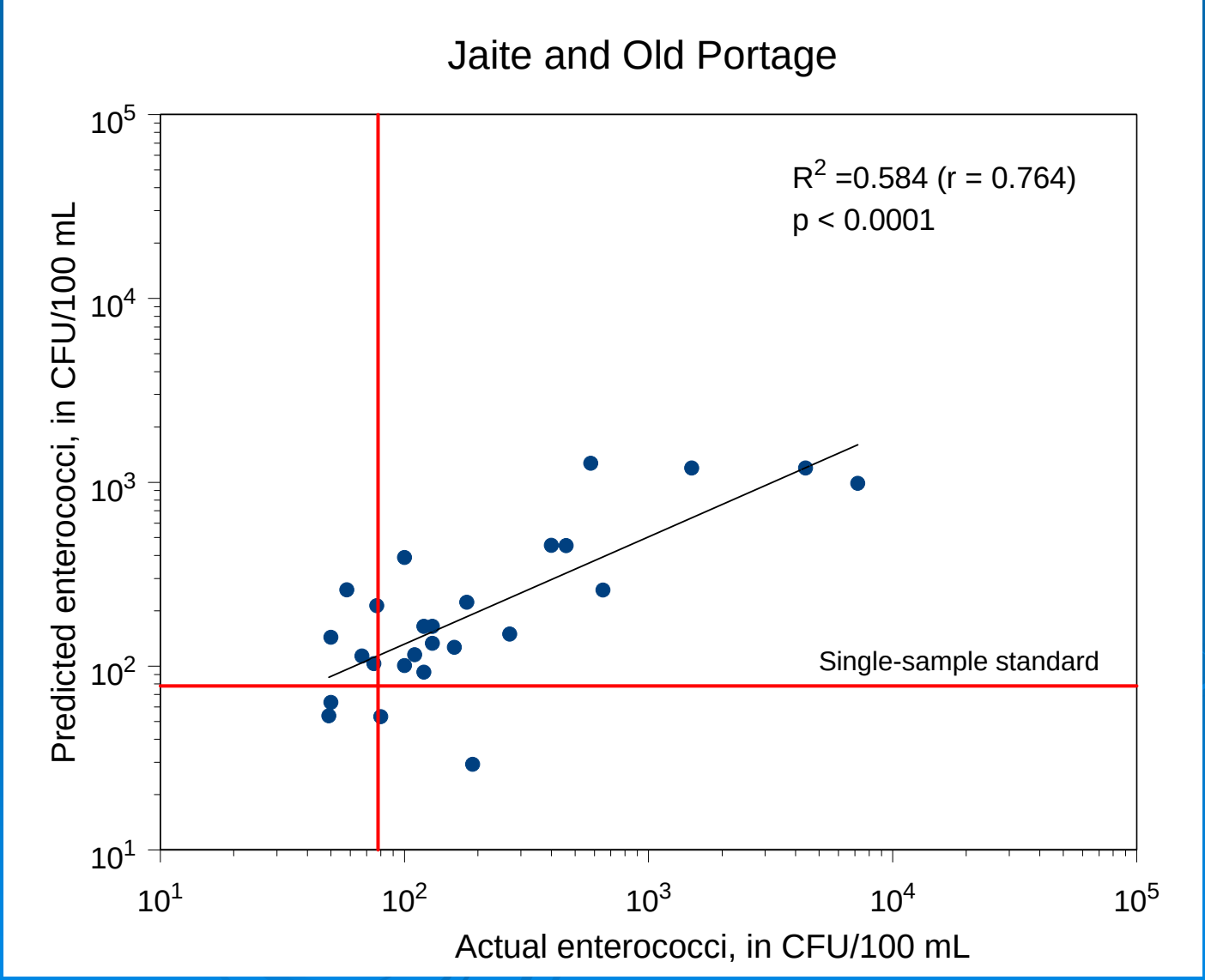
SLR results for enterococci

2004

Model variable:

log RLUs

(direct analysis)



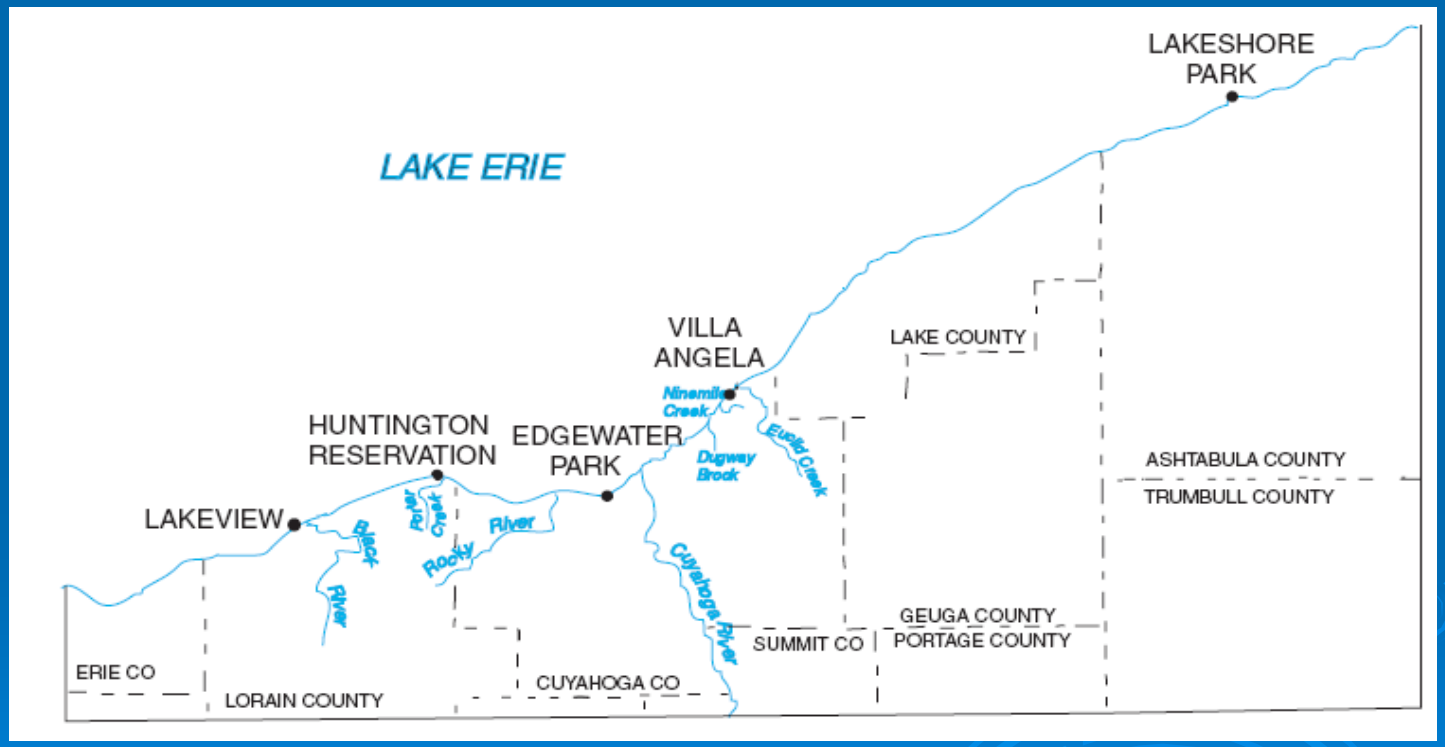
SLR vs yesterday's enterococci

	Correct Responses	False Positive	False Negative
Jaite/Old Portage			
SLR	73.1% (19)	19.2% (5)	7.7% (2)
Yesterday's count	76.9% (20)	11.5% (3)	11.5% (3)

Field Testing of IMS/ATP Method

Preliminary study in 2005

USGS and Northeast Ohio Regional Sewer District



Lake Erie beaches — Edgewater and Villa Angela, Cleveland, Ohio

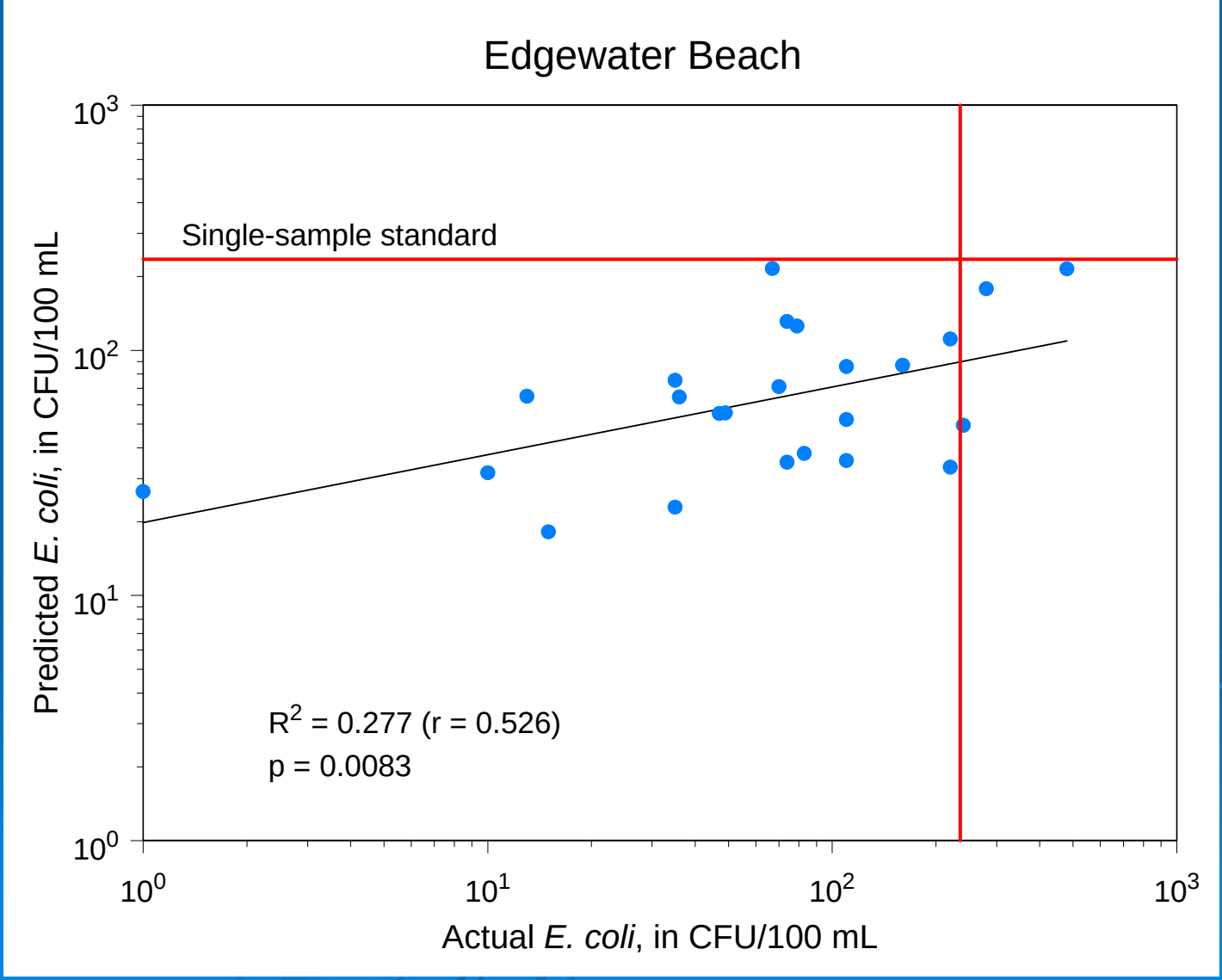
SLR results for *E. coli*

2005

Model variable:

log RLUs

(single filtration)



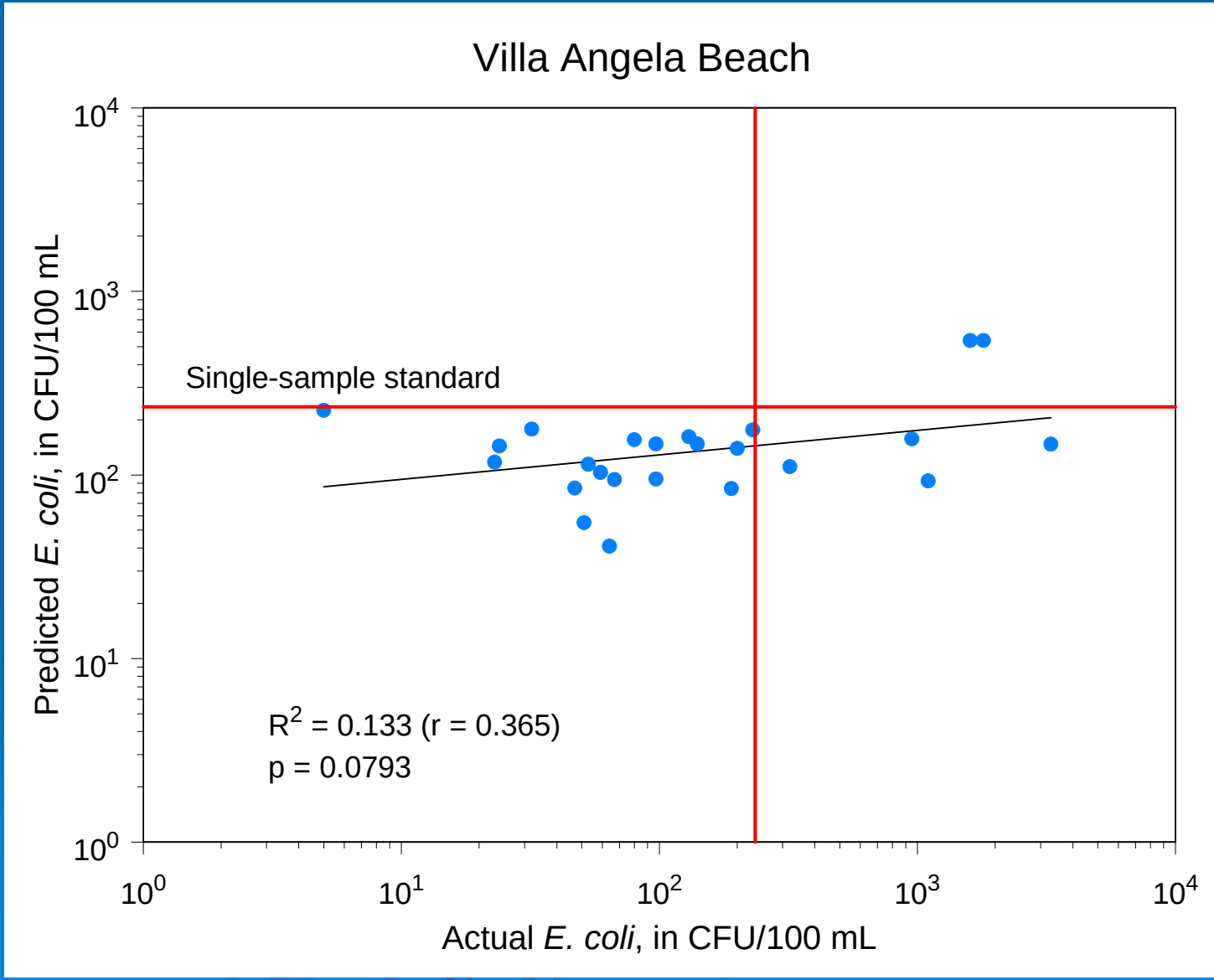
SLR results for *E. coli*

2005

Model variable:

log RLUs

(single filtration)



SLR versus yesterday's *E. coli*

	Correct Responses	False Positive	False Negative
Edgewater			
SLR	88.0% (21)	0% (0)	13.0% (3)
Yesterday's count	73.9% (17)	13.0% (3)	13.0% (3)
Villa Angela			
SLR	83.0% (20)	0% (0)	17.0% (4)
Yesterday's count	73.9% (17)	13.0% (3)	13.0% (3)

Next steps

- Continue optimization of method
  - Test new antibody-bead complex
  - Bacteriophage to release ATP

- 2006-2008 study at Lake Erie beaches
  - Develop MLR models
  - Include measurements for chlorophyll
- 2006 study at Cuyahoga Valley National Park
  - Test MLR models

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