

Effects of Biogas and Septic Tank Sludge Centrate on Microalgae Cultivation

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Introduction (Microalgae and Anaerobic digestion)

- Integration of microalgae-based anaerobic digestion (AD) and wastewater treatment (WWT) can provide many environmental and economic advantages:

- ✓ Electrical power and heat for AD
- ✓ Offset energy consumption for algae cultivation, processing and extraction
- ✓ Nutrients removal in WWT
- ✓ Potential to replace activated sludge
- ✓ Potential to minimize CO₂ emission

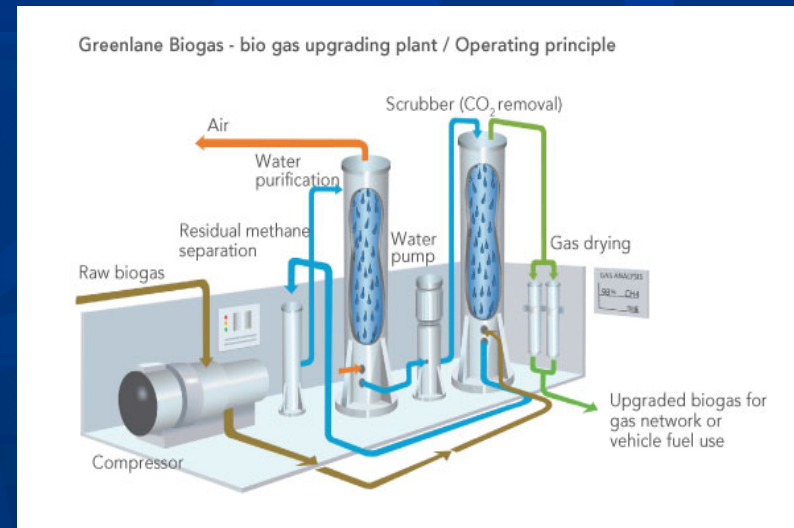


Algae cultivation pond in Israel for biofuel

<https://www.greenprophet.com/2011/09/seabiotic-biofuel-al>

Introduction (Biogas & biogas purification)

- Main components of biogas:
 - ✓ Methane CH_4 (40-70%)
 - ✓ Carbon dioxide CO_2 (30-60% downgrade engine efficiency)
 - ✓ Hydrogen sulfide H_2S (trace levels, contributes to corrosion of pipelines and engines)
 - ✓ Water vapor
- Current purification methods (physical absorption, chemical solvents, membrane separation, etc.) are energy intensive.

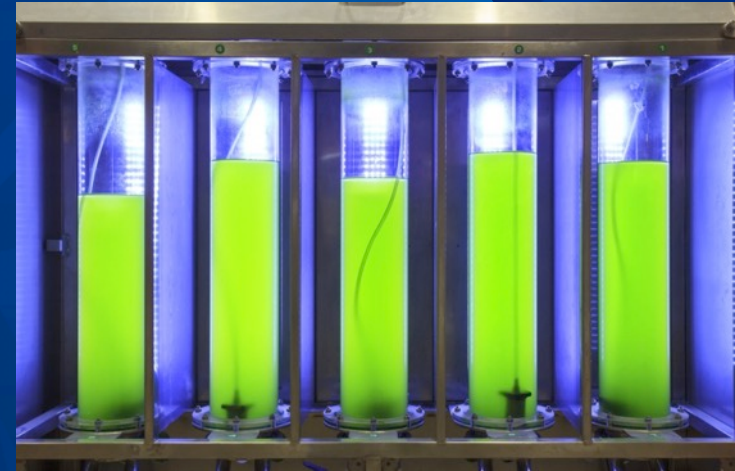


Biogas purification system by pressurized water (physical treatment)

http://www.sarlin.com/sarlin_products/Biogas-upgrading-plants/z3awigjh/2a12e6e6-e2e0-4eb1-a63f-9399f4ca1b25

Introduction (Biogas purification by microalgae)

- Algae can scrub CO_2 and H_2S from biogas:
 - ✓ Low cost
 - ✓ High efficiency
 - ✓ Increases microalgal productivity and nutrient assimilation
 - ✓ CO_2 from biogas can control the pH (~ 8.0) in culture solution (prevent NH_3^{\uparrow} and PO_4^{\downarrow})
 - ✓ First proposed by Oswald and Golueke in 1960
 - ✓ Minimize the CO_2 emission to atmosphere



Biogas purification system by algae
(biological treatment)

<https://www.israel21c.org/is-2016-the-year-of-the-algae/>

Introduction (microalgae and WWT)

- Cultivation of microalgae in different types of wastewater :

- ✓ Reduce nutrients (N and P)
- ✓ Reduce soluble chemical oxygen demand (sCOD)
- ✓ Increased algae production
- ✓ Currently tested on primary effluent, industrial effluent and high-strength wastewater (Wang, et al. 2010; Wang, et al 2014)



The use of algae in wastewater treatment

[https://doi.org/10.1016/S0015-1882\(14\)70180-6](https://doi.org/10.1016/S0015-1882(14)70180-6)

Introduction (Septic tank sludge and its centrate)

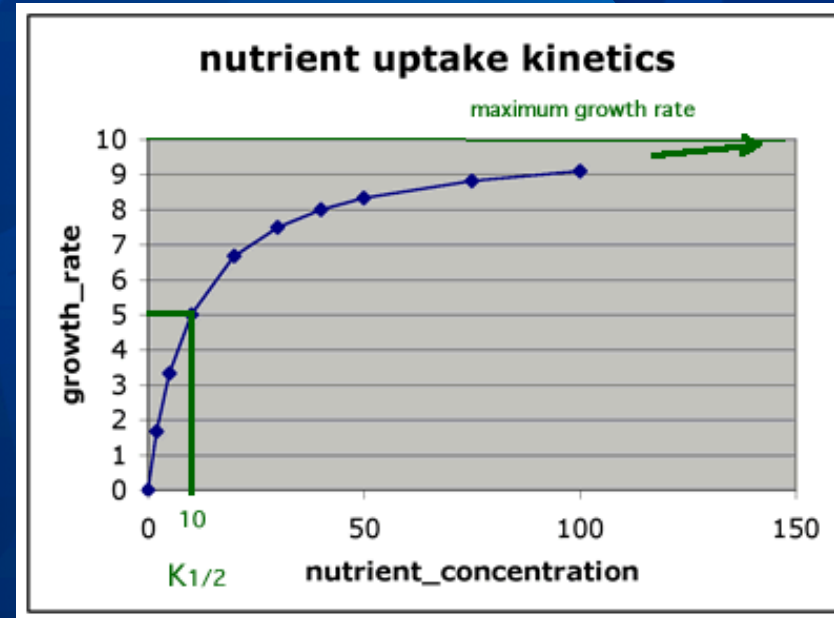
- Septic tank sludge (STS):
 - ✓ Is the final product of onsite wastewater treatment system (septic system)
 - ✓ 20 % population in the US relies on septic systems
 - ✓ Usually has high concentrations of organic carbon (~10,000 mg/L), ammonia nitrogen (~250 mg/L), and phosphate (~55 mg/L) (Diaz-Valbuena et al. 2011).
- Septic tank sludge centrate (STSC):
 - ✓ Rarely studied
 - ✓ Potentially a good medium for algae culture



STSC

Introduction (algae growth kinetics)

- Ideal microalgae growth kinetics can be determined if cells are grown under similar conditions (as shown in curve)
- The growth rate is always controlled by the most limited nutrient
- The nutrient levels can also affect the lipid content in cells (Khozin-Goldberg and Cohen 2006, Rodolfi et al. 2009, Xin et al. 2010)



Graphical representations of ideal microalgae growth

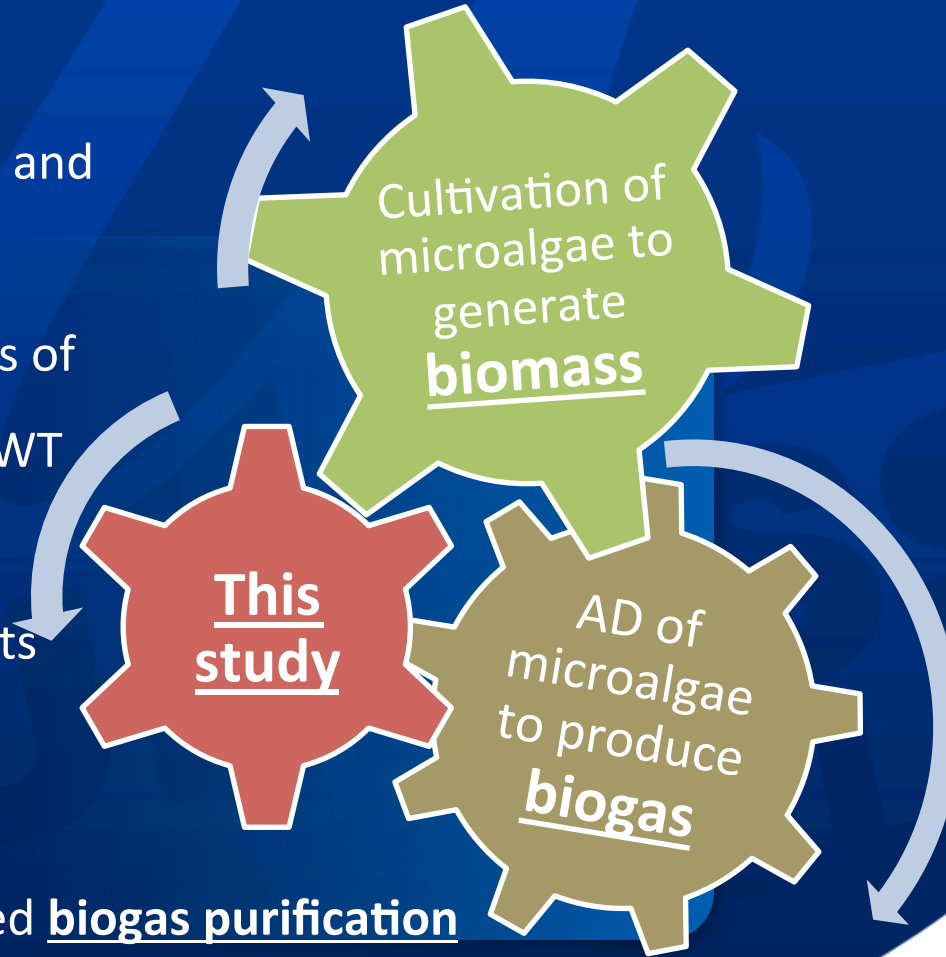
<http://web.pdx.edu/~rueterj/courses/esr472/>

Objectives

- Evaluate the effects of using biogas and septic tank sludge centrate (STSC) as the major carbon sources and growth medium for microalgal cultivation.
 1. Study the effect of biogas on microalgal biomass growth (autotrophic metabolism);
 2. Study the effect of STSC on microalgal biomass growth (heterotrophic metabolism);
 3. Analyze the effect of biogas on nutrient (N and P) removal from STSC;
 4. Study the kinetics of nutrient removal based on biological assimilation;
 5. Study the effect of biogas and STSC on microalgal lipid content.

Significance

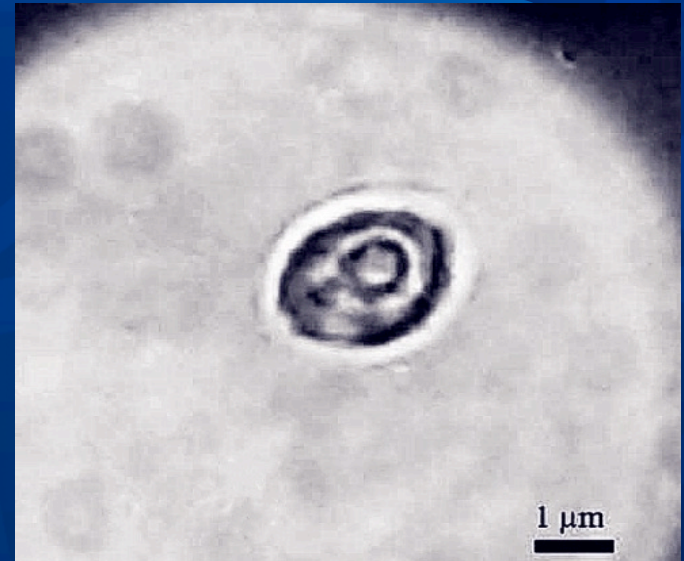
- AD of microalgae has been widely studied and proven to produce clean energy – methane
- Cultivation of microalgae in different types of wastewater for biomass production and WWT is also receiving attention
- This study aims to integrate the two aspects
- We believe:



➤ If the processes of microalgae-based biogas purification and anaerobic centrate reuse can be integrated, we could redefine the role of microalgae in environmental engineering

Materials (STSC and microalgae)

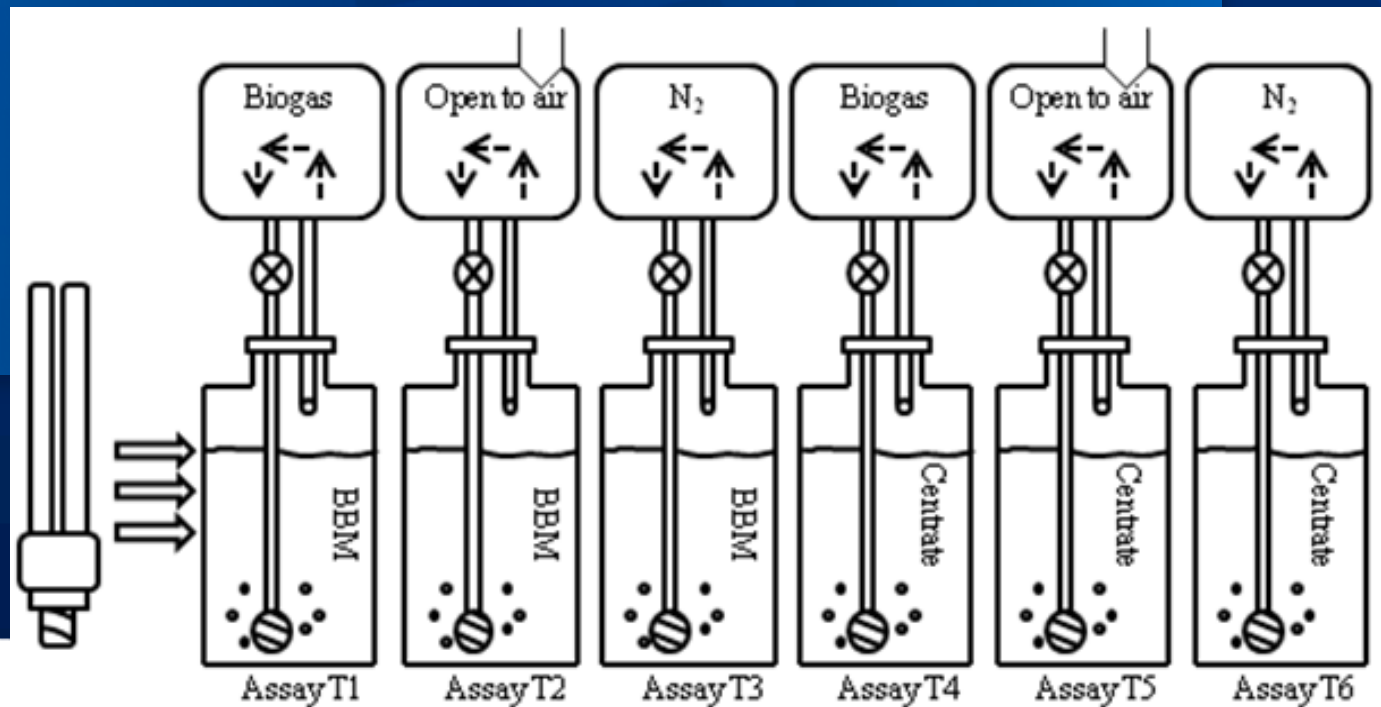
- STCS:
 - ✓ Filtered using glass fiber filters
 - ✓ Autoclaved at 120°C for 30 min
- Microalgae:
 - ✓ Strain was isolated from the primary clarifiers at the Lowell Regional Wastewater Utility
 - ✓ Cultivated in the modified Bold Basal's medium (BBM)



Single microalgal cell image obtained in Umass Lowell Environmental Lab

Methods (experimental design)

- 500 mL culture bottles with 400 mL working volume for 10 days
- Metal cap with two stainless steel fittings and rubber washer
- Biogas circulation from a Tedlar bag to culture solution, forced by a peristaltic pump (10mL/min)
- 30°C and 24 hr illumination at roughly 3,000 Lux



Schematic diagram for microalgae cultivation

Cultivation Apparatus



Methods (group set-up)

1. Assay (T1) : biogas and BBM
2. Assay (T2): air and BBM
3. Assay (T3): N₂ and BBM
4. Assay (T4) : biogas and STSC
5. Assay (T4) : air and STSC
6. Assay (T4) : N₂ and STSC

- Comparison between T1 and T2 can show the advantage of high concentration of CO₂ from biogas
- Comparison between T3 and T6 can test the potential of heterotrophic pathway of microalgae in STSC and can show the advantages of joint effect from biogas and STSC

Results (T1 vs. T2)

- Biomass Production (measured by volatile suspended solids VSS):

- ✓ T1's biomass production (890 mg/L)

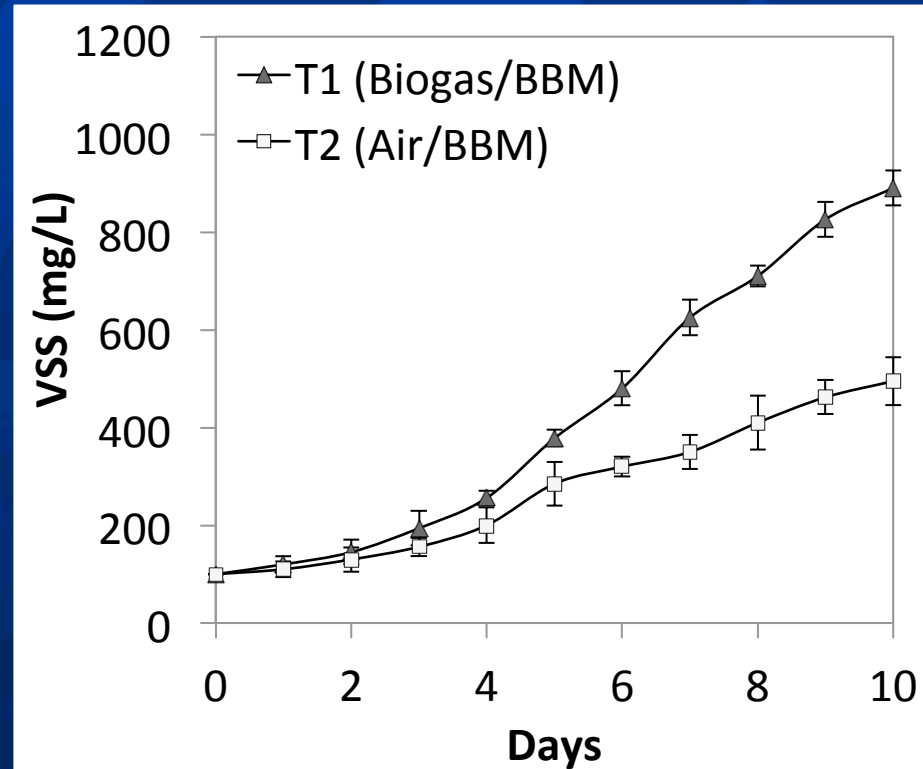
- ✓ T2's biomass production (495 mg/L)

- Specific growth rate (μ) (unit: day^{-1})

- ✓ T1 and T2 both had the highest μ at day 4-5

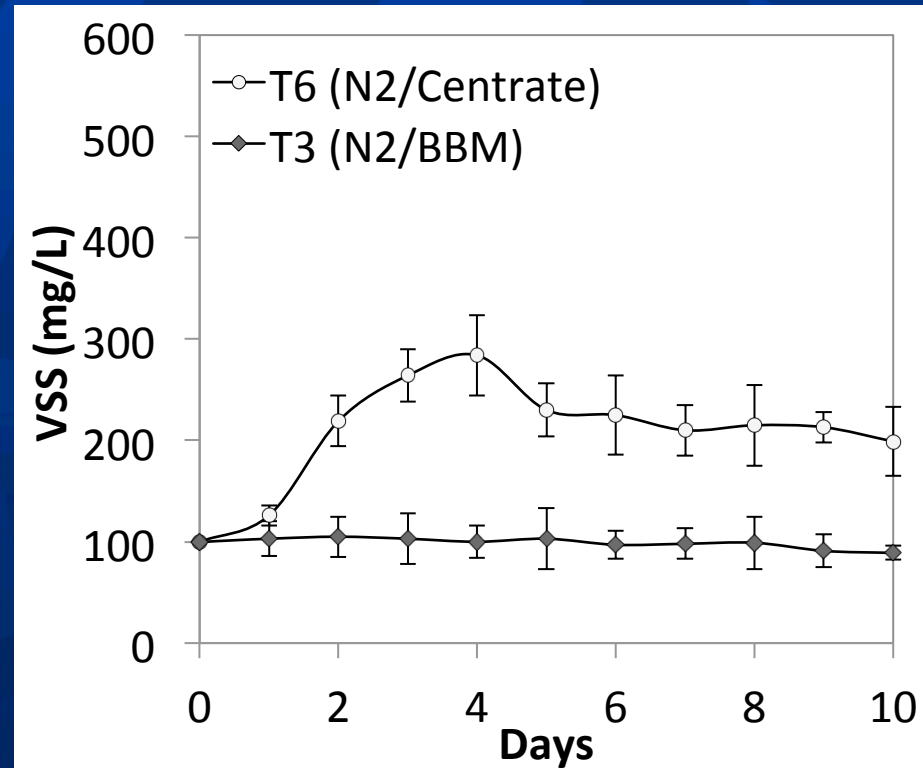
- ✓ T1 μ (0.53 d^{-1})

- ✓ T2 μ (0.35 d^{-1})



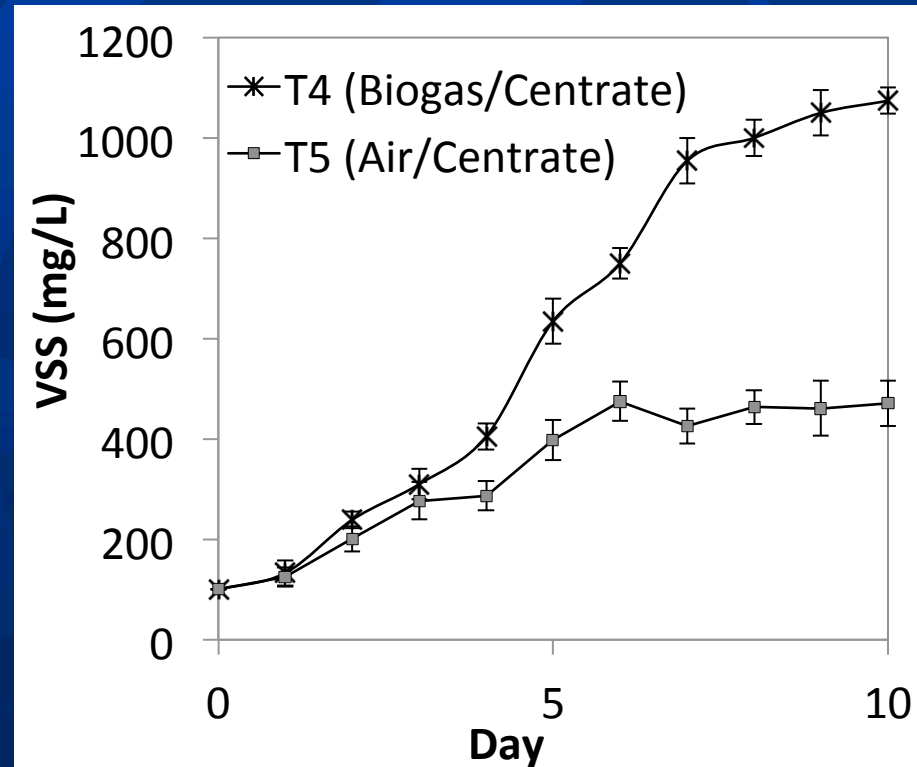
Results (T3 vs. T6)

- Biomass Production (measured by VSS):
 - ✓ T3's biomass production (negative)
 - ✓ T6's biomass production (306 mg/L)
- Specific growth rate (μ) (unit: day^{-1})
 - ✓ T6 had the highest μ at day 1-2
 - ✓ T6 μ (0.55 d^{-1})
- Results show a short hyper-growth period, indicating increased heterotrophic metabolism of microalgae in STSC



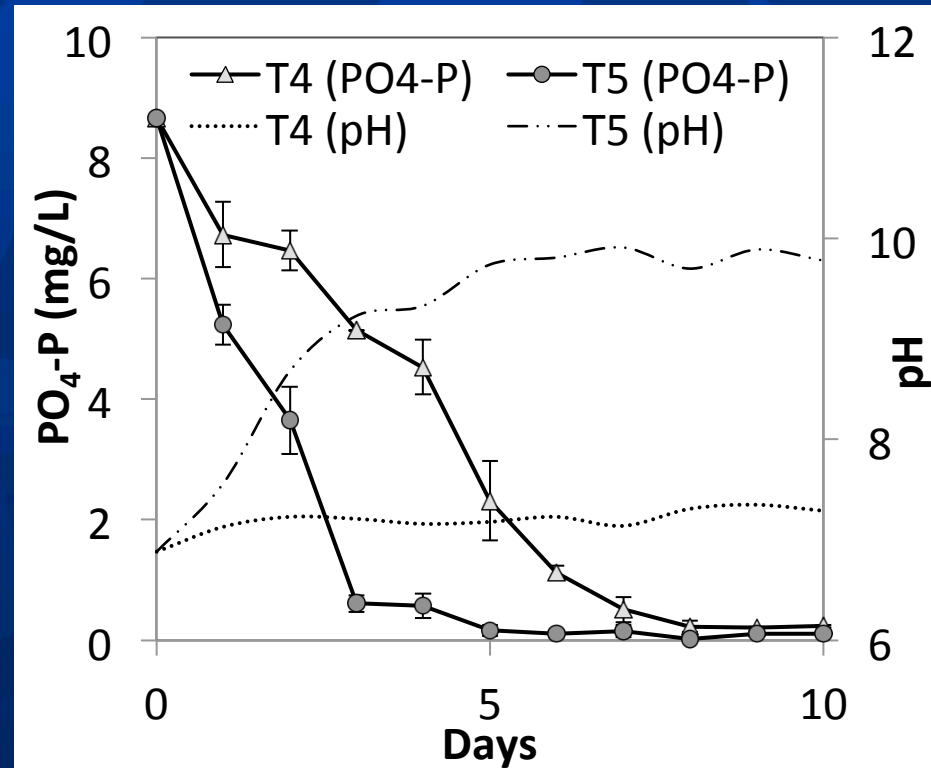
Results (T4 vs. T5)

- Biomass Production (measured by VSS):
 - ✓ T4's biomass production (1,074 mg/L)
 - ✓ T5's biomass production (471 mg/L)
- Specific growth rate (μ) (unit: day^{-1})
 - ✓ T4 had the highest μ at day 1-2 (0.59 d^{-1})
 - ✓ T5 had the highest μ at day 4-5 (0.45 d^{-1})
- Results show a maximum biosynthetic rate governed by mixotrophic metabolism



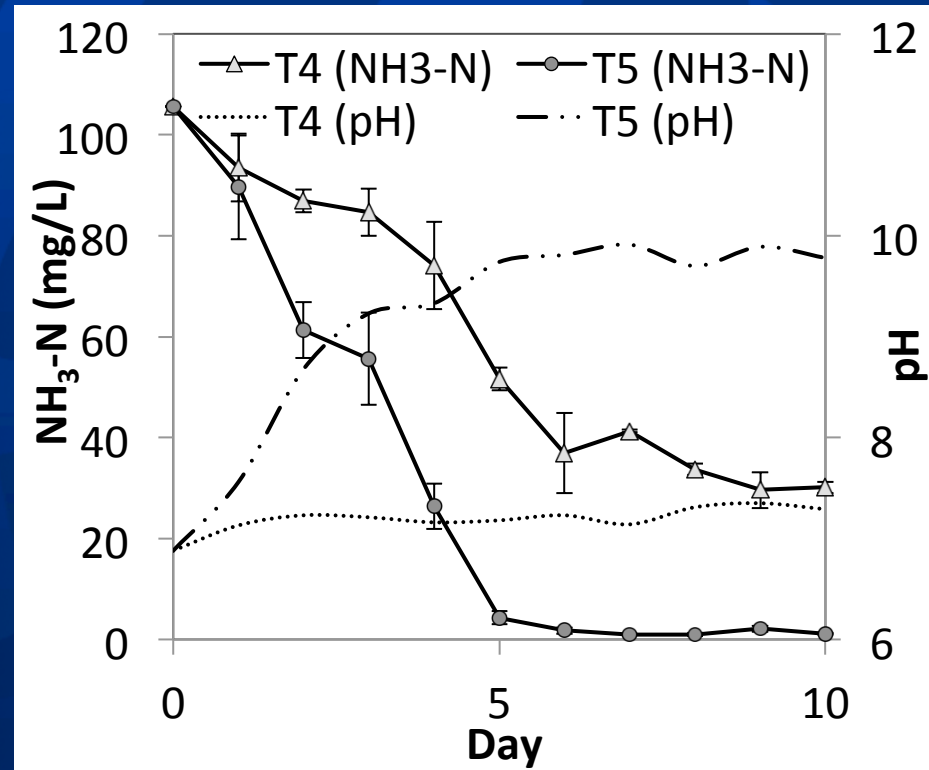
Results (Phosphate removal for T4 and T5)

- Both assays show significant reduction of phosphate (T4 = 97.27%, T5 = 98.87%)
- T4 had slow reduction rate $4.14 \text{ mg/L} \cdot \text{d}^{-1}$
- T5 had increased reduction rate of $8.10 \text{ mg/L} \cdot \text{d}^{-1}$
- Increase in pH with time of T5 indicates phosphate precipitation
- T4's pH was buffered due to high CO_2 concentration in biogas



Results (Ammonia removal for T4 and T5)

- T5 had greater ammonia removal than T4 (T4 = 71.50 %, T5 = 98.96%)
- Portion of volatilization of ammonia in T5 can be calculated by equation
 - $FAN/TAN = 1/(1+10^{(pK_a-pH)})$
- In T5, 80.92% of ammonia was volatilized (only 18.04% was used by microalgae)
- In T4, ammonia volatilization was neglected
- Again, in T4, the pH was buffered due to the presence of biogas



Results (Algae growth kinetics of nutrient removal for T4)

- (Y) Yield of biomass with respect to the nutrient consumption of phosphorus or nitrogen:

$$\text{➤ } Y_P = (B - B_0) / (P_0 - P); Y_N = (B - B_0) / (N_0 - N) \quad \{B \text{ is biomass}\}$$

- Y_P / Y_N is 8.95
- Ideal N/P ratio (Y_{ideal}) for microalgae is 7.2 ($C_{106}H_{181}O_{45}N_{16}P$) (Grobbelaar, 2003)
- $Y_P / Y_N > Y_{\text{ideal}}$ indicates the phosphorus was the limiting nutrient
- Kinetics of nutrient removal should focus on limiting nutrient P.

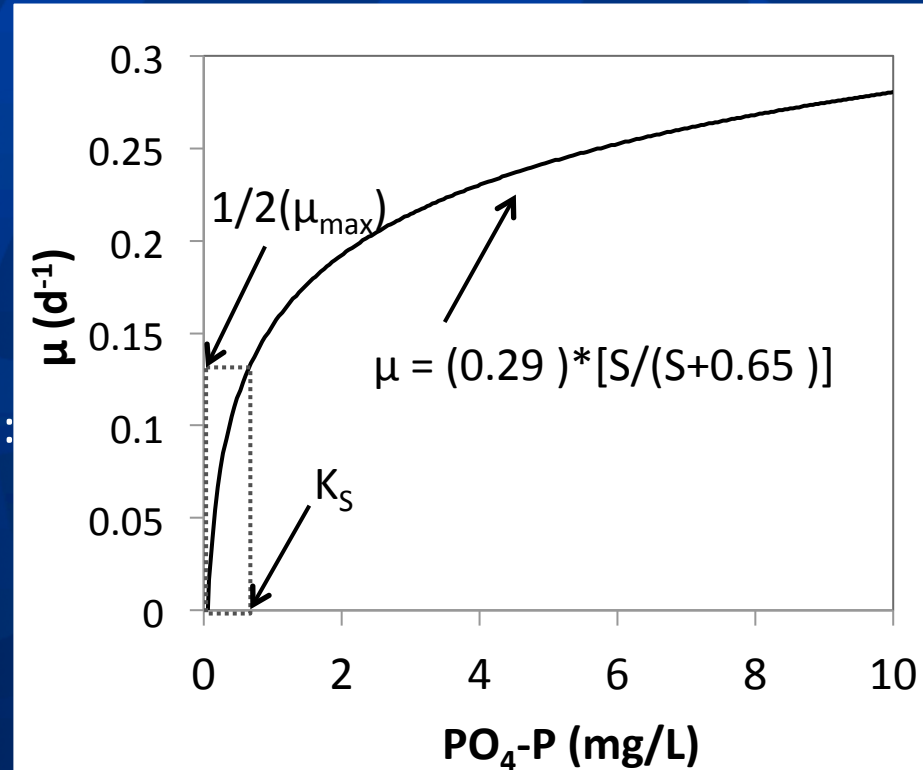
Results (Algae growth kinetics based on phosphate removal for T4)

- Monod equation was used

➤ $dx/x = [\mu_{\max} * S / (K_s + S)] * dt$

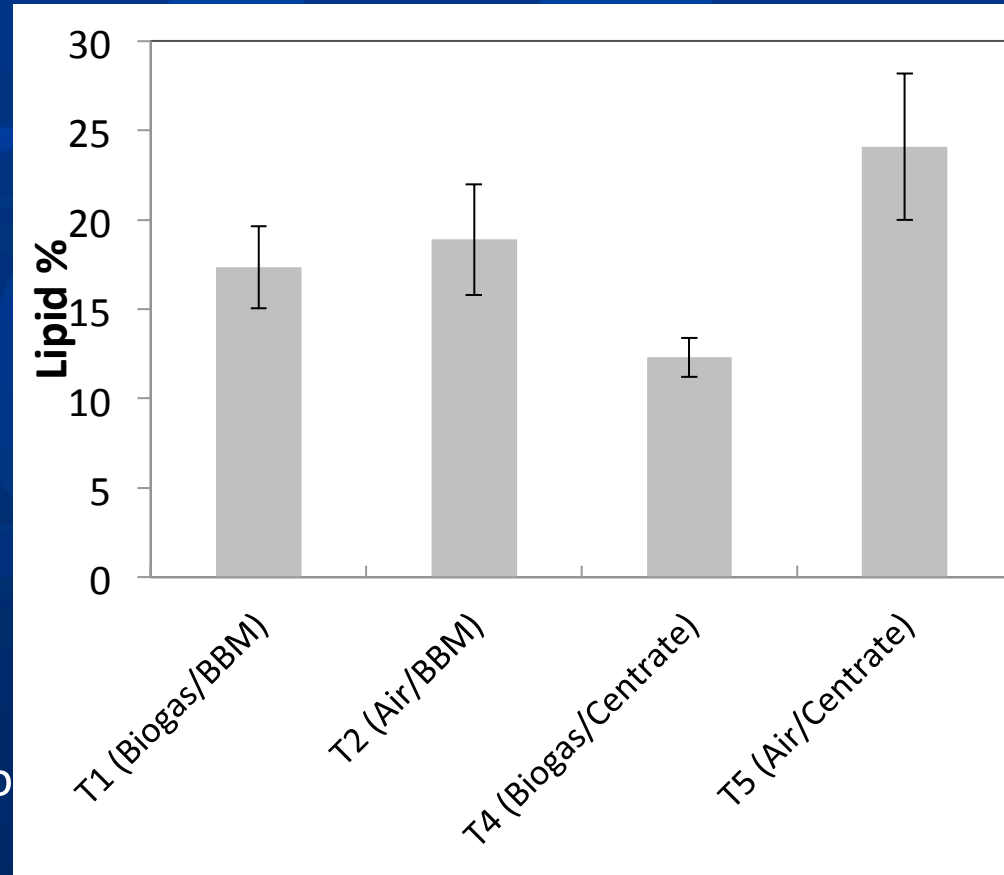
- Microsoft Excel Solver was used to determine the Monod equation coefficients:

➤ $\mu_{\max} = 0.29 \text{ (d}^{-1}\text{)}; K_s = 0.65 \text{ (mg/L)}$



Results (Lipid analysis)

- T1 and T2 had similar lipid contents
 - ✓ T1 (17.3 %), T2 (18.9 %)
- T5 had higher lipid content than T4
 - ✓ T4 (12.3 %), T5 (24.1 %)
- The volatilization of NH_3 and precipitation of PO_4 in T5 caused the “**nutrients deficient condition**”, and significantly triggered the microalgae to accumulate lipid.



- This situation was widely observed by other researchers (Khozin-Goldberg and Cohen 2006, Converti et al. 2009, Rodolfi et al. 2009, Xin et al. 2011)

Conclusion

- Biogas and septic tank sludge centrate can cause microalgae to shift to mixotrophic metabolism, and significantly increase biomass production
- Biogas contains high concentration of CO₂, can buffer the pH of culture solution and prevent ammonia volatilization and phosphate precipitation
- Phosphate is the limiting nutrient in this cultivation system, and its concentration can be used to predict the microalgae growth
- Nutrient deficient condition can cause the microalgae to accumulate more lipid

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Questions?