

## Abstract

Cellulosic ethanol is a potential alternative to petroleum-based fuels and, because it is created from lignocellulose found in woody plant materials, it does not compete directly with food production, unlike corn-based ethanol. One obstacle in cellulosic ethanol production is dealing with the stillage by-product that is typically high in nutrients and chemical oxygen demand. Growing algae for biofuels and high-value pigments requires high nitrogen inputs to sustain growth and produce more biomass, providing a possible bioremediation option for stillage. The objective of this study was to cultivate algae using stillage as a nutrient source. Sugarcane bagasse stillage from the UF-IFAS Stan Mayfield Biorefinery Pilot Plant was characterized. A strain of microfilamentous cyanobacterial algae, *Spirulina* sp., was isolated using standard culture medium, then inoculated into flasks with 2% dilutions of stillage supplemented with *Spirulina* nutrients. The experimental group using the 2% stillage dilutions produced more biomass than the control under the same conditions, as measured by optical density. Lipid analysis using nuclear magnetic resonance showed that the algae biomass had low oil content and was not ideal for biodiesel production. However, the biomass growth under experimental conditions points towards a potential use of stillage as a nutrient source in algae production.

## Introduction

- Stillage is an obstacle to the sustainability of Cellulosic Ethanol fuels with high levels of Chemical Oxygen Demand (COD) and Total Nitrogen (TN).
- Algae can be used as a bioremediation tool and can effectively lower TN and COD.
- A bottleneck in the production of algae is finding a sustainable Nitrogen source.

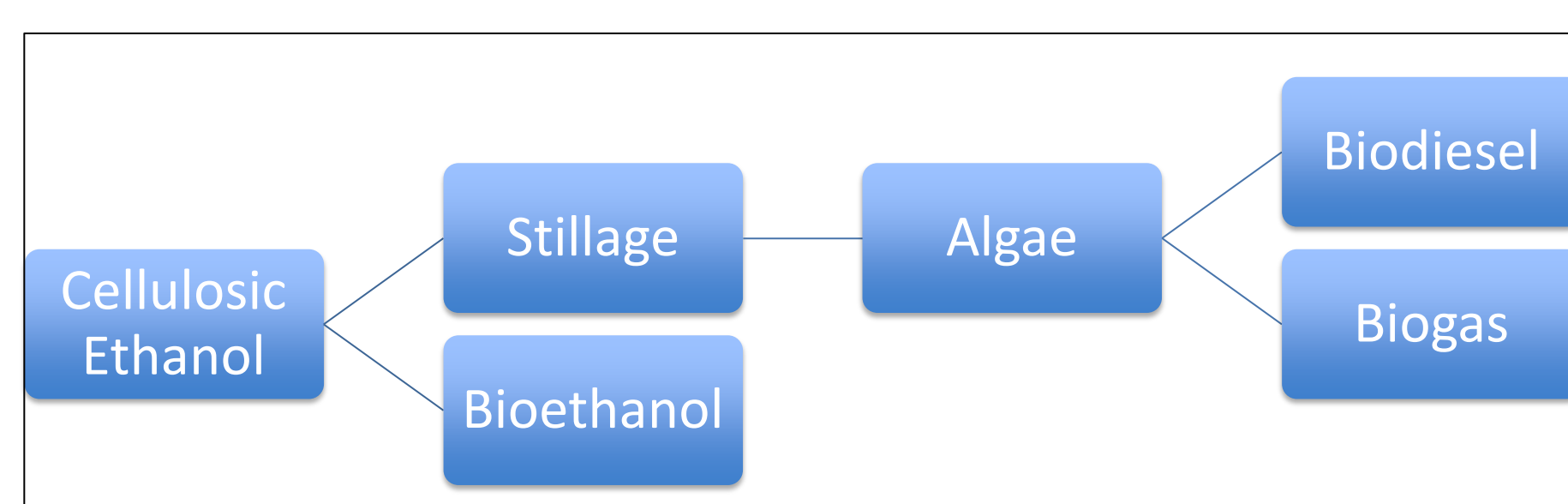


Figure 1: Flow chart showing holistic bioenergy creation that results from the production of cellulosic ethanol and its stillage by-product.

## Hypothesis

A dilution of cellulosic ethanol stillage can be used as a nitrogen source for a culture medium for *Spirulina* sp. when supplemented with bicarbonate and micronutrients.

## Methodology

- Culture of mixed morphology curly and straight *Spirulina* sp. was obtained (figure) and spiral strain was isolated by serial dilutions.
- The algae was cultured using *Spirulina* Medium (Andersen 2005).
- Cellulosic Ethanol Stillage was obtained from the UF-IFAS Stan Mayfield Biorefinery Pilot Plant and characterized.
- pH, Electrical Conductivity, Optical Density (Absorbance), Total and Soluble COD, TN, Total Ammoniacal Nitrogen (TAN) and Total Phosphorus were measured (APHA 2005).
- 250ml Erlenmeyer flasks were filled with *Spirulina* Medium and 2% dilutions of homogenized stillage then inoculated with *Spirulina* from isolated culture.



Figure 2: *Spirulina* algae with curly and straight morphologies. Photograph taken at 40x magnification.

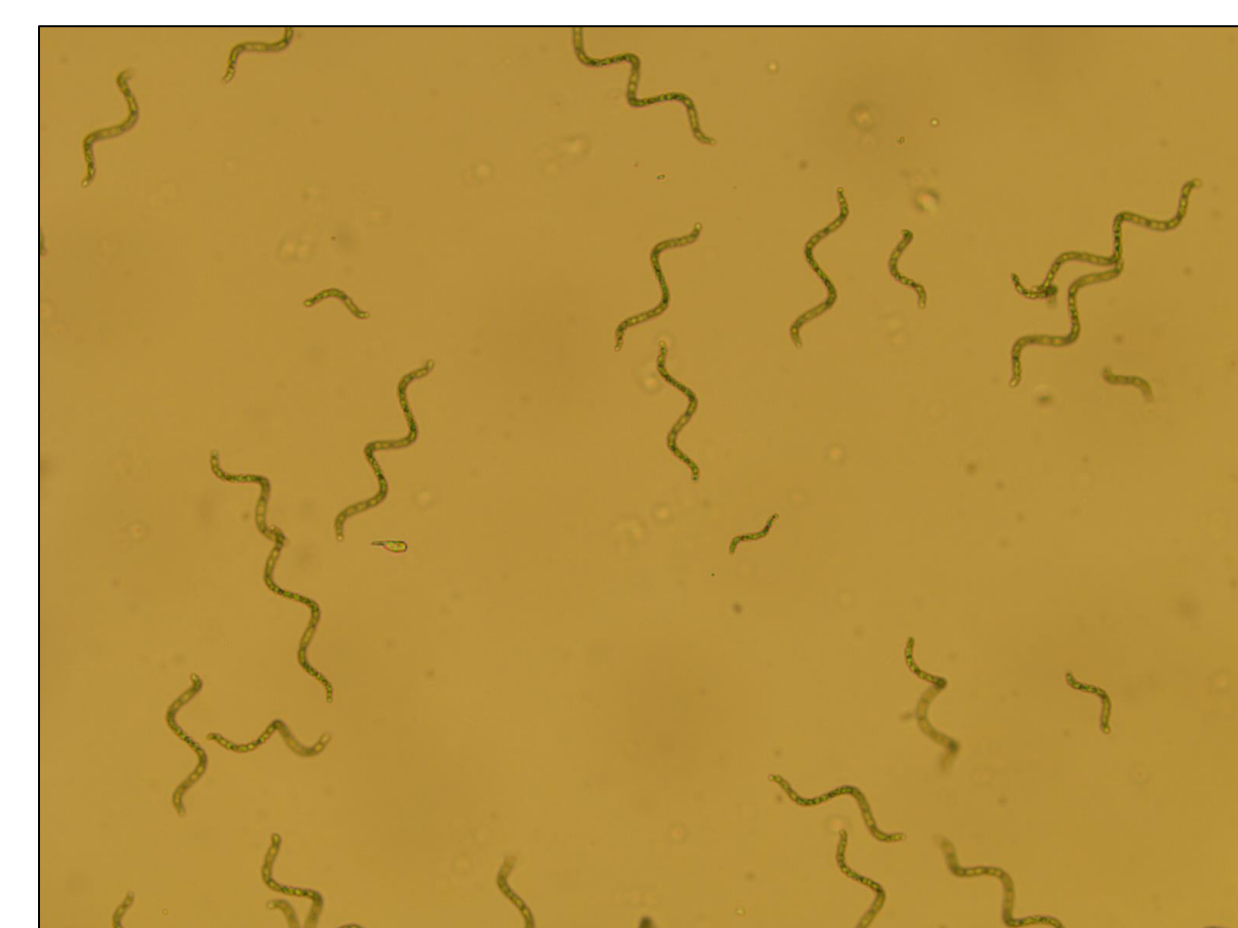


Figure 3: Curly morphology *Spirulina* isolated from the mixed culture by serial dilution.

## Results

	mg/L	mg/L	mg/L	mg/L	mg/L	mS	%	mg/L
	TN ^	TP ^	TCOD*	SCOD*	PH *	TAN*	EC*	TrarSRP
AVG	1167	577.3	104533	82700	6.5	602	9	52
STD	115	5.51	1021.4	1967	0	7	0	0

Figure 4: Table summarizing the characterization of Cellulosic Ethanol Stillage.

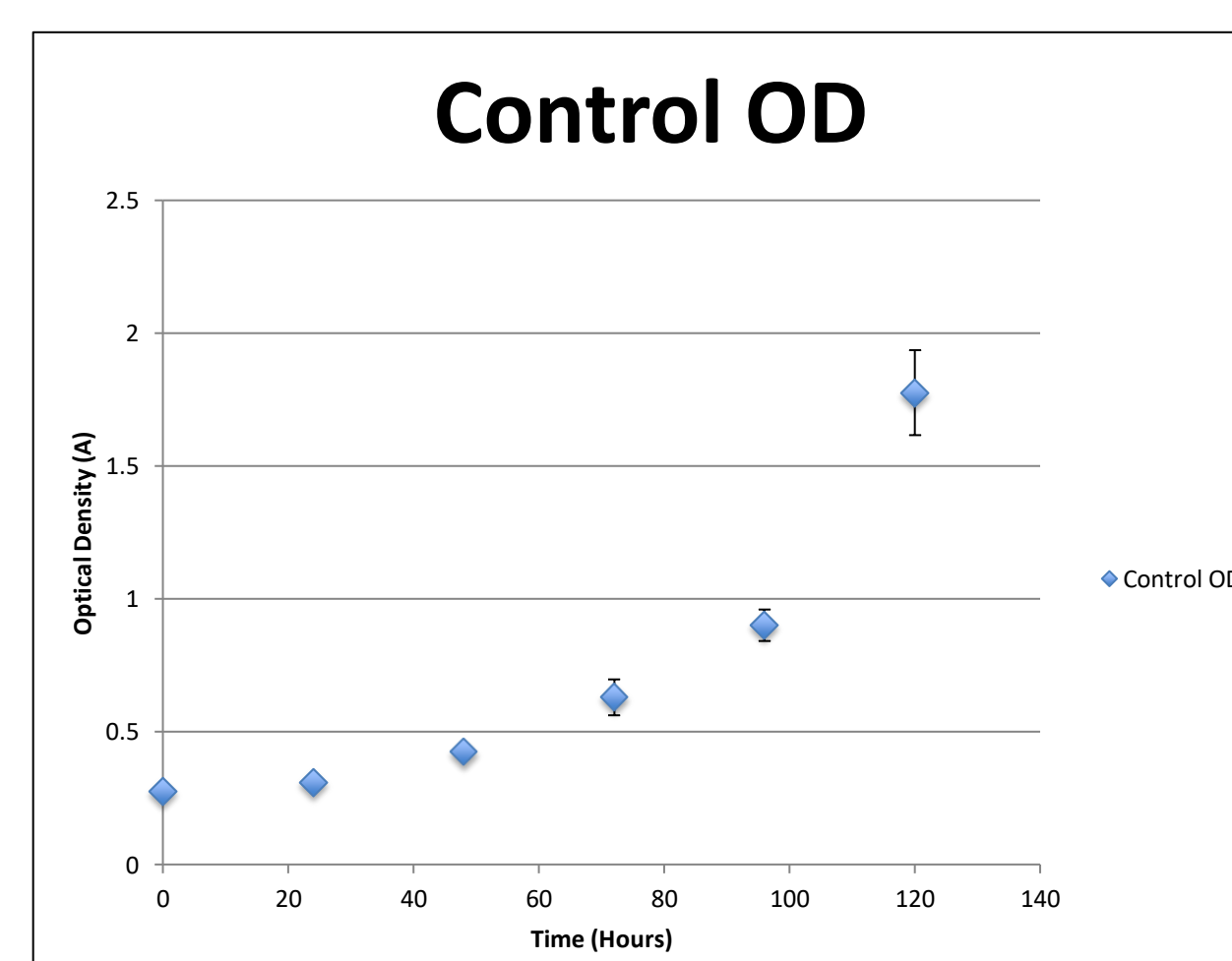


Figure 5: Graph showing increase in biomass in the control as measured by increase of optical density.

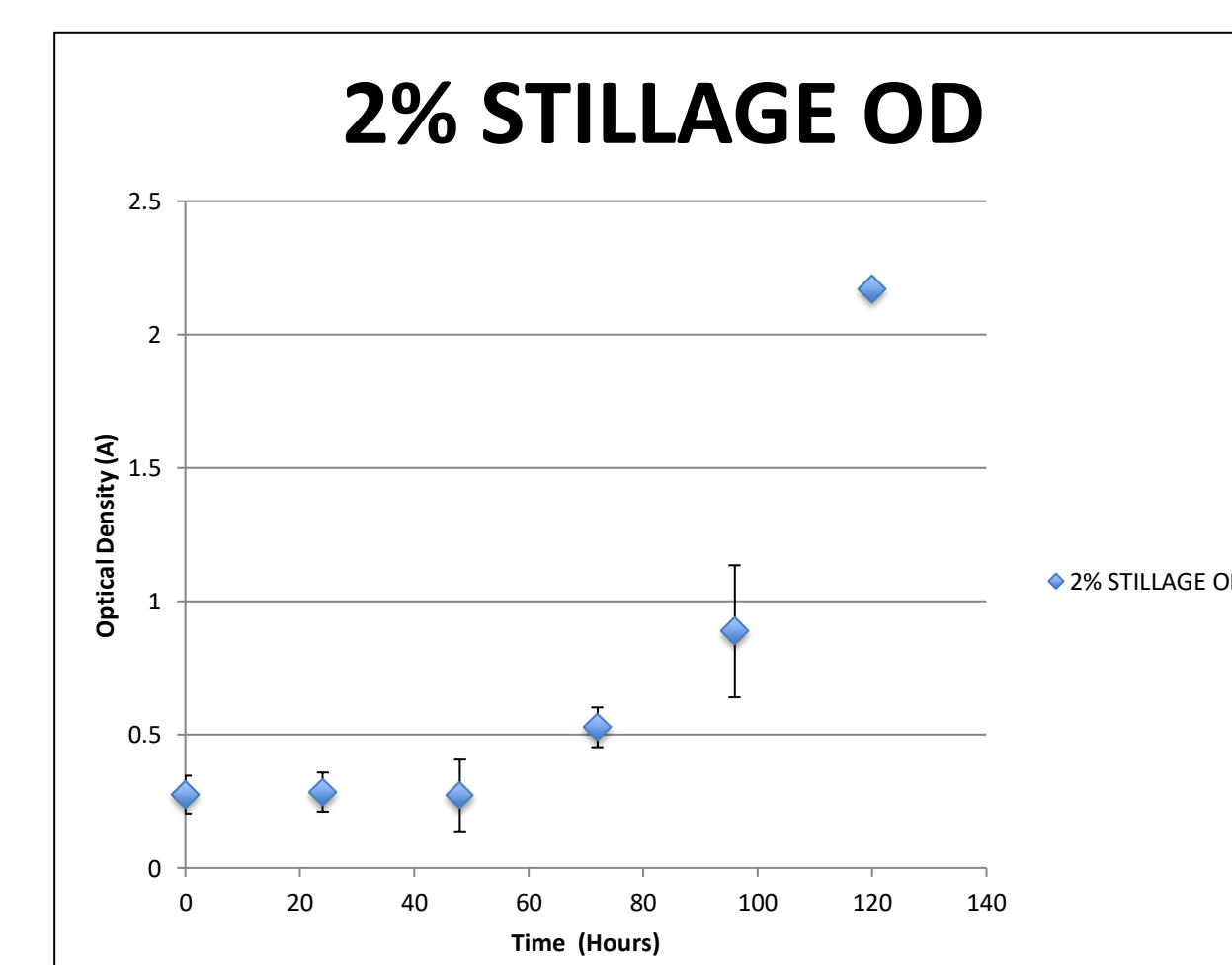


Figure 6: Graph showing increase in biomass in the experimental group using 2% dilutions of stillage as nitrogen source.

## Conclusions

- Preliminary tests show that algae can grow on the stillage and that it may even be a preferred medium.
- Chemical Oxygen Demand is mainly in the soluble phase meaning that the nutrients are available.
- While nuclear magnetic resonance testing did not show substantial lipid content, there may still be ample opportunity to produce biogas from algae via anaerobic digestion.

## Future Research

- Future experiments should concentrate on the effectiveness of bioremediation.
- Trials should be run to test the viability of the stillage as a nutrient source for other strains of algae potentially higher in oil content.
- Experiments should look at the viability of *Spirulina* as a feedstock for biogas through anaerobic digestion.

## Acknowledgements

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## References

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