Waste Activated Sludge as a Feedstock and Inoculum for Anaerobic Digestion

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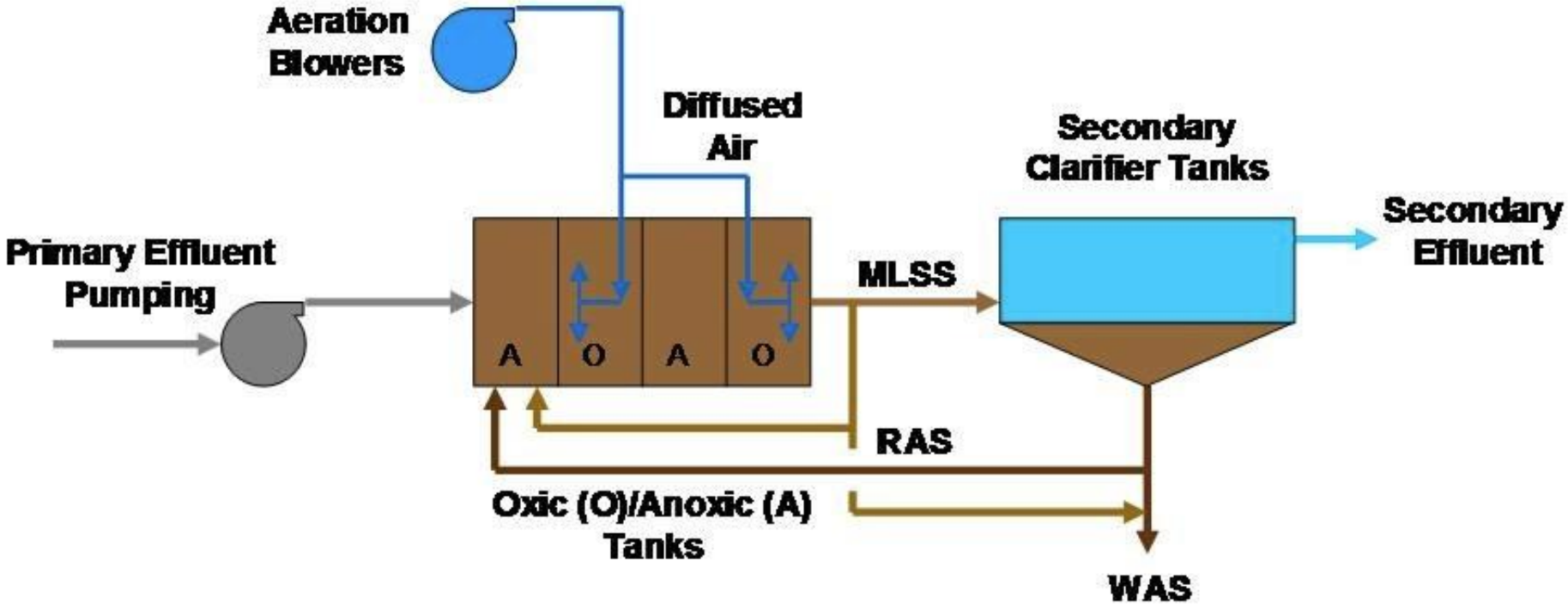
Introduction: Waste Activated Sludge (WAS)

- WAS: Excess biosolids produced in the activated sludge process of waste water treatment



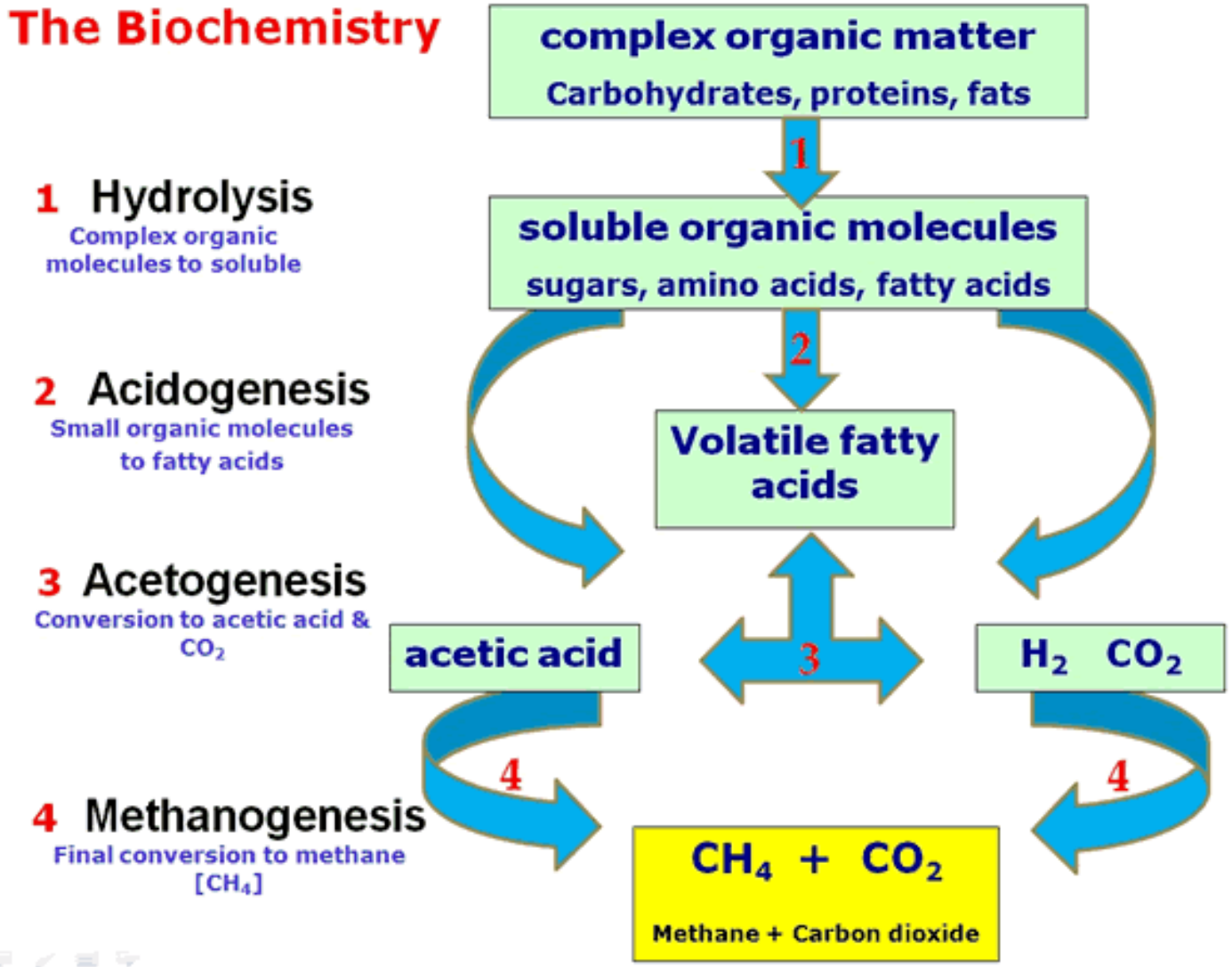
The Problem

- WAS management accounts for 60% of the total operating cost
- Current disposal methods:
  - Land application
  - Landfill
  - Incineration
  - Anaerobic Digestion

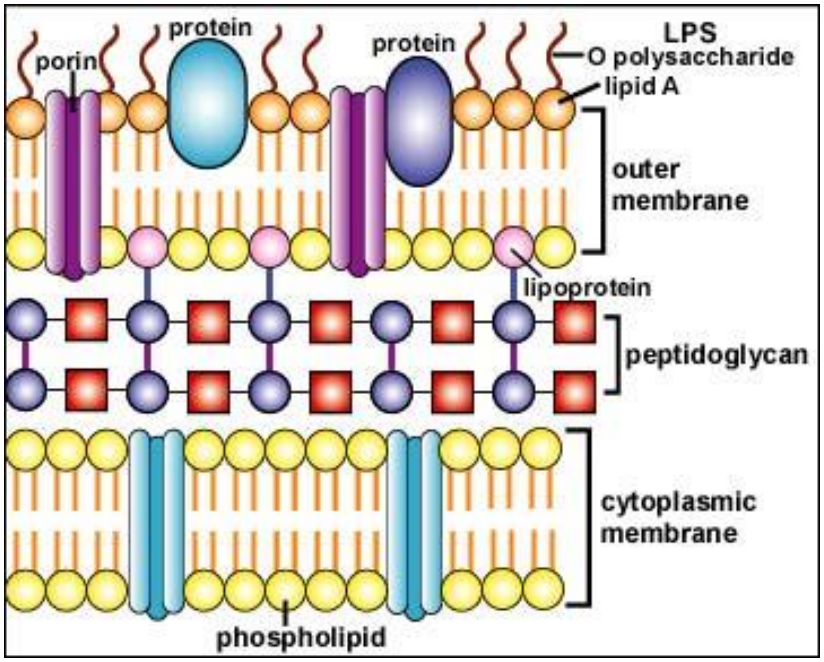
Waste Water Treatment at UF

- The University of Florida treats its own wastewater
  - 3 million – 5 million gallons/day
- WAS picked up once a week by GRU
  - Treated and land applied

Anaerobic Digestion



Anaerobic Digestion of WAS



- WAS is composed of Gram Negative bacterial cells
  - Thin layer of peptidoglycan covered by outer membrane
  - Limits hydrolysis
  - Pretreatment required to release soluble, digestible contents

WAS as an inoculum

- Waste water treatment: anaerobic and aerobic conditions
- Methanogens: anaerobic conditions
- If methanogens are present, WAS could inoculate itself
- Eliminate need for external inoculum → lower operating cost

Objectives

- Test pretreatment methods of WAS and compare levels of cell destruction
- Determine the potential of using WAS as an inoculum for anaerobic digestion

Objective One

Pretreatment of WAS

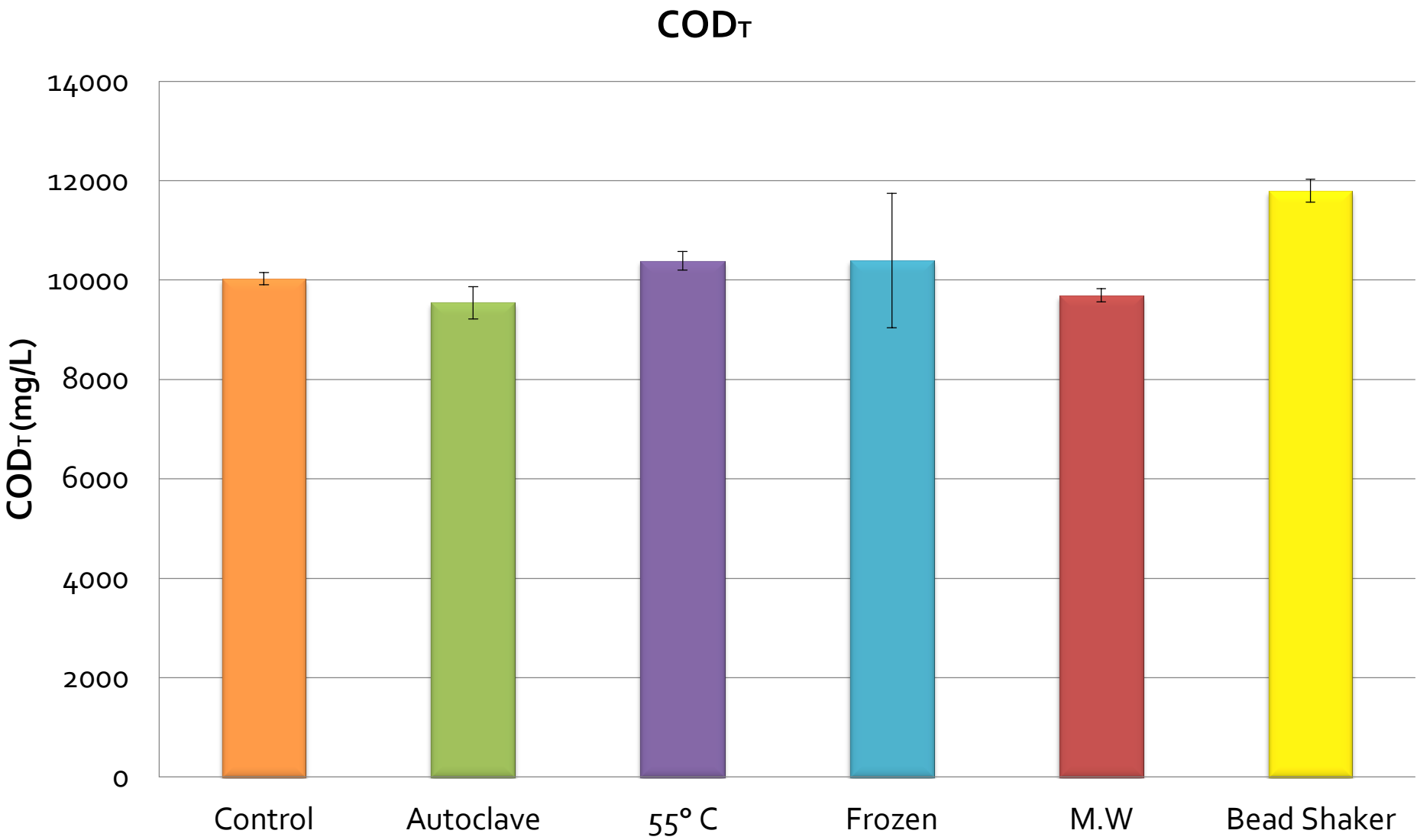
Methods

- Samples collected from the University of Florida Waste Water Treatment Plant
- 5 pretreatments performed in triplicate:
  - Microwave (30 seconds at 1500 W)
  - Autoclave (30 minutes)
  - Bead Shaker (5 minutes)
  - Frozen (24 hours, 20°C)
  - Incubator at 55°C (24 hours)

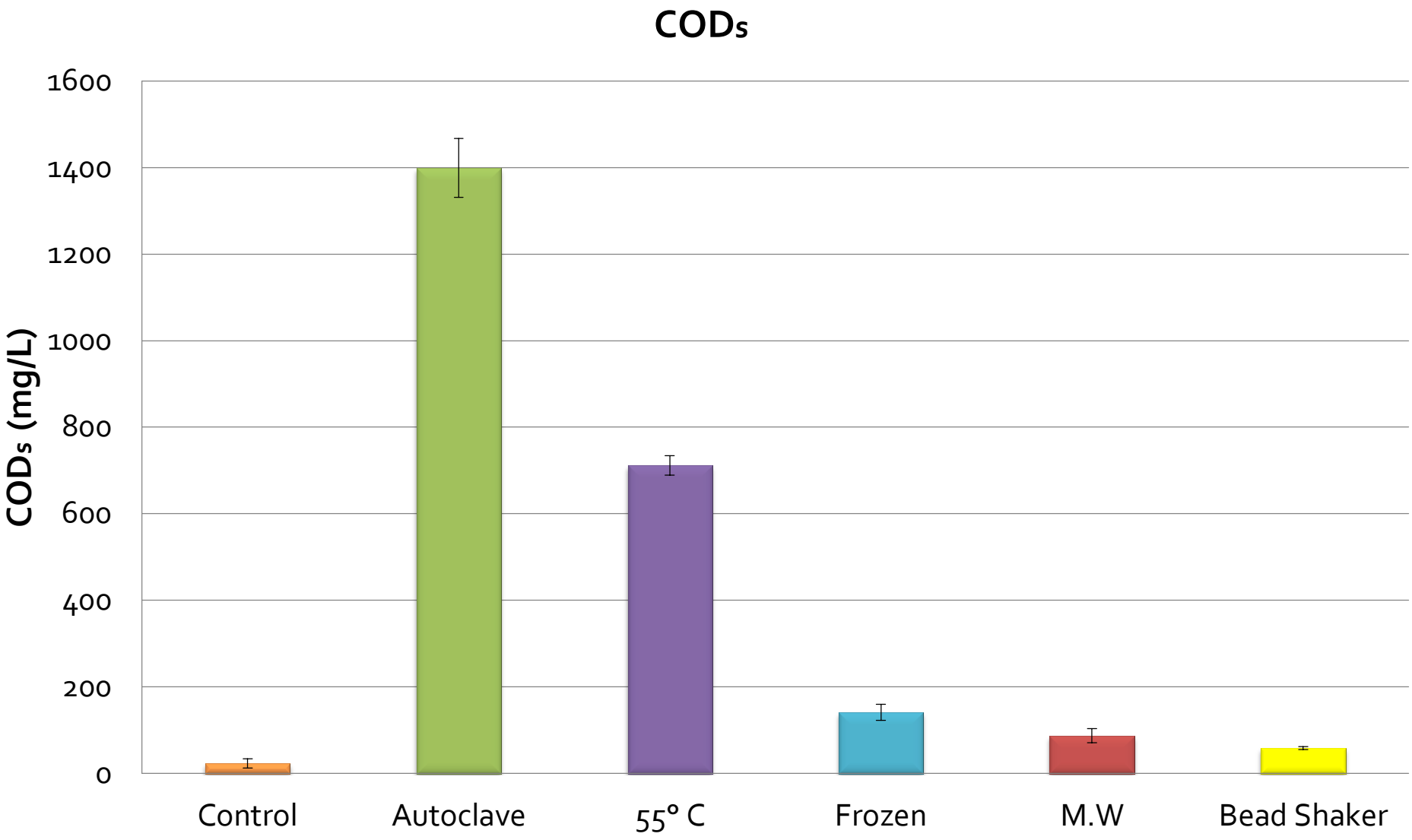
Methods

- Total Chemical Oxygen Demand (COD_T) measured
  - Samples (triplicate) diluted 1:10 with deionized water
- Soluble Chemical Oxygen Demand (COD_S) measured
  - Centrifuged at 4,000 rpm for 30 minutes, supernatant tested
- Microscopic examination for fractured cells

Results


COD$_T$
COD$_T$ (mg/L)
14000
12000
10000
8000
6000
4000
2000
0
Control
Autoclave
55° C
Frozen
M.W
Bead Shaker

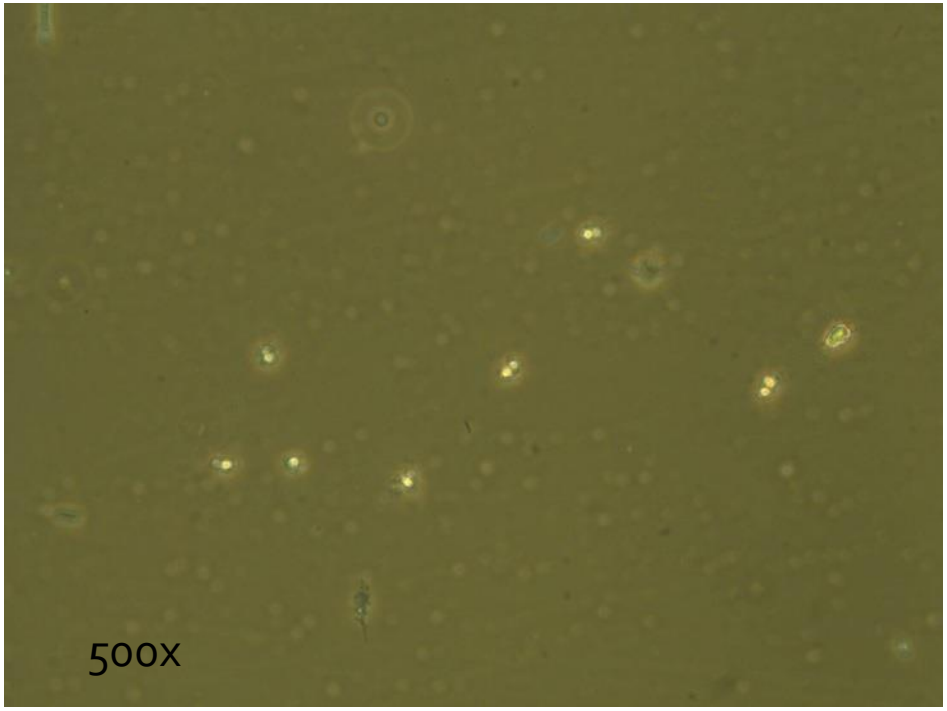
Results

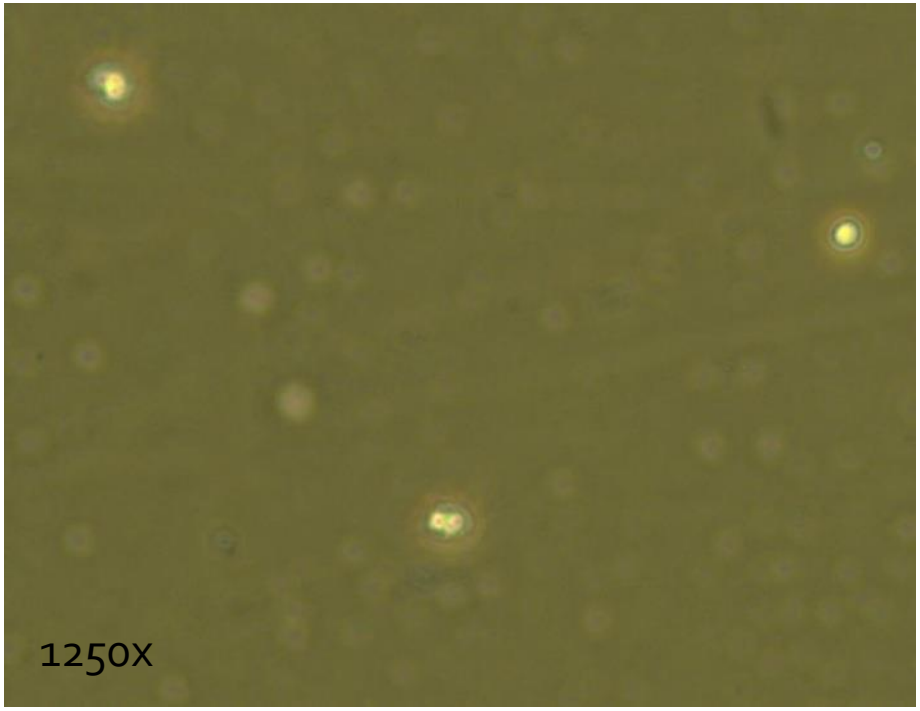

CODs
CODs (mg/L)
1600
1400
1200
1000
800
600
400
200
0
Control
Autoclave
55° C
Frozen
M.W
Bead Shaker

Results

Control (Untreated WAS)

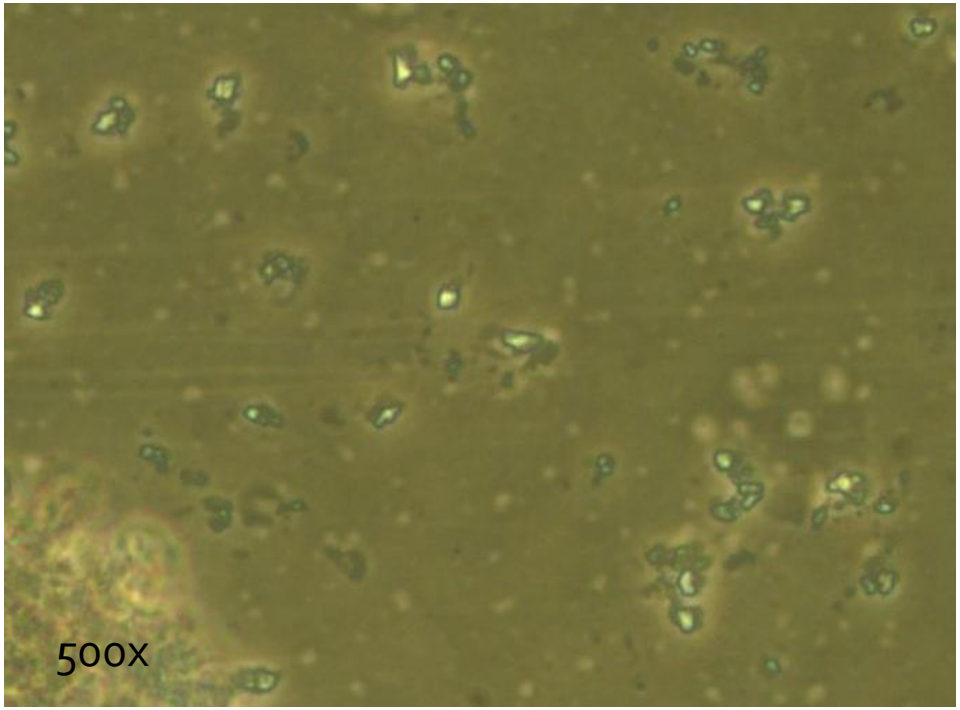
Intact cells under phase contrast illumination

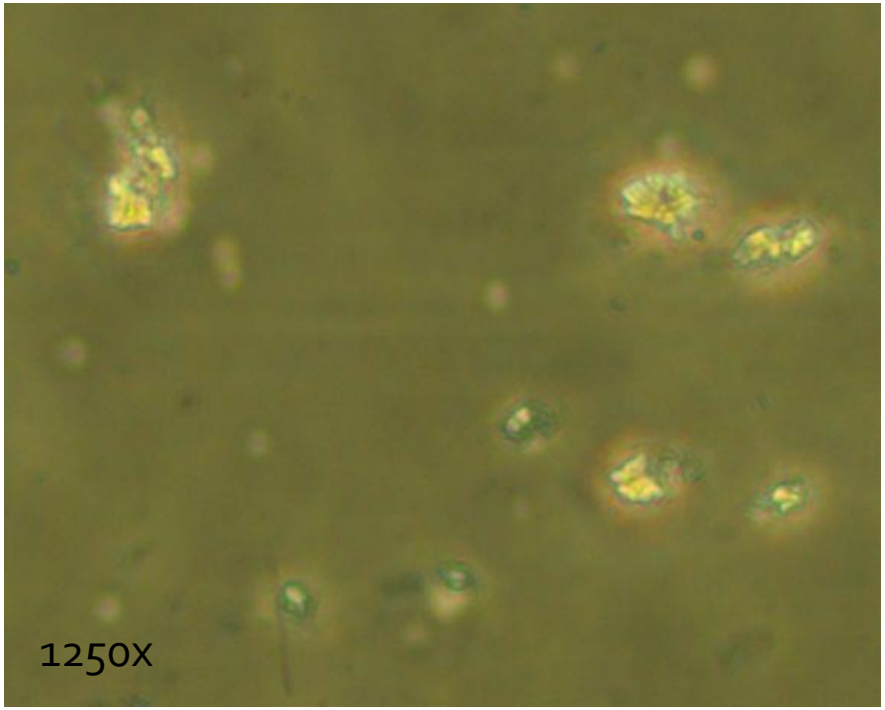




Results

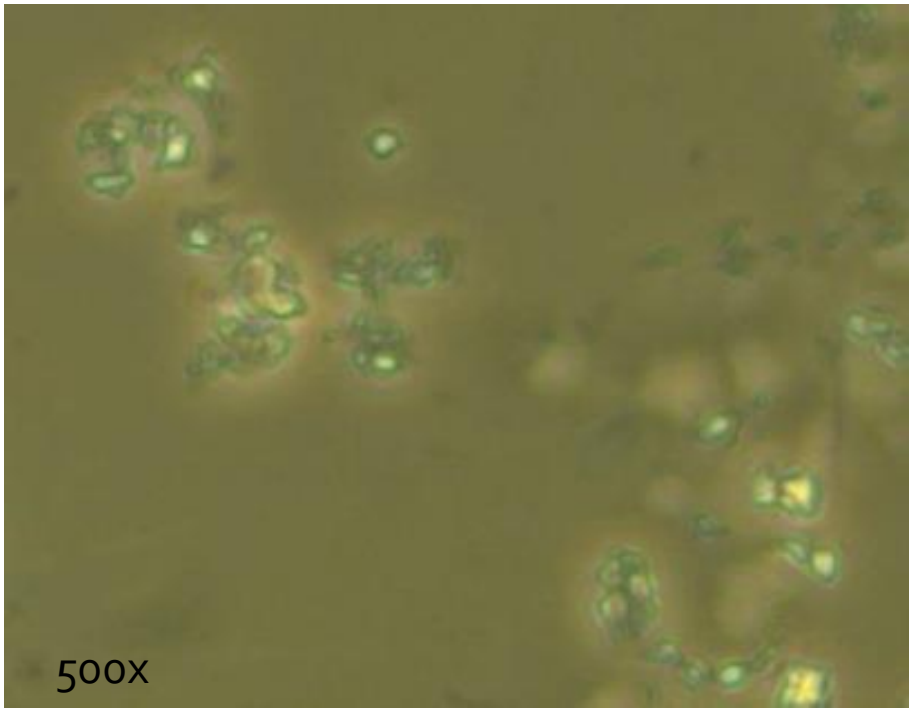
Pretreatment: Autoclave for 30 minutes

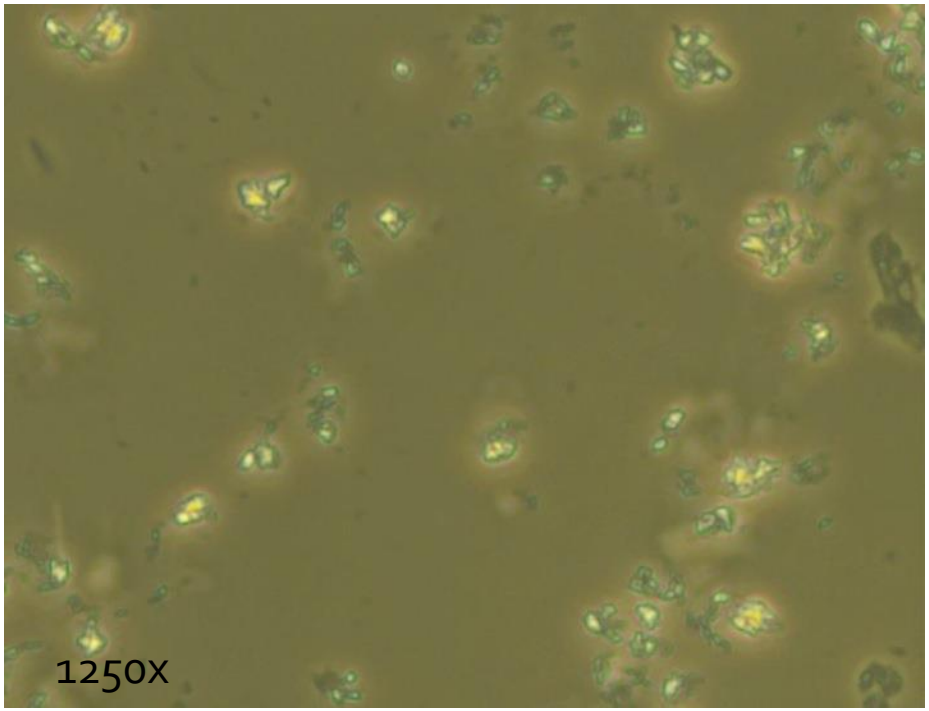




Results

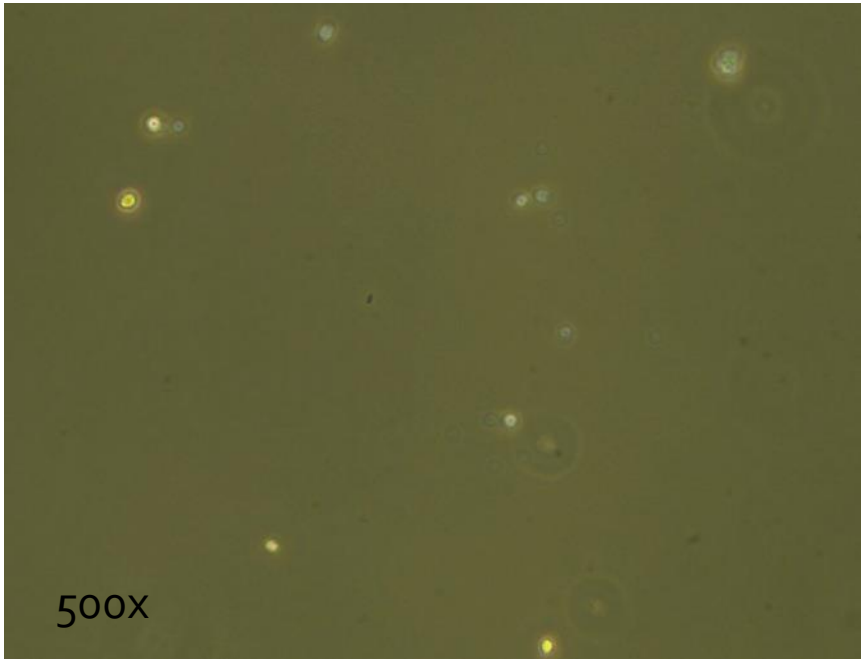
Pretreatment: 55°C for 24 hours

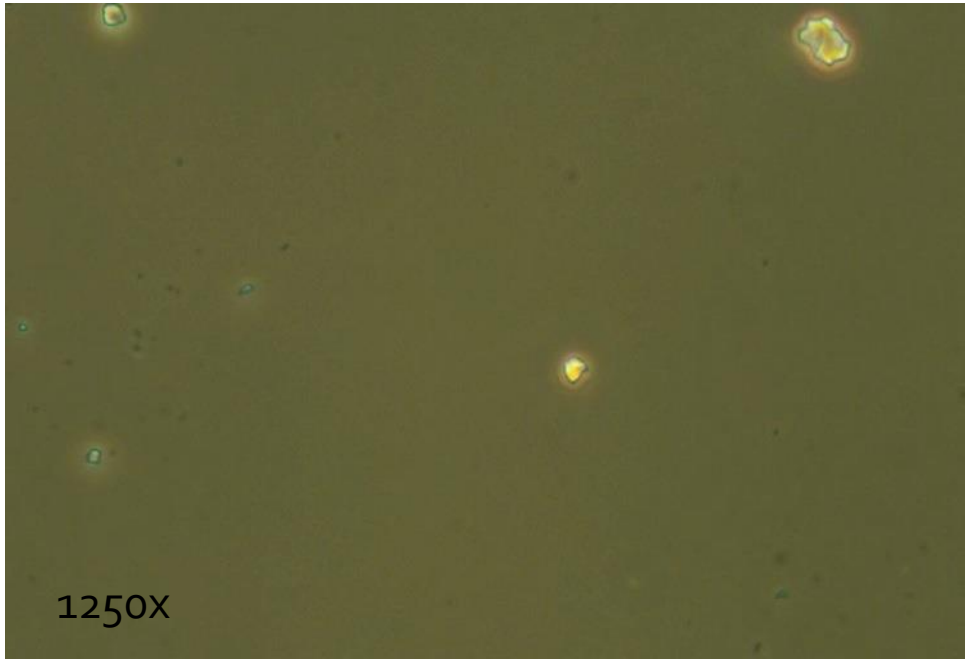




Results

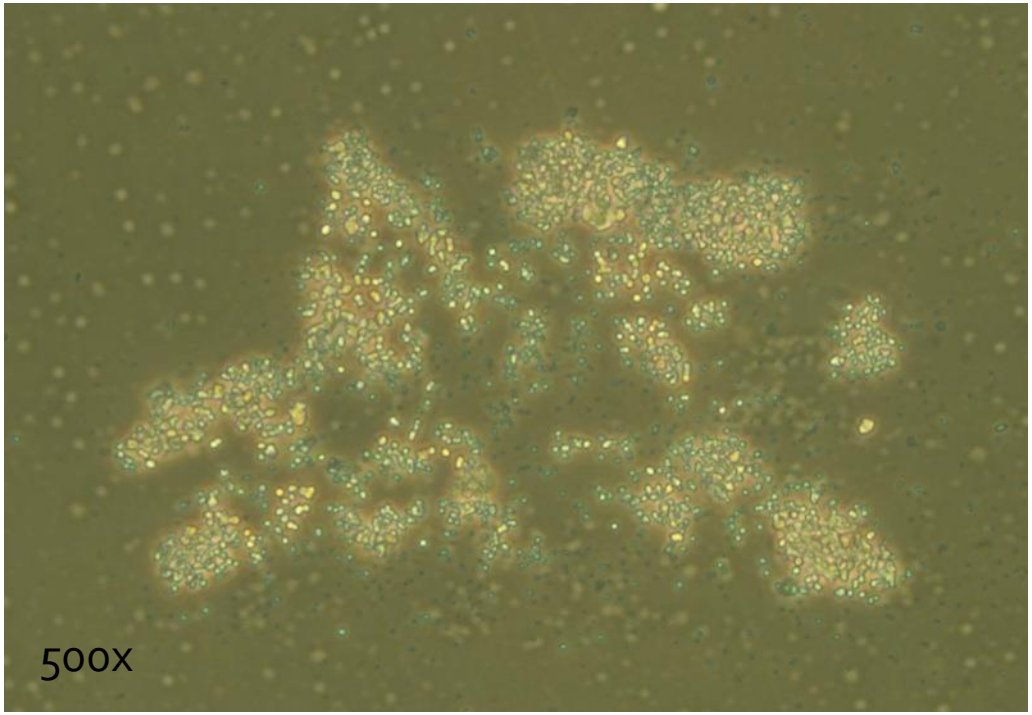
Pretreatment:
Frozen for 24 hours

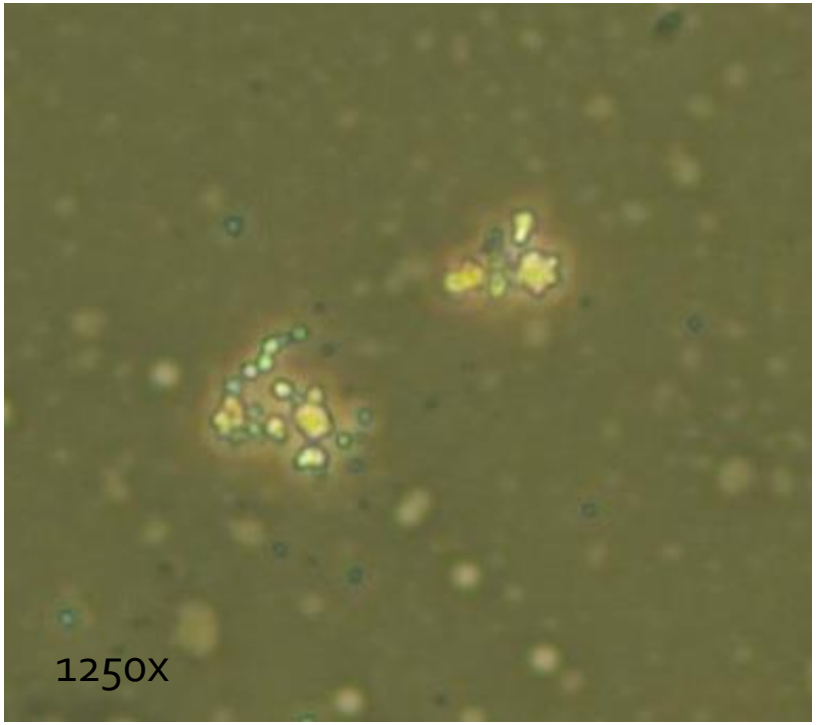




Results

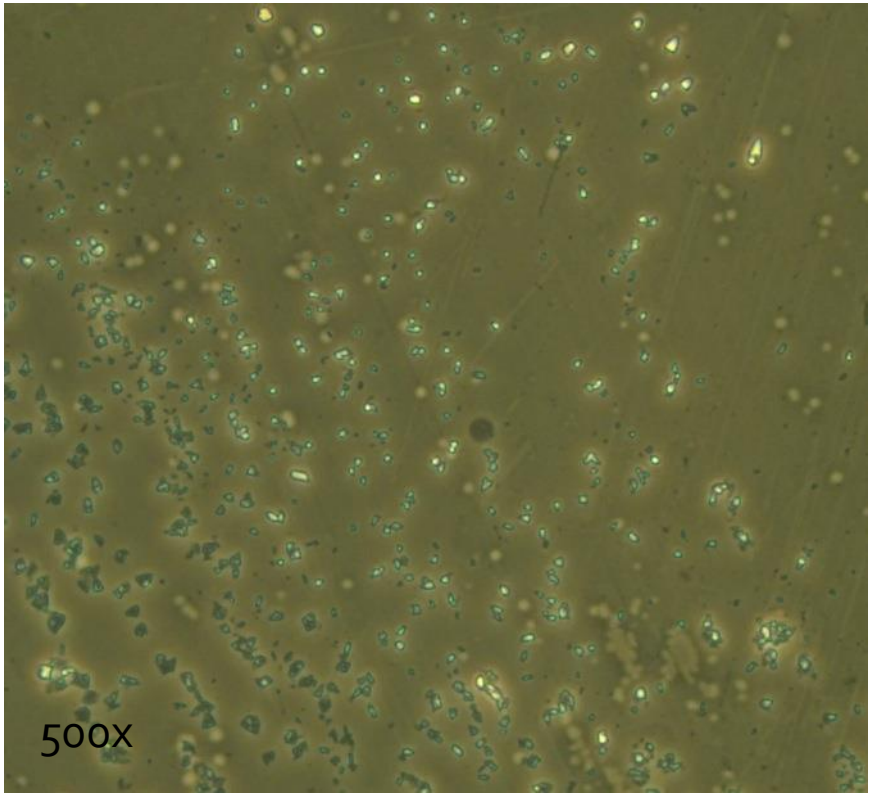
Pretreatment: Microwaved for 30 seconds (1500 W)

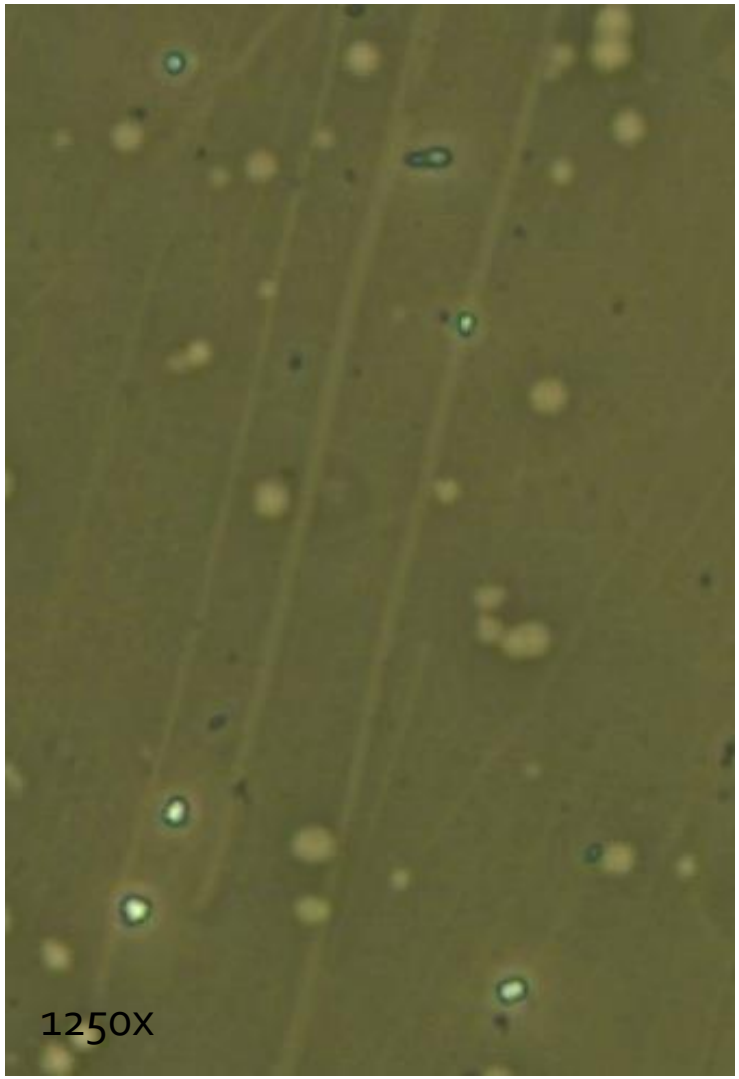




Results

Pretreatment: Bead Shaker for 5 minutes





Objective Two

WAS as an Inoculum

Methods

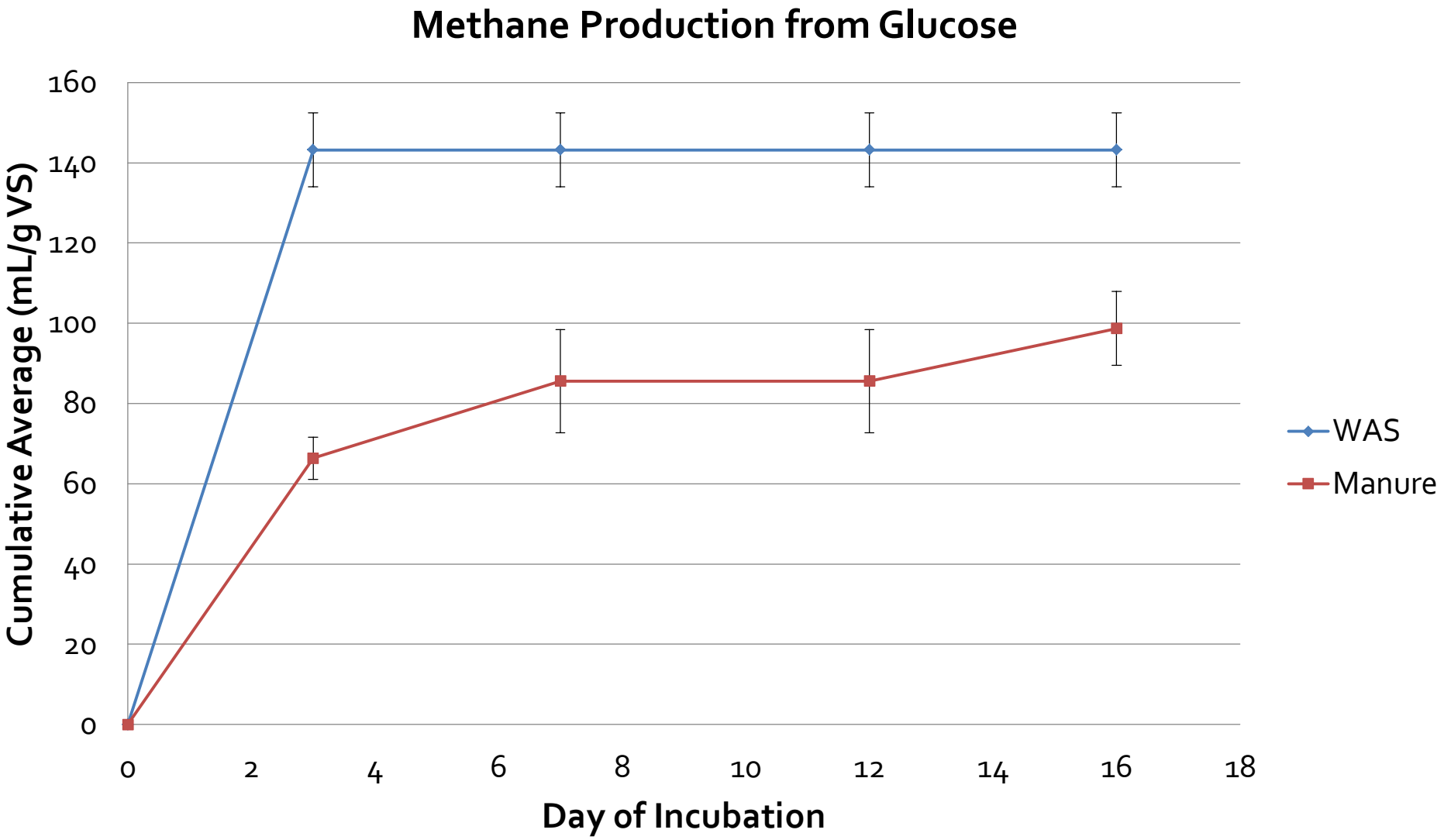
- Incubation Test:
  - WAS and Dairy Manure used to digest glucose and cellulose
  - Incubated at 35°C for 16 days
  - Biochemical Methane Potential (BMP) assays measured

Inocula	Substrate
WAS (200 mL)	Glucose (0.4 g)
	Cellulose (0.4 g)
Dairy Manure (200 mL)	Glucose (0.4 g)
	Cellulose (0.4 g)

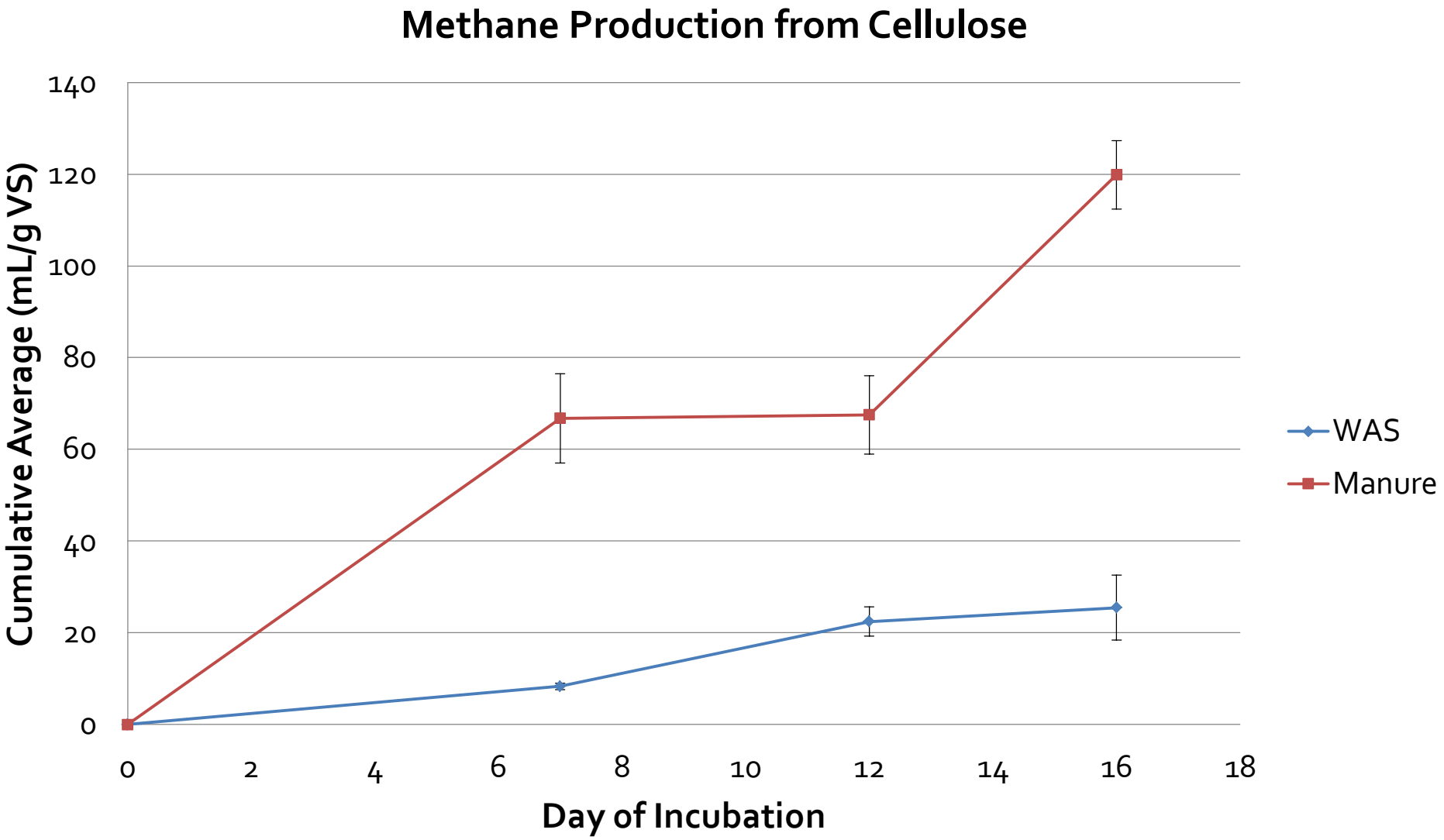
Methods

- pH measured after incubation test
- A fluorospectrometer (Nanodrop 3300) was used for methanogen detection before and after incubation test
  - Methanogens fluoresce blue (F_{420} Coenzyme)
  - 2μL of sample exposed to UV light and fluorescence recorded at 470nm

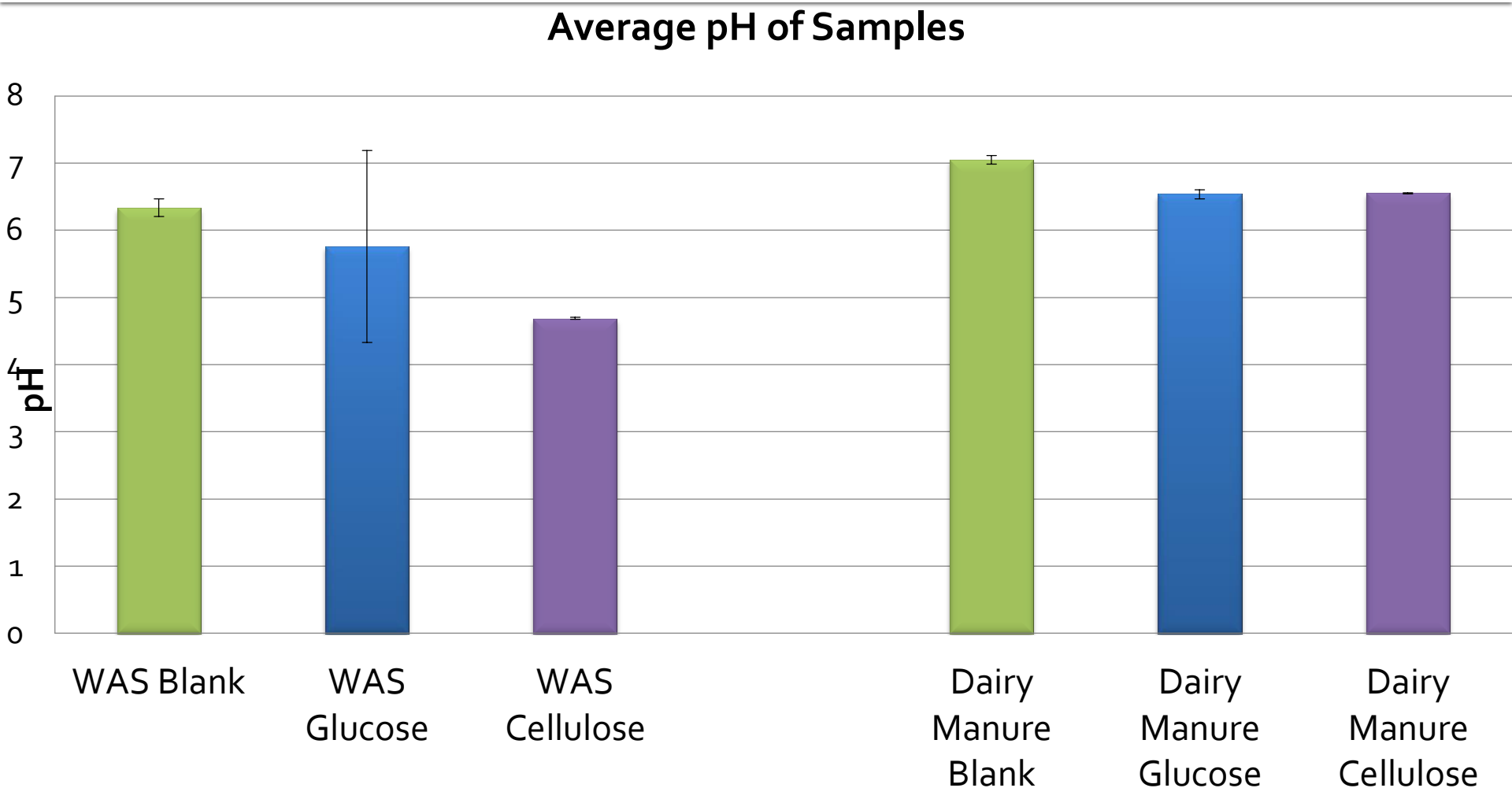
Results


Methane Production from Glucose
Cumulative Average (mL/g VS)
160
140
120
100
80
60
40
20
0
0
2
4
6
8
10
12
14
16
18
Day of Incubation
WAS
Manure

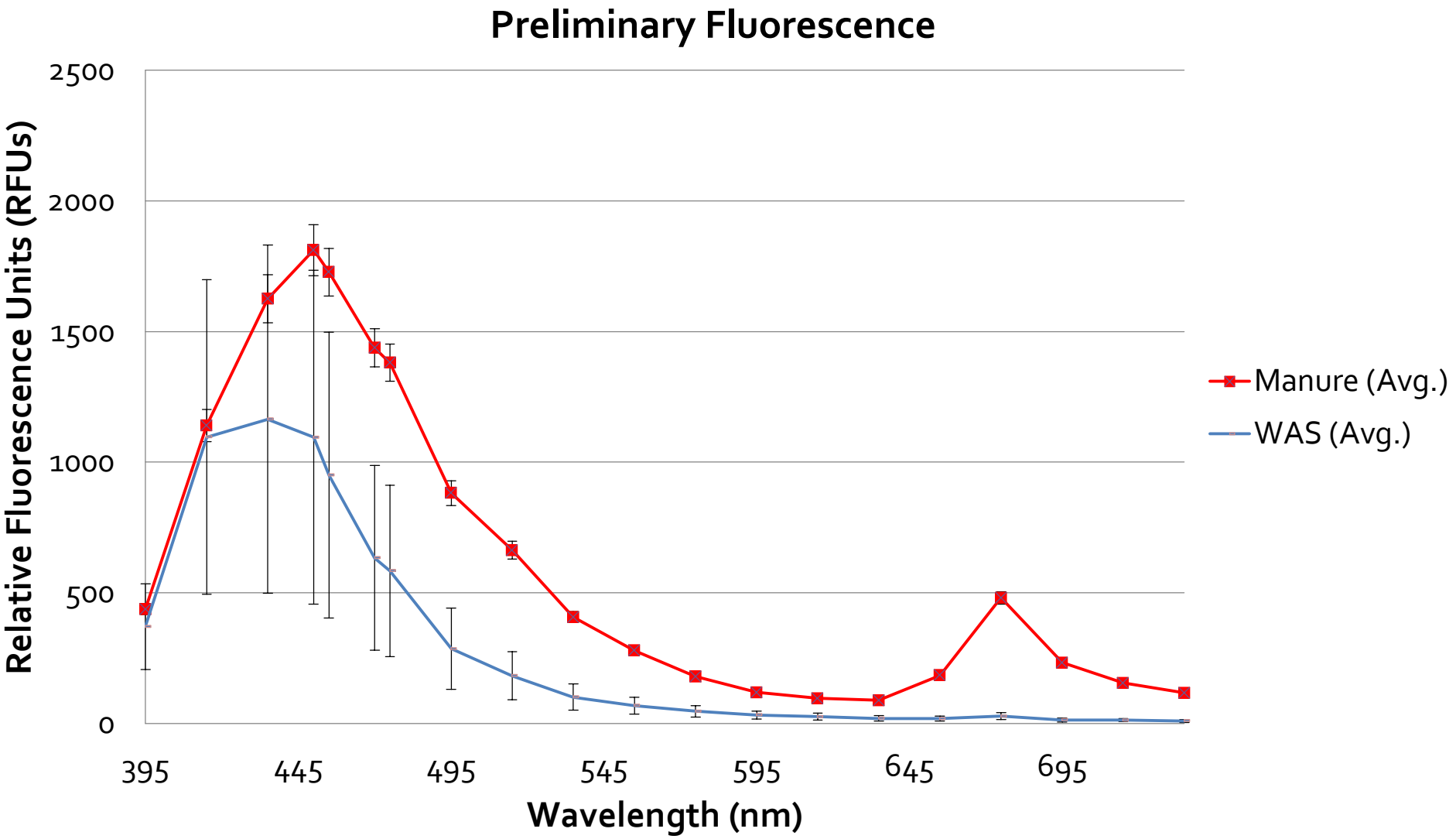
Results


Methane Production from Cellulose
Cumulative Average (mL/g VS)
140
120
100
80
60
40
20
0
0
2
4
6
8
10
12
14
16
18
Day of Incubation
WAS
Manure

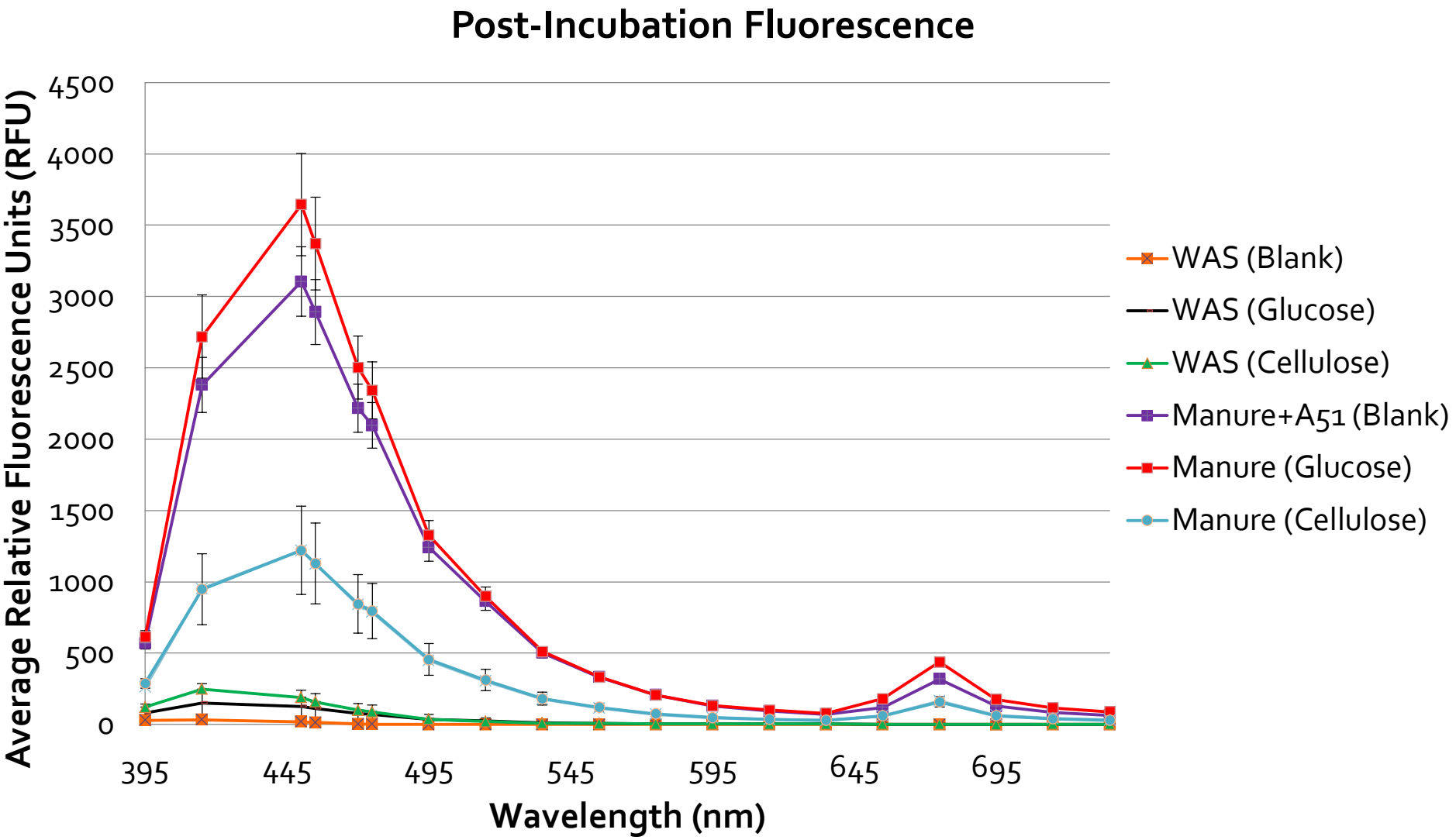
Results


Average pH of Samples
pH
8
7
6
5
4
3
2
1
0
WAS Blank
WAS Glucose
WAS Cellulose
Dairy Manure Blank
Dairy Manure Glucose
Dairy Manure Cellulose

Results


Preliminary Fluorescence
Relative Fluorescence Units (RFUs)
2500
2000
1500
1000
500
0
395
445
495
545
595
645
695
Wavelength (nm)
Manure (Avg.)
WAS (Avg.)

Results


Post-Incubation Fluorescence
Average Relative Fluorescence Units (RFU)
4500
4000
3500
3000
2500
2000
1500
1000
500
0
395
445
495
545
595
645
695
Wavelength (nm)
WAS (Blank)
WAS (Glucose)
WAS (Cellulose)
Manure+A51 (Blank)
Manure (Glucose)
Manure (Cellulose)

Conclusions

- WAS Pretreatment:
  - Cell lysis achieved under pretreatments
  - The autoclave was the best form of pretreatment, followed by heating at 55°C
- WAS as an inoculum:
  - Not as effective as the control
  - Became rapidly too acidic

Future Studies

- WAS Pretreatment:
  - Optimization of pretreatment techniques
  - Comparison of energy requirements for each pretreatment
- WAS as an Inoculum
  - Determine optimal loading rate
  - Full scale testing using pretreated W.A.S. as a feedstock

Thank you!

- Any questions?