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Spatial Light Dilution as a Technique for Conversion of Solar Energy to Algal Biomass

Daniel J. Dye
Utah State University

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SPATIAL LIGHT DILUTION AS A TECHNIQUE FOR CONVERSION
OF SOLAR ENERGY TO ALGAL BIOMASS

by

Daniel J. Dye

A dissertation submitted in partial fulfillment
of the requirements for the degree

of

DOCTOR OF PHILOSOPHY

in

Biological Engineering

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UTAH STATE UNIVERSITY
Logan, Utah

2010
ABSTRACT

Spatial Light Dilution as a Technique for Conversion of Solar Energy to Algal Biomass

by

Daniel J. Dye, Doctor of Philosophy
Utah State University, 2010

Major Professors: Dr. Ron Sims and Dr. Byard Wood
Department: Biological Engineering

A photobioreactor has been designed and developed to efficiently utilize solar irradiance through spatial dilution of sunlight. The concept of spatial light dilution is simple: incident sunlight is spread over a large surface area, thus reducing the photon flux density of the light. The implementation of this technique, however, is difficult. The reactor described within uses a new approach to spatial light dilution, utilizing recently-developed optical components to diffuse concentrated sunlight inside an algae culture. Preliminary productivity tests indicate a 2-3 fold increase in productivity per unit aperture (sunlight collection area) over a control reactor with direct-sunlight. Aperture productivity of up to 15 gm m$^{-2}$ day$^{-1}$ and total solar efficiency of 2% were achieved.

A new parameter and yield coefficient are introduced. The parameter total light delivered is defined as the quantity of photons delivered per unit volume per day. The coefficient for yield of biomass on photons is also introduced. For the organism studied in this research, *Neochloris oleoabundans*, the yield of biomass on photons is approximately 1.09 gm mass per mol photons. The total light delivered to a culture over
24 hours, multiplied by the yield coefficient, provides an estimate of the volumetric productivity of the reactor in sequential-batch operation. In a series of laboratory studies, the total light delivered ranged from 0.097 to 0.945 mol photons L\(^{-1}\) day\(^{-1}\), and the volumetric productivity ranged from 0.11 to 0.945 gm L\(^{-1}\) day\(^{-1}\).

A reactor productivity model, integrating reactor geometry and optics with the biomass yield coefficient and volumetric productivity model, predicts that the model organism in the proposed reactor can produce an annual average of 40 gm biomass per square meter of collector area. The model predicts an annual aperture yield of 14.6 kg m\(^{-2}\), at 3% efficiency. This predictive model can be applied to any location that solar data exists, and the techniques can be applied to other types of organisms and reactors to provide productivity estimates.
DEDICATION

I would like to dedicate this work to my son, Braedan James. Braedan was born on the first day of the first semester I joined the Biological Engineering department at USU, and he will be starting pre-school about the same time I’m graduating with my Ph.D. Unbeknownst to him, he has made untold sacrifices caused by my decision to return to school, and I truly hope that my achievements and contributions will make him proud.
ACKNOWLEDGMENTS

This work would not be possible without the support of my wife, my son, and my family. I truly am indebted to all my family members for providing me with the support and patience I have needed to make the commitment to this work.

I am grateful for the years of support and guidance provided by Dr. Byard Wood. It has been an honor to study under Dr. Wood for both my M.S.M.E. and my Ph.D. I have been very fortunate to have the unique opportunity to study under Dr. Ron Sims as well. Dr. Sims’ excitement and love for his work is contagious. I feel very grateful for having had the opportunity to study under these two professionals.

I would like to thank my committee for their service and support. Dr. Lance Seefeldt, Dr. Conly Hansen, and Dr. Charlie Miller are all well respected professors, researchers, and individuals and I feel very fortunate to have such a rich committee. Mr. Jeff Muhs, the former director of the USU energy lab, has been instrumental in the development of the prototype reactor described within. Jeff conceived the idea of using planar waveguides directly lit by linear concentrators, which helped increase the efficiency of spatial light dilution. Early discussions with him about the design concept and his ability to garner funding for the project were greatly appreciated.

There have been numerous people who have assisted me in my research, and to them I am extremely grateful. I’ve been lucky to have several great undergraduate assistants, including Jared Pike, Candace Clark, Mikey Morgan, Nathan Philips, and Damien Bellos. The Biofuels Center has fantastic technical support provided by Mike Morgan, and his work ethic and expertise in machining has proven invaluable. Several students, faculty, and staff have had an impact on my education and research at USU. Pete Zemke, Shaun Dustin, Brett Barney, Sridhar Viamajala, Brad Wahlen, Stephen Merrigan, and too many others to list have all been helpful throughout this research.

This work was funded in part by the Utah Science, Technology, and Research Initiative. Development of the prototype photobioreactor was supported by DARPA contract numbers 4000067218 and 4000067505. The Space Dynamics Lab provided
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Dan Dye
## CONTENTS

<table>
<thead>
<tr>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>ABSTRACT</td>
</tr>
<tr>
<td>DEDICATION</td>
</tr>
<tr>
<td>ACKNOWLEDGMENTS</td>
</tr>
<tr>
<td>LIST OF TABLES</td>
</tr>
<tr>
<td>LIST OF FIGURES</td>
</tr>
<tr>
<td>NOMENCLATURE</td>
</tr>
</tbody>
</table>

### CHAPTER

#### 1 INTRODUCTION

1.1. Introduction ........................................... 1
1.2. Light Dilution ........................................... 4
1.3. Objectives of this Study ................................... 7

#### 2 LITERATURE REVIEW

2.1. Introduction ........................................... 8
2.2. Temporal Dilution ........................................... 8
2.3. Spatial Dilution ........................................... 11
2.4. Conclusions ............................................ 14

#### 3 SPATIALLY DILUTED PHOTOBIOREACTOR DESIGN AND CONCEPT

3.1. Introduction ........................................... 16
3.2. Spatial Light Dilution Relationships, Efficiency, and Geometry ........................................... 17
3.3. Optical Component Selection ................................... 21
3.4. Prototype sdPBR ........................................... 35
3.5. Conclusions ............................................ 37

#### 4 PROTOTYPE sdPBR PRODUCTIVITY TESTS

4.1. Introduction ........................................... 39
4.2. Methods ................................................. 39
4.3. Experimental Results .................................... 47
4.4. Conclusions ............................................ 54
<table>
<thead>
<tr>
<th>Chapter</th>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>VOLUMETRIC PRODUCTIVITY WITH sdPBR-SIMULATING REACTORS</td>
<td>56</td>
</tr>
<tr>
<td></td>
<td>5.1. Introduction</td>
<td>56</td>
</tr>
<tr>
<td></td>
<td>5.2. Parameters Investigated</td>
<td>56</td>
</tr>
<tr>
<td></td>
<td>5.3. Methods</td>
<td>59</td>
</tr>
<tr>
<td></td>
<td>5.4. Sample Experimental Data</td>
<td>64</td>
</tr>
<tr>
<td></td>
<td>5.5. Results of Study #1: Effects of Light Intensity, Reactor Width, and Dilution Fraction on Volumetric Productivity</td>
<td>68</td>
</tr>
<tr>
<td></td>
<td>5.6. Results of Study #2: Effects of Light Intensity, Diurnal Cycle, and Dilution Fraction on Volumetric Productivity</td>
<td>73</td>
</tr>
<tr>
<td></td>
<td>5.7. Combined Factors and Response</td>
<td>78</td>
</tr>
<tr>
<td></td>
<td>5.8. Conclusions</td>
<td>80</td>
</tr>
<tr>
<td>6</td>
<td>PRODUCTIVITY MODEL FOR THE sdPBR</td>
<td>82</td>
</tr>
<tr>
<td></td>
<td>6.1. Introduction</td>
<td>82</td>
</tr>
<tr>
<td></td>
<td>6.2. Solar and Environmental Data for System Productivity Model</td>
<td>83</td>
</tr>
<tr>
<td></td>
<td>6.3. Maximum Performance Predicted with the sdPBR and <em>Neochloris oleoabundans</em></td>
<td>87</td>
</tr>
<tr>
<td></td>
<td>6.4. Conclusions</td>
<td>90</td>
</tr>
<tr>
<td>7</td>
<td>SUMMARY</td>
<td>92</td>
</tr>
<tr>
<td>REFERENCES</td>
<td></td>
<td>94</td>
</tr>
<tr>
<td>APPENDICES</td>
<td></td>
<td>101</td>
</tr>
<tr>
<td>A</td>
<td>METHODS AND PROTOCOLS</td>
<td>102</td>
</tr>
<tr>
<td></td>
<td>A.1. Culture Density</td>
<td>102</td>
</tr>
<tr>
<td></td>
<td>A.2. Algal Energy Content</td>
<td>104</td>
</tr>
<tr>
<td></td>
<td>A.4. Sequential-Batch Operation</td>
<td>106</td>
</tr>
<tr>
<td>B</td>
<td>UNCERTAINTY ANALYSIS</td>
<td>107</td>
</tr>
<tr>
<td></td>
<td>B.1. General Uncertainty Analysis</td>
<td>107</td>
</tr>
<tr>
<td></td>
<td>B.2. Confidence Interval on Pvol</td>
<td>107</td>
</tr>
<tr>
<td>C</td>
<td>PARAMETRIC STUDY DATA</td>
<td>109</td>
</tr>
<tr>
<td>CURRICULUM VITAE</td>
<td></td>
<td>138</td>
</tr>
</tbody>
</table>
## LIST OF TABLES

<table>
<thead>
<tr>
<th>Table</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.1. Biomass Productivity Realized in Temporally Diluted Photobioreactors</td>
<td>11</td>
</tr>
<tr>
<td>2.2. Biomass Productivity Realized in Spatially Diluted Photobioreactors</td>
<td>14</td>
</tr>
<tr>
<td>4.1. Performance of <em>N. oleoabundans</em> on Sodium Nitrate [46]</td>
<td>42</td>
</tr>
<tr>
<td>4.2. SE+ Media Recipe</td>
<td>43</td>
</tr>
<tr>
<td>4.3. Sequential-Batch Test Results for <em>N. oleoabundans</em></td>
<td>53</td>
</tr>
<tr>
<td>5.1. Study 1, Salient Data for Regression Analysis</td>
<td>72</td>
</tr>
<tr>
<td>5.2. Study 2, Salient Data for Regression Analysis</td>
<td>77</td>
</tr>
<tr>
<td>6.1. Parameters Used in the Productivity Model</td>
<td>82</td>
</tr>
<tr>
<td>B.1. Estimates of Bias and Precision Error for Various Instruments</td>
<td>108</td>
</tr>
<tr>
<td>C.1. Parametric Study Experimental Conditions</td>
<td>109</td>
</tr>
</tbody>
</table>
# LIST OF FIGURES

<table>
<thead>
<tr>
<th>Figure</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.1. Representative response of the net rate of photosynthesis (as oxygen evolution) to light intensity. Commonly referred to as the Photosynthesis/Irradiance (P/I) response [20, 23, 24]</td>
<td>4</td>
</tr>
<tr>
<td>1.2. Instantaneous and time-averaged intensity experienced by an algae cell in a PBR designed for temporal light-dilution</td>
<td>5</td>
</tr>
<tr>
<td>1.3. Instantaneous and time-averaged intensity experienced by an algae cell in a PBR designed for spatial light-dilution</td>
<td>6</td>
</tr>
<tr>
<td>2.1. Vertical flat-plate Photobioreactor, with turbulent mixing to induce high-frequency light/dark cycle</td>
<td>10</td>
</tr>
<tr>
<td>2.2. Simple reactor geometry for demonstrating concept of spatial dilution: both reactors could have the same volume and light collection area perpendicular to the incident light, but the cylindrical reactor has a larger illuminated surface area, thus the PPFD is diluted over the surface</td>
<td>13</td>
</tr>
<tr>
<td>3.1. Spatially-diluted photobioreactor developed at the USU energy laboratory, which utilizes linear concentrators directly coupled to planar waveguides [32]</td>
<td>17</td>
</tr>
<tr>
<td>3.2. Line drawing of the proposed spatially-diluted photobioreactor geometry. ( I_{DN} ) and ( I_i ) are the direct normal and internal PPFD, respectively. ( w ) is the collector or chamber width, ( V ) is the chamber volume, and ( h ) is the chamber height</td>
<td>19</td>
</tr>
<tr>
<td>3.3. Relationship between reactor volume and width for fixed optical efficiency and light dilution ratio</td>
<td>20</td>
</tr>
<tr>
<td>3.4. Fresnel lens back focal length curves for grooves facing towards the sun (out) or towards the image (in)</td>
<td>23</td>
</tr>
<tr>
<td>3.5. Device for measuring focal-line width of Fresnel lens</td>
<td>23</td>
</tr>
<tr>
<td>3.6. Predicted and measured focal zone profiles for the Edmund Optics NT46-616 Fresnel lens</td>
<td>24</td>
</tr>
</tbody>
</table>
3.7. Parabolic primary and hyperbolic secondary solar concentrator. F1 is the primary focal point, F2 is the near focal point of the hyperbolic reflector, and F3 is the far focal point of the hyperbolic reflector ................................................................. 25

3.8. Parabolic primary and elliptical solar concentrator. F1 is the primary focal point, F2 is the near focal point of the elliptical reflector, and F3 is the far focal point of the elliptical reflector .................................................................................. 25

3.9. Profiles of concentrated light at the focal plane for the Fresnel lens and parabolic/hyperbolic systems ......................................................................................................................... 26

3.10. The effect of tracking error on collection efficiency for three different concentrators ........................................................................................................................................ 27

3.11. Line-drawing of the prototype planar waveguide ........................................... 28

3.12. Waveguide test device on-sun ...................................................................... 30

3.13. Side-view of test device ................................................................................ 30

3.14. Comparison of performance between the five waveguide prototypes tested at USU ........................................................................................................................................ 31

3.15. PAR flux emitted from the side of the prototype planar waveguide .......... 32

3.16. Laser light exiting the waveguide ............................................................... 33

3.17. Sunlight exiting the waveguide ..................................................................... 33

3.18. Prototype sdPBR ........................................................................................... 36

3.19. Side-view of reactor chamber, showing location of ports for supporting systems .................................................................................................................................... 37

3.20. sdPBR mounted on two-axis solar tracker .................................................. 38

3.21. sdPBR tracking the sun ................................................................................ 38

4.1. Control reactor. Dimensions: 10 in. wide, 10.5 in. tall, and 13.75 in. long ....... 40

4.2. Side view of control reactor ........................................................................... 40

4.3. Example of the density of a culture either operated in 1) batch or 2) sequential-batch mode ........................................................................................................................................ 45
4.4. Batch growth curves, which shows that the culture density in the sdPBR is more affected by cloudy weather than in the control reactor ................................. 48
4.5. Accumulative energy from direct normal irradiance as measured by the NIP, and total hemispherical irradiance on a horizontal plane as measured by the PSP ... 48
4.6. Culture density in the sdPBR and control reactors over the course of a 16-day sequential-batch test .................................................................................................. 50
4.7. Culture density before and an estimate of density after each harvest during the sequential-batch test. Confidence interval bars withheld for clarity, but are similar to the previous figure ............................................................................................. 50
4.8. Daily accumulative energy input during the sequential-batch test as measured by the NIP and PSP ................................................................................................. 51
4.9. Nutrient concentration relative to the media concentration over the course of the test. *N. oleoabundans* in SE+ media .............................................................................. 51
4.10. Aperture productivity from the sdPBR and control reactors over 10 days of sequential-batch run. *N. oleoabundans*, SE+ media, pH = 7.2, temp. = 29 ± 1 °C ........................................................................................................................................ 52
4.11. Energy conversion efficiency of *N. oleoabundans* in the sdPBR operated in sequential-batch mode. Data corresponds to efficiency over the full solar spectrum (left vertical axis) and efficiency over the PAR portion of the solar spectrum (right vertical axis) .............................................................................................. 54
5.1. Averaged PAR Transmission vs. Depth and Culture Density for *N. oleoabundans* ........................................................................................................................................... 58
5.2. Experimental laboratory reactor lighting sketch................................................. 60
5.3. Light profile across reactor cross-section.............................................................. 61
5.4. Normalized SPD of the ASTM Air Mass 1.5 Direct Normal Irradiance (Sunlight), a SoLux lamp, and a Philips Plant & Aquarium Fluorescent Lamp .................... 62
5.5. Distribution of PPFD on reactor lit surface............................................................. 63
5.6. PPFD vs. light spacing for bench-top reactors ..................................................... 63
5.7. Culture density response to harvesting started when the density is below the steady-state operating density ................................................................. 65
5.8. Culture density response to harvesting started when the density is above the steady-state operating density ............................................................. 65
5.9. Culture density response to harvesting started when the density is at the steady-state operating density ................................................................. 65
5.10. Triplicate test of w = 50mm, \( I_i = 175 \mu\text{mol m}^{-2}\text{ sec}^{-1} \), DC = 12/12, and \( \Delta = 0.7 \). DW at end of day period is shown. Diluted DW after each harvest withheld for clarity .......................................................................................................................... 67
5.11. Representative change in DW (days 7 & 8) and actual DW over 24 hour period (day 9) ................................................................................................................. 68
5.12. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 75 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 69
5.13. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 125 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 69
5.14. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 175 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 70
5.15. Maximum volumetric productivity of *N. oleoabundans* vs. light intensity ...... 70
5.16. Maximum volumetric productivity of *N. oleoabundans* obtained at each reactor width. Lines represent light intensities. ................................................................. 71
5.17. Maximum volumetric productivity obtained with N. oleoabundans vs. total light delivered ......................................................................................... 73
5.18. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 75 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 74
5.19. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 125 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 74
5.20. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_i = 175 \mu\text{mol m}^{-2}\text{ sec}^{-1} \) .......................................................................................................................... 75
5.21. Maximum volumetric productivity of *N. oleoabundans* obtained at each light intensity. Lines represent diurnal cycles. .......................................................... 75
5.22. Maximum volumetric productivity of *N. oleoabundans* obtained at each diurnal cycle. Lines represent light intensities. .......................................................... 76
5.23. Maximum volumetric productivity obtained with *N. oleoabundans* vs. total light delivered ......................................................................................................... 77
5.24. Measured volumetric productivity vs. total light delivered for all sets of experimental conditions ......................................................................................................... 79
5.25. Second-order model for biomass productivity of *N. oleoabundans* on TLD ......................................................................................................................... 79
5.26. Correlation between predicted and measured volumetric productivity of *N. oleoabundans* ........................................................................................................ 80
6.1. Measured direct normal irradiance and corresponding direct normal PPFD ........ 84
6.2. Direct normal and total hemispherical PPFD on a horizontal plane over a day .. 85
6.3. Average annual DNI for a selection of U.S. cities ........................................... 85
6.4. Monthly averages of daily energy flux for different collector types in Las Vegas, NV (adapted from [55]) ................................................................................................. 86
6.5. Representative TMY3 data for Las Vegas, Nevada on 7/11/91 ...................... 87
6.6. Predicted aperture productivity for each day of the year; LDR=10, w=50mm, \( \mu_{opt} = 85\% \), Las Vegas, NV ................................................................. 88
6.7. Monthly averages for biomass productivity per unit aperture area; LDR=10, w=50mm, \( \mu_{opt} = 85\% \), Las Vegas, NV ................................................................. 89
6.8. Maximum and average aperture biomass productivity vs. reactor width; DR=10, \( \mu_{opt} = 85\% \), Las Vegas, NV ................................................................. 89
6.9. Maximum and average aperture biomass productivity vs. reactor width; DR=10, \( \mu_{opt} = 85\% \), Las Vegas, NV ................................................................. 90
A.1. Strength of the correlation between DW and OD between the wavelengths 300 – 900 nm ................................................................. 103
A.2. Comparison between DW and OD$_{750}$ for *N. oleoabundans* from several different reactors and growth curves ............................................................. 103

A.3. Schematic of dilutions and cuvettes used to measure spectral light attenuation vs. path-length ........................................................................................................... 105

A.4. Spectral transmission through *N. oleoabundans* over a 1 mm path-length for different concentrations of a 1.76 gm/L culture .......................................................... 106

C.1. 20 mm reactor, condition #1 ............................................................................ 111

C.2. 20 mm reactor, condition #2 ............................................................................ 112

C.3. 20 mm reactor, condition #3 ............................................................................ 112

C.4. 20 mm reactor, condition #4 ............................................................................ 113

C.5. 20 mm reactor, condition #5 ............................................................................ 113

C.6. 20 mm reactor, condition #6 ............................................................................ 114

C.7. 20 mm reactor, condition #7 ............................................................................ 114

C.8. 20 mm reactor, condition #8 ............................................................................ 115

C.9. 20 mm reactor, condition #9 ............................................................................ 115

C.10. 20 mm reactor, condition #10 ...................................................................... 116

C.11. 35 mm reactor, condition #11 ...................................................................... 116

C.12. 35 mm reactor, condition #12 ...................................................................... 117

C.13. 35 mm reactor, condition #13 ...................................................................... 117

C.14. 35 mm reactor, condition #14 ...................................................................... 118

C.15. 35 mm reactor, condition #15 ...................................................................... 118

C.16. 35 mm reactor, condition #16 ...................................................................... 119

C.17. 35 mm reactor, condition #17 ...................................................................... 119

C.18. 35 mm reactor, condition #18 ...................................................................... 120

C.19. 35 mm reactor, condition #19 ...................................................................... 120

C.20. 35 mm reactor, condition #20 ...................................................................... 121

C.21. 50 mm reactor, condition #21 ...................................................................... 121

C.22. 50 mm reactor, condition #22 ...................................................................... 122
C.23. 50 mm reactor, condition #23................................................................. 122
C.24. 50 mm reactor, condition #24................................................................. 123
C.25. 50 mm reactor, condition #25................................................................. 123
C.26. 50 mm reactor, condition #26................................................................. 124
C.27. 50 mm reactor, condition #27................................................................. 124
C.28. 50 mm reactor, condition #28................................................................. 125
C.29. 50 mm reactor, condition #29................................................................. 125
C.30. 50 mm reactor, condition #30................................................................. 126
C.31. 50 mm reactor, condition #31................................................................. 126
C.32. 50 mm reactor, condition #32................................................................. 127
C.33. 50 mm reactor, condition #33................................................................. 127
C.34. 50 mm reactor, condition #34................................................................. 128
C.35. 50 mm reactor, condition #35................................................................. 128
C.36. 50 mm reactor, condition #36................................................................. 129
C.37. 50 mm reactor, condition #37................................................................. 129
C.38. 50 mm reactor, condition #38 (replicate #1).......................................... 130
C.39. 50 mm reactor, condition #38 (replicate #2).......................................... 130
C.40. 50 mm reactor, condition #38 (replicate #3).......................................... 131
C.41. 50 mm reactor, condition #39................................................................. 131
C.42. 50 mm reactor, condition #40................................................................. 132
C.43. 50 mm reactor, condition #41................................................................. 132
C.44. 50 mm reactor, condition #42................................................................. 133
C.45. 50 mm reactor, condition #43................................................................. 133
C.46. 50 mm reactor, condition #44................................................................. 134
C.47. 50 mm reactor, condition #45................................................................. 134
C.48. 50 mm reactor, condition #46................................................................. 135
C.49. 50 mm reactor, condition #47................................................................. 135
C.50. 50 mm reactor, condition #48................................................................. 136
C.51. 50 mm reactor, condition #49 ................................................................. 136
C.52. 50 mm reactor, condition #50 ................................................................. 137
### NOMENCLATURE

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Δ</td>
<td>Dilution fraction [L/L]</td>
</tr>
<tr>
<td>δ</td>
<td>Declination [degrees]</td>
</tr>
<tr>
<td>φ</td>
<td>Latitude [degrees]</td>
</tr>
<tr>
<td>λ</td>
<td>Wavelength [nm]</td>
</tr>
<tr>
<td>η&lt;sub&gt;FS&lt;/sub&gt;</td>
<td>Full-spectrum-solar-to-biomass energy conversion efficiency</td>
</tr>
<tr>
<td>η&lt;sub&gt;PS&lt;/sub&gt;</td>
<td>Photosynthetic, or PAR-spectrum-solar-to-biomass energy conversion efficiency</td>
</tr>
<tr>
<td>η&lt;sub&gt;opt&lt;/sub&gt;</td>
<td>Optical efficiency</td>
</tr>
<tr>
<td>η&lt;sub&gt;FS&lt;/sub&gt;</td>
<td>Full-solar-spectrum-to-biomass energy conversion efficiency</td>
</tr>
<tr>
<td>μ</td>
<td>Specific growth rate [day&lt;sup&gt;−1&lt;/sup&gt;]</td>
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<td>A&lt;sub&gt;apt&lt;/sub&gt;</td>
<td>Solar collector lit area; aperture [m&lt;sup&gt;2&lt;/sup&gt;]</td>
</tr>
<tr>
<td>A&lt;sub&gt;waveguide&lt;/sub&gt;</td>
<td>Light distribution area [m&lt;sup&gt;2&lt;/sup&gt;]</td>
</tr>
<tr>
<td>ASTM</td>
<td>American Society of Testing and Materials</td>
</tr>
<tr>
<td>CO₂</td>
<td>Carbon dioxide</td>
</tr>
<tr>
<td>d</td>
<td>Depth of reactor, along the linear optical axis [m]</td>
</tr>
<tr>
<td>DC</td>
<td>24 hour diurnal cycle [hours of daylight / hours of darkness]</td>
</tr>
<tr>
<td>DNI</td>
<td>Direct normal irradiance [W m&lt;sup&gt;−2&lt;/sup&gt;]</td>
</tr>
<tr>
<td>DW</td>
<td>Dry weight [gm L&lt;sup&gt;−1&lt;/sup&gt;]. gm of algae always refers to dry weight in this text</td>
</tr>
<tr>
<td>F/#</td>
<td>Ratio of focal length to lens width</td>
</tr>
<tr>
<td>FAME</td>
<td>Fatty acid methyl ester, aka biodiesel</td>
</tr>
<tr>
<td>G&lt;sub&gt;biomass&lt;/sub&gt;</td>
<td>Energy stored in biomass [KJ m&lt;sup&gt;−2&lt;/sup&gt; day&lt;sup&gt;−1&lt;/sup&gt;]</td>
</tr>
<tr>
<td>Symbol</td>
<td>Definition</td>
</tr>
<tr>
<td>--------</td>
<td>------------</td>
</tr>
<tr>
<td>GDN</td>
<td>Total direct normal energy delivered over time [KJ m⁻² day⁻¹], or [KW-hr m⁻² day⁻¹]</td>
</tr>
<tr>
<td>GTH</td>
<td>Total hemispherical energy delivered over time [KJ m⁻² day⁻¹], or [KW-hr m⁻² day⁻¹]</td>
</tr>
<tr>
<td>GSC</td>
<td>“Green Solar Collector”</td>
</tr>
<tr>
<td>h</td>
<td>Height of culture chamber inside reactor [mm]</td>
</tr>
<tr>
<td>H</td>
<td>Energy content of algae biomass [KJ gm⁻¹]</td>
</tr>
<tr>
<td>HRT</td>
<td>Hydraulic retention time, the effective time a unit volume of water is inside the reactor</td>
</tr>
<tr>
<td>IDN</td>
<td>Direct normal PPFD [µmol m⁻² sec⁻¹]</td>
</tr>
<tr>
<td>Iᵢ</td>
<td>PPFD emitted by the waveguides inside the PBR [µmol m⁻² sec⁻¹]</td>
</tr>
<tr>
<td>Iᵢ,avg</td>
<td>Average Iᵢ over a 24-hour period [µmol m⁻² sec⁻¹]</td>
</tr>
<tr>
<td>Isat</td>
<td>PPFD at saturation [µmol m⁻² sec⁻¹]</td>
</tr>
<tr>
<td>Isolar</td>
<td>PPFD of sunlight [µmol m⁻² sec⁻¹]</td>
</tr>
<tr>
<td>LDR</td>
<td>Light dilution ratio</td>
</tr>
<tr>
<td>Lx</td>
<td>Luminous flux [lumens/m²]</td>
</tr>
<tr>
<td>NIP</td>
<td>Normal incidence pyrheliometer</td>
</tr>
<tr>
<td>NREL</td>
<td>National Renewable Energy Lab</td>
</tr>
<tr>
<td>OAR</td>
<td>Optical area ratio</td>
</tr>
<tr>
<td>ODₗ</td>
<td>Optical density measured at λ</td>
</tr>
<tr>
<td>Papt</td>
<td>Aperture productivity (gm mass per aperture area) [gm m⁻² day⁻¹]</td>
</tr>
<tr>
<td>Pareal</td>
<td>Areal productivity [gm m⁻² day⁻¹]</td>
</tr>
<tr>
<td>Pvol</td>
<td>Volumetric productivity [gm L⁻¹ day⁻¹]</td>
</tr>
</tbody>
</table>
PAR Photosynthetic active radiation (400 nm – 700 nm)

PBR Photobioreactor

PPFD Photosynthetic photon flux density [µmol photons m⁻² sec⁻¹], typically [µmol m⁻² sec⁻¹]

PSP Precision spectral pyrheliometer

R² Goodness-of-fit; variance explained by a model

sdPBR Spatially-diluted photobioreactor

SE Soil extract, microalgal growth media

SPD Spectral power distribution

t time [day]

TAG Triacylglycerol, an ester of glycerol bound to three fatty acids, which are convertible to FAME

tdPBR Temporally-diluted photobioreactor

TLD Total light delivered [mol photons/L]

TMY Typical meteorological year

USU Utah State University

UTEX University of Austin, Texas algal culture collection

Vₜ Volume harvested [L]

Vₐ Reactor volume [L]

vvm Gas flow ratio: volume gas per volume reactor per minute [L gas / L reactor / min]

w Width of culture chamber inside reactor, center-center of the waveguides [mm]

X Algal culture density [gm L⁻¹]
$Y_{X/P}$  

Yield of biomass on photons [gm mass/mol photons]
1.1. Introduction

Photosynthetic organisms such as single-celled algae and cyanobacteria, colloquially referred to as microalgae, are the source of a significant portion of the world’s annual biomass produced. However, despite the massive quantity of microalgae growing each year across the planet, only a small fraction are grown for commercial use. The biomass produced by microalgae can be used as a feedstock for pharmaceuticals or nutraceuticals, feed for aquaculture, or feedstock for an energy conversion process where the biomass is converted to gas or liquid fuels or burned directly [1-4]. Microalgae are able to absorb CO₂ [5, 6] as well as contaminants from wastewater [7, 8], which means they can be used for services in addition to the products they generate. In addition, some microalgae are used for human consumption, such as *Aphanizomenon flos-aqua*, a cyanobacterium that grows in natural lakes such as Upper Klamath Lake (Oregon, USA). In 1998, 10⁶ kg of it was harvested, with a commercial market value of $100 million [9]. Other than in a few cases, it is probable that in order for mass culturing of phototrophic microalgae to be economical, a variety of products and services will need to be exploited.

Despite the great potential, commercial development of microalgae as a biomass feedstock has not met expectations [10], due to both economic and technical hurdles. However, several factors have led to the resurgence of focus on microalgae by academic and private research organizations as well as large venture capital investments in technologies associated with producing and processing algal biomass [11]. It is expected that the proper techniques will be developed to efficiently convert solar energy into algal biomass, from which a variety of products can be developed, thus making the process economically viable.

In order to make large-scale production of microalgae biomass feasible, culturing systems must achieve a balance between productivity and cost. The culturing system
can be viewed as simply a means to convert solar energy into biomass. In order to achieve high areal yields, the culturing system must effectively utilize the ground surface area and efficiently utilize solar irradiance. To date, the majority of commercial culturing of algal biomass is done in raceway ponds [12]. However, while extremophiles such as *Spirulina platensis* may be able to thrive in open ponds, most species will be out-competed by local species, so the number of species that can be grown as a pure culture in ponds is limited. Unfortunately, while ponds may be relatively inexpensive, their productivity is too low to make them cost effective except for a limited number of high-value products, or where an extremophile is used [12].

Enclosed culturing systems, commonly referred to as photobioreactors (PBR), have been the focus of research for decades but still have not managed to enter the commercial mass-production market. Besides some small share of the market such as in preparing feedstock for aquaculture, PBRs have not reached high yields at a low enough cost to make them economical for production of biofuels or commodities.

There are a variety of PBRs that have been designed to grow microalgae, and some do so effectively. However, very few PBR’s have been designed to efficiently distribute sunlight to the culture at an intensity below the saturation intensity of the microorganism, which would potentially increase its photosynthetic efficiency [13]. Delivering photosynthetic active radiation (PAR) to the culture at an intensity below the saturation intensity of the culture, which is typically 10% that of full-intensity sunlight [14], allows the organism to utilize more of the photons and therefore operate at a higher efficiency. One method for reducing the photosynthetic photon flux density (PPFD) below 10% of full sunlight is by collecting and distributing the photons over a larger surface area. This lighting design concept, called spatial-dilution of light, has been put to practice in a variety of reactors using fiber optic cables to deliver light to the culture [15-19]. The problem with using multiple optical components and fiber optics is that optical losses add up quickly and rapidly degrade the solar-to-biomass energy conversion efficiency.
While microalgae will typically grow in any type of PBR where solar or artificial light is delivered to the culture, most PBRs are not optimized for efficient utilization of light. Microalgae have a toxic limit with sunlight that is much lower than the average intensity of solar irradiance [14]. Figure 1.1 shows that the saturation intensity of sunlight is about an order of magnitude lower than the intensity delivered by solar irradiance. The effects of these kinetics are that sunlight is typically delivered to microalgae at an intensity too high, thus either wasting the solar energy or causing photoinhibition and damaging the photosynthetic apparatus.

The process of oxygenic photosynthesis uses the energy delivered in photosynthetically active radiation (PAR) photons to remove electrons from water in an oxidation reaction, where light is considered a substrate because the energy of the light is stored in the products [20]. The coupled reduction reaction is the transfer of electrons to carbon dioxide, thereby reducing (“fixing”) it into an organic carbon form. There is a complex process of biochemical steps that determines the rate that the photosynthetic apparatus inside the cell can accept photons. From a minimal PPFD up to the saturation intensity (I_sat), the photosynthetic apparatus can accept the photons efficiently and the growth rate increases with an increase in photon flux, as shown in Figure 1.1. However, when the PPFD reaches I_sat, there are not enough open reaction centers to utilize the incident photons, and therefore any further increase in PPFD does not result in an increase in specific growth rate. Further increases in PPFD intensity above I_sat can result in damage to the organism due to photoinhibition and photooxidation [14].

The maximum specific growth rate typically occurs around 200 µmol m^{-2} sec^{-1} [21, 22]. Unfortunately for microalgae in sunny regions of the US, solar irradiance is delivered at a flux rate an order of magnitude greater than that of I_sat. This much sunlight directed onto algae causes photoinhibition and possibly irreparable damage to the photosynthetic apparatus [14]. Fortunately for engineers, this produces an interesting problem in photobioreactor design.
1.2. Light Dilution

In order to take advantage of the high rate of sunlight delivery and eliminate photoinhibition, a method must be used to dilute the PAR inside the PBR. There are two design methods used for distributing the energy: (1) temporal dilution and (2) spatial dilution. Temporal dilution relies on the phenomenon that light/dark frequency caused by turbulent mixing can dilute the PPFD over time (i.e. temporally) by exposing the cells to short periods of high intensity light followed by longer periods in the dark, thus reducing the time-averaged intensity below the saturation intensity [13, 25-27]. The alternative method, known as spatial dilution, is where the incident sunlight is spread over a larger surface area, either by means of optical systems or techniques involving reactor geometry [17, 28, 29]. Both methods have been shown to reduce the average
PPFD below $I_{sat}$, which increases the energy conversion efficiency. To date, most PBR's have not been designed to utilize either dilution technique, except for a select few.

The effects of temporal and spatial dilution are demonstrated in Figure 1.2 and Figure 1.3, respectively. As shown, a microalga cell inside a PBR utilizing temporal dilution would experience short periods of high light intensity, up to $I_{solar}$, followed by longer periods in the dark. The time-averaged light intensity is then below $I_{sat}$. This scheme is a simple concept, but the duration of the exposure time and dark time are critical to the effectiveness of operating in this manner. A PBR utilizing spatial dilution can be designed such that the maximum instantaneous and the average intensity at any point inside the culture is below $I_{sat}$.

![Figure 1.2. Instantaneous and time-averaged intensity experienced by an algae cell in a PBR designed for temporal light-dilution](image-url)
Photobioreactors utilizing temporal dilution have been shown to efficiently utilize solar irradiance [30, 31], but to date the spatial dilution concept and techniques have been underdeveloped. Spatial dilution techniques require knowledge of optical systems design and analysis. The use of optical systems to dilute sunlight can effectively eliminate saturation intensities, but it can also have a negative impact on the overall efficiency with which sunlight is delivered to the culture. While it appears that temporal dilution systems may be preferable because they use simpler optical components (e.g. a single flat or curved transparent wall) and require less capital cost, the operating costs could be significantly higher due to the requirement of turbulent mixing to induce the high-frequency light/dark cycling inside the PBR. The energy required for pumping air is linearly proportional to the flow rate. Considering that the gas flow rate used for
temporal dilution have been reported as high as 3 vvm [31], and as low as 0.25 vvm for spatial dilution [32], the energy required to mix a temporally-diluted reactor would be an order of magnitude higher than for a spatially-diluted reactor of the same volume. Since spatial dilution does not require such high mixing rates, the operating costs could be significantly less. However, spatial dilution techniques could incur relatively high capital costs, depending on the complexity of the optical systems. The balance between capital costs and operating costs is unknown yet, and should not be a deciding factor in photobioreactor design until the methods and efficacy of both dilution options are thoroughly analyzed.

1.3. Objectives of this Study

The objectives of this study are to (1) design and test an optics-integrated spatially-diluted PBR, (2) develop laboratory-scale PBRs to study the operational characteristics and biomass yields of the selected organism and solar PBR, (3) determine a volumetric productivity model for the selected organism in solar-simulating reactors, and (4) establish a model which incorporates solar and environmental data for a site and predicts daily and annual biomass yield. This represents new solar and PBR technology, practical laboratory techniques for determining organism kinetics, and methods for combining environmental data, optical relationships, and organism kinetics into a predictive model.
CHAPTER 2
LITERATURE REVIEW

2.1. Introduction

There is a large body of literature on photobioreactor design concepts, but the bulk of the literature focuses on different types of geometry, not the way in which the light is delivered [33-37]. While most reactors are designed to utilize temporal dilution of PAR to induce light-limited conditions, and there are several PBR geometries that use this dilution technique, little attention is paid to spatial dilution of light. This could very well be due to the fact that optical engineers typically do not get involved with biological engineering problems, but it is this type of interdisciplinary effort that is needed to solve the problem of designing an efficient photobioreactor system.

Typically, it is assumed that the only method to utilize high light intensity in outdoor PBRs is through the temporal dilution technique [33, 38]. This assumption could be due to the lack of awareness of spatial dilution techniques, or it could be due to the failure of previous attempts at utilizing the spatial dilution technique to efficiently convert solar irradiance into biomass. It is suggested that photobioreactor designs should be divided into two main categories based on their light dilution technique, i.e. temporal and spatial light dilution.

There are two mechanisms for diluting light: temporal and spatial dilution. Both have advantages and disadvantages for increasing the productivity of the reactor, and both have different economic impacts on reactor capital and operating expenses. Both concepts are introduced and discussed, and literature on reactors utilizing each technique is presented.

2.2. Temporal Dilution

The rate of photosynthesis is inhibited when the photosynthetic reaction centers absorb photons at a rate faster than they can transfer the electrons away from the reaction centers, which is a phenomenon known as photoinhibition [14]. Prolonged
exposure to high light intensities may cause irreparable damage to the photosynthetic apparatus such as bleaching of pigments, a phenomenon typically referred to as photooxidation [14]. Exposing cells to a PPFD above $I_{\text{sat}}$ for even short periods of time can cause some degree of photoinhibition, and when the cells are stressed due to environmental conditions the photoinhibition effect could be more severe [14]. It has been found that the effects of photoinhibition on oxygen evolution are realized very rapidly. For example, a steady-state level of oxygen evolution was reached in less than 30 minutes with the cyanobacterium *Anacystis nidulans* (*Synechococcus*) after exposure to an inhibitory light intensity [39]. Fortunately, it has been shown that temporal dilution of light can mitigate or eliminate photoinhibition [13, 26, 27].

The average light intensity incident on a cell as it moves throughout a reactor volume can be diluted over time, or temporally diluted, by exposing the cell to a quick pulse of high-intensity light followed immediately by moving the cell to the dark, where it has a chance to transfer the energy away from the photon absorption centers.

Several experiments have been performed to elucidate the effects of flashing light on the efficiency of photon utilization. One significant report on the phenomenon [13] demonstrated that for light-limited illumination intensities, the rate and efficiency of photosynthesis was identical whether the incident light was delivered constantly at a low level or flashed at an intensity and frequency such that the average intensity was at the same low level. However, temporal dilution only works for specific flashing light patterns. For instance, if the saturation intensity is $1/10^{th}$ of full sunlight, then the algal cells must spend $1/10^{th}$ of the time exposed to the light and $9/10^{th}$ of the time in the dark. Also, the duration of the flash must be sufficiently short. For example, for *Chlorella pyrenoidosa* at 25 °C in a laboratory reactor, the flash time has to be less than 4 msec to attain the maximum efficiency obtained with constant light below $I_{\text{sat}}$ [13].

While it has been proven that temporal dilution can work, it appears obvious that in order for the technique to work well reactor mixing characteristics must be designed very well, such that the high-frequency mixing is attained and the time a cell is exposed to $I_{\text{solar}}$ is very short. Effectively utilizing temporal dilution in a reactor requires
exact mixing characteristics, such that the light-dark cycle frequency and intensity meet the recommendations provided in the literature [13, 26, 27]. It must be recognized, however, that it is possible that different organisms would respond more favorably to different light-dark cycles.

Photobioreactors have been built which utilize temporal dilution. Probably the most well-studied temporally-diluted PBR (tdPBR) is the flat, inclined model developed in the 1990’s (see e.g. [31]). A reactor of this style, shown in Figure 2.1, has a large aperture directed South and is thin relative to its width and height. It lets direct and diffuse sunlight enter through the transparent surfaces, and uses the temporal-dilution effect to obtain high-efficiency. Several studies have been performed on this reactor style to determine the optimal reactor thickness [31, 40], population density, gas sparging rate, and reactor tilt angle [30]. Some of the results are presented in Table 2.1.

Figure 2.1. Vertical flat-plate Photobioreactor, with turbulent mixing to induce high-frequency light/dark cycle

1 Photograph courtesy of Pete Zemke
Table 2.1. Biomass Productivity Realized in Temporally-Diluted Photobioreactors

<table>
<thead>
<tr>
<th>Organism</th>
<th>Optimum Culture Density [gm/L]</th>
<th>Volumetric Productivity [gm L(^{-1}) day(^{-1})]</th>
<th>Aperture Productivity [gm m(^{-2}) day(^{-1})]</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Nannochloropsis</em> sp.</td>
<td>2.0 - 2.3</td>
<td>0.24</td>
<td>12.1</td>
<td>[40]</td>
</tr>
<tr>
<td><em>Spirulina platensis</em></td>
<td>15.8</td>
<td>4.3</td>
<td>51.1</td>
<td>[31]</td>
</tr>
<tr>
<td><em>Spirulina platensis</em></td>
<td>8</td>
<td>-</td>
<td>60-70</td>
<td>[30]</td>
</tr>
</tbody>
</table>

While it is obvious that vigorous mixing is critical to the effectiveness of temporal dilution, it has been shown that mixing also is critical for increasing mass transfer rates and decreasing the thickness of boundary layers around individual cells [25]. One potential advantage to spatial dilution is that the mixing does not need to be vigorous to induce temporal dilution of high-intensity light, but it is expected there will still be a minimal mixing regime to maximize mass transfer.

Experiments performed by Otto Pulz and Qiang Hu have demonstrated yields up to 120 gm m\(^{-2}\) day\(^{-1}\) of ground surface area (unpublished). Such high rates have been achieved placing the flat-plate reactors at a close spacing, such that they shade each other and therefore the incident light is partly direct and partly diffuse. This is counter-intuitive, as it is usually assumed that surface-shading would decrease productivity. However, while the volumetric productivity would decrease, the biomass yield per unit of ground area can actually increase.

2.3. Spatial Dilution

Spatial dilution is simply any technique whereby, using the reactor geometry and/or optical components, the PPFD is decreased (diluted) by spreading it over a larger surface area. Therefore, the illuminated surface-to-volume ratio of a given reactor is
increased. For instance, tubular PBRs can be defined as utilizing spatial dilution since the curved surface allows for a greater illuminated area as compared to a rectangular surface with the same volume [28]. However, a simple 2:1 spatial dilution does not reduce the incident PPFD below the saturation intensity of the algae, so tubular reactors still require turbulent mixing to induce a flashing light effect to operate at a high efficiency. In this manner, a tubular reactor is better classified as a hybrid temporal-spatial dilution PBR. To obtain spatial dilution of light at an intensity below the saturation intensity, optical concentrators and diffusers would most likely need to be used, as discussed in Chapter 3.

As demonstrated in Figure 2.2, two reactors with the same cross-sectional area, and hence same volume, can have different lit surface areas. The cylindrical tube will have a 60 % increase in lit surface area compared with the rectangular reactor. This approach to spatial dilution has some obvious drawbacks, such as the fact that the intensity across the lit surface will vary greatly, and there could be significant optical losses from the tube area that is skewed to the incident angle of light. However, this design does not require any type of tracking system and it has the ability to collect both direct and diffuse light, both of which are significant advantages.

In order for a tubular PBR to work well, it must employ temporal dilution as well since a significant amount of the culture will be illuminated at a PPFD greater than \( I_{\text{sat}} \). In essence, a tubular PBR is a hybrid reactor, utilizing both spatial and temporal dilution, though probably not utilizing either very well. The geometric spatial dilution of light induced by this design is very minimal, and in order to temporally dilute the light, a highly turbulent and well-mixed flow regime must be established and maintained. Inducing such a flow regime in long, small-diameter tubes is not a trivial task.

More sophisticated (i.e. complicated) designs have been developed that use optical systems to concentrate and sequentially distribute the light over a larger surface area. Such designs have made use of Fresnel lenses as well as concentrating reflectors for the primary concentrator. Some systems have used fiber optics to carry the light to the PBR [15-19]. While optical fibers allow flexibility in the design and separation of the
reactor from the concentrator optics, delivering light through optical fibers can be very inefficient [41]. Other spatial-dilution designs have coupled the concentrating optics directly to the distributing optics to reduce optical coupling losses and system complexity [29, 32]. With the development of optical waveguides capable of carrying light deep within the culture and substantially increasing the illuminated surface area, much higher spatial dilution ratios are possible.

![Diagram of reactors](image)

Figure 2.2. Simple reactor geometry for demonstrating concept of spatial dilution: both reactors could have the same volume and light collection area perpendicular to the incident light, but the cylindrical reactor has a larger illuminated surface area, thus the PPFD is diluted over the surface.

The “Green Solar Collector” (GSC) design [29, 42] utilizes Fresnel lenses, which couple light to the top edge of a solid planar waveguide. The planar waveguide carries the light down to a light distributor, which is an inverted triangular extension of the waveguide. The increased incident angle caused by the triangular portion of the waveguide causes a decrease in total internal reflection and an increase in refracted light escaping the waveguide. Roughening of the surface accentuates the effects of this. This concept is similar to the one developed at USU, with the big differences being that the GSC was designed for a 2:1 dilution of sunlight, which is still above the saturation intensity of most microalgae. The construction of the planar waveguides and the method with which tracking alignment is maintained is also different than the USU
design. In the GSC, the Fresnel lenses hover above the reactor and waveguides, and are adjusted in two axes to keep the light focused on the waveguide. In the USU design presented below, the reactor assembly rotates with the Fresnel lenses in at least one degree of tracking motion. This method could incur higher costs due to the increased weight of moving components, but the optical gains from reduced reflection losses could make it worthwhile. A detailed techno-economic analysis would need to be performed to compare the variety of system variations possible.

Little productivity data exists for spatially-diluted PBRs. The bulk of the literature discussed the design, but not the performance. Representative data is presented in Table 2.2.

Table 2.2. Biomass Productivity Realized in Spatially-Diluted Photobioreactors

<table>
<thead>
<tr>
<th>Design</th>
<th>Organism</th>
<th>Volumetric Productivity [gmL⁻¹ day⁻¹]</th>
<th>Aperture Productivity [gm m⁻² day⁻¹]</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.4 cm ID tubular, horizontal orientation.</td>
<td><em>Arthrossira platensis</em></td>
<td>1.26</td>
<td>30</td>
<td>[28]</td>
</tr>
<tr>
<td>Reflector-based solar concentrating system, fiber optics, side-lighting fibers, and vertical biofilms.</td>
<td><em>Cyanobacteria</em></td>
<td>NA</td>
<td>50</td>
<td>[17]</td>
</tr>
<tr>
<td>Fresnel lens-based solar concentrating system, fiber optics, and illumination sheets in a fermentor.</td>
<td><em>Chlorella sp.</em></td>
<td>0.02</td>
<td>-</td>
<td>[16]</td>
</tr>
</tbody>
</table>

2.4. Conclusions

The literature survey reveals that temporal dilution is a well-established, well-studied light dilution technique. PBRs designed to utilize temporal dilution through turbulent mixing have produced some of the highest biomass productivity rates of
enclosed PBRs. The downside to the design is the intensive mixing and high energy requirement to induce temporal light dilution.

The literature on spatial light dilution is not nearly as comprehensive as it is on temporal dilution. There have been a few attempts to integrate optical concentrators and light diffusers with a PBR, but the optically-complex systems suffered from high optical losses. The literature survey did not reveal where laboratory-based data had been used in the design of the PBR. Very little biomass productivity data is available from prototype systems, so it is difficult to make realistic estimates of daily or annual biomass productivity potential with spatially-diluted PBRs. Essentially, despite a few discreet data points, the technique of spatial-light dilution has not been thoroughly studied. A comprehensive study with lab-based reactors, an optical design which increases system efficiency, and a system model used for estimating productivity would be unique additions to the literature on spatial-light dilution.
CHAPTER 3
SPATIALLY-DILUTED PHOTOBIOREACTOR CONCEPT AND DESIGN

3.1. Introduction

Spatial distribution of sunlight is performed by using an optical system to collect and subsequently distribute sunlight over a larger surface area, thus reducing it from 2000 μmol m\(^{-2}\) sec\(^{-1}\) to a more reasonable number such as 200 μmol m\(^{-2}\) sec\(^{-1}\). This gives a 10:1 dilution of light, assuming no optical losses.

The objective, to effectively collect direct normal irradiance (DNI) and deliver it to a microalgal culture at an intensity below the saturation intensity, can be accomplished in a compact package with a minimal number of optical components. If the optics are designed correctly, the collection and delivery efficiency are high and there is potential for high aperture and volumetric yields. There are simply two optical components in the reactor design presented here: 1) the solar concentrators, and 2) the planar waveguides (Figure 3.1).

This design has minimized the number of optical components as compared to most other spatially-diluted photobioreactors, since spatially-diluted PBRs typically use fiber optic cables to transmit the light from the solar concentrators to the light diffusers. One significant aspect of this design is that the light intensity exiting the waveguides between each chamber can be tailored to any desired PPFD. However, the actual intensity delivered to the culture will fluctuate daily and annually because it is a function of the variable sunlight intensity incident on the reactor.

With this design, as the designed light intensity is increased the depth of the waveguide must be decreased, which means that the reactor chamber volume decreases as well. The optimal reactor chamber width, light intensity, and corresponding reactor volume must all be taken into consideration when designing this system. Maximizing productivity from this reactor then is a combination of experimental biomass yield data as well as optical design constraints.
3.2. Spatial Light Dilution Relationships, Efficiency, and Geometry

3.2.a. Spatial Light Dilution Relationships

The light dilution ratio (LDR), a key parameter of the sdPBR, is

\[
LDR = \frac{I_{DN}}{I_i}
\]  
Equation 1

where \(I_{DN}\) is the incident direct normal PPFD, and \(I_i\) is the PPFD delivered by the light diffusing optics (e.g. planar waveguides) to the algae.

The optical area ratio (OAR) is the ratio of light diffuser area (\(A_{\text{diffuser}}\)) to the collector aperture area normal to the sun (\(A_{\text{apt}}\))

\[
OAR = \frac{A_{\text{diffuser}}}{A_{\text{apt}}} = \eta_{\text{opt}} LDR
\]  
Equation 2
where $\eta_{\text{opt}}$ is the optical system efficiency. With the proposed design presented here, where waveguides are used to diffuse the light, $A_{\text{waveguide}}$ is the area of the light diffusing waveguides ($A_{\text{diffuser}} = A_{\text{waveguide}}$). For example, if the desired maximum $I_i$ is 200 $\mu$mol m$^{-2}$ sec$^{-1}$, the direct normal PPFD is assumed to be 2000 $\mu$mol m$^{-2}$ sec$^{-1}$, and the optical system has an efficiency of 80%, then $LDR = 10$, but $OAR = 8$. With 100% efficient optics, $LDR = OAR$, but that is not achievable. Knowledge of the optical system efficiency can make design of the light dilution ratio more accurate, since the $OAR$ can be designed for a targeted $LDR$.

3.2.b. Optical System Efficiency

The efficiency of the optical system ($\eta_{\text{opt}}$) is defined as the ratio of incident direct normal PPFD on the solar collector ($I_{\text{DN}}$) to the average PPFD emitted by the waveguide ($I_i$). Optical system efficiency is calculated as the ratio of the $OAR$ to the $LDR$.

$$\eta_{\text{opt}} = \frac{(I_i)(\text{Area}_{\text{waveguide}})}{(I_{\text{DN}})(\text{Area}_{\text{apt}})} = \frac{OAR}{LDR} \quad \text{Equation 3}$$

For example, the prototype optical system has effectively reduced the solar PPFD from 1650 $\mu$mol m$^{-2}$ sec$^{-1}$ to 120 $\mu$mol m$^{-2}$ sec$^{-1}$. The prototype collectors had $A_{\text{apt}} = 0.0155$ m$^2$, and $A_{\text{waveguide}} = 0.155$ m$^2$, with $OAR = 10$. Using the correlation in eq. 3, this represents an effective $LDR = 13.6$ at an overall system efficiency of 73%.

3.2.c. Reactor Geometry and Delivered Light Intensity

The reactor chamber volume between each pair of waveguides is driven by the light dilution ratio and the collector aperture width. The relationships for reactor volume ($V_R$), chamber height ($h$), and collector aperture area ($A_{\text{apt}}$), respectively, are as follows:

$$V_R = (h)(w)(d) = \frac{(I_{\text{DN}})(d)(w^2)(\eta_{\text{opt}})}{2I_i} = \frac{(LDR)(d)(w^2)(\eta_{\text{opt}})}{2} \quad \text{Equation 4}$$
\[ h = \frac{(w)(LDR)(\eta_{opt})}{2} \]  

\[ A_{apt} = (w)(d) \]

where \( w \) is the chamber width from the center of the waveguides (same as collector width) and \( d \) is the chamber depth (See Figure 3.2).

Figure 3.2. Line drawing of the proposed spatially-diluted photobioreactor geometry. \( I_{DN} \) and \( I_i \) are the direct normal and internal PPFD, respectively. \( w \) is the collector or chamber width, \( V \) is the chamber volume, and \( h \) is the chamber height.
These relationships are used to calculate the volume of the reactor based on an assumed or measured light dilution ratio, optical system efficiency, and chamber width. The geometric relationships are used to calculate aperture productivity of the reactor given data or estimates of volumetric productivity.

One important consideration is that

\[ V_R \propto w^2 \quad \text{Equation 7} \]

which means that the reactor width results in a 4x increase in volume. The relationship between reactor chamber volume and width is shown graphically in Figure 3.3. This relationship is important when considering the relationship between volumetric and aperture productivity, as is discussed later.

![Figure 3.3. Relationship between reactor volume and width for fixed optical efficiency and light dilution ratio](image-url)
Reactor Internal PPFD

The PPFD inside the reactors \( (I_i) \), emitted by the planar waveguides, is

\[
I_i = \frac{(I_{DN})(A_{opt})(\eta_{opt})}{A_{waveguide}} = \frac{(I_{DN})(\eta_{opt})}{OAR} = \frac{i_{DN}}{LDR} \left[ \frac{\mu\text{moles}}{m^2 \text{sec}} \right] \quad \text{Equation 8}
\]

\( I_{DN} \) varies daily and seasonally, since it is the direct normal PPFD delivered by sunlight. The delivered PPFD, \( I_i \), could be selected based on a maximum acceptable PPFD inside the culture, an average value during the year, a value that maximizes productivity over a given time period, or by some other standard.

3.3. Optical Component Selection

3.3 a. Concentrating Optics

A variety of reflective and refractive concentrating optics have been investigated for use in the sdPBR. Fresnel lenses are commercially available, so that option was thoroughly investigated. Different designs for reflector optics have been investigated, which allow for utilization of IR energy in addition to the PAR energy delivered to the culture. The concentrators considered in this study are presented below and recommendations are made.

Fresnel Lenses

Four different companies were identified that can produce Fresnel lenses with the necessary aperture for a prototype reactor. Edmund Optics, RHK, and Fresnel Technologies, Inc., have lenses available for purchase. The fourth company, 3M, does not have lenses available, instead requiring design specifications in order to make a mold for the desired lens. One potential advantage to working with 3M is their capability to produce an array of Fresnel lenses up to 1 meter wide by 3 meters long. They also report the acrylic material used has demonstrated acceptable performance in
the field for a period of 17 years, and they are developing coatings that could be used to reflect the infrared.

The lens selected for the prototype was available from Edmund Optics, with the following specifications: *Overall width: 57.15 mm; effective width: 50.8 mm; length: 304.8 mm; effective focal length: 50.8 mm; center thickness: 1.524 mm; F/#: 1; NA: 0.45; grooves/mm: 7.874; Edmund Optics part #: NT46-616.*

The Fresnel lens design is a thin lens, with similar performance to a thicker, cylindrical lens. One side of the lens is flat while the other has a series of angled planes cut into the surface, which is commonly referred to as the grooved surface.

The lens was tested on-sun and was found to have a back focal length of 47 mm (1.85 in.) with the grooves facing the sun (grooves out), and 45.7 mm (1.8 in.) with the grooves facing the image (grooves in). The lenses were also found to have a higher irradiance concentration ratio with the grooves facing the sun instead of the image, as shown in Figure 3.4. The focal zone width was measured by sweeping a 100 micron fiber optic cable across the focal zone, perpendicular to the length of the lens, shown in Figure 3.5.

The focal zone measured agreed very favorably with the focal zone predicted by TracePro, as shown in Figure 3.6. As evident in Figure 3.6, the bulk of the concentrated light falls within a 1.5 mm wide focal line. TracePro predicts a collection efficiency of 89%, but it is likely that the actual efficiency will be lower due to the scattering of light caused by imperfections in the material and the geometry of the lens.
Figure 3.4. Fresnel lens back focal length curves for grooves facing towards the sun (out) or towards the image (in)

Figure 3.5. Device for measuring focal-line width of Fresnel lens
Parabolic Troughs

Reflecting optics were investigated as an alternative to lenses. Parabolic troughs were considered in particular, due to their widespread use as a method for concentrating solar energy. With the parabolic trough, both hyperbolic and elliptical secondary reflectors were modeled, as shown in Figure 3.7 and Figure 3.8. Either can be used as a band pass filter to separate the infrared from the visible light, which could be necessary for thermal management of the waveguide entrance region. The hyperbolic secondary has the distinct advantage over the elliptical in that the focal point of the primary mirror is placed behind the secondary, which would allow for the infrared energy passing through the secondary to be concentrated onto a photovoltaic array. The elliptical secondary is mounted behind the focal point of the primary mirror; therefore the infrared transmitted through it is too diffuse for a concentrated photovoltaic array.
Figure 3.7. Parabolic primary and hyperbolic secondary solar concentrator. F1 is the primary focal point, F2 is the near focal point of the hyperbolic reflector, and F3 is the far focal point of the hyperbolic reflector.

Figure 3.8. Parabolic primary and elliptical solar concentrator. F1 is the primary focal point, F2 is the near focal point of the elliptical reflector, and F3 is the far focal point of the elliptical reflector.
Comparison Between Fresnel Lenses and Parabolic Troughs

Both solar concentrating options have their advantages and disadvantages. Fresnel lenses are simple, compact, lightweight devices that can easily be produced in a large array that can be placed on the front-end of a photobioreactor. Parabolic troughs with a secondary mirror may be slightly more complex to produce and assemble, but have a distinct advantage in only requiring tracking in one axis, as opposed to the need for two axis tracking for the Fresnel lens. Conversely, the Fresnel lens has a tighter focal point, as shown in Figure 3.9. This may allow for a thinner waveguide; however, one disadvantage to this is that the flux may not be spread over the waveguide entrance as uniformly as it would be with the parabolic system. Another advantage that the Fresnel lens has is that it is less susceptible to tracking errors, as evident in Figure 3.10. The fast focus of the parabolic trough selected as a potential prototype concentrator results in less tolerance to tracking error, but the projected tracking accuracy required of 0.15 degrees is attainable.

![Figure 3.9. Profiles of concentrated light at the focal plane for the Fresnel lens and parabolic/hyperbolic systems](image)
Figure 3.10. The effect of tracking error on collection efficiency for three different concentrators

Conclusions

The Fresnel lens appears to be an acceptable concentrator for the prototype reactor which was used as a proof-of-concept. For future studies, it is recommended that the parabolic trough system be pursued further. A side-by-side comparison of actual collection efficiency and delivery into the waveguides, as well as economics of the collectors, needs to be performed. The advantages that the parabolic/hyperbolic system provides, such as single-axis tracking, thermal management, and potential for electricity or heat generation from the infrared, are very appealing and could offset the additional costs and complexity of that type of concentrator.

3.3.b. Diffusing Optics

Thin planar waveguides are an integral component in the sdPBR. Planar waveguides are responsible for taking the concentrated flux from the PBR primary optics and uniformly distributing it inside the reactor culture, in order to distribute solar
irradiance to the algal culture at a desirable intensity. The planar waveguides act as a side-lighting optical fiber, where the flux exits the planar waveguide perpendicular to the entrance aperture. An industry partner (Lumitex, Inc., Strongsville, Ohio) with experience in backlighting optics was contracted to use proprietary techniques to build the waveguide core. Several prototypes were tested before a functional prototype was selected for use in the PBR.

Prototype Design

The prototype waveguide (Figure 3.11) had to meet the following specifications:

- ±10% flux variation across surface
- <10% light loss from entrance through exit apertures
- Watertight seal on the three non-lit edges
- Polished entrance aperture
- Reflective surface on non-lit edges to retain escaping light

![Figure 3.11. Line-drawing of the prototype planar waveguide](image)
Prototype Development

The prototype waveguides were developed by a collaborative effort between the USU Biofuels Center and Lumitex, Inc. USU developed the design and specifications to meet the needs of the prototype advanced sdPBR under development, and Lumitex, Inc. built the hardware and used proprietary technology to etch the waveguide core for light extraction. The desired traits and performance of the waveguides were specified initially, but as testing of five successive prototypes ensued, the design was modified with each prototype. The testing technique and design iterations are discussed below.

Testing Technique

Each prototype waveguide was tested at Lumitex, Inc. under artificial lighting conditions and at USU on-sun, with the actual concentrator used in the prototype sdPBR. At USU, the test device consisted of a linear Fresnel lens to concentrate sunlight into the waveguide, an enclosure for the waveguide, and a two-axis solar tracker to point the device at the sun (Figure 3.12 and Figure 3.13). The enclosure was opaque, such that no light entered except for the light emitted from the waveguide. The enclosure also had several ports along one side, where the quantum sensor was placed and readings were taken.

Design Iterations

Five different prototypes were tested. The details of each prototype are not available due to the proprietary nature of the technology, but performance observations for each prototype are discussed.
Figure 3.12. Waveguide test device on-sun

Figure 3.13: Side-view of test device
Prototype IV was selected as the best prototype since it had the highest efficiency and average flux, despite the fact that it did not have the highest minimum/maximum (min/max) flux ratio. Prototype IVa was similar to Prototype IV in design, except that it had a reflective treatment adjacent to the perimeter of the waveguides in order to reflect light back into the waveguides. The reflective treatment didn’t increase the performance as expected, so for the balance of the prototypes delivered it was not included. Prototype III also performed well and is similar in design to Proto IV; the main difference being Prototype IV has a polycarbonate envelope that wraps around the bottom edge of the waveguide. Prototypes III, IV, and IVa all had a different surface treatment than Prototypes I and II, which significantly increased their performance as is evident in Figure 3.14.

![Figure 3.14: Comparison of performance between the five waveguide prototypes tested at USU](image-url)
Each design was tested on-sun in the device shown in Figure 3.12. The data were compiled and the average flux emitted from the waveguide, the min/max flux ratio, and the overall efficiency of each waveguide is presented in Figure 3.14. A surface plot of the flux intensity emitted from one side of the planar waveguide is shown in Figure 3.15, and two images of light exiting the planar waveguide are shown in Figure 3.16 and Figure 3.17.

Figure 3.15. PAR flux emitted from the side of the prototype planar waveguide
Figure 3.16. Laser light exiting the waveguide

Figure 3.17. Sunlight exiting the waveguide
Performance Problems and Resolutions

Once a prototype was selected with desirable optical properties, leak-testing commenced. The waveguides were submersed in water for 24 hours, and to our dismay six out of ten leaked. The polycarbonate envelopes used to contain the waveguide core were sealed around the perimeter with a UV-cured adhesive, which was very brittle and prone to failing. Several attempts were made by Lumitex, Inc. to build an envelope that did not leak, but they were not successful.

In order to remedy the problem, a fixture was built at the Biofuels Center to make envelopes out of 0.01 inch thick clear polycarbonate. Three sides of the envelope were double seamed with a heat-sealing device. This approach proved very successful, as all of the waveguides were rebuilt with this envelope and none of them failed in the PBR.

Conclusions

A prototype planar waveguide was developed which met the important requirements and specifications in order to be used in the prototype sdPBR. The planar waveguides could be bolted into the reactor and removed for maintenance, and the envelope around the waveguide core minimally affects light transmission and was watertight.

When tested with a Fresnel lens on a two-axis solar tracking system pointed at the sun, the waveguide delivered the following performance:

- Efficiency from concentrator aperture to waveguide exit aperture: 77%
- Average light intensity exiting the waveguide: 135 μmol m⁻² sec⁻¹
- Standard deviation of flux exiting the waveguide: 25%

The flux deviation and efficiency were not as good as expected, but were sufficient for the prototype reactor. Future versions of the waveguides will require much less development effort, since the groundwork has been laid with these prototypes.
3.4. Prototype sdPBR

The prototype sdPBR developed at the USU Biofuels Center, shown in Figure 3.18, used linear Fresnel lenses to focus sunlight onto the edge of planar waveguides which carry the light into the algal culture. This system was designed with an OAR of 10:1. Experiments with the solar concentrators and planar waveguides demonstrated an optical system efficiency of 77% [32, 43]. With the addition of a cover glazing to prevent dust and moisture from affecting the Fresnel lenses, the overall light delivery efficiency was approximately 69%.

The reactor was comprised of multiple chambers with identical lighting conditions. The chamber width was selected based on screening experiments performed at the Biofuels Center (unpublished). For the tests reported here, the inner four reactor chambers were used.

Supporting Systems

The sdPBR utilized active control of pH and temperature to enhance algal growth, and there were several ports for introducing gas and media. The location of the various hardware components in each of the reactor chambers is shown in Figure 3.19.

The reactor had a simple sparging system designed to provide mixing via induced air-lift while the reactor was tracking the sun. The gas was introduced through the bottom corner of the reactor (see Figure 3.19), so that as the angle of the reactor changed throughout the day the gas was always introduced at the lowest point in the reactor. VWR acrylic flowmeters were used to control the gas flow rate at 0.25 vvm.

The pH was monitored with a PHCN-201 meter (Omega Engineering, Inc, Stamford, Connecticut), and pH was controlled by metering CO₂ into the culture as needed (see section 4.2 for details). CO₂ was blended with the air and introduced through the gas port.
Temperature was controlled within the reactor by transferring heat to/from a submerged serpentine stainless-steel coil, which carried a propylene glycol antifreeze fluid. The heat transfer fluid temperature was controlled by a refrigerated circulator, which utilized a thermocouple immersed in the reactor to provide feedback to the circulator’s built-in control system. Insulation was added around 4 sides of the reactor to reduce heat exchange with the environment as well as block all ambient light from entering the reactor.

The sdPBR was mounted on a two-axis solar tracking system, which was a modified hybrid solar lighting system tracker (Model HSL3010) supplied by Sunlight.
Direct (Figure 3.20 and Figure 3.21. A counter-balance was used to offset the weight of the reactor as the tracker pointed the concentrators at the sun, and the tracking system was calibrated over the course of a day with all components installed to ensure accurate tracking within the capabilities of the tracker.

![Diagram of reactor chamber](image)

**Figure 3.19.** Side-view of reactor chamber, showing location of ports for supporting systems

3.5. Conclusions

The concept of spatial light dilution is simple, but implementation is much more difficult. The geometric relationships were presented, a study of concentrating and diffusing optical components and prototype selection was discussed, and the prototype PBR design was described. Most of the components of the prototype sdPBR were commercially available. The exception is the sdPBR housing, tracker, and the planar waveguides. The planar waveguides are typically used for laptop display backlighting, so they are used in a new and unique way here.
Figure 3.20. sdPBR mounted on two-axis solar tracker

Figure 3.21. sdPBR tracking the sun
CHAPTER 4
PROTOTYPE sdPBR PRODUCTIVITY TESTS

4.1. Introduction

A set of prototype performance experiments using the sdPBR is described. The goals of these experiments were to test the technical feasibility of distributing light inside a PBR and investigate the potential for an increase in productivity over a control reactor without spatial light distribution. Reactor performance is related to location and weather conditions, in addition to the organism and culture environment, therefore a control reactor was built for comparative purposes. The control reactor provided for the ability to test the value of using a spatial-dilution system in an algal culture. The culture conditions such as media, temperature, and light intensity were not fully optimized for the organism used in the experiments, there is little outdoor productivity data with *Neochloris oleoabundans*, and environmental conditions vary greatly for different locations and seasons, so comparing the experimental productivity in these first experiments to published values in the literature has little value. However, operating a control reactor simultaneously with the sdPBR with similar culture conditions provides a basis for comparison. These preliminary tests were designed to illustrate the strengths and weaknesses of the sdPBR concept.

4.2. Methods

*Control reactor design*

A control reactor was designed that was similar in shape to the sdPBR but transmitted sunlight directly to the culture through the aperture, without the use of concentrators and diffusers to dilute the light. The culture in the control reactor was maintained at the same conditions as the sdPBR. Both reactors were operated at the same temperature, pH, gas flow rate, and sequential-batch conditions. Temperature and pH were controlled in the same manner in both reactors, as described previously. The control reactor, without its insulating wrap, is shown in Figure 4.1 and Figure 4.2.
Figure 4.1. Control reactor. Dimensions: 10 in. wide, 10.5 in. tall, and 13.75 in. long

Figure 4.2. Side view of control reactor
Environmental sensors and data acquisition

DNI was measured with a normal incidence pyrheliometer (NIP) (The Eppley Laboratory, Inc, Newport, RI, USA) mounted on a two-axis solar tracking system (Sunlight Direct, Inc, San Diego, CA, USA). Total hemispherical irradiance was measured with a precision spectral Pyrheliometer (PSP) (The Eppley Laboratory). Temperature inside the culture was measured with K-type thermocouples sheathed in stainless steel. A datalogger (Campbell Scientific CR1000, Logan, UT, USA) was used to record data. A voltage signal from each sensor was measured every second, multiplied by a conversion factor, and the average was recorded over 60 second intervals. The 60-second average of the solar irradiance [W m\(^{-2}\)], was then multiplied by time to give the energy flux [kJ m\(^{-2}\)] and summed over a 24 hour period between culture data points to calculate G [J m\(^{-2}\) day\(^{-1}\)], the total energy delivered to the microalgal culture between growth measurements.

Organism and Culture Environment

The green alga *Neochloris oleoabundans* (UTEX 1185) [44], a member of the Chlorophyceae class, was selected for use in this bioreactor because of its potential for high biomass and lipid productivity rates. This organism is used extensively in the aquaculture industry for growing bivalves [3, 45]. Lipid mass fractions of 30 – 40 % are achievable [3, 46] under optimized conditions. The mean concentration of unsaturated fatty acids has been demonstrated at 85%, with the most abundant fatty acids being 16:1, 18:2, and 18:3 [3, 47]. The biomass and lipid productivity of *N. oleoabundans* was optimized in a laboratory photobioreactor using sodium nitrate as the nitrogen source [46], and the results are summarized in Table 4.1. Based on a compositional analysis performed by the USU Analytical Lab, the C:N:P molar ratio of *N. oleoabundans* grown on SE+ media is 42 : 5.5 : 1.

The saturation intensity of *Neochloris oleoabundans* is expected to be between 5 - 10 % of full sunlight, based on experimental results with several other Chlorophyta
This means that it would have a saturation intensity on the order of 100 – 200 µmols m\(^{-2}\) sec\(^{-1}\), assuming full sunlight is approximately 2000 µmols m\(^{-2}\) sec\(^{-1}\).

| Table 4.1. Performance of *N. oleoabundans* on Sodium Nitrate [46] |
|---------------------------------|-----------------|-----------------|
| **Concentration** | **Productivity** | **Nitrate Conc.** |
| **[gm DW/L]** | **[gm DW/L-day]** | **[mM]** |
| Biomass | 3.15 | 0.63 | 10 |
| Lipids | - | 0.133 | 5 |

*Culture Maintenance and Inoculum Preparation*

Sterile techniques were used to maintain clean cultures on streak plates. A 500 mL baffled Erlenmeyer flask was inoculated every three days from a Petri dish and placed on an illuminated shaker table. The reactors were inoculated at a 1% throw volume from a shaker flask between 7-12 days old.

*Media Composition*

The media recipe (SE+, Table 4.2) was adapted from the modified soil extract (SE) media which is very similar to various versions of Bristol media used extensively for *N. oleoabundans* cultures. The defined SE media has been modified to include ferric ammonium citrate as an iron source, which based on qualitative tests helped increase growth rates. It is interesting to note that the biochemical composition of *N. oleoabundans* was previously compared when it was grown in fresh-water versus marine media, and the marine media resulted in an increase in neutral lipid content as well as an increase in dry cell weight [48]. If lipids were the target, marine media would need to be investigated.
Table 4.2: SE+ Media Recipe

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<th>Species</th>
<th>MW [gm/mol]</th>
<th>Mass Concentration [mg/L]</th>
<th>Molar Concentration [mM/L]</th>
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</thead>
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<td>850</td>
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</tr>
<tr>
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</tr>
<tr>
<td>Sodium Chloride</td>
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<td>50</td>
<td>0.86</td>
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<td>NA</td>
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<td>0.039</td>
<td>0.00003</td>
</tr>
</tbody>
</table>

Nutrient Availability

An ion chromatograph (Dionex DC-10, Sunnyvale, CA, USA) was used to monitor the ion concentration in the media to ensure nutrient availability in the reactor during sequential-batch mode. NaF was used as an internal standard.

pH Control With CO2 Injection

The pH value of the media of a growing microalgal culture tends to rise as CO2 is absorbed by the culture during photosynthesis at a rate faster than the influx rate from atmospheric CO2. Because of precipitate formation and variation in growth kinetics, it is critical to maintain a constant pH in the media to generate meaningful productivity data [49]. Therefore, an active pH control system was used to maintain static pH in the culture.

Gaseous carbon dioxide dissolves in water, forming carbonic acid (H2CO3). As the pH of the solution rises, carbonic acid becomes the hydrogen carbonate ion (HCO3⁻), and finally the carbonate ion (CO3²⁻) [50]:

\[
CO_2 + H_2O \leftrightarrow H_2CO_3 \leftrightarrow H^+ + HCO_3^- \leftrightarrow 2H^+ + CO_3^{2-}
\]  

Equation 9
Injecting CO₂ into the culture drives the equation to the left. Maintaining the pH at a constant value in this manner ensures a consistent concentration of the inorganic carbon species.

The control system injected short bursts of 0.2 micron filtered 99.995% pure CO₂ into the culture to bring the pH down below a set point. In both the solar sdPBR and all of the laboratory reactors, a Sensorex pH electrode connected to an Omega PHCN-201 mini panel-mount pH controller was used to monitor the pH and control the CO₂ solenoid valve.

The culture temperature and pH were maintained constant at 29 ± 1 °C and 7.2 ± 0.1, respectively. These set points were determined by a combination of screening tests and from recommended values in the literature [51].

**Productivity experiment design**

Experiments were performed to compare culture density and productivity between the sdPBR and control reactors. To obtain an initial assessment of the productivity potential of both reactors, the reactors were operated in sequential-batch mode. In sequential-batch mode, 10% of the reactor volume was removed every 24 hours and replaced with fresh media. This results in a 10-day hydraulic retention time (HRT), which was used to operate at late-log growth and at a high culture density. Figure 4.3 demonstrates the concept and the differences in culture density between batch and sequential-batch operation of a PBR.

Both reactors were operated at the same temperature, gas flow rate, and pH, and they had the same media composition and inoculum. Both reactors were located on the roof of the USU Biofuels Center two meters distance apart, so they were exposed to identical environmental conditions. The only difference was in the solar energy collected by each reactor: the sdPBR collected DNI while the control reactor collected both beam and diffuse irradiance. The control reactor was mounted on a fixed-angle and not tracking the sun, since it does not need to be pointed directly at the sun to collect irradiance. Therefore, the control reactor could not collect as much beam
irradiance as the sdPBR. However, the control reactor could deliver diffuse irradiance to the culture, which the sdPBR could not, due to the differences in the optical system. The ability to collect diffuse irradiance proved to be one significant disadvantage for the sdPBR, and an advantage for the control reactor, especially on cloudy days.

Figure 4.3. Example of the density of a culture either operated in 1) batch or 2) sequential-batch mode

**Growth rate, yield, and efficiency calculations**

The specific growth rate ($\mu$) is calculated with the expression

$$\mu = \frac{\ln(X_2) - \ln(X_1)}{t_2 - t_1} \quad [time^{-1}]$$  \hspace{1cm} Equation 10

where $X_2$ and $X_1$ are the biomass dry weights measured at times $t_2$ and $t_1$, respectively.
The volumetric ($P_{vol}$) and aperture ($P_{apt}$) biomass productivity are calculated using Eq.s (10) and (11), respectively.

$$P_{vol} = \frac{x_2 - x_1}{t_2 - t_1} = \frac{x_2 V_H}{(t_2 - t_1)V_R} = \frac{x_2}{(t_2 - t_1)} \Delta \text{ [mass volume}^{-1} \text{time}^{-1} \text{]}$$  \hspace{1cm} \text{Equation 11}

$$P_{apt} = \frac{(x_2 - x_1)V_R}{(t_2 - t_1)A_{apt}} = P_{vol} \frac{V_R}{A_{apt}} \text{ [mass area}^{-1} \text{time}^{-1} \text{]}$$  \hspace{1cm} \text{Equation 12}

where $V_R$ is the volume of the reactor, $V_H$ is the volume of the reactor harvested between times 1 and 2, and $\Delta$ is the dilution fraction ($V_H/V_R$).

Typically, areal productivity is incorrectly defined as the biomass productivity relative to the aperture (lit surface area) of the reactor. This is not to be confused with the biomass produced per unit of ground area, which areal productivity should technically refer to. The metric used here is called aperture productivity ($P_{apt}$), the biomass produced per unit aperture area (light collection area), which is calculated with Eq. (12).

The solar energy to biomass conversion efficiency ($\eta_{FS}$) of the reactor is calculated with Eq. (13)

$$\eta_{FS} = \frac{(P_{apt})(H)}{G_{DN}} = \frac{G_{biomass} [kJ \text{ m}^{-2} \text{day}^{-1}]}{G_{DN} [kJ \text{ m}^{-2} \text{day}^{-1}]}$$  \hspace{1cm} \text{Equation 13}

where $(H)$ is the energy content of the algae, $(G_{DN})$ is the accumulative solar energy delivered to the sdPBR over the same time period the aperture productivity is calculated, and $(G_{biomass})$ is the energy stored in the algae biomass over the same time period.

Note that this is the energy conversion efficiency over the whole solar spectrum. The photosynthetic efficiency ($\eta_{PS}$) is typically defined as the energy stored in the
biomass divided by the energy delivered in the PAR spectrum, which is approximately 40% of the entire solar spectrum of DNI. Either metric is a valid efficiency, depending on what the comparative benchmark is. The photosynthetic efficiency is useful for comparing one organism to another, or for comparing one reactor to another. When comparing the potential efficiency of biological conversion of solar energy with the efficiency of photovoltaics or solar thermal systems, $\eta_{FS}$ is the correct form to use.

4.3. Experimental Results

Specific growth rates in batch tests

A typical batch curve for the two reactors is presented in Figure 4.4. A drop in the daily accumulative energy, shown in Figure 4.5, corresponds to the drop in culture density in Figure 4.4 on day seven. It is evident that a lack of beam irradiation caused by cloud cover significantly affected the sdPBR.

The batch tests indicate that the sdPBR produces a higher specific growth rate than the control reactor. It is important to note that the specific growth rate calculation is over 24 hours, which includes dark time where mass is lost; the instantaneous specific growth rate during the daylight period would be higher.

During the exponential growth phase, the maximum specific growth rates ($\mu_{\text{max}}$) of the sdPBR and control reactor were 1.34 day$^{-1}$ and 0.94 day$^{-1}$, respectively. This shows a 30% increase in the specific growth rate of the algae inside the sdPBR. This could be due to the flux distribution inside the sdPBR, but it could also be due to the fact that the sdPBR collects approximately 30% more energy than the control reactor since it collects DNI.
Figure 4.4. Batch growth curves, which shows that the culture density in the sdPBR is more affected by cloudy weather than in the control reactor.

Figure 4.5. Accumulative energy from direct normal irradiance as measured by the NIP, and total hemispherical irradiance on a horizontal plane as measured by the PSP.
Sequential-batch tests

After the preliminary batch growth tests were performed, the reactors were inoculated and operated in a sequential-batch mode. A 16-day test was performed, where daily harvesting of 10% of the reactor and addition of new media started after 4 days of growth. The results are shown in Figure 4.6 and Figure 4.7, where Figure 4.6 shows the culture densities every 24 hours, while Figure 4.7 shows the culture densities before and after each harvest and dilution.

As in the batch test, at one point during the test there was a significant dip in the culture density of the sdPBR, but not a noticeable dip in the control reactor density. This corresponded to cloud cover that blocked beam irradiance. Daily accumulative energy, where the irradiant power from each sensor is integrated over a 24-hour period, is shown in Figure 4.8. It appears that just as the culture was adapting to the reactor and reaching steady state, there was a partially-sunny day followed by a fully cloudy day, and the response of the sdPBR was immediate.

In addition to culture density and solar irradiance, the concentrations of three key nutrients (nitrate, phosphate, and sulfate) were measured. The concentrations of these ions were monitored over the test to ensure that the culture was nutrient-sufficient. Nutrient limitation would have reduced the biomass production rate, therefore it was necessary to monitor the media to ensure light-limited conditions. The concentration in the sdPBR of the three ions which were monitored, relative to their original concentration, are shown in Figure 4.9. The results were similar for the control reactor.
Figure 4.6. Culture density in the sdPBR and control reactors over the course of a 16-day sequential-batch test.

Figure 4.7. Culture density before and an estimate of density after each harvest during the sequential-batch test. Confidence interval bars withheld for clarity, but are similar to the previous figure.
Figure 4.8. Daily accumulative energy input during the sequential-batch test as measured by the NIP and PSP

Figure 4.9. Nutrient concentration relative to the media concentration over the course of the test. *N. oleoabundans* in SE+ media
Biomass productivity rates in sequential-batch mode

The biomass aperture yield from both reactors during sequential-batch operation is shown in Figure 4.10. The productivity of the sdPBR changed much more noticeably on a daily basis compared to the control reactor, due to the influence of clouds on the growth kinetics in the sdPBR. It is apparent that disturbances in direct beam radiation did not affect the control reactor as significantly. The specific growth rates and yields from clear-sky days during the month of September are shown in Table 4.3.

![Figure 4.10](image.png)

Figure 4.10. Aperture productivity from the sdPBR and control reactors over 10 days of sequential-batch run. *N. oleoabundans*, SE+ media, pH = 7.2, temp. = 29 ± 1 °C

A paired t-test was used to determine if the productivity of the sdPBR was significantly different than that of the control reactor over the course of nine days of testing. The productivity data was paired by time, and the resulting p-value was 7.69e-
7, indicating that the difference in productivity between the two reactors is statistically significant.

<table>
<thead>
<tr>
<th>Table 4.3. Sequential-Batch Test Results for <em>N. oleoabundans</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameter</td>
</tr>
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<tr>
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</tr>
<tr>
<td>Max. Volumetric Yield [gm L&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;]</td>
</tr>
<tr>
<td>Avg. Volumetric Yield [gm L&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;]</td>
</tr>
<tr>
<td>Max. Aperture Yield [gm m&lt;sup&gt;-2&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;]</td>
</tr>
<tr>
<td>Avg. Aperture Yield [gm m&lt;sup&gt;-2&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;]</td>
</tr>
</tbody>
</table>

These results indicate that during the sequential-batch tests the sdPBR was only operating at 16% of the maximum specific growth rate achieved during the batch growth test (µ = 1.34 day<sup>-1</sup> for batch, and µ = 0.21 day<sup>-1</sup> for sequential batch). This demonstrates the need to optimize the reactor and sequential-batch operating parameters. Even with such a low specific growth rate relative to the maximum, the sdPBR still managed to yield up to 15 gm m<sup>-2</sup> day<sup>-1</sup> (Table 4.3).

*Energy Efficiency of the sdPBR*

Both the total energy efficiency and the photosynthetic efficiency of the sdPBR over a portion of the test are shown in Figure 4.11. A large spike is noted around day 14, which corresponds with a higher growth rate after a drop in the reactor culture density due to cloud cover. The increase in productivity after a decrease in density could indicate that a lower culture density is preferable, which means a larger fraction of the reactor should be harvested each day. The reason that a lower density would
increase efficiency is that the light would penetrate farther into the culture. This indicates that it may be possible to increase the density by reducing the chamber width to allow the light to penetrate farther into the culture, without negatively affecting productivity. The average overall efficiency over 10 days was approximately 1%, while the maximum was 2%.

![Energy conversion efficiency of N. oleoabundans in the sdPBR operated in sequential-batch mode. Data corresponds to efficiency over the full solar spectrum (left vertical axis) and efficiency over the PAR portion of the solar spectrum (right vertical axis).](image)

4.4. Conclusions

From the results obtained with these experiments, it is apparent that there may be value in using an optical system to spatially-dilute PAR inside an algal culture. When compared with a control reactor without spatial light dilution under similar culture conditions, the sdPBR produced up to three times as much biomass per day with only an extra 30% energy input.
The sdPBR was able to produce biomass at $P_{apt} = 15 \text{ gm m}^{-2} \text{ day}^{-1}$ in September at latitude 41.735°N. In a sunnier climate with longer days, more biomass could be produced. In addition, this was the first attempt at producing algae with this reactor, therefore the implementation of optimized culture conditions and operating conditions is expected to have higher yields. There is also room for improvement in the optical system of this reactor. Since DNI is required, care must be taken to ensure accurate solar tracking and alignment of the optical components, both of which may have decreased the efficiency of the prototype reactor. Tests demonstrated an optical system efficiency of 69%, with accurate alignment of the optics with the sun, but if the alignment is off or if it changes throughout the day the efficiency would be substantially lower. Tracking error could have made a significant contribution to the low solar energy conversion efficiency obtained in this experiment.

Solar energy conversion efficiency of up to approximately 9% (calculated over the entire solar spectrum) has been obtained with solar photobioreactors [30]. The prototype sdPBR operated at approximately 1% solar energy conversion efficiency. It is clear that there is room for improvement, which is to be expected with the un-optimized operating conditions and inefficiencies in the prototype optical system.
5.1. Introduction

A parametric study was designed to investigate the net volumetric productivity ($P_{\text{vol}}$) of reactors that simulate a chamber inside the spatially-diluted photobioreactor, where $P_{\text{vol}}$ is the net production of biomass per liter of reactor over a 24-hour period. Experimental designs were developed to investigate several parameters which affect the light environment inside the reactor, such as reactor width, diurnal cycle, light intensity, and volumetric dilution fraction. Analysis of the data lead to a statistical model of $P_{\text{vol}}$ for different reactor and lighting conditions, which was incorporated with an optical model of the sdPBR to predict aperture yield.

There are two main studies that were performed. Study 1 focused on how the light profile across the reactor width affects $P_{\text{vol}}$ by experimentally determining and maximizing $P_{\text{vol}}$ with three different reactor widths and light intensities. Study 2 focused on examining the effects of light intensity and diurnal cycle on $P_{\text{vol}}$ in a reactor that simulates a culture chamber inside the prototype sdPBR. In both studies, the reactor was operated at three of four different volumetric dilution fractions, in order to determine the maximum volumetric productivity for each combination of parameters under investigation. All of the different conditions tested are given in Appendix C (Table C.1.).

5.2. Parameters Investigated

Study 1: Effects of reactor width ($w$), light intensity ($I_i$), and dilution fraction ($\Delta$) on $P_{\text{vol}}$

It was expected that $P_{\text{vol}}$ would respond linearly to light intensity and reactor width, and that dilution fraction would have a non-linear affect on $P_{\text{vol}}$. The linear response in $P_{\text{vol}}$ to light intensity is due to the linear nature of the $P/I$ curve below the saturation limit Figure 1.1. Similarly, reactor width will affect the average light intensity
inside the reactor, so it was expected to have a linear response as well. The volumetric dilution fraction ($\Delta$), the volume of culture harvested daily relative to the total reactor volume, was expected to affect $P_{\text{vol}}$ non-linearly, as seen previously in reactor optimization studies [31]. At very low $\Delta$ (e.g. 0.1), the culture becomes very dense but the specific growth rate is low due to the age of the culture and poor light penetration. At very high $\Delta$ (e.g. 0.9), the growth rate is high due to the high light penetration, but the culture density is very low and thus productivity is low. The maximum productivity is obtained between the high and low $\Delta$, where the combination of a strong growth rate and good culture density are combined.

The light intensity and reactor width were each varied at three levels. $P_{\text{vol}}$ was optimized by testing at different dilution fractions for a given light intensity and reactor width pair. To do this, the dilution fraction was varied at three of four levels in order to bracket the maximum $P_{\text{vol}}$. This design allowed for a fully parametric study, where the maximum $P_{\text{vol}}$ versus light intensity and reactor width was generated.

The reactor widths selected for testing were 20, 35, and 50 mm. The selection of these widths was based on preliminary studies at the USU Biofuels Center. Absorbance vs. depth was measured for multiple densities, and the results are shown in Figure 5.1 (see Appendix A for methods). Since the reactors were lit from both sides, the effective light-path was only half of the width (e.g. 10 mm in the 20 mm reactor). According to the data presented in Figure 5.1, over 90% of the PAR would be absorbed by 5 mm depth for typical culture densities obtained in these reactors, so it was expected that all of the incident light on the reactor was absorbed.

The average light intensity incident on the side of the reactor was fixed at three different levels: 75, 125, and 175 $\mu$mol m$^{-2}$ sec$^{-1}$. These levels were chosen based on the previous discussion on the saturation intensity of *N. oleoabundans*, which is expected to be between 100 and 200 $\mu$mol m$^{-2}$ sec$^{-1}$ [21].

The dilution fraction was varied at three or four levels, in order to obtain an estimate for maximum productivity. The dilution fractions used in this study were: 0.1, 0.3, 0.5, 0.7, and 0.9.
The data analysis was performed using the Analysis ToolPak in Microsoft Excel, v. 2007. A regression model was applied to the different factors and a combination of factors to test for significance and linearity of the response variable, $P_{\text{vol}}$. The experiment had two main factors ($w$ and $I_i$), which varied over three levels each, as well as three or four levels of $\Delta$. Each of the possible 27$(+)$ combinations of factors was tested once, which means there were not true replicates of the data. However, during each experiment, the productivity observation was based on three consecutive days when steady-state conditions were been met, so that each observation was an average of three values. This may be considered a pseudo-replication. The photobioreactor was assumed to be at steady-state when the standard deviation of $P_{\text{vol}}$ varied by less than 5% over three consecutive days.
Study 2: Effects of diurnal cycle (DC), light intensity (I), and dilution fraction (Δ) on \( P_{\text{vol}} \)

The \( P_{\text{vol}} \) of the 50 mm wide reactor, which is approximately the same width as the prototype solar sdPBR, was determined at three different light intensities and three different diurnal cycles, and maximized with respect to dilution fraction. This range of factors was selected in order to simulate the conditions inside an sdPBR in the natural environment.

The average daily light intensity and daylight period (diurnal cycle) was based on outdoor conditions in the desert southwest. The light intensity was set at three levels: 75, 125, and 175 \( \mu \text{mol m}^{-2} \text{sec}^{-1} \), which represent realistic values that the constructed sdPBR optics can deliver to the culture throughout the year. The diurnal cycle (hours on / hours off) was set at three levels: 9/15, 12/12, and 15/9, which are typical minimum, average, and maximum daily light cycles. The different pairs of combinations were operated at three or four different dilution fractions, in order to maximize \( P_{\text{vol}} \). The statistical analysis was performed similarly to Study 1.

5.3. Methods

Experimental Reactor Design

The design of the experimental reactors used in the laboratory for organism and reactor kinetics was based on the design of an individual chamber inside the sdPBR. All design elements were considered and made to simulate the conditions of a reactor on-sun, which meant a unique sunlight-simulating light source had to be identified.

The reactors were square in shape, with 250 x 250 mm parallel transparent polycarbonate walls perpendicular to the direction of the light path, and were lit from both sides as shown in Figure 5.2. The reactors had widths of 20, 35, and 50 mm in the direction of the light path, and the light profile has the form shown in Figure 5.3. The reactors were shaped as such so they would be similar to the chamber configuration of the outdoor spatially-diluted PBR, and culturing chambers with plane parallel lit surfaces have an easily resolved light intensity profile. Cylindrical reactors such as stirred-tank
reactors or Erlenmeyer-style flasks on a shaker table have a poorly-defined light profile as well as a long optical path-length, making them unrealistic for determination of culture kinetics on light [52]. Square or rectangular reactors, such as the ones used here, have a light profile which is simple to measure.

The reactors were tipped at a shallow angle to induce airlift mixing and to simulate the effects of tracking the sun. Mixing induced by a gas flow is sufficient for a variety of gas flow rates above 0.1 vvm, based on the results of qualitative dye tests. Additional tests determined that a flow rate of 0.5 vvm was preferable to minimize biofilm formation on the walls of the reactor.

![Figure 5.2. Experimental laboratory reactor lighting sketch](image)

**Operating Conditions**

The following culture operating conditions were used for all experiments. Typical conditions for reactor operation were obtained from the literature or determined with screening tests with *N. oleoabundans*. Media and culture maintenance information is the same as in section 4.2.
Light Source

SoLux brand incandescent lamps, which have a very similar spectrum to DNI, were used with different diurnal cycles in order to simulate solar lighting conditions. As is readily apparent in Figure 5.4, the SoLux lamp delivers a visible light spectral power distribution (SPD) very similar to that of the ASTM E891-82 Air Mass 1.5 Sunlight. The SPD of a plant and aquarium style fluorescent bulb is also shown in the figure, for comparative purposes. Fluorescents, metal halides, high pressure sodium, or other typical light sources generally do a poor job of simulating sunlight.

The SoLux bulb is a 12 V, 50 W, MR-16 bulb which has been tuned with reflecting filters to match sunlight. The 4700 K model with the 36 degrees beam spread was used. SoLux cages were used to hold the light bulbs, and Malibu 12 V / 600 W power supplies were used to power eight bulbs (400 W total power required) for each reactor. A light-diffusing acrylic sheet (Acrylite Satin Ice) was placed between the bulbs and the reactor in order to help spread the light evenly over the reactor surface as well as attenuate some of the infrared energy that would put an additional heat load on the reactors.

The PPFD across the surface of the reactor was measured at 25 points on the inside of the lit surface with a Li-Cor 2\pi quantum sensor. The average PPFD of all 25 measurements was used to determine the spacing between the light bank and reactor,
and this was performed for each lit surface of the reactor. A typical flux profile is shown in Figure 5.5. The distance between the reactor and light bank were adjusted to give the desired intensity. All reactors were configured individually for each light intensity, and the average PPFD vs. distance curve is shown in Figure 5.6.

![Figure 5.4. Normalized SPD of the ASTM Air Mass 1.5 Direct Normal Irradiance (Sunlight), a SoLux lamp, and a Philips Plant & Aquarium Fluorescent Lamp](image)

**Temperature Control**

Temperature in the reactor was maintained by using temperature-controlled distilled water from a VWR Signature™ heated/refrigerated circulator through a stainless steel coil inside each culture chamber, and pulling ambient air over the reactors and light bulbs with a fan. A K type thermocouple was immersed in each reactor, which was monitored by a National Instruments cDAQ-9172 signal acquisition board. The circulating fluid flowed through all reactors in parallel, and changes in the fan speed were used to fine-tune the operating temperature of the reactor.
Figure 5.5. Distribution of PPFD on reactor lit surface

Figure 5.6. PPFD vs. light spacing for bench-top reactors
The cultures were operated at a temperature of 29 °C ±1 °C. This was chosen based on the results of simple shaker-flask tests in our lab, as well as reported cultivation temperatures ranging from 28 to 34 °C [46, 47].

Gas Delivery and Mixing

A pair of 0.025 in. diameter ports in the bottom corner of the reactor was used for delivering gas to the reactor. Introducing gas at the bottom corner of the reactor induces air-lift of the fluid, which creates thorough mixing within the reactor. Laboratory air, which has been filtered and dried, was run through a pressure regulator and delivered at 10 psig to a manifold feeding all reactors. Bottled CO₂, obtained from the Utah State University gas sales, was fed to a manifold at a slightly higher pressure than the air pressure. The pH control system operates the solenoid valves on the CO₂ manifold to control the pH in each reactor (see section 4.2).

Screening tests and qualitative dye tests were used to determine the operating gas flow rate of 0.5 vvm. This was higher than the flow rate used in the outdoor sdPBR tests, but the sdPBR has the additional mixing imparted by the sun-tracking motion of the reactor. At 0.5 vvm, biofouling was kept to a minimum in all reactor widths. At lower gas flow rates, the 20 mm reactor would occasionally form a biofilm on the lit surfaces, thus ruining the test.

5.4. Sample Experimental Data

Sample data sets are presented to demonstrate the response of a photobioreactor to sequential harvesting. The daily harvesting and addition of fresh media was started when the culture appeared to be well-established, with a density of at least 100 mg/L. Once sequential-batch harvesting was initiated, it would usually take three to seven days to reach steady-state. The harvesting would be initiated below, above, or right at the steady-state density, and the culture would respond accordingly. Examples of responses to the various cases are shown below.
Figure 5.7. Culture density response to harvesting started when the density is below the steady-state operating density.

Figure 5.8. Culture density response to harvesting started when the density is above the steady-state operating density.
In order to gain confidence in the experiment design and ensure reliability of running each set of conditions once, a set of parameters was selected at random to run in triplicate. The resulting DW data is shown in Figure 5.10. The average DW at the end of the three tests was 0.45 gm/L, with a standard deviation of 2%. The average $P_{vol}$ was 0.31 gm L$^{-1}$ day$^{-1}$, with the same standard deviation. The low standard deviation in the data between three replicates, which were run several weeks apart, builds confidence in the methods and the ability of the organism to reach steady-state in the reactors.

The density immediately after harvesting and addition of fresh media, the lower density during sequential-batch operation (see e.g. Figure 5.7, Figure 5.8, and Figure 5.9) is an estimate based on the fraction harvested and the DW immediately before harvesting. The line connecting the lower DW to the upper DW at the end of a 24 hour period is simply representative of the increase in DW over time. The actual culture density changed very little immediately following a harvest and dilution, since this occurred at the end of the light period. In this environment, there was minimal loss due
to respiration during the dark period, and growth resumed when the light period began (Figure 5.11).

![Graph showing dry weight over time for different conditions](image)

**Figure 5.10.** Triplicate test of \( w = 50 \text{mm}, \; I_i = 175 \mu\text{mol m}^{-2} \text{sec}^{-1}, \; DC = 12/12, \) and \( \Delta = 0.7. \) DW at end of day period is shown. Diluted DW after each harvest withheld for clarity.

The important data point recorded from each experimental setup was the density at the end of the light period. This value was the key indicator of steady state conditions, and the density multiplied by the volume of culture removed provides the volumetric productivity – the response variable in these studies.
5.5. Results of Study #1: Effects of Light Intensity, Reactor Width, and Dilution Fraction On Volumetric Productivity

The results of Study 1 are presented. In Figure 5.12, Figure 5.13, and Figure 5.14, error bars represent the standard deviation of the steady-state productivity data for a given set of conditions. In Figure 5.15 and Figure 5.16, the error bars represent the 95% confidence interval that the mean productivity value lies within. If error bars are not visible, the confidence interval is covered by the marker. All data presented was obtained with N. oleoabundans, with the pH, temperature, and media defined in the preceding section.
Figure 5.12. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at $I_1 = 75 \, \mu\text{mol m}^{-2} \text{sec}^{-1}$

Figure 5.13. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at $I_1 = 125 \, \mu\text{mol m}^{-2} \text{sec}^{-1}$
Figure 5.14. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at \( I_l = 175 \, \mu\text{mol m}^{-2}\,\text{sec}^{-1} \)

![Graph showing volumetric productivity vs. dilution fraction](image)

Figure 5.15. Maximum volumetric productivity of *N. oleoabundans* vs. light intensity

![Graph showing volumetric productivity vs. light intensity](image)
Figure 5.16. Maximum volumetric productivity of *N. oleoabundans* obtained at each reactor width. Lines represent light intensities.

*Study 1 Model*

Regression analysis was performed on the two independent variables, light intensity and reactor width. Both factors were statistically significant, and \( P_{\text{vol}} \) responded linearly to each. The two factors were then lumped together into a common factor: the quantity of photons per unit volume per day. The definition of this parameter led to a simple model with a constant biomass yield term.

A simple yield model is proposed, where

\[
P_{\text{vol}} = Y_{X/P} \cdot \text{TLD}
\]

Equation 14

TLD is the “total light delivered,” or total number of photons delivered per unit volume over one day [mol photons/L], and \( Y_{X/P} \) is the yield of biomass on photons [gm mass/mol photons]. TLD is calculated as follows
where \( I_{i,avg} \) is the average internal PPFD over the course of a day.

A linear regression analysis was performed in Excel to determine the value of \( Y_{X/P} \) based on the TLD and corresponding maximum productivity data from Study 1 (Table 5.1). The analysis showed a strong correlation between \( P_{vol} \) and TLD over a 24 hour period with different reactor volumes, such that

\[
Y_{X/P} = 1.08 \frac{gm \text{ mass}}{mol \text{ photons}} \tag{Equation 16}
\]

\[
P_{vol} = TLD \left( 1.08 \frac{gm \text{ mass}}{mol \text{ photons}} \right) \tag{Equation 17}
\]

The relationship between \( P_{vol,max} \) and TLD is shown in Figure 5.17. This data demonstrates the linear response of daily volumetric productivity to total light delivered.

<table>
<thead>
<tr>
<th>( I_i ) [( \mu mol \text{ m}^{-2} \text{ sec}^{-1} )]</th>
<th>w [mm]</th>
<th>( I_{avg} ) [( \mu mol \text{ m}^{-2} \text{ sec}^{-1} )]</th>
<th>TLD [( \mu mol \text{ L}^{-1} )]</th>
<th>( P_{vol} ) [gm L(^{-1}) day(^{-1})]</th>
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</thead>
<tbody>
<tr>
<td>75</td>
<td>20</td>
<td>46.9</td>
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<tr>
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<tr>
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<td>35</td>
<td>109.4</td>
<td>0.540</td>
<td>0.612</td>
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<tr>
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<td>50</td>
<td>109.4</td>
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</table>

\(^2\) \( I_i \) = internal PPFD; \( w \) = reactor width; \( I_{avg} \) = 24-hour average PPFD; TLD = total light delivered; \( P_{vol} \) = volumetric productivity
Figure 5.17. Maximum volumetric productivity obtained with N. oleoabundans vs. total light delivered

5.6. Results of Study #2: Effects of Light Intensity, Diurnal Cycle, and Dilution Fraction on Volumetric Productivity

The results of Study 2 are presented in Figure 5.18, Figure 5.19, and Figure 5.20. Error bars represent the standard deviation of the steady-state productivity data for a given set of conditions. In Figure 5.21 and Figure 5.22, the error bars represent the 95% confidence interval that the mean productivity value lies within. If error bars are not visible, the confidence interval is covered by the marker. The methods used to determine the confidence interval are discussed in Appendix B.
Figure 5.18. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at $I_1 = 75 \, \mu \text{mol m}^{-2} \text{sec}^{-1}$

Figure 5.19. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at $I_1 = 125 \, \mu \text{mol m}^{-2} \text{sec}^{-1}$
Figure 5.20. Volumetric productivity of *N. oleoabundans* vs. dilution fraction at $I_l = 175 \mu\text{mol m}^{-2} \text{sec}^{-1}$

Figure 5.21. Maximum volumetric productivity of *N. oleoabundans* obtained at each light intensity. Lines represent diurnal cycles.
Study 2 Model

A linear regression analysis was performed in Excel to determine the significance and linearity of the volumetric productivity response to light intensity and diurnal cycle. In addition, a linear regression was performed to determine the value of $Y_{X/P}$ based on the TLD and corresponding maximum productivity data from Study 2 (see Table 5.2). The analysis showed a strong correlation between biomass yield and total delivered light over a 24-hour period with a light and dark cycle, such that

$$Y_{X/P} = 1.15 \frac{gm \text{ mass}}{mol \text{ photons}}$$  
Equation 18

$$P_{vol} = TLD \left( 1.15 \frac{gm \text{ mass}}{mol \text{ photons}} \right)$$  
Equation 19
Table 5.2. Study 2, Salient Data for Regression Analysis

<table>
<thead>
<tr>
<th>$I_i$</th>
<th>$N_{DL}$</th>
<th>$I_{avg}$</th>
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<td>46.9</td>
<td>0.16875</td>
<td>0.184</td>
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<tr>
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<td>0.172</td>
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<td>0.225</td>
<td>0.257</td>
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<td>0.315</td>
<td>0.313</td>
</tr>
<tr>
<td>75</td>
<td>15</td>
<td>46.9</td>
<td>0.16875</td>
<td>0.207</td>
</tr>
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<td>78.1</td>
<td>0.28125</td>
<td>0.318</td>
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<tr>
<td>175</td>
<td>15</td>
<td>109.4</td>
<td>0.39375</td>
<td>0.444</td>
</tr>
</tbody>
</table>

The relationship between $P_{vol,max}$ and TLD is shown in Figure 5.23. This data demonstrates the linear response of daily volumetric productivity to total light delivered, as was seen in Figure 5.17.

---

$R^2 = 0.966$

Figure 5.23. Maximum volumetric productivity obtained with *N. oleoabundans* vs. total light delivered

---

$I_i$ = internal PPFD; $N_{DL}$ = number of daylight hours; $I_{avg}$ = 24-hour average PPFD; TLD = total light delivered; $P_{vol}$ = volumetric productivity
5.7. Combined Factors and Response

*Linear Model*

The maximum volumetric productivity and factor data from both studies can be combined into one data set. TLD is calculated as shown previously for each combination of light intensity, number of daylight hours, and reactor width, and a statistical analysis is performed on the complete data set. The regression analysis for the entire data set results in a biomass yield on photons value of

\[ Y_{X/P} = 1.09 \frac{g m \text{ mass}}{mol \text{ photons}} \]  

\[ P_{\text{vol}} = TLD \left( 1.09 \frac{g m \text{ mass}}{mol \text{ photons}} \right) \]  

Equation 20

Equation 21

There is a highly significant correlation between TLD and \( P_{\text{vol}} \) as expected (p-value = 2.4x10^{-20}). The measured and predicted volumetric productivity are shown in Figure 5.24. Error bars represent the 95% confidence interval.

*Second-Order Model*

While the linear model and corresponding \( Y_{X/P} \) yield acceptable results, a second-order polynomial relationship between \( P_{\text{vol}} \) and TLD fits the data better. This is due to the fact that the oxygenic photosynthetic response of algae to increasing light intensity is non-linear. The curve (Figure 5.25) takes on the shape of the P/I curve (Figure 1.1), which is nonlinear below \( I_{\text{sat}} \).

The second-order model is represented by the equation

\[ P_{\text{vol}} = -0.239 TLD^2 + 1.23 TLD \]  

Equation 22
Figure 5.24. Measured volumetric productivity vs. total light delivered for all sets of experimental conditions

Figure 5.25. Second-order model for biomass productivity of *N. oleoabundans* on TLD
The strength of the correlation \( (R^2) \) between the predicted \( P_{\text{vol}} \) and the measured \( P_{\text{vol}} \) is 0.9915 for the second-order equation, while it is 0.9776 for the linear equation (Figure 5.26). Because of the stronger fit to the data, the second-order model is used as the productivity model for the sdPBR for the data presented in Chapter 6.

![Figure 5.26. Correlation between predicted and measured volumetric productivity of *N. oleoabundans*](image)

5.8. Conclusions

The relationship between maximum volumetric productivity total light delivered to a culture of *N. oleoabundans* was determined for light intensities below the saturation limit, typical diurnal cycles, and reactor widths of 20, 35, and 50 mm. The relationship can be estimated with a linear model, but a second-order nonlinear curve fits slightly better. This slight non-linearity is due to the non-linearity of the photosynthesis/irradiance curve shown in Figure 1.1.

The maximum productivity for each pair of conditions was obtained by adjusting the dilution fraction to bracket in the maximum productivity. These curves represent
the significance of the dilution fraction on maximizing the productivity of a given reactor. The way in which a reactor is operated, and not simply how much light is delivered to it, can greatly increase its productivity.
6.1. Introduction

A reactor productivity model combines the response of *N. oleoabundans* to different environmental conditions obtained experimentally with the optical design, configuration, and experimental performance results of the sdPBR. To calculate biomass productivity for each day of the year, the parameters shown in Table 6.1 were used.

### Table 6.1. Parameters Used in the Productivity Model

<table>
<thead>
<tr>
<th>Reactor Optical Parameters</th>
<th>Units</th>
<th>Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Light dilution ratio, LDR</td>
<td>NA</td>
<td>Constant</td>
</tr>
<tr>
<td>Reactor width, w</td>
<td>m</td>
<td>Constant</td>
</tr>
<tr>
<td>Reactor volume, V_R</td>
<td>L</td>
<td>Calculation</td>
</tr>
<tr>
<td>Aperture &amp; waveguide area, A</td>
<td>m²</td>
<td>Calculation</td>
</tr>
<tr>
<td>Optical efficiency, η_opt</td>
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<td>Constant</td>
</tr>
</tbody>
</table>

**Environmental Parameters**

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<th></th>
<th>W/m²</th>
<th>TMY3⁴</th>
</tr>
</thead>
<tbody>
<tr>
<td>Direct Normal Irradiance, DNI</td>
<td>µmol m⁻² sec⁻¹</td>
<td>Calculation</td>
</tr>
<tr>
<td>Date / Time</td>
<td>Local Standard Time</td>
<td>TMY3</td>
</tr>
<tr>
<td>Declination, δ</td>
<td>Degrees</td>
<td>Calculation</td>
</tr>
<tr>
<td>Day of the year, n</td>
<td>Day #</td>
<td>TMY3</td>
</tr>
<tr>
<td>Number of daylight hours, N_DL</td>
<td>Hours</td>
<td>Calculation</td>
</tr>
</tbody>
</table>

**Reactor Biological Parameters**

<table>
<thead>
<tr>
<th></th>
<th>mol photons/L</th>
<th>Calculation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Light Delivered, TLD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Biomass yield on photons, Y_X/P</td>
<td>gm mass/mol photons</td>
<td>Constant</td>
</tr>
</tbody>
</table>

6.2. Solar and Environmental Data for System Productivity Model

*Calculation of the Number of Daylight Hours*

The productivity model for the prototype reactor requires knowledge of the daily average light intensity inside the reactor \((I_{i,avg})\) and TLD, which both require the number of hours of daylight \(N_{DL}\) to be calculated. The TMY3 data set does not include \(N_{DL}\) so it must be calculated as follows

\[
N_{DL} = \frac{2}{15} \cos^{-1}(-\tan \phi \tan \delta)
\]

Equation 23

where \(\phi\) is the latitude and \(\delta\) is the declination angle [54]

\[
\delta = 23.45 \text{ deg} \sin \left(360 \frac{284 + n}{365}\right)
\]

Equation 24

where \(n\) is the day of the calendar year.

*Estimation of Direct Normal PPFD from Direct Normal Irradiance Data*

The ratio of \(I_{DN}\) to DNI (eq. 25) has been estimated using the ASTM standard spectral power distribution for Air Mass 1.5. Over the spectra between 400 nm and 700 nm, there are approximately 1390 \(\mu\text{mol}\) photons m\(^{-2}\) sec\(^{-1}\) and 299.6 W m\(^{-2}\).

\[
I_{DN}/\text{DNI} = \frac{\int_{\lambda=400\text{nm}}^{\lambda=700\text{nm}} \frac{\text{DNI}(\lambda)}{h_{\lambda}^2} d\lambda}{\int_{\lambda=400\text{nm}}^{\lambda=700\text{nm}} \text{DNI}(\lambda) d\lambda}
\]

\[
I_{DN}/\text{DNI} \approx \frac{1390}{299.6} \frac{\mu\text{mol}}{\text{m}^2\text{sec}} = 4.64 \frac{\mu\text{mol}}{\text{W/m}^2\text{sec}}
\]

Equation 25
where $h$ is Planck’s constant ($6.626068 \times 10^{-34}$ J sec) and $c$ is the speed of light in a vacuum ($2.998 \times 10^8$ m/s). Figure 6.1 shows measured DNI data at the USU Biofuels Center in Logan, Utah, and the corresponding $I_{DN}$. Figure 6.2 demonstrates the difference between $I_{DN}$ and $I_{TH}$, the total hemispherical PPFD for clear-sky conditions. It is important to note that the ballpark number of 2000 µmol m$^{-2}$ sec$^{-1}$ is typically used for PPFD of “sunlight,” but this is for total hemispherical PPFD. Direct normal PPFD from beam radiation is approximately 20% less. This is an important consideration.

![Figure 6.1. Measured direct normal irradiance and corresponding direct normal PPFD](image)

**Solar Data for Model Site: Las Vegas, Nevada**

The NREL Redbook [55] was consulted to select a model city for biomass productivity estimates. The annual average DNI was the salient factor, and it is presented for a selection of cites in Figure 6.3. Las Vegas, Nevada was chosen as the model city for the biomass productivity estimates due to its high level of direct normal irradiance. The low DNI of Western Washington is shown for comparison.
Figure 6.2. Direct normal and total hemispherical PPFD on a horizontal plane over a day

Figure 6.3. Average annual DNI for a selection of U.S. cities
Figure 6.4 shows the daily average flux for energy delivered to a flat-plate, a 1-axis tracker aligned along a N-S axis, and two a 2-axis tracker. Note that the energy flux plotted for both of the trackers here is the beam component of sunlight only, which is what the sdPBR can collect.

![Figure 6.4. Monthly averages of daily energy flux for different collector types in Las Vegas, NV (adapted from [55])](image)

**Las Vegas Hourly Data**

Typical Meteorological Year 3 (TMY3) data was used to provide estimates of hourly solar and environmental conditions [53]. The data set is based off of measured data over the years 1961 to 1990 and 1991 to 2005, and provides statistically typical hourly conditions for the location for every hour in the year. Irradiance and illuminance for a representative day are graphed in Figure 6.5.

Since neither global horizontal nor direct normal PPFD were measured, estimates were made based on the direct normal irradiance.
6.3. Maximum Performance Predicted with the sdPBR and *Neochloris oleoabundans*

The volumetric productivity model was combined with the TMY3 data for Las Vegas, Nevada, in order to estimate the daily and annual biomass productivity. The aperture and volumetric productivity for a two-axis tracking sdPBR, with different reactor widths, are presented.

The estimated total biomass produced is 14.6 kg m\(^{-2}\) yr\(^{-1}\) for a reactor with an LDR of 10, 50 mm width, and an 85% optical efficiency. The daily biomass productivity per unit aperture is shown in Figure 6.6, and monthly averages are given in Figure 6.7. The maximum aperture productivity is estimated at 69 gm m\(^{-2}\) day\(^{-1}\), with an annual average of 40 gm m\(^{-2}\) day\(^{-1}\).
The biomass yield per area of sunlight collected changes only minimally based on reactor chamber width, but the yield per unit volume of reactor \( P_{\text{vol}} \) changes much more significantly. This is simply due to the higher photon flux per liter of culture in the thinner reactor widths. The increase in volumetric productivity is inversely proportional to the square of the reactor width, over the range of reactor widths tested in this study. The annual maximum and average aperture and volumetric productivities for \( w = 20\text{mm} \), \( 35\text{mm} \), and \( 50\text{mm} \) are given in Figure 6.8 and Figure 6.9, respectively. An economic study would be required to compare the increase in aperture yield vs. the substantial decrease in volumetric productivity, since both have economic implications.
Figure 6.7. Monthly averages for biomass productivity per unit aperture area; LDR=10, w=50mm, $\mu_{opt} = 85\%$, Las Vegas, NV

Figure 6.8. Maximum and average aperture biomass productivity vs. reactor width; DR=10, $\mu_{opt} = 85\%$, Las Vegas, NV
To estimate the full-spectrum solar energy conversion efficiency, the maximum aperture yield of 14.6 kg m$^{-2}$ yr$^{-1}$ was multiplied by an estimated energy content of H = 21.1 KJ/gm for the model organism, and then divided by the total energy delivered per year of $G_{DN} = 9.3$ MJ/m$^2$.

$$
\eta_{FS} = \frac{(P_{apt})(H)}{G_{DN}} = \frac{(14.6 \text{ kg m}^{-2} \text{ yr}^{-1})(21.1 \text{ KJ/gm})}{9.3 \text{ MJ/m}^2} = 3.3\% \quad \text{Equation 26}
$$

With LDR = 10, w = 50 mm, and $\eta_{opt} = 85\%$, the estimated full spectrum efficiency is $\eta_{FS} = 3.3\%$. From this, the estimated photosynthetic efficiency is $\eta_{PS} = 8.5\%$.

6.4. Conclusions

A method for combining a biological yield model with historical solar data has been described. This simple method allows for the estimation of biomass productivity
for different locations throughout the U.S. It is preferable to use TMY data over frequently used ballpark numbers such as 2,000 μmol photons m$^{-2}$ sec$^{-1}$ or 1,000 W m$^{-2}$, because those figures are not what a reactor would actually be exposed to throughout the day or year.

The model does have limitations. One is that the model assumes that the reactor would be operated in sequential-batch mode at the maximum volumetric productivity point. This is possible though, since the density just before harvesting should indicate the fraction of the reactor to be harvested. Instead of focusing on a fixed dilution fraction, it would probably be advantageous to operate at a fixed daily starting density. Another limitation of the model is that it doesn't account for the loss in algal culture due to endogenous metabolism which would be experienced during several days of cloud-cover. The model assumes there is zero productivity (no net positive culture growth) at that time, but it does not assume the viability of the culture would be reduced over a cloudy period. The assumption is that the culture would resume normal growth rates and biomass yield when direct normal irradiance returned. Daily and even hourly changes in the photon flux may or may not cause the volumetric productivity model to deviate from steady-state conditions, and therefore the response of the culture to hourly changes in photon flux should be investigated in future studies.
Utah State University is developing an extensive knowledge base in photobioreactor and raceway design and operational optimization, aimed at developing high-yield reactors. The work presented here represents advancements in photobioreactor design, algae productivity assessment, and biomass production systems modeling. This effort in particular delves into a unique photobioreactor development, as well as integrating optical systems and biomass production rates into a photobioreactor design.

A prototype spatially-diluted photobioreactor has been designed, built, and preliminary tests have been performed. This sdPBR represents advancements in photobioreactor design as well as lighting technology. The prototype sdPBR provided invaluable operational knowledge as well as preliminary biomass production kinetics on sunlight. However, the prototype was not an optimized design, which is why a parallel study was performed in the laboratory to investigate the variable parameters of this design. An overall optical efficiency of 69% was achieved with the prototype optical system.

A set of laboratory reactors was designed to simulate conditions inside the sdPBR; most importantly they use a light spectrum very comparable to sunlight. The lights used for the laboratory reactors have a substantially better spectrum than typical lights used in laboratory photobioreactors, making the kinetics data obtained with these reactors more realistic. The bench-top reactors were used to investigate light intensity, light path length, diurnal cycle, and volumetric dilution fraction. A simple yield model was developed, which can be used for estimates of biomass on the quantity of photons delivered per day. The linear model fit to the data produced a biomass yield on photons term of $Y_{x/p} = 1.09 \text{ gm mass/mol photons}$. A second-order model was also developed, which fits the data better at the upper range of TLD values. The maximum volumetric productivity obtained from the solar-simulating reactors was $0.95 \text{ gm L}^{-1} \text{ day}^{-1}$. In the
literature, the maximum volumetric productivity value found for *Neochloris* sp. in a photobioreactor is 0.63 gm L⁻¹ day⁻¹ [46], and ranges down to 0.018 gm L⁻¹ day⁻¹ [56]. An sdPBR system performance model was developed that incorporates the optical design attributes of the reactor with the experimentally-determined parameters. The yield models determined by the laboratory experiments were combined with the optics in the system model and TMY data, so that aperture and volumetric yields can be investigated. The estimated maximum aperture productivity of the sdPBR is 14.6 kg m⁻² yr⁻¹. This represents an approximate full-spectrum efficiency of 3%.

The design concepts and experimental techniques developed here can be applied to future generations of sdPBR reactor designs, as well as to further studies of algae kinetics. The use of lamps with near-sunlight spectra is quite unique in the laboratory, and it is hoped that the work here will demonstrate how to effectively use this type of laboratory photobioreactor to obtain practical kinetics data. The development of thin planar waveguides for delivering light inside a photobioreactor has helped reduce the number of optical components in, the volume of, and the footprint of an sdPBR.

It is suggested that future studies investigate even thinner reactors, in order to increase the volumetric productivity further. The 20 mm reactors were more prone to biofouling than the thicker reactors, so it is expected that even thinner reactors will be more challenging to operate. It is expected that there will be an economic tradeoff between reactor volume and volumetric productivity, which would be a necessary study before implementing this reactor on a large scale.
REFERENCES


APPENDICES
A.1. Culture Density

Two techniques were used for determining the culture density: optical density (OD) and total suspended solids (TSS). The TSS test is a modified version of the Gravimetric Method 2540 D [57]. For this study, dry weight (DW) is the preferred terminology used in place of TSS.

A Shimadzu UV-1800 spectrophotometer with a 1 nm resolution was used to measure the absorbance across a 1 cm path-length plastic disposable cuvettes, and the solution was diluted so that absorbance < 0.5 for a reading.

The density of the culture in reactors was measured every 24 hours, when either a sample reading was taken or part of the culture was harvested and replenished with fresh media. Growth was measured as optical density at 750 nm (OD$_{750}$). A periodic verification was made against a DW measurement. The wavelength was chosen based on correlating DW to OD at all wavelengths between 300 and 900 nm, and then scanning the results for the highest statistical R-squared correlation. This was conducted for a batch-growth test from the inoculation of a reactor through the end of the exponential growth phase. The correlation of the relationship between OD and DW for one growth data set is shown in Fig. A.1. At 750 nm, $R^2 = 0.9998$.

Once the wavelength value of 750 nm was chosen, the DW and OD$_{750}$ data from multiple reactors was compared. The results, shown graphically in Fig. A.2, confirmed that 750 nm is an optimal wavelength for comparing OD to DW. It gives an R-squared value of 0.987 for the entire data set, and the relationship between DW and OD$_{750}$ is $DW = (0.183 \text{ gm/L})(OD_{750}) + 0.0402 \text{ gm/L}$. 
Figure A.1. Strength of the correlation between DW and OD between the wavelengths 300 – 900 nm

Figure A.2. Comparison between DW and OD$_{750}$ for *N. oleoabundans* from several different reactors and growth curves
A.2. Algal Energy Content

Periodic samples of algae were collected from the reactors for an analysis of energy or lipids content. After centrifugation, the sample was placed in a -80°C freezer overnight, and then freeze-dried on a lyophilizer. A bomb calorimeter (Parr 1241, Moline, IL, USA) was used to measure the energy content of lyophilized algae samples. The energy content of a dried *N. oleoabundans* sample, obtained during steady-state operation in the prototype sdPBR, was determined to be 21.1 KJ/gm.


In order to find the relationship between spectral transmission versus depth, light transmission was measured with several concentrations of *N. oleoabundans* (up to 1.76 gm/L) over several different path-lengths. An Ocean Optics JAZ spectrophotometer, which has a 1 nm resolution and is capable of scanning over the 250-850 nm range, was used to measure the percent transmission. To vary the path-length, a series of Starna quartz cuvettes were used, with path-lengths of 1, 2, 5, 10, 20, 40, and 50 mm (Fig. A.3). An Ocean Optics deuterium-tungsten halogen light source was used to deliver light through fiber optics to an adjustable cuvette holder, where a collection lens collimates the light before passing it through the cuvette for the transmission measurement. Spectrasuite software was used to acquire the data. The combination of culture dilutions and path-lengths is shown in Fig. A.4.

Once the transmission data is obtained, MathCAD was used to fit an exponential curve to the transmission data for each concentration. If spectral transmission was required, an equation could be derived for each wavelength.

A general equation for the average transmission of 400 -700 nm light through a culture of *Neochloris oleoabundans* was derived and has the form

\[ T(z, X) = e^{\left(-\frac{1.44_{16} m^2}{gm}\right)z \cdot X} \]

Equation 27
where $z$ is the depth in mm, and $X$ is the culture density in gm/L.

Figure A.3. Schematic of dilutions and cuvettes used to measure spectral light attenuation vs. path-length
A.4. Sequential-Batch Operation

While comparing the relative performance between the different reactor widths and investigating the effects of daily volumetric dilution fraction versus productivity, the reactors were operated in a sequential-batch mode. In sequential-batch mode, a fraction of the reactor volume is harvested every 24 hours and replaced by fresh media [40, 58].

With a variable energy source (e.g. outdoor sunlight), it may be more efficient and productive to select a culture density to dilute down to at the time of the harvest, such that the culture begins each 24 hour period at the same density. The volume to harvest can be calculated from the equation:

\[ V_H = \frac{(X_2 - X_1)V_R}{X_2} \]  

Equation 28
APPENDIX B
UNCERTAINTY ANALYSIS

B.1. General Uncertainty Analysis

The method for the general uncertainty analysis is thoroughly discussed in [59]. Some of the specific equations used are presented here. The reader is referenced to [59] for a thorough treatment of the topic.

In general, the uncertainty of a data reduction equation is desired. The data reduction equation, \( r \), is a function of \( J \) variables \( X_i \): \( r = r(X_1, X_2, \ldots, X_j) \). The general uncertainty equation is

\[
U_r = \left[ \sum_{i=1}^{J} \left( \frac{\partial r}{\partial X_i} \right)^2 (U_{X_i})^2 \right]^{1/2}
\]

Equation 29

The overall uncertainty in a measurement, \( U_x \), is the root-sum-square of the precision \( (P) \) and bias \( (B) \) error estimates.

\[
U_x = [B^2 + P_X^2]^{1/2}
\]

Equation 30

The 95% confidence interval is used throughout the study. Bias and precision error estimates were obtained from manufacturer’s specifications whenever possible, or estimated from experience with the instrument. Several instrument error estimates are presented in Table B.1.

B.2. Confidence Interval on \( P_{vol} \)

In determination of \( P_{vol} \) in the solar-simulating reactors, \( N = 3 \) consecutive data points were taken. The 95% confidence limit, \( P_X \), is calculated with the expressions [59]
Table B.1. Estimates of Bias and Precision Error for Various Instruments

<table>
<thead>
<tr>
<th>Description</th>
<th>Brand/Model</th>
<th>Measurement</th>
<th>Precision error</th>
<th>Bias error</th>
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<td>Pipette, 1000 µL</td>
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<td>Volume</td>
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<td>0.058% of full scale</td>
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<td>Pipette, 200 µL</td>
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<td>Volume</td>
<td>0.1 µL</td>
<td>0.096% of full scale</td>
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<td>±0.002 Abs (0.5Abs)</td>
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<td>±0.1 mL</td>
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<td>Corning pyrex</td>
<td>Volume</td>
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<td>±2 mL</td>
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<td>0.0003 gm</td>
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<td>1 min</td>
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<tr>
<td>Aperture</td>
<td></td>
<td>Length</td>
<td>5%</td>
<td></td>
</tr>
</tbody>
</table>

\[
P_{X} = \frac{tS_{X}}{\sqrt{N}} \tag{Equation 31}
\]

\[
S_{X} = \left[ \frac{1}{N-1} \sum_{i=1}^{N} (X_{i} - \bar{X})^2 \right]^{0.5} \tag{Equation 32}
\]

where \( S \) is the sample standard deviation, \( X \) is a sample value, and \( N \) is the number of samples. \( t \) is from the t distribution. For \( N = 3 \), the value of \( t \) is 4.303. The uncertainty based on instrument error was observed to negligible compared to the variance due to the nature of the biological system.
APPENDIX C

PARAMETRIC STUDY DATA

The dry weight data from the parametric tests are presented here. The charts are presented in order of reactor width, light intensity, and diurnal cycle. All of the pertinent data is included on each graph, including the final averaged volumetric productivity obtained at steady state. Table C.1 lists all of the conditions used and references the figure numbers below.

<table>
<thead>
<tr>
<th>Condition Number</th>
<th>Reactor Width [mm]</th>
<th>Light Intensity [µmol m⁻² sec⁻¹]</th>
<th>Diurnal Cycle 15/9</th>
<th>Dilution Fraction</th>
<th>Figure Number</th>
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Figure C.1. 20 mm reactor, condition #1
Figure C.2. 20 mm reactor, condition #2

Figure C.3. 20 mm reactor, condition #3
Figure C.4. 20 mm reactor, condition #4

Figure C.5. 20 mm reactor, condition #5
Figure C.6. 20 mm reactor, condition #6

Figure C.7. 20 mm reactor, condition #7
Figure C.8. 20 mm reactor, condition #8

Figure C.9. 20 mm reactor, condition #9
Figure C.10. 20 mm reactor, condition #10

Figure C.11. 35 mm reactor, condition #11
Figure C.12. 35 mm reactor, condition #12

Figure C.13. 35 mm reactor, condition #13
Figure C.14. 35 mm reactor, condition #14

Figure C.15. 35 mm reactor, condition #15
Figure C.16. 35 mm reactor, condition #16

Figure C.17. 35 mm reactor, condition #17
Figure C.18. 35 mm reactor, condition #18

Figure C.19. 35 mm reactor, condition #19
Figure C.20. 35 mm reactor, condition #20

Figure C.21. 50 mm reactor, condition #21
Figure C.22. 50 mm reactor, condition #22

Figure C.23. 50 mm reactor, condition #23
Figure C.24. 50 mm reactor, condition #24

Figure C.25. 50 mm reactor, condition #25
Figure C.26. 50 mm reactor, condition #26

Figure C.27. 50 mm reactor, condition #27
Figure C.28. 50 mm reactor, condition #28

Figure C.29. 50 mm reactor, condition #29
Figure C.30. 50 mm reactor, condition #30

Figure C.31. 50 mm reactor, condition #31
Figure C.32. 50 mm reactor, condition #32

Figure C.33. 50 mm reactor, condition #33
Figure C.34. 50 mm reactor, condition #34

Figure C.35. 50 mm reactor, condition #35
Figure C.36. 50 mm reactor, condition #36

Figure C.37. 50 mm reactor, condition #37
Figure C.38. 50 mm reactor, condition #38 (replicate #1)

Figure C.39. 50 mm reactor, condition #38 (replicate #2)
Figure C.40. 50 mm reactor, condition #38 (replicate #3)

Figure C.41. 50 mm reactor, condition #39
Figure C.42. 50 mm reactor, condition #40

Figure C.43. 50 mm reactor, condition #41
Figure C.44. 50 mm reactor, condition #42

Figure C.45. 50 mm reactor, condition #43
Figure C.46. 50 mm reactor, condition #44

Figure C.47. 50 mm reactor, condition #45
Figure C.48. 50 mm reactor, condition #46

Figure C.49. 50 mm reactor, condition #47
Figure C.50. 50 mm reactor, condition #48

Figure C.51. 50 mm reactor, condition #49
Figure C.52. 50 mm reactor, condition #50

- N. oleoabundans
- 50 mm reactor
- SE+ Media
- 175 μmol m⁻² sec⁻¹
- 0.9 dilution fraction
- 15/9 diurnal cycle
- Pvol = 0.22 gm L⁻¹ day⁻¹
EDUCATION

Ph.D. Degree, Biological Engineering  
Utah State University, Logan, UT  
August 2010  
4.0 GPA

M.S. Degree, Mechanical Engineering  
University of Nevada, Reno, NV  
May 2003  
3.9 GPA

B.S. Degree, Mechanical Engineering  
Saint Martin’s University, Olympia, WA  
May 2001  
3.7 GPA

- Passed FE Exam, June 2001

BIOFUELS RESEARCH EXPERIENCE

Graduate Research Assistant  
Biological Engineering Dept., USU  
Logan, UT  
Jan. 2007 – Aug. 2010

- Project lead on the Advanced Photobioreactor Development and Testing project, sponsored by the Defense Advanced Research Projects Agency
- Developed unique solar and laboratory photobioreactors to evaluate algal growth kinetics and productivity rates
- Coordinated with engineers, biochemists, and microbiologists in a research environment to develop intellectual property and peer-reviewed papers on algal-based biofuels and products
- Directly trained and supervised undergraduate students in the lab
- Developed chemical hygiene plans, including safe storage and disposal, and inventoried and managed equipment in the Biofuels Center
- Presented research results at professional conferences and meetings

SOLAR ENERGY RESEARCH EXPERIENCE

Graduate Research Assistant  
Mechanical Engineering Dept., USU  
Logan, UT  

- Performed optical modeling with TracePro; completed various analyses on solar concentrators and light distribution systems
- Installed, calibrated, and maintained an array of solar tracking and concentrating systems
Research Engineer
June 2003 – Aug. 2005
Mechanical Engineering Dept., University of Nevada, Reno, NV
• Directed research and served as Co-PI of a DOE-sponsored solar lighting research project
• Designed fiber optic and photovoltaic-testing experiments; published the results at a professional conference and in a peer-reviewed journal
• Coordinated student research projects, managed laboratory space, and designed/built websites

Graduate Research Assistant
June 2001 – May 2003
Mechanical Engineering Dept., University of Nevada, Reno, NV
• Designed and built a unique infrared optical system through extensive optical modeling with TracePro
• Contributed to research on solar light concentrating and hybrid solar lighting
• Presented research at several professional conferences; published articles in peer-reviewed conference proceedings and a journal

TEACHING EXPERIENCE
Volunteer Lecturer/Tutor, Utah State University, 2008 - 2010
• Provided special lectures for undergraduate and graduate students in Biochemical Engineering, Heat and Mass Transfer, and Bioinstrumentation
• Developed and delivered introductory seminars to MathCAD
• Supervised teaching assistant in Bioinstrumentation
• Organized study sessions and provided one-on-one tutoring assistance

INTERNSHIP & CONSULTING EXPERIENCE
Undergraduate Research Assistant
Summer 2000
Pacific Northwest National Laboratory (PNNL), Richland, Washington
• Tested techniques for forming special glass mixtures for the Hanford waste vitrification project
• Cultivated laboratory and team-working skills in a professional environment

Independent Contractor / Consultant
Spring 2004
Sustainable Energy Solutions, LLC, Reno, NV
• Evaluated energy consumption of various pumping systems in a gold ore refining facility
• Proposed alternative system designs to reduce energy demand and cost
SELECTED ENGINEERING PROJECTS

- Served as team leader for senior design project to design and build an instrumented bench-top air-conditioning apparatus for experimentation and teaching
- Designed instrumentation systems using National Instruments and Campbell Scientific hardware
- Developed a lab project using a special “photographic” bacteria

COMPUTER SKILLS

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AWARDS AND ACHIEVEMENTS

Nominated by the College of Engineering for the Utah State University Robin’s Award for Graduate Research Assistant of the Year, 2010
Session Chair for the Biofuels and Bioprocesses Session, Institute of Biological Engineering Regional Conference, 2008 and 2009
Recipient of the 1st Place Poster Award, Institute of Biological Engineering Regional Conference, 2008
Graduate Student Representative for the Biological Engineering Department at USU, 2007/2008
Recipient of the 1st Place Oral Presentation Award, USU Graduate Student Symposium, 2007
Recipient of the Tomorrow Fellowship, Space Dynamics Lab, 2005
Recipient of the 1st Place Poster Award, American Solar Energy Society Conference, 2002
Recipient of the Mechanical Engineering Student of the Year award, Saint Martin’s University, 2001
Member of the Society of Fellows Invitational Honor Society, Saint Martin’s University, 2000/20001

PROFESSIONAL AFFILIATIONS

- Institute of Biological Engineers (IBE)
- American Society of Mechanical Engineers (ASME)
- American Solar Energy Society (ASES)
- National Society of Professional Engineers (NSPE)
PUBLICATIONS

Journals:


Conference Proceedings:


