

USING THE FLUID-IMAGING FLOWCAM® TO ANALYZE PHYTOPLANKTON
COMMUNITIES IN FLORIDA FRESHWATERS OF DIFFERENT TROPHIC STATUS

By

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To my family and friends for all their love and support

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LIST OF ABBREVIATIONS

ABD	Area based diameter (ABD) is an algorithm used by the FlowCAM® to calculate bio-volumes. ABD measures a diameter of a circle by arranging the pixels that comprised the imaged particle into a solid circle.
CHL	Chlorophyll (CHL) is a green pigment, present in all green plants and in nearly all algae, responsible for the absorption of light to provide energy for photosynthesis. Measurements of chlorophyll concentration are often used to estimate algal biomass in a water body and assess biological productivity.
ESD	Equivalent spherical diameter (ESD) is an algorithm used by the FlowCAM® to calculate bio-volumes. ESD measures the mean measurement of an object size along a specific direction conducted every 5° of the particle based on 36 sample measurements.
FlowCAM®	The Fluid Imaging Technologies Inc. Flow Cytometer and Microscope (FlowCAM®) combines capabilities of flow cytometer, microscopy, and fluorescence detection. It is an integrated system for rapidly analyzing particles in a moving fluid. The FlowCAM® automatically counts, images, and analyzes the particles or cells in a sample or a continuous flow.
TN	Total nitrogen (TN) consists of inorganic and organic forms. It is a measure of all forms of nitrogen found in a water sample. Nitrogen stimulates growth of aquatic plants and algae in water bodies.
TP	Total phosphorus (TP) is a measure of all forms of phosphorus found in a water sample. Phosphorus is an element that, in its different forms, stimulates the growth of aquatic plants and algae in water bodies.

Abstract of Thesis Presented to the Graduate School
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USING THE FLUID-IMAGING FLOWCAM® TO ANALYZE PHYTOPLANKTON
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This study examined the ability of the FlowCAM® relative to the inverted microscope to distinguish different algal community metrics (taxonomic ID, cell counts, and bio-volumes) in Florida freshwaters of different trophic status. A total of 32 algal samples preserved with Lugol's solution were analyzed to assess phytoplankton community composition and abundance. Samples (N=23) were obtained from two lakes, Lake Santa Fe and Lake Lochloosa in Alachua County and archived samples (N=9), counted by a professional phycologist, were obtained from the St. John's River. Lake samples (N=198) collected by Florida LAKEWATCH citizen scientists were analyzed with the FlowCAM®.

The FlowCAM® and microscope effectively identified (visual identification) phytoplankton communities at the genera level. Comparatively, FlowCAM® was faster for obtaining phytoplankton metrics. Speed of the FlowCAM® was especially evident when analyzing citizen scientists' samples. FlowCAM® bio-volume and cell count estimates for higher taxonomic algal levels (above class) were not significantly different ($p < 0.05$) from microscope estimates. Estimates for the lowest taxonomic level, genera,

were not significantly different when comparing results from the microscope and FlowCAM® among stations at each lake, but occasional differences were detected for individual samples. The FlowCAM® model used in this study had limitations imposed by the optics used, clearly indicating a microscope would be needed if species identification is required. In addition to providing information on algal communities, the FlowCAM® quickly calculated the volume of total seston using either volume area based diameter (ABD) or volume equivalent spherical diameter (ESD).

CHAPTER 1 INTRODUCTION

Measurement of chlorophyll (CHL) concentrations as an index of algal biomass has become routine in limnological and water quality studies (Hoyer et al. 2004; Carlson 2007; Bachmann et al. 2012) due to its ease of measurement, which translates to increased ability to analyze large numbers of water samples. Chlorophyll measurements have been used to classify the trophic status of lakes (Carlson 1977; Forsberg and Ryding 1980; Ryding and Rast 1989; Carlson 2007) and have been quantitatively linked to nutrients relationships like in-lake chlorophyll-TP concentrations (Dillon and Rigler 1974; Jones and Bachmann 1976; Brown et al. 1998; Hoyer et al. 2002), water clarity (Canfield and Hodgson 1983), zooplankton (Canfield and Watkins 1984), fish (Bachmann et al. 1996), aquatic bird abundance (Hoyer and Canfield 1994) and water quality as defined by lake users (Hoyer et al. 2004). Kratzer and Brezonik (1981) used chlorophyll measurements to establish the Florida Trophic State Index and Florida LAKEWATCH established a long-term chlorophyll/nutrient database (up to 24 years) that has been used to identify trends in water quality in hundreds of Florida lakes (Biggam 2012). Given all the scientific information developed for this important environmental variable, the United States Environmental Protection Agency and the Florida Department of Environmental Protection now use chlorophyll concentration as part of the numeric nutrient criteria (NNC) established for Florida's freshwaters in 2013 (Rule 62-302 and Rule 62-303, Florida Administrative Code).

Prior to the chlorophyll-phosphorus relationship established in the 1970s, scientific studies of algal communities primarily used microscopes. Visual identification of phytoplankton was necessary because phytoplankton species were used as

indicators of water quality (Rawson 1956). Biological methods for assessing specific water quality problems developed over time, but relationships between water quality and different types of algae remained largely narrative (APHA 1998). Duarte et al. (1992), however, established an empirical quantitative relationship demonstrating the percent contribution of algal genera to the phytoplankton community in Florida lakes changed with lake productivity; green algae dominate oligotrophic waters, diatoms mesotrophic waters, and cyanobacteria eutrophic to hypereutrophic waters. Microscopic counts of the algal communities in Florida lakes were also used to explain the observation that only a small percentage of algal communities in Florida lakes were at their maximal achievable density (Agustí et al. 1990).

Algal populations close to their maximal densities had algal biomass values > 10 mg/L and chlorophyll concentrations > 10 μ g/L. It was suggested that as non-nutrient constraints became more important, algal communities shifted from small-celled, diatom-green algal communities to communities dominated by large blue-greens algae. Based on 308 samples from 165 Florida lakes, Duarte et al. (1992) and Agustí et al. (1990) noted the development of the quantitative empirical algal relationships were limited by the time and expense of counting algal communities with a microscope, thus future advancements using large numbers of lakes would be difficult to achieve. Limnologists have long recognized that the time it takes to analyze microorganisms in a sample with a microscope does not enhance an understanding of algal relationships or permit the establishment of long-term records (Needham and Lloyd 1915).

Microscopic counts of the phytoplankton community, however, are needed because this permits a determination of species richness and diversity in phytoplankton

populations. Understanding the spectrum of phytoplankton interactions under different edaphic (i.e., geology and soils) conditions and during different seasons (Rundquist et al. 1996) as well as determining how different types of algae influence chlorophyll per unit biomass ratios are important for understanding the limnology of lakes (Canfield et al. 1985). Therefore, Fluid-imaging Technology was developed at the Bigelow Laboratory in 1999 with intent to fill the gaps in phytoplankton research that exists due to the limitations imposed by use of microscopes.

This tool, the FlowCAM®, originated to allow oceanographers to count large numbers of algal samples in real-time and thus reduce the need for tedious and expensive microscopic counts (Nelson et al. 2014). The usability of the FlowCAM® was quickly established for marine water (e.g., Sierack et al. 1998; Lavrentyey et al. 2004; Vaillancourt et al. 2004; See et al. 2005; Buskey and Hyatt 2006), but use in phytoplankton-rich freshwaters, like those in Florida, has not been widely tested.

Determining relative abundance of different algal groups in a phytoplankton community is important in lake management because community composition is a reflection of changing trophic status and nutrient levels. Determining the algal type and biomass of algae is useful in not only assessing a system's biological productivity, but in determining how users assess the quality of different water bodies.

My primary research objective evaluated the ability of the FlowCAM®, compared to the traditional inverted microscope, to distinguish different algal communities preserved with Lugol's in Florida freshwaters of different trophic status. I was particularly interested in the amount of time it would take to process the Lugol's samples and determine if the FlowCAM® provided results similar those obtained by use of an

inverted microscope; I specifically compared estimates of microscopic algal cell counts and biomass at different taxonomic levels to estimates obtained by using the FlowCAM®. I also recruited volunteers from the Florida LAKEWATCH program to collect bimonthly surface water samples from their lakes and preserve samples for phytoplankton analysis with Lugol's solution. These samples were then analyzed to determine additional relationships including total algal bio-volume to the dominant phylum in Florida lakes of different trophic states.

CHAPTER 2 METHODS

Water and Phytoplankton Sampling

Phytoplankton communities in Florida lakes differing in trophic status were assessed. Five lakes in Alachua County, Florida were sampled bimonthly with trophic status determined by Florida LAKEWATCH using average chlorophyll concentrations (Forsberg and Ryding 1980): Lakes Alto, Santa Fe and Little Santa Fe (mesotrophic), Lochloosa and Newnan (hypereutrophic). At each lake, three open-water sites were sampled for TP ($\mu\text{g/L}$), TN ($\mu\text{g/L}$) and chlorophyll *a* ($\mu\text{g/L}$) at 0.5 m following Florida LAKEWATCH sampling protocol (Canfield et al. 2002). Additionally, Secchi disk depth was measured at each site and phytoplankton samples were collected at 0.5 m in 120-mL amber glass bottles. Algal samples were preserved with roughly 0.5 mL of Lugol's solution to 100 mL of water. Water chemistry samples were placed on ice until return to the University of Florida's Fisheries and Aquatic Sciences' water chemistry laboratory. All water samples were stored frozen until water chemistry analyses were completed. Algal samples were placed in a dark container until return to the water chemistry laboratory and stored in the container at room temperature until processed.

To expand the number of lakes sampled and to encompass a wider range of lake trophic status, citizen scientists from Florida LAKEWATCH collected water chemistry samples following Florida LAKEWATCH protocol (Canfield et al. 2002) and water samples for phytoplankton. Six citizen scientists were trained by Florida LAKEWATCH to collect and preserve algal samples, which included preservation with Lugol's solution. Water samples from the five lakes in Alachua County and seven lakes throughout Florida (one citizen scientist sampled two lakes) were collected twice monthly at 0.5 m

at three stations in each lake for one year to the time sampling started for each citizen scientist. The lakes included three oligotrophic lakes (Lake Ola in Orange County, Lake Jem in Lake County, and Lake Bear in Seminole County), three eutrophic lakes (Lake Bay and Lake Florence in Lake County and Lake Talquin in Gadsen County), and a hypereutrophic lake (Lake Boca Cove in Polk County). Water chemistry samples were frozen prior to shipment to University of Florida's Fisheries and Aquatic Sciences' water chemistry laboratory and preserved algal samples were placed into a dark container prior to and during shipment.

Sample Analyses

Water chemistry

Water chemistry samples from all twelve lakes were stored frozen until processed using Florida LAKEWATCH protocols (Canfield et al. 2002). TP concentrations ($\mu\text{g/L}$) were determined using procedures of Murphy and Riley (1962) following persulfate digestion (Menzel and Corwin 1965). TN concentrations ($\mu\text{g/L}$) were determined by oxidizing water samples with persulfate and measuring nitrate-nitrogen with second derivative spectroscopy (D'Elia et al. 1997; Simal et al. 1985; Wollin 1987). Chlorophyll *a* concentrations ($\mu\text{g/L}$) were determined spectrophotometrically following pigment extraction with hot 90% ethanol (Method 10200 H, APHA 1992; Sartory and Grobbelarr 1984). Canfield et al. (2002) and Hoyer et al. (2012) have shown Florida LAKEWATCH laboratory and field protocols provide credible data comparable to data from other water quality laboratories and satisfactory for use in research and assessment evaluations (Hoyer et al. 2012, 2013).

The inverted microscope

Phytoplankton samples preserved with Lugol's were stored at room temperature and in the dark until processed. Prior to processing samples, I had to learn how to count and identify individual alga (based solely on visual observation), measure algal sizes, and calculate bio-volumes using an inverted microscope. I was trained by a University of Florida phycologist (Mary Cichra) with more than 26 years of experience in counting Florida algae. Counting procedures followed standard methods (APHA, 1985) and samples from two lakes, Lake Santa Fe (mesotrophic) and Lake Lochloosa (hypereutrophic), were used for training.

For inverted microscopic analyses, a Nikon phase-contrast inverted microscope was used. Individual preserved algal samples were dispensed into a 19-mm diameter, cylindrical-settling chamber and the phytoplankton cells were allowed to settle for 24 hrs. Phytoplankton cells were then counted at 400X and 100X and identified to species where possible (visual observation only). A minimum of 30 ocular μm grids was counted at 400X. If 100 cells were not counted within 30 grids, up to a maximum of 100 grids were counted or until 100 cells of a single taxa was reached. All large-celled taxa ($>30 \mu\text{m}$ maximal linear dimension) in the chamber were counted at 100X. Cell bio-volumes were then calculated from measurements of the geometric shape associated with an individual cell.

Recognizing different individuals counting algae can introduce variability into reported results, nine archived algal samples were obtained from a St. John's River study that had previously been counted by a University of Florida phycologist. To ensure consistency in counting, two of these samples were counted by myself and compared to the phycologist's results. All nine samples were subsequently counted

using the FlowCAM® and compared to the microscope counts and bio-volumes reported by the phycologist. Six of the samples were from Lake George in Putnam County, two samples were from Lake Crescent, in Putnam/Flagler counties and one sample was from St. John's River just North of Palatka. Unlike the surface algal samples collected by the citizen scientist and myself, these samples involved the use of an intergraded tube sampler to collect algae throughout the water column. The different phytoplankton collection methods, however, do not affect the comparative analyses between the inverted microscope and FlowCAM®.

The Fluid-Imaging FlowCAM®

Fluid Imaging Technologies loaned a Benchtop B3 Series FlowCAM® to test the ability of the FlowCAM® to provide algal taxonomic ID, cell counts, and bio-volumes. Information from the FlowCAM® was then compared to information obtained by use of an inverted microscope. Additional water chemistry information obtained through the efforts of the citizen scientists were analyzed to determine relationships between total algal bio-volume and the dominant algal phyla in Florida freshwaters of different trophic status. The Vice President of Fluid Imaging Technologies provided a two-day training session for the FlowCAM® to ensure proper and effective use of the machine. Due to the limited time with the FlowCAM®, 10 days, priority was placed on counting as many samples as possible that spanned different months of the year and lakes of varying trophic status. During those 10 days, 198 individual samples were processed. This included 120 algal samples from lakes Alto, Santa Fe, Little Santa Fe, Lochloosa, and Newnan sampled between May to August 2013. Lakes Ola and Bear had 48 algal samples from June through September 2013, and Lake Talquin had a total of 21 algal

samples from June to October 2013. Finally, the nine archived samples from the St. Johns River were processed.

The FlowCAM®'s procedures and operations followed the Fluid Imaging Technologies manual (Fluid Imaging Technologies FlowCAM® Manual 2011). The FlowCAM®, an imaging particle analysis system for the identification and classification of aquatic microorganisms and particles, combines aspects of microscopy and flow cytometer (Sieracki et al. 1998). It essentially represents an automated microscope for detailed morphological analysis of algal cells. A simplified block diagram is shown in Figure 2-1.

Sample fluid from the Lugol's preserved algal samples was pumped through a glass flow chamber. The fluid then passed through the field of view of the FlowCAM®'s camera. This caused an activation of a LED flash to provide back lighting and permitted the camera to capture an image of each particle or algal cell that passed through the field of view of a 10X objective microscope lens. The camera was triggered synchronously with the flash, essentially freezing the sample for the camera to acquire the image of the flowing sample. A collage of images was produced once the fluid sample had completely passed (Figure 2-2). The depth of the flow chamber set the upper size limit for the particles that could be analyzed and the lower size limit was determined by the FlowCAM®'s magnification.

Once each image from the field of focus was attained, the FlowCAM®'s Visual Spreadsheet software automatically stored the images within a collage. This was done in the AutoImage mode and the camera was set to capture images synchronously at a constant flow rate (0.4mL/min) regardless of the concentration of particles in the

sample. The Visual Spreadsheet is statistical-pattern recognition software and permits the operator to filter particles based on shapes, size range, and color intensity to name a few. Visual Spreadsheet also is used to automatically classify the different types of algae or other particles found in the samples that were photographed. The operator first creates a library of cell images of particular taxa. When the Visual Spreadsheet's pattern recognition is applied, the software's statistical-pattern recognition program separately analyzes each particle image to determine which library class is closest to the particle being analyzed. Each cell image had up to 26 different measurements that could indexed to a specific algal cell. The measurements were then be placed into morphological measurements (diameter, length, width, perimeter, circularity, etc.), gray-scale measurements (intensity, transparency, color information, etc.), and spectral measurements (peak area and width) from the signals collected in channels of fluorescence. By combining the morphological, gray-scale measurements, and the spectral measurements and the statistical pattern recognition software, the instrument could preliminarily (pending acceptance by the investigator) differentiate, discriminate and obtain an estimate of community structure in the environment.

Due to the Lugol's solution in the samples, gray-scale measurements and spectral measurements could not be effectively obtained. If these types of measurements are needed, the samples would need to be either live or preserved with formaldehyde; neither approach was suitable for this project. Classifying to the lower taxonomic level, however, was possible due to the FlowCAM[®]'s option 'Like Selected Particles'. This option applies statistical filters to the whole database based on one individual image for a specific genus, but by using the supplied 10X lens provided, the

size range of particles that could be counted was limited to those longer than 10 μm . Prior to running the samples, each sample was pre-filtered with a 100- μm mesh net to prevent clogging of the flow cell resulting in only particles $<100\ \mu\text{m}$ being counted during this study. After preliminary tests, a particle limit of 1,000 counts was set to determine when most individual sample-runs would end.

Particle concentration (particles/mL) was calculated using the FlowCAM®'s Visual Spreadsheet software and the AutoImage Mode. The volume of each field of view was determined by the height and width imaged, times the depth of the flow cell. Dividing the total number of particles imaged by the total volume of sample processed provided the particle concentration of the sample (Fluid Imaging Technologies 2013).

FlowCAM® software used two algorithms to calculate bio-volumes: area based diameter (ABD) and equivalent spherical diameter (ESD). ABD measured a diameter of a circle by arranging the pixels that comprised the imaged particle into a solid circle. ESD measured the mean measurement of an object size along a specific direction conducted every 5° of the particle based on 36 sample measurements (e.g., makes the shape of a 3-dimensional object on a 2-dimensional plane). The Visual Spreadsheet then generated an extensive (30) list of individual particle properties.

Statistical Analyses

JMP statistical package edition Pro 9.0. was used for all statistical analyses (SAS, 2000). Statements of statistical significance were for p-values ≤ 0.05 .

Prior to analyses of phytoplankton abundances, all data were logarithmically (base 10) transformed to accommodate heteroscedasticity (Sokal and Rohlf, 1981). Because there were zero values for some algal genera (visual identification) in the

resulting individual counts, a value of one was added to all individual counts prior to transformation.

All data were plotted to provide a visual interpretation of relationships. A Tukey-Kramer HSD was used to compare the replicated samples of the FlowCAM®. A paired t-test was completed to compare FlowCAM® community metrics with metrics obtained by use of the inverted microscope among lakes, dates and stations. Linear regression analysis was used to determine whether statistically significant linear relationships existed between the FlowCAM® bio-volumes (ABD) and measured in-lake chlorophyll *a* concentrations. Additionally, linear regression analyses were used to assess statistical significance in total algal bio-volume, detritus and dominating algal phyla relationships in Florida lakes of different trophic status and on different dates.

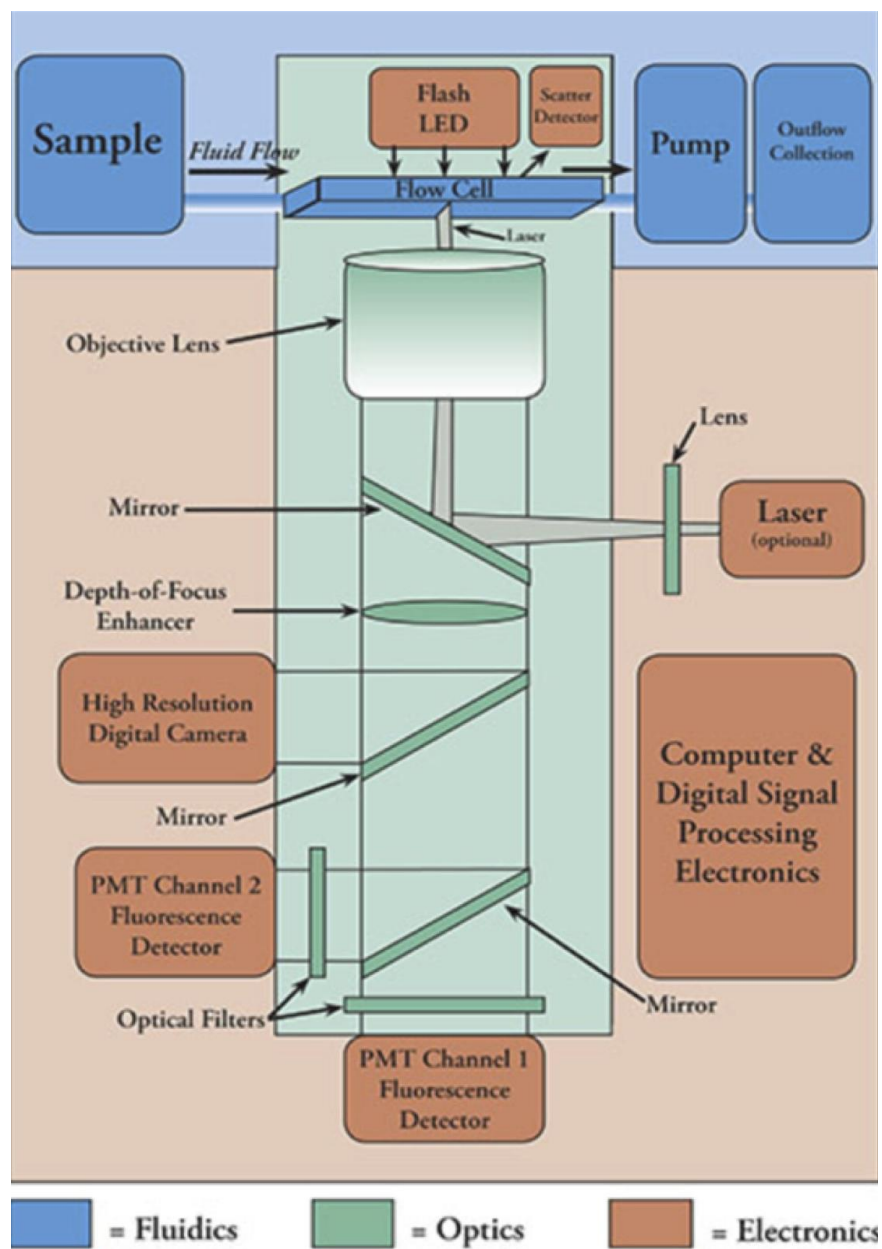


Figure 2-1. Simplified block diagram of FlowCAM® functions (Fluid Imaging Technologies, USA; www.fluidimaging.com).

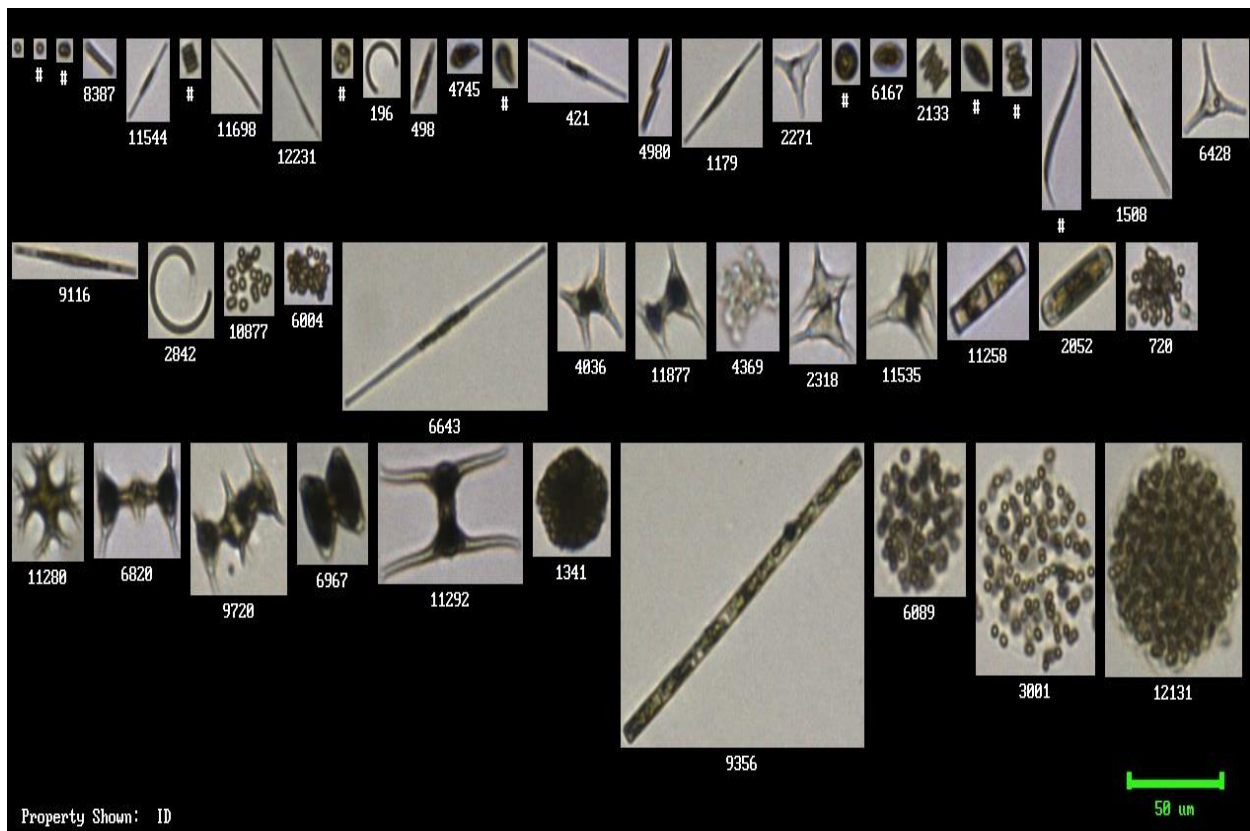


Figure 2-2. Images and particle ID numbers (algae and detritus) provided by FlowCAM® for a water sample collected on May 9, 2013 from Lake Lochloosa, Alachua County, Florida.

CHAPTER 3 RESULTS

Fluid-Imaging FlowCAM® Counts

The FlowCAM® is an imaging particle counter that provides visual and quantitative information on particles (algae and detritus) in a water sample (Figure 3-1). An operator of the FlowCAM® must determine how many particles in each sample should be counted prior to use. Setting a particle limit determines how much sample volume must be processed and how long it takes to complete the counts and classification of each sample.

Four lakes were selected to examine how many particles should be counted (100, 500, 1000 or 2000 particles) for the lakes included in the study. Each particle category limit had phytoplankton samples from three different stations per date (9 total samples per each particle count category). Samples were analyzed for Lake Lochloosa and Newnan Lake (May 9, 23 and June 13, 2013), two dates from Lake Santa Fe (May 9 and 23, 2013) and one date from Lake Ola (June 10, 2013). A one-way ANOVA followed by a Tukey-Kramer HSD test suggested, on each date of sampling, there were no statistical differences within any lake among the estimated total number of particles per mL for each particle count category (100, 500, 1000 or 2000 particles). The volume of the sample analyzed was dependent on trophic state and the date of sampling. The least volume analyzed was at Newnan Lake, approximately 0.02 mL for 100 particles, 0.14 mL for 500 count, 0.25 mL for 1,000 count and 0.57 mL for 2,000 counts. The time it took 2,000 particles to be imaged by the FlowCAM®, was approximately 1.5 min and roughly 1 min to have 1,000 particles imaged. The most sample volume analyzed for any sample was at Lake Ola, approximately 0.48 mL for 100 particles, 2.2 mL for 500

count, 5.4 mL for 1,000 count and 8.6 mL for the 2,000 count. The FlowCAM® imaged 2,000 particles in about 21 min and 13 min to image 1,000 particles. Once the operator became familiar with the algal genera encountered in the samples, classification of individual particle images required approximately 3-min for 100 particle counts, 5-min for 500 particle counts, 10-min for 1,000 particle counts and 20-min for 2000 particle counts for oligotrophic/mesotrophic waters. For productive waters, classification of 1000 counts took about 30-min and the 2,000 count took about 45-min because a greater number of alga genera were encountered in these waters.

The sample volume processed and the time needed for the FlowCAM® to process a sample was largest and longest at oligotrophic Lake Ola. However, the sample-volume processed could be reduced in half by pre-settling the phytoplankton sample (similar to the inverted microscopic samples) and thereby permitting the FlowCAM® to finish imaging the particles in less time.

For the Bench B3 Series FlowCAM® the optics did not allow the definitive identification of an alga to species, but did allow the taxonomic identification to genus for most algal via the Visual Spreadsheet output (Figure 3-1). The number of genera identified in any specific sample was dependent on the volume of water (Figure 3-2 and Figure 3-3). The greater the volume of water analyzed, the more genera encountered; likewise, the more particles counted, the greater number of genera encountered (Figure 3-4). In the case of Lake Lochloosa, the number of genera encountered continued to rise even when 12,000 particles (2 mL of sample) were classified (Figure 3-5). When using a microscope, a much greater volume of water (5 to 10 mL) is typically settled than the volume used by the FlowCAM®. For Lake Lochloosa and Lake Santa Fe,

microscope counts identified a greater number of genera than the FlowCAM® (Figure 3-2 and Figure 3-3).

Encountering rare genera is a challenge for users of both the FlowCAM® and the inverted microscope. The probability of encountering a rare alga is dependent on the volume of sample analyzed. To ensure the FlowCAM identified the greatest number of genera in each sample, a particle count of 1,000 was used, as approximately 95% of all genera were encountered with a count of 1,000 (Table 3-1)(Figure 3-5). Therefore, Florida LAKEWATCH algal samples were processed with a particle count limit of 1,000 because this provided the shortest processing time and allowed the capturing of the most abundant algal genera. It also provided the ability to identify detrital particles (Figure 3-1).

During the time period (10 days) that the FlowCAM® was available, 481 samples (including the replicated samples) were processed (exclusive of particle classification; see above for amount of time needed for particle classification). Once, the 1,000-particle limit was selected, the FlowCAM® was used for five days to process the 198 water samples from nine different water bodies (8 lakes from citizen scientist 1 from St. Johns River). For the lakes sampled by Florida LAKEWATCH volunteers and myself (12 lakes total), water samples collected were bimonthly from May 2013 to October 2014.

Microscope Counts

A total of 32 samples were analyzed with both the FlowCAM® and the inverted microscope. The samples came from two lakes (Lake Lochloosa and Lake Santa Fe) and were collected between the months of May to July 2013. Individual samples from Lake Lochloosa required approximately 1.5 hrs to completely count by use of the

microscope, whereas Lake Santa Fe samples took approximately 45 min to complete due to fewer algae.

Assessing the amount of processing time required for an individual to analyze an algal sample and determine if densities per mL or bio-volumes per mL for results from replicated samples differed; I first made three replicate counts on samples collected on two separate dates from Lake Lochloosa (six samples) and on a single date for Lake Santa Fe (three samples). Time for analysis was the same for each replicate, thus given my level of training, counting time did not decrease with the number of replicates. Analyzing the replicates from each lake and date using Tukey-Kramer HSD test at alpha 0.05, showed no significant difference in densities per mL or bio-volumes per mL for each lake and date (Lochloosa, May 9 and June 13, 2013 and Santa Fe May 23, 2013).

To determine difference between person-to-person reported microscope densities and bio-volumes, I recounted two samples from the St. John's River archived samples that were previously counted by the University of Florida phycologist. Paired t-tests between the bio-volumes ($p=0.764$) and density concentrations ($p=0.245$) reported by the phycologist and myself resulted in no significant differences. However, the phycologist identified more genera in each sample than I did in.

Fluid-Imaging FlowCAM® and Microscope Counts

To compare estimates of microscopic algal bio-volume and genera composition to estimates obtained by the FlowCAM®, I analyzed the same sample-volume (mL) using the inverted microscope and the FlowCAM® for three individual samples. Two samples, collected on different dates, were obtained from Lake Lochloosa (May 9 and June 13, 2013) and 1mL was analyzed with the FlowCAM® and the microscope. One sample was obtained from Lake Santa Fe (May 23, 2013) and 3mL were analyzed.

Paired t-tests demonstrated no significant difference between genera bio-volume and density estimates obtained by the FlowCAM® or the microscope for each lake and date (Table 3-2 and Table 3-3). However, more genera were identified using the inverted microscope (See previous discussion regarding sample volumes).

Continuing to compare the inverted microscope and the FlowCAM®, I examined the estimated algal density of all 32 microscopic algal samples (13 samples from Lake Lochloosa, 10 samples from Lake Santa Fe, and 9 samples from St. Johns River). Density was estimated as individual cell count per mL or colony count per mL. The paired t-test was used to compare the individual cell count per mL and colony count per mL in each sample from the microscope to the density estimates from the FlowCAM®. For the 13-Lake Lochloosa samples examined, individual cell counts per mL were significantly higher using the inverted microscope for 11 samples at the genera and families level, nine samples at the order level and three samples at class level. For Lake Santa Fe (10 samples analyzed) significant differences were detected in five samples at the genera and family level, four samples had higher microscopic counts at the order level and one sample at the class level. In the St. Johns River, (9 samples analyzed) significantly higher microscope counts at the genera (seven samples), family (six samples), order (five samples), and class (four samples) levels (Table 3-4) were obtained. When using colony counts per mL instead of individual cell counts per mL (Table 3-5), Lake Lochloosa had four samples that were significantly different at the genera and family level and two samples significantly different at the order level. The St. Johns River only had one sample that was significantly different at the genera level. However, microscope and FlowCAM® algal density (individual cells or colonies)

estimates were not significantly different for most samples at Class level and all samples at the Phylum level for all water bodies (Appendix A and B).

Algal biomass estimates (32 samples), based on the bio-volumes calculated with the microscope and the ABD and ESD FlowCAM® methods, were compared (as with the algal density estimates). ABD and ESD FlowCAM® bio-volumes were significantly different from each other. The bio-volumes obtained by use of the inverted microscope were consistently higher for the lower taxonomic levels. However, microscope estimates and FlowCAM® bio-volume estimates, regardless of whether ABD or ESD bio-volumes were used, were not significantly different (Paired t-test) at the Phylum taxonomic level (Table 3-6 and Table 3-7). The FlowCAM® however, provides another particle estimate, the aspect ratio, which provides information on whether the particle is circular or linear. Where linear shaped algal genera dominated (e.g., Lake Santa Fe) over circular shape genera, FlowCAM® ESD bio-volumes were comparable to the microscope bio-volumes (Table 3-8; Appendix C-E).

Florida Lakes

After the empirical tests were completed, five days were available to process bimonthly phytoplankton samples collected in eight Florida lakes from May to October 2013. A total of 198 lake samples were processed and algal ABD bio-volumes were compared by use of linear regression analyses to water quality estimates that were obtained from water samples that were simultaneously collected. There was a strong relationship ($R^2 = 0.88$) between total algal bio-volume and cyanobacteria bio-volume (Figure 3-6), but cyanobacteria bio-volume was not correlated with TP, TN or CHL. Blue-green algae also dominated over time in the individual lakes (Figure 3-7). For the lakes sampled, predicted chlorophyll a concentrations were calculated using the

chlorophyll-biomass regression equation developed by Canfield et al. (1985) and the FlowCAM® ABD bio-volume (converted to biomass). There was a significant positive correlation with measured chlorophyll concentrations, but the relationship was weak ($R^2 = 0.22$; Figure 3-8). Besides looking at algal community metrics, the FlowCAM® provided an opportunity to examine detritus bio-volumes in individual lakes over time (Figure 3-7). There, however, was not a correlation between total detritus and total algal bio-volume.

The FlowCAM® also provided the ability to look at correlations among each Phylum. Charophyta had the strongest correlation with Cyanobacteria ($R^2 = 0.71$) and the weakest correlation with Ochrophyta ($R^2 = 0.41$). However, Chlorophyta's strongest correlation was with the Ochrophyta and Euglenophyta ($R^2 = 0.5$) and weakest with Dinophyta ($R^2 = 0.27$).

Examining relationships with the measured water quality parameters demonstrated that the Phylum Ochrophyta had the strongest correlations with TP ($R^2 = 0.9$), TN ($R^2 = 0.77$) and chlorophyll ($R^2 = 0.86$). Weakest negative correlations were TP with the Charophyta ($R^2 = 0.4$). TN had a weak negative relationship with Dinophyta ($R^2 = 0.59$), and Chlorophyll was weakly negatively correlated with both Charophyta and Dinophyta ($R^2 = 0.4$).

Table 3-1. Number of genera found with different particle counts using the FlowCAM® for Lake Ola (Orange County, Florida), Lake Lochloosa and Lake Santa Fe (Alachua County, Florida). Total of 36 samples were analyzed with 9 samples per particle counts. The percent genera identified is the upper 95% confidence limit estimate divided by the maximum number of genera encountered.

Particle Counts	Maximum Number of Genera Found	Upper 95% Confidence Limit	Percent Genera Identified
100	11	9	83
500	20	15	76
1000	20	19	97
2000	24	22	93

Table 3-2. Paired t-test comparing estimates of ABD algal bio-volumes ($\mu\text{g}^3/\text{mL}$) from the FlowCAM® to algal bio-volumes estimates ($\mu\text{g}^3/\text{mL}$) from the inverted microscope. The same volume (mL) of sample was analyzed for three samples at the genera taxonomic level.

Lake	Date	Volume settled (mL)	N	p-Value
Lochloosa	May 9, 2013	1	18	0.4033
Lochloosa	June 13, 2013	1	20	0.1512
Santa Fe	May 23, 2013	3	10	0.6483

Table 3-3. Paired t-test comparing algal densities estimates (cell/mL) of the FlowCAM® to algal densities estimates (cell/mL) from the inverted microscope. The same volume (mL) of sample was analyzed for three samples at the genera taxonomic level.

Lake	Date	Volume settled (mL)	N	p-Value
Lochloosa	May 9, 2013	1	18	0.2716
Lochloosa	June 13, 2013	1	20	0.2916
Santa Fe	May 23, 2013	3	10	0.5562

Table 3-4. Paired t-test results for algal density using individual cell counts per mL for 32 microscopic and FlowCAM® algal samples at different taxonomic levels. Significant differences for individual comparisons within each sample site were identified at $p < 0.05$.

Taxa Level	Lochloosa	Santa Fe	St. Johns River
Genus	11 different 2 not	5 different 5 not	7 different 2 not
Family	11 different 2 not	5 different 5 not	6 different 3 not
Order	9 different 4 not	4 different 6 not	5 different 4 not
Class	3 different 10 not	1 different 9 not	4 different 5 not
Phylum	13 none	10 none	9 none

Table 3-5. Paired t-test results for algal density counting a colony as a single unit per mL for 32 microscopic and FlowCAM® algal samples at different taxonomic levels. Significant differences for individual comparisons within each sample site were identified at $p < 0.05$.

Taxa Level	Lochloosa	Santa Fe	St. Johns River
Genus	4 different 9 not	10 none	1 different 8 not
Family	4 different 9 not	10 none	9 none
Order	2 different 11 not	10 none	9 none
Class	13 none	10 none	9 none
Phylum	13 none	10 none	9 none

Table 3-6. Paired t-test results for algal bio-volumes of the 32 microscopic and FlowCAM® algal samples using FlowCAM®'s Area Based Diameter (ABD) at different taxonomic levels. Significant differences for individual comparisons within each sample site were identified at $p < 0.05$.

Taxa Level	Lochloosa	Santa Fe	St. Johns River
Genus	7 different 6 not	2 different 8 not	8 different 1 not
Family	7 different 6 not	2 different 8 not	8 different 1 not
Order	6 different 8 not	2 different 8 not	5 different 4 not
Class	2 different 11 not	1 different 9 not	6 different 3 not
Phylum	13 none	10 none	2 different 7 not

Table 3-7. Paired t-test results for algal bio-volumes of the 32 microscopic and FlowCAM® algal samples using FlowCAM®'s Equivalent Spherical Diameter (ESD) at different taxonomic levels. Significant differences for individual comparisons within each sample site were identified at $p < 0.05$.

Taxa Level	Lochloosa	Santa Fe	St. Johns River
Genus	4 different 9 not	10 none	4 different 5 not
Family	4 different 9 not	10 none	3 different 6 not
Order	3 different 10 not	10 none	3 different 6 not
Class	13 none	10 none	3 different 6 not
Phylum	13 none	10 none	1 different 8 not

Table 3-8. Paired t-test results for algal bio-volumes of the 32 microscopic and FlowCAM® algal samples using FlowCAM®'s aspect ratio to combine Area Based Diameter (ABD) and Equivalent Spherical Diameter (ESD) bio-volumes at different taxonomic levels. Significant differences for individual comparisons within each sample site were identified at $p < 0.05$.

Taxa Level	Lochloosa	Santa Fe	St. Johns River
Genus	6 different 7 not	10 none	5 different 4 not
Family	6 different 7 not	10 none	5 different 4 not
Order	3 different 10 not	10 none	3 different 6 not
Class	2 different 11 not	10 none	4 different 5 not
Phylum	13 none	10 none	1 different 8 not

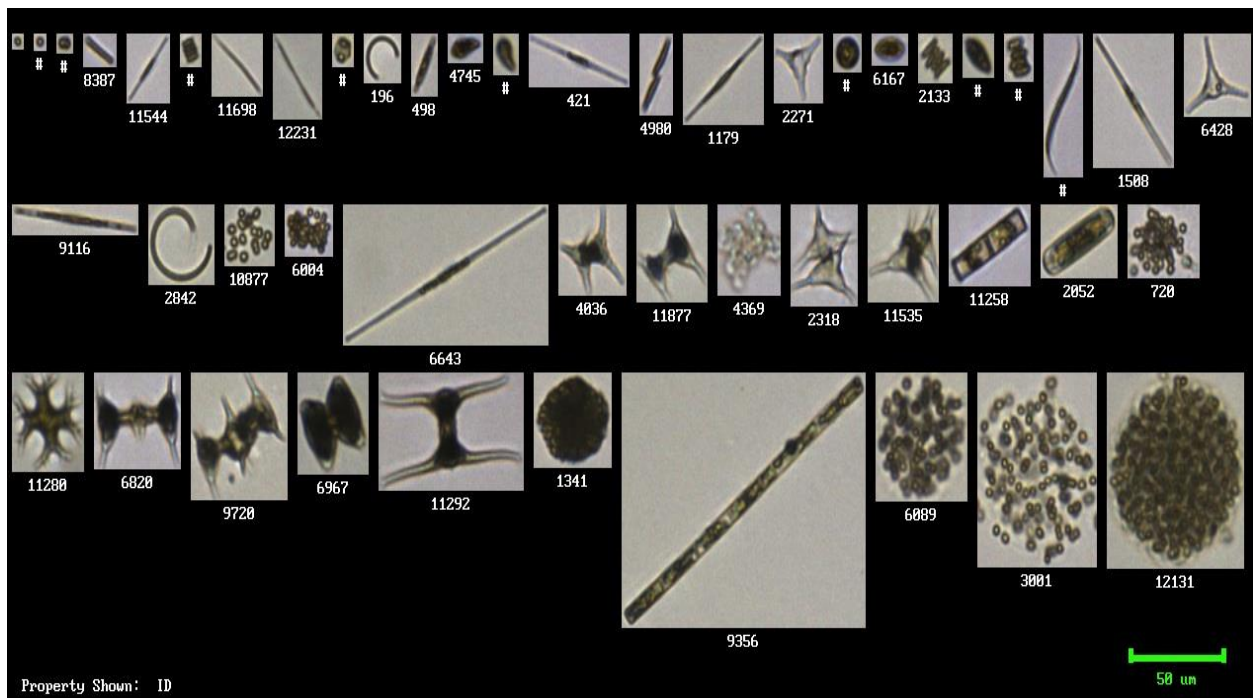


Figure 3-1. Images and particle numbers (algae and detritus) provided by FlowCAM® for a water sample collected on May 9, 2013 from Lake Lochloosa, Alachua County, Florida.

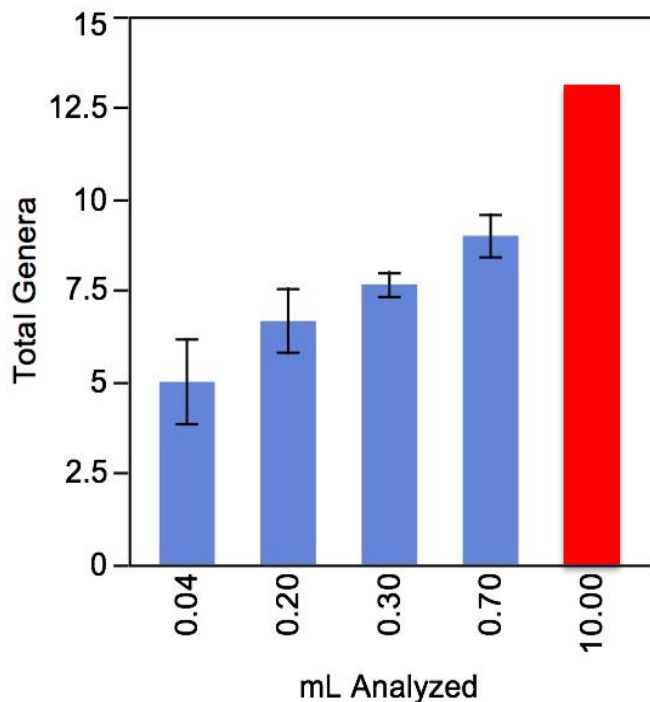


Figure 3-2. Volume of sample analyzed by the FlowCAM® (Blue) and inverted microscope (Red) and the total number of genera identified May 23, 2013, in Lake Santa Fe, Alachua County, Florida.

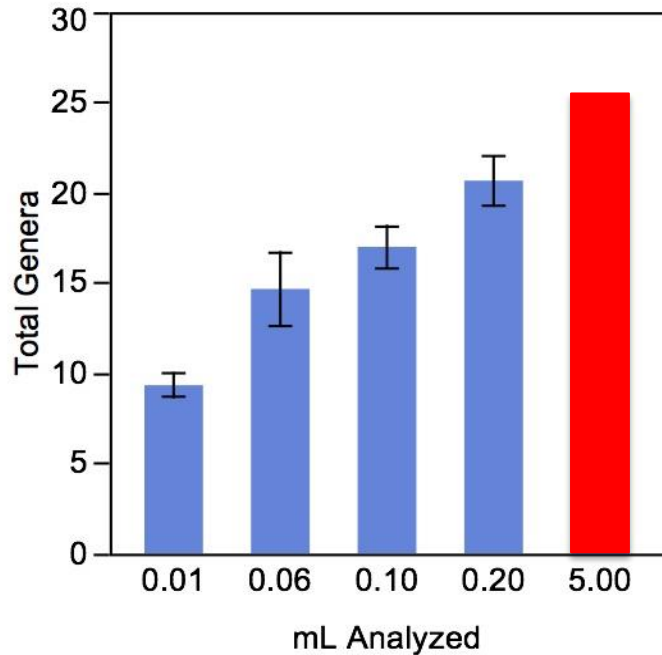


Figure 3-3. Volume of sample analyzed by the FlowCAM® (Blue) and inverted microscope (Red) and the total number of genera identified May 9, 2013, in Lake Lochloosa, Alachua County, Florida.

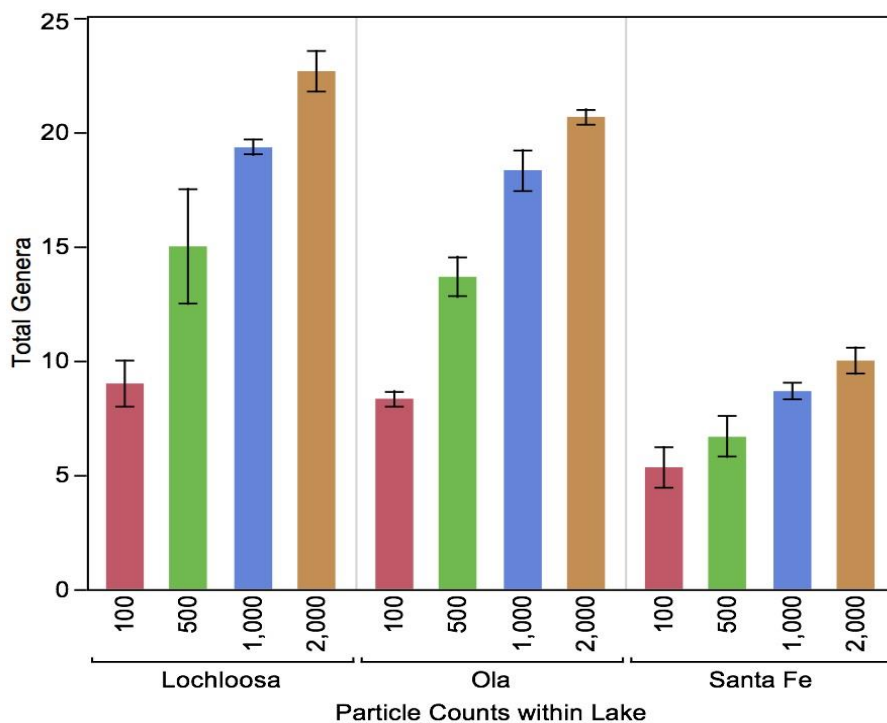


Figure 3-4. FlowCAM® replicate particle counts from 100 to 2,000 and number of genera for lakes of different trophic status, Lake Lochloosa (Hypereutrophic), May 9, 2013, Lake Ola (Oligotrophic), June 10, 2013, and Lake Santa Fe (Mesotrophic), May 23, 2013.

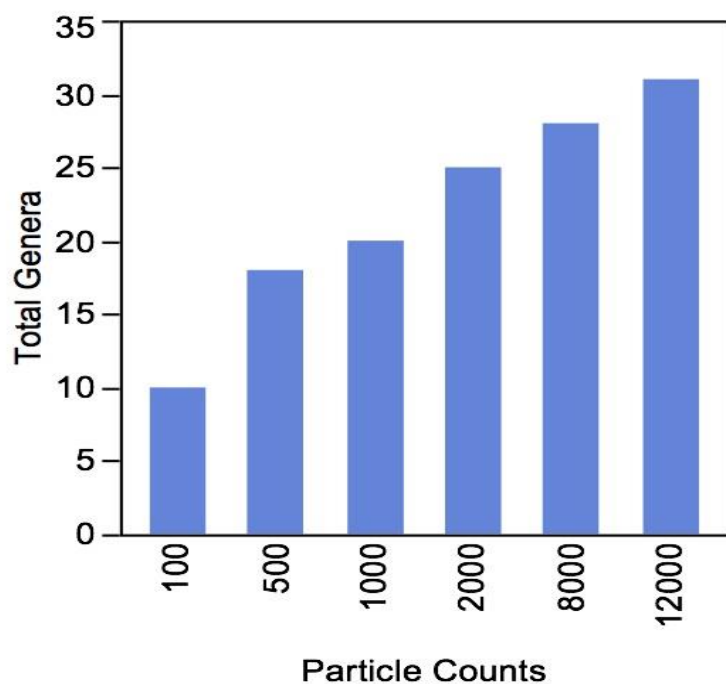


Figure 3-5. FlowCAM® replicate particle counts from 100 to 12,000 and number of genera, May 23, 2013, in Lake Lochloosa, Alachua County, Florida.

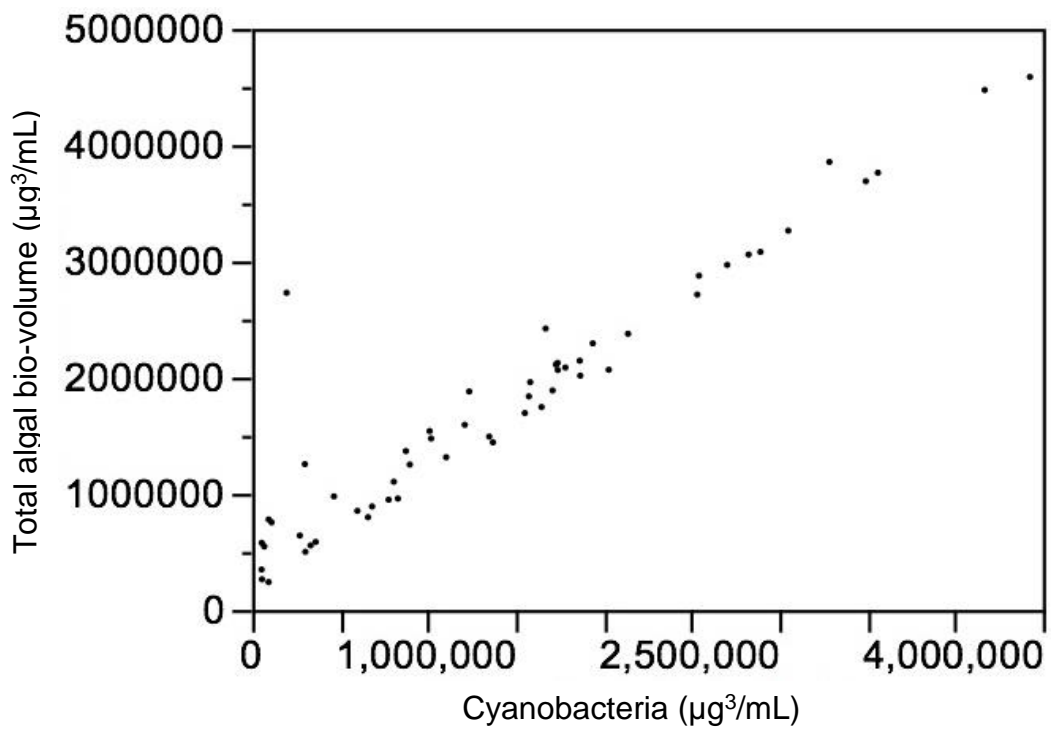


Figure 3-6. Relationship between total algal bio-volume and cyanobacteria bio-volume as measured by FlowCAM®'s Area Based Diameter (ABD) bio-volume estimates for 198 water samples from eight Florida lakes collected from May-October 2013.

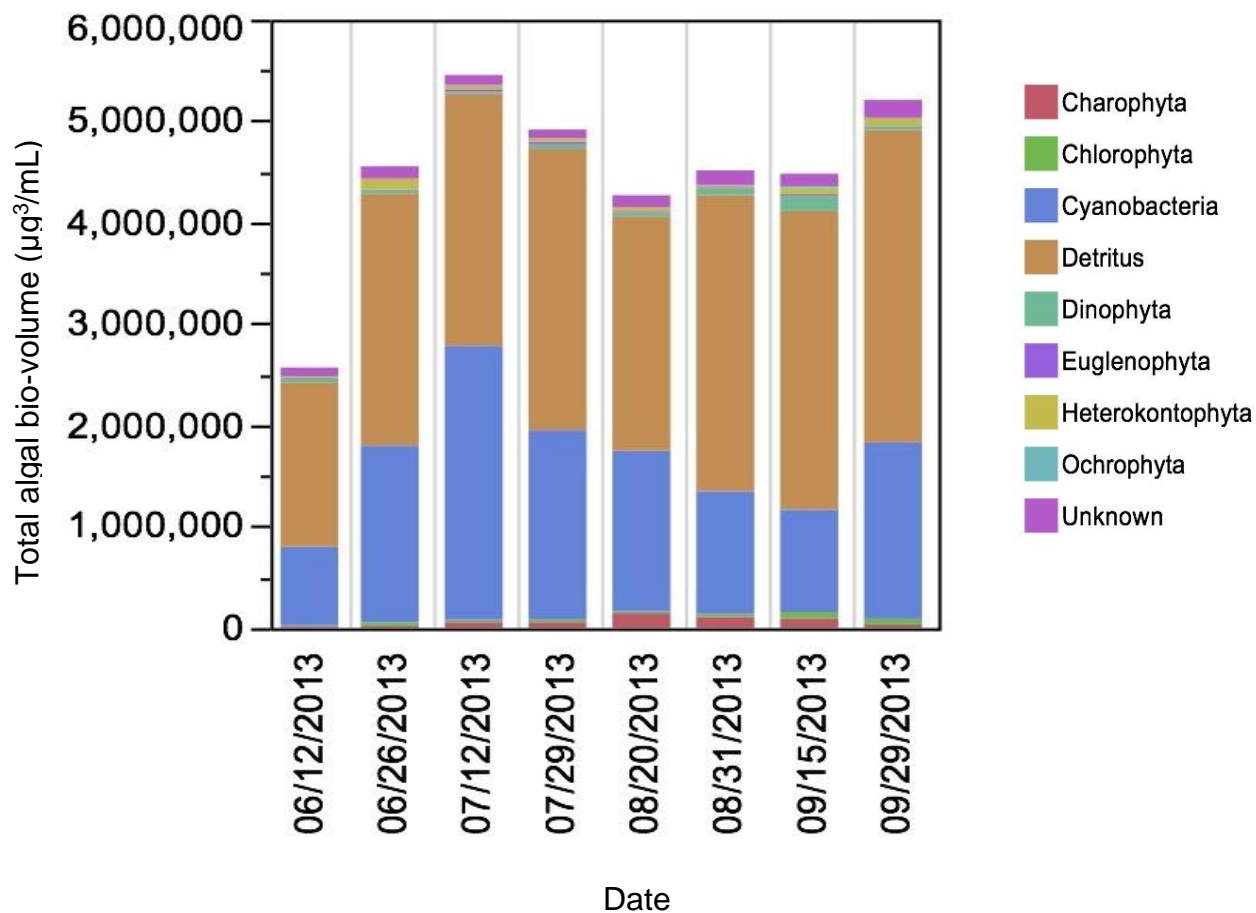


Figure 3-7. FlowCAM®'s phytoplankton Area Based Diameter (ABD) total bio-volumes ($\mu\text{g}^3/\text{mL}$) in Lake Bear, Seminole County, Florida, by category, June-September 2013.

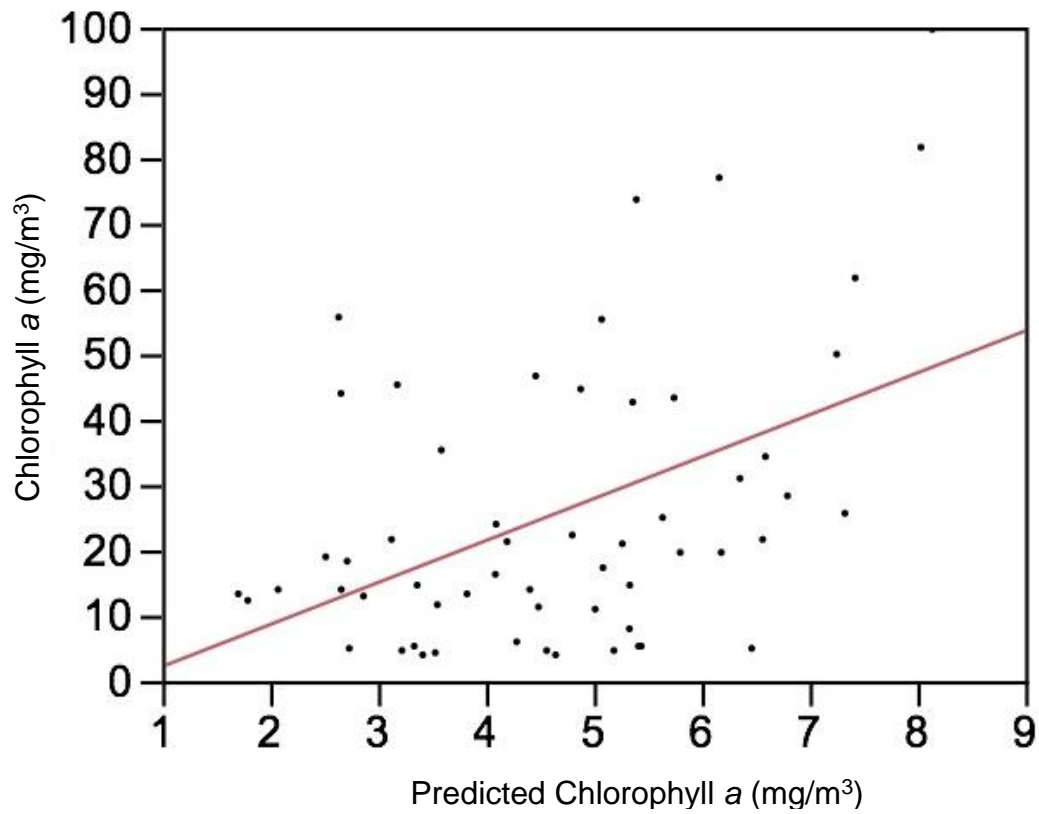


Figure 3-8. Predicted chlorophyll a (mg/m^3) concentrations compared to chlorophyll a measurements (mg/m^3) for 198 water samples from eight Florida lakes collected from May-October 2013. Predicted chlorophyll a was calculated from Canfield et al. (1985).

CHAPTER 4 DISCUSSION

Determining the number of samples needed to describe a biological community (e.g., the phytoplankton community of a lake) within specific statistical limits is critical. Wasted time and money can occur when either too many or too few samples are collected. Too many samples waste time and money, but too few samples can introduce errors in interpretations and make the sampling meaningless (Calhoun 1966). Consequently, too few samples are often analyzed when using a microscope to study phytoplankton communities. This is due to the microscopic work being time consuming, tedious, expensive, and the variation in measurements and counts can differ between observers (e.g., Canfield et al. 1985; Schmid et al. 1998; Carpentier et al. 1999; Buskey and Hyatt 2006; Alvarez et al. 2011). Given the suite of potential problems, lake managers and many regulatory groups like the United States Environmental Protection have chosen to rely on chlorophyll *a* measurements to help understand the algal trends because a large number of samples can be collected and samples can be processed quickly.

Duarte et al. (1990) demonstrated that phytoplankton microscope counting errors decreases when algal biomass increases in a sample. After many decades of using the microscope, standardization of quantitative phytoplankton analyses, however, are still evolving because of multiple types of counting errors (Brierley et al. 2007). For instance, when calculating algal bio-volumes with a microscope, many geometric shapes are used (Hillebrand et al. 1999) and choosing the wrong geometric shape strongly influences the calculated bio-volumes (Sun and Liu 2003). Li et al. (2014) stated a limitation of microscope counting, in analysis of large and irregular colonies, is that

internal cells are shaded by the peripheral cells potentially resulting in either an over estimate if the cell count was derived by multiplying the area or an underestimate. This too could occur with the FlowCAM® and must manually be accounted for.

Although chlorophyll is commonly used, it is acknowledged that the chlorophyll per unit biomass ratio varies with the type of algae present and the major factor influencing chlorophyll content in Florida phytoplankton is nitrogen (Canfield et al. 1985). Thus, phycologists and limnologists recognize that it is often desirable to understand the type and abundance of algae that occur over trophic status and seasons (Rundquist et al. 1996). The use of microscopes will remain as a standard tool for many years, but can be used in tandem with a faster method for analyzing phytoplankton communities to address emerging questions about algal community structure and ecology. Allen et al. (1994) stated, using the FlowCAM® in combination with microscope counts and measurements of the algal dimensions is the fastest and most accurate way to determine bio-volumes on a routine bases.

Use of advanced automated measuring systems have increased in the last few decades to overcome some drawbacks in using traditional inverted microscopic methods (Carpentier et al. 1999; Babin et al. 2005; Benfield et al. 2007). The FlowCAM® is a combination of the flow cytometer and microscope (Sieracki et al. 1998) and can address the need for processing large number of phytoplankton samples quickly as demonstrated in this study. The FlowCAM® offers many advantages over traditional microscopic methodologies. Algal samples can be analyzed whether the sample is live or preserved. It is not necessary for the FlowCAM® samples to be pre-settled in chambers, as is done for samples examined with the inverted microscope. Completing

sample analyses with the FlowCAM® takes roughly half the time than with the microscope and data entry is automatic.

Automatic data entry prevents errors associated with human data entry. Collected information is exported to Excel spreadsheets and permits computer summary analyses to be completed quickly and statistical information (e.g., mean, minimums, maximums, standard deviations) is provided for the output parameters (Appendix F). The Excel files of statistical information and digitized pictures can also be readily transmitted from phycologist to phycologist when the assistance of other experts is needed. Another advantage to having digitized pictures and computerized data, corrections could be easily made if the taxonomy of an algal cell was misidentified or if in the future a change in taxonomy occurs (Reynolds 1998).

Another benefit of the FlowCAM®'s is the cell recognition software, which helps reduce the number of cells that must be sorted and classified. Although individual images still need to be examined by a trained operator/phycologist to identify algae (Buskey and Hyatt 2006), the processing time is rapid compared to the traditional inverted microscope method. Besides seeking advice from an outside expert, the FlowCAM® provides a platform where a phycologist can train numerous individuals to identify phytoplankton being displayed by the FlowCAM®.

Since 2000, the FlowCAM® has been used successfully in marine systems (Lavrentyey et al. 2004; Vaillancourt et al. 2004; See et al. 2005; Buskey and Hyatt 2006) and in a freshwater lake (Wang et al. 2015). These studies have also clearly verified that using a FlowCAM® overcomes many of the limitations imposed by using the microscope. Of importance to this study, Alvarez et al. (2011) demonstrated the

relationship between FlowCAM's® algal counts and counts obtained by use of a microscope on preserved (Lugol's) marine samples was not significantly different as long as the factory-defined limitations of the FlowCAM® were considered. Wang et al. (2015) also demonstrated that colony cell density estimates obtain by use of a microscope were not significantly different from those of the FlowCAM® in a eutrophic lake, much like this study.

In this study of Florida water bodies, the FlowCAM® provided various estimates of the phytoplankton abundance at higher taxonomic levels (Class and Phylum) that were not significantly different from those obtained by use of the inverted microscope. Significant differences in cell density and bio-volume estimates were occasionally found at the Genus, Family, and Order levels, but these differences were related to the presence of algal colonies. My data demonstrated that by counting a colony as a single unit per mL resulted in a fewer significant differences at a lower taxonomic level between the FlowCAM® and inverted microscope. Bellinger and Sigee (2015) noted numerous studies of phytoplankton community dynamics and successional changes provided a better understanding of the environmental changes when phylum-level data were used verses species data. Dietmar et al. (2013) concluded, when examining long-term monitoring data of phytoplankton diversity, that reanalysis of the data at the species, genus and family taxonomic level is insufficient due to changes in taxonomic literature or expertise. Therefore, the FlowCAM® is applicable given no class or phylum differences in densities and bio-volumes per mL between the two tools.

These findings suggest estimates of cell density (individual cell count per mL and colony count per mL) and bio-volume (FlowCAM®'s ABD, ESD and the microscope's

geometric bio-volume calculations) between the inverted microscope and the FlowCAM® are similar at higher taxonomic levels encouraging the use of more-rapid analytical methods, like the FlowCAM®. The FlowCAM® automatically counts algal colonies not cells within a colony. But as can be done with a microscope, an operator can manually count individual cells if need be. In a eutrophic lake, Wang et al. (2015) demonstrated the FlowCAM® can accurately monitor estimates of plankton population and colony size distribution of bloom forming *Microcystis*. Not being able to count individual cells within a colony may not constitute a problem for some algal studies because cyanobacteria colonies have an adaptive importance in phytoplankton ecology (Agusti and Philips 1992). For Florida's shallow-productive waters, colony forming algal genera are more abundant than genera with limited-size plasticity (Duarte and Canfield 1990), suggesting colony counts are probably more appropriate for community ecology studies and the ability of the FlowCAM® to count colonies can be advantageous.

Sun and Liu (2003) and Jakobsen and Carstensen (2011) suggested that choosing the wrong geometric shape or the wrong particle-size algorithm strongly influenced the calculated algal bio-volumes. Traditional microscopic methods for calculating biovolumes allow only measurements of a limited number of cells or use previously published cell dimensions (Alvarez et al. 2014). The FlowCAM® generates two algorithms to estimate the bio-volume of particles counted (Area Based Diameter, ABD and Equivalent Spherical Diameter, ESD). However the FlowCAM® additionally calculates 30 individual measurements (length, width, etc.) with mean, minimum, maximum, and standard error parameters provided for the particles counted (Appendix F). Several marine studies have shown that the FlowCAM® accurately measures the

size of particles in water samples (Sieracki et al. 1998; Sterling et al. 2004; Buskey and Hyatt 2006; Tauxe et al. 2006; Meunier et al. 2012). Thus, the FlowCAM® operator could use generated size information to calculate their own bio-volume estimates (e.g., Alvarez et al. 2012) if the FlowCAM's® ABD or ESD bio-volumes are deemed unacceptable.

Comparing the microscopic-estimated Florida freshwater bio-volumes to those of the FlowCAM® demonstrated the bio-volumes estimates for the higher taxonomic levels (Phylum and Class) were not significantly different. Differences, however, were detected for Family and Genera estimates within a station of the lake. Alvarez et al. (2012) concluded that the two algorithms currently used in the FlowCAM® provided a good approximation of estimated microscopic bio-volumes in the case of spherical or elliptical algal cells, but poorly in the case of cylindrical cells. This was especially true, as demonstrated in this project, when non-linear cylindrical algal cells dominated algal samples. When this was the case, FlowCAM® bio-volumes estimates compared to those of the microscope tended to be over estimates when using the ESD algorithm, but under estimates when using the ABD algorithm.

Results obtained during this project also must be viewed in their proper context because the Bench B3 Series FlowCAM® does not represent the most up-to-date equipment available from Fluid Imaging Technologies. The loaned Bench B3 Series FlowCAM® had one objective lens (10X), only AutoImaging mode was used, and prior to running the Lugol's samples into the flow cell, the samples were filtered with a 100µm mesh net to prevent clogging. The objective lens limited the size of the particle that could be processed and it also made it difficult to visually identify algal cells to the

species level. The Bench B3 Series FlowCAM® also had an upper size limit that caused some particles, such as large algal colonies and filamentous algae, to be cut off in the captured images (e.g., Busky and Hyatt 2006), thereby reducing counts and bio-volume estimates. The AutoImaging mode had a lower sample volume processed, sometimes contributing to the identification of fewer genera, but the AutoImaging is most useful in collecting the whole plankton community assessment within the alleged size range (Jakobsen and Carstensen 2011). Additionally, the 100µm mesh could have prevented algal of the correct size range from entering the sample due to accumulation of the bigger particles in the straining mesh. Fluid-Imaging Technologies has apparently made advancements in expanding the size range from 2µm to 2,000µm and adding additional algorithms to accommodate diverse samples.

The FlowCAM® can certainly become a valuable tool in studies of phytoplankton community ecology and lake ecology, but it can also become a useful tool for studying the role of detritus due to the FlowCAM's® ability to capture detrital images and calculate detrital bio-volume. Detritus (Figure 3-7) is normally ignored when using the microscope. Algal biomasses as estimated by chlorophyll measurements are typically readily available, but chlorophyll measurements provide no information on algal community structure or detritus. Bigham (2012) recently established for Florida lakes seasonal patterns in chlorophyll concentrations and seasonal patterns within individual lakes due to the influence of temperature and rainfall. Bigham's (2012) work, however, suggest algal community structure and detritus could be influencing CHL/TP ratios in Florida lakes.

The FlowCAM[®] offers an opportunity for limnologists to expedite their efforts to test existing empirical algal models and develop new models to address broad-based ecological questions. There is great potential, as shown in the marine studies, for the FlowCAM[®] to increase the ability to count phytoplankton more frequently and aid in faster processing of large numbers of phytoplankton samples. Monitoring algal populations is essential when monitoring freshwater bodies because phytoplankton biomass is a proxy for primary productivity in many aquatic ecosystems and the FlowCAM[®] can advance water research covering time and space in a manageable time frame. The ability to collect surface water algal samples frequently from a large number of lakes also now exists because of the establishment of citizen monitoring programs like Florida LAKEWATCH. Algal samples collected by citizen scientists and preserved with Lugol's solution can be processed effectively by the FlowCAM[®], thus long-term records of the phytoplankton could be established at many individual water bodies, ending the dilemma identified by Needham and Lloyd (1915).

CHAPTER 5 CONCLUSION

Combining flow cytometer and microscope optics (FlowCAM®) represents a major advance in the development of tools for the study of phytoplankton ecology as well as the role of non-living particles in water. The FlowCAM® and the inverted microscope are two tools with advantages and disadvantages. The microscope shall never be eliminated due to the need for many studies to identify phytoplankton to the species-level. Yet, using the FlowCAM® in combination with microscope counts, increases efficiency and consistency to determine bio-volume on a routine basis (Allen et al. 1994). At this point, the FlowCAM® is a tool that can definitely provide reliable algal community metrics at the phylum level. Given the advantages of the FlowCAM®, it is now possible to frequently count large numbers of samples that scientists have identified as needed for advancements in the understanding of phytoplankton community ecology.

The ability of the FlowCAM® to process large numbers of Lugol's preserved algal samples offers the scientific community an opportunity to process samples from large number of aquatic systems when working with citizen scientists. In the case of Florida LAKEWATCH, long-term information on CHL, TP, TN and water clarity as measured by a Secchi disk has been collected by citizen scientists sampling their lakes monthly at different stations and the data are comparable to those collected by professionals. Integrating a FlowCAM® into the program would establish a long-term database on the phytoplankton in the biological community compared to chlorophyll samples.

Knowledge of the food-chain base could be most useful in limnological and marine studies. Equally important is the role the FlowCAM® can play as an outreach

educational tool and as a motivational tool for citizen scientists. Most citizen-scientists have no knowledge of what microorganisms live in “their” water. The citizen scientists that I worked with became more enthused and interested once they saw the FlowCAM® pictures of phytoplankton. With this enthusiasm, the citizen scientists are even more encouraged to collect samples. The ability of the FlowCAM® to process large numbers of Lugol’s preserved algal samples offers the scientific community an opportunity to process samples from large number of aquatic systems when working with citizen scientists and can detect changes in phytoplankton community composition at “their” lake. The use of the FlowCAM® and citizen scientists also represents the most cost-effective method for collecting to number of algal samples needed over space and time for scientific and lake management advancements (Canfield et al. 2002). Furthermore, analyses of the FlowCAM® provides a faster tool than the inverted microscope and provides important algal community information that is not available by using chlorophyll measurements alone.

APPENDIX A
RESULTS OF PAIRED T-TEST BETWEEN FLUID-IMAGING FLOWCAM® AND
MICROSCOPE INDIVIDUAL CELL COUNT PER ML

Table A-1. Individual cell count per mL for Lake Lochloosa at different taxonomic levels using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	22	0.0068*
Genus	May 9, 2013	1	28	0.0041*
Genus	May 9, 2013	2	29	0.0070*
Genus	May 9, 2013	3	30	0.0018*
Genus	May 23, 2013	1	26	0.1143
Genus	May 23, 2013	2	27	0.1113
Genus	May 23, 2013	3	25	0.0188*
Genus	June 13, 2013	1	26	0.0304*
Genus	June 13, 2013	2	30	0.0005*
Genus	June 13, 2013	3	29	0.0007*
Genus	June 27, 2013	1	31	<0.0001*
Genus	June 27, 2013	2	30	0.0034*
Genus	June 27, 2013	3	28	0.0241*
Family	March 17, 2013	1	18	0.0112*
Family	May 9, 2013	1	23	0.0035*
Family	May 9, 2013	2	23	0.0097*
Family	May 9, 2013	3	26	0.0018*
Family	May 23, 2013	1	21	0.1354
Family	May 23, 2013	2	23	0.1072
Family	May 23, 2013	3	21	0.0263*
Family	June 13, 2013	1	21	0.0368*
Family	June 13, 2013	2	23	0.0008*
Family	June 13, 2013	3	24	0.0011*
Family	June 27, 2013	1	25	0.0001*
Family	June 27, 2013	2	24	0.0049*
Family	June 27, 2013	3	22	0.0189*
Order	March 17, 2013	1	13	0.0630
Order	May 9, 2013	1	17	0.0097*
Order	May 9, 2013	2	17	0.0638
Order	May 9, 2013	3	20	0.0126*
Order	May 23, 2013	1	15	0.1189
Order	May 23, 2013	2	18	0.1311
Order	May 23, 2013	3	16	0.0269*
Order	June 13, 2013	1	16	0.0354*
Order	June 13, 2013	2	18	0.0033*
Order	June 13, 2013	3	19	0.0095*
Order	June 27, 2013	1	19	0.0062*
Order	June 27, 2013	2	20	0.0039*
Order	June 27, 2013	3	17	0.0463*

Table A-1. Continued

Taxa Level	Date	Station	N samples	p-Value
Class	March 17, 2013	1	11	0.1054
Class	May 9, 2013	1	11	0.0435*
Class	May 9, 2013	2	12	0.1142
Class	May 9, 2013	3	14	0.0420*
Class	May 23, 2013	1	11	0.2098
Class	May 23, 2013	2	12	0.1981
Class	May 23, 2013	3	11	0.1095
Class	June 13, 2013	1	11	0.1644
Class	June 13, 2013	2	11	0.0587
Class	June 13, 2013	3	12	0.0993
Class	June 27, 2013	1	13	0.0419*
Class	June 27, 2013	2	13	0.0653
Class	June 27, 2013	3	11	0.1788
Phylum	March 17, 2013	1	9	0.2395
Phylum	May 9, 2013	1	7	0.1628
Phylum	May 9, 2013	2	9	0.2171
Phylum	May 9, 2013	3	9	0.1861
Phylum	May 23, 2013	1	9	0.3126
Phylum	May 23, 2013	2	8	0.4257
Phylum	May 23, 2013	3	7	0.2700
Phylum	June 13, 2013	1	8	0.2152
Phylum	June 13, 2013	2	8	0.1713
Phylum	June 13, 2013	3	9	0.2734
Phylum	June 27, 2013	1	9	0.1235
Phylum	June 27, 2013	2	9	0.1587
Phylum	June 27, 2013	3	9	0.2890

Table A-2. Individual cell count per mL for Lake Santa Fe at different taxonomic levels using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	18	0.2631
Genus	May 9, 2013	1	18	0.0258*
Genus	May 9, 2013	2	17	0.0072*
Genus	May 9, 2013	3	18	0.0406*
Genus	May 23, 2013	1	14	0.1868
Genus	May 23, 2013	2	14	0.1332
Genus	May 23, 2013	3	12	0.3198
Genus	June 27, 2013	1	15	0.0108*
Genus	June 27, 2013	2	17	0.0090*
Genus	June 27, 2013	3	18	0.0586
Family	March 17, 2013	1	18	0.2631
Family	May 9, 2013	1	17	0.0264*
Family	May 9, 2013	2	17	0.0072*
Family	May 9, 2013	3	18	0.0406*
Family	May 23, 2013	1	14	0.1868
Family	May 23, 2013	2	14	0.1332
Family	May 23, 2013	3	12	0.3198
Family	June 27, 2013	1	15	0.0108*
Family	June 27, 2013	2	17	0.0090*
Family	June 27, 2013	3	18	0.0586
Order	March 17, 2013	1	16	0.2490
Order	May 9, 2013	1	14	0.0623
Order	May 9, 2013	2	13	0.0112*
Order	May 9, 2013	3	15	0.0171*
Order	May 23, 2013	1	12	0.2010
Order	May 23, 2013	2	12	0.1795
Order	May 23, 2013	3	10	0.3462
Order	June 27, 2013	1	12	0.0169*
Order	June 27, 2013	2	15	0.0102*
Order	June 27, 2013	3	14	0.0795
Class	March 17, 2013	1	11	0.2408
Class	May 9, 2013	1	9	0.1633
Class	May 9, 2013	2	8	0.0656
Class	May 9, 2013	3	10	0.0486*
Class	May 23, 2013	1	9	0.3762
Class	May 23, 2013	2	7	0.3458
Class	May 23, 2013	3	7	0.4834
Class	June 27, 2013	1	8	0.0749
Class	June 27, 2013	2	9	0.0579
Class	June 27, 2013	3	10	0.0876
Phylum	March 17, 2013	1	8	0.2467
Phylum	May 9, 2013	1	8	0.1549
Phylum	May 9, 2013	2	7	0.0735

Table A-2. Continued				
Taxa Level	Date	Station	N samples	p-Value
Phylum	May 9, 2013	3	7	0.1167
Phylum	May 23, 2013	1	7	0.3609
Phylum	May 23, 2013	2	6	0.3117
Phylum	May 23, 2013	3	6	0.4686
Phylum	June 27, 2013	1	7	0.1004
Phylum	June 27, 2013	2	7	0.0862
Phylum	June 27, 2013	3	8	0.1379

Table A-3. Individual cell count per mL for St. Johns River at different taxonomic levels using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	November 11, 2012	Leo	31	0.1162
Genus	March 13, 2013	Creslm	30	0.0349*
Genus	May 7, 2013	Leo	35	0.0123*
Genus	July 9, 2013	Creslm	33	0.0008*
Genus	July 9, 2013	Leo	28	0.0038*
Genus	July 22, 2013	Leo	25	0.0348*
Genus	August 13, 2013	Leo	27	0.0036*
Genus	August 27, 2013	Leo	22	0.1092
Genus	October 1, 2013	Pipe	30	0.0121*
Family	November 11, 2012	Leo	24	0.1566
Family	March 13, 2013	Creslm	24	0.0516
Family	May 7, 2013	Leo	29	0.0137*
Family	July 9, 2013	Creslm	28	0.0027*
Family	July 9, 2013	Leo	22	0.0080*
Family	July 22, 2013	Leo	21	0.0353*
Family	August 13, 2013	Leo	20	0.0222*
Family	August 27, 2013	Leo	17	0.1500
Family	October 1, 2013	Pipe	26	0.0089*
Order	November 11, 2012	Leo	18	0.1322
Order	March 13, 2013	Creslm	17	0.1386
Order	May 7, 2013	Leo	21	0.0306*
Order	July 9, 2013	Creslm	21	0.0047*
Order	July 9, 2013	Leo	17	0.0148*
Order	July 22, 2013	Leo	16	0.0546
Order	August 13, 2013	Leo	16	0.0439*
Order	August 27, 2013	Leo	13	0.1520
Order	October 1, 2013	Pipe	20	0.0304*
Class	November 11, 2012	Leo	11	0.1472
Class	March 13, 2013	Creslm	12	0.1346
Class	May 7, 2013	Leo	14	0.0489*
Class	July 9, 2013	Creslm	15	0.0214*
Class	July 9, 2013	Leo	11	0.0535
Class	July 22, 2013	Leo	11	0.0371*
Class	August 13, 2013	Leo	11	0.0847
Class	August 27, 2013	Leo	8	0.3145
Class	October 1, 2013	Pipe	14	0.0287*
Phylum	November 11, 2012	Leo	8	0.1323
Phylum	March 13, 2013	Creslm	9	0.2537
Phylum	May 7, 2013	Leo	10	0.1808
Phylum	July 9, 2013	Creslm	10	0.0680
Phylum	July 9, 2013	Leo	8	0.1210
Phylum	July 22, 2013	Leo	7	0.1544
Phylum	August 13, 2013	Leo	9	0.0985

Table A-3. Continued

Taxa Level	Date	Station	N samples	p-Value
Phylum	August 27, 2013	Leo	6	0.3265
Phylum	October 1, 2013	Pipe	10	0.0761

APPENDIX B
RESULTS OF PAIRED T-TEST BETWEEN FLUID-IMAGING FLOWCAM® AND
MICROSCOPE COLONY COUNT PER ML

Table B-1. Colony count per mL for Lake Lochloosa at different taxonomic levels
using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	22	0.1316
Genus	May 9, 2013	1	28	0.1194
Genus	May 9, 2013	2	29	0.1160
Genus	May 9, 2013	3	30	0.0470*
Genus	May 23, 2013	1	26	0.7682
Genus	May 23, 2013	2	27	0.6221
Genus	May 23, 2013	3	25	0.3355
Genus	June 13, 2013	1	26	0.2977
Genus	June 13, 2013	2	30	0.0044*
Genus	June 13, 2013	3	29	0.0223*
Genus	June 27, 2013	1	31	0.0007*
Genus	June 27, 2013	2	30	0.0808*
Genus	June 27, 2013	3	28	0.2444
Family	March 17, 2013	1	18	0.1368
Family	May 9, 2013	1	23	0.1229
Family	May 9, 2013	2	23	0.1207
Family	May 9, 2013	3	26	0.0463*
Family	May 23, 2013	1	21	0.7688
Family	May 23, 2013	2	23	0.6216
Family	May 23, 2013	3	21	0.3561
Family	June 13, 2013	1	21	0.2759
Family	June 13, 2013	2	23	0.0048*
Family	June 13, 2013	3	24	0.0247*
Family	June 27, 2013	1	25	0.0017*
Family	June 27, 2013	2	24	0.0922
Family	June 27, 2013	3	22	0.2155
Order	March 17, 2013	1	13	0.1613
Order	May 9, 2013	1	17	0.0879
Order	May 9, 2013	2	17	0.2190
Order	May 9, 2013	3	20	0.0691
Order	May 23, 2013	1	15	0.7452
Order	May 23, 2013	2	18	0.6219
Order	May 23, 2013	3	16	0.3070
Order	June 13, 2013	1	16	0.2186
Order	June 13, 2013	2	18	0.0077*
Order	June 13, 2013	3	19	0.0550
Order	June 27, 2013	1	19	0.0143*
Order	June 27, 2013	2	20	0.0635
Order	June 27, 2013	3	17	0.2663
Class	March 17, 2013	1	11	0.2081
Class	May 9, 2013	1	11	0.1372

Table B-1. Continued				
Taxa Level	Date	Station	N samples	p-Value
Class	May 9, 2013	2	12	0.2488
Class	May 9, 2013	3	14	0.1432
Class	May 23, 2013	1	11	0.7385
Class	May 23, 2013	2	12	0.6968
Class	May 23, 2013	3	11	0.4035
Class	June 13, 2013	1	11	0.3414
Class	June 13, 2013	2	11	0.0671
Class	June 13, 2013	3	12	0.2035
Class	June 27, 2013	1	13	0.0593
Class	June 27, 2013	2	13	0.1589
Class	June 27, 2013	3	11	0.3637
Phylum	March 17, 2013	1	9	0.3474
Phylum	May 9, 2013	1	7	0.3315
Phylum	May 9, 2013	2	9	0.3898
Phylum	May 9, 2013	3	9	0.3333
Phylum	May 23, 2013	1	9	0.7793
Phylum	May 23, 2013	2	8	0.7802
Phylum	May 23, 2013	3	7	0.5664
Phylum	June 13, 2013	1	8	0.3964
Phylum	June 13, 2013	2	8	0.2119
Phylum	June 13, 2013	3	9	0.4046
Phylum	June 27, 2013	1	9	0.1696
Phylum	June 27, 2013	2	9	0.2817
Phylum	June 27, 2013	3	9	0.4871

Table B-2. Colony count per mL for Lake Santa Fe at different taxonomic levels using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	18	0.7288
Genus	May 9, 2013	1	18	0.3790
Genus	May 9, 2013	2	17	0.1031
Genus	May 9, 2013	3	18	0.3775
Genus	May 23, 2013	1	14	0.9989
Genus	May 23, 2013	2	14	0.6201
Genus	May 23, 2013	3	12	0.7047
Genus	June 27, 2013	1	15	0.0685
Genus	June 27, 2013	2	17	0.0744
Genus	June 27, 2013	3	18	0.2862
Family	March 17, 2013	1	18	0.7288
Family	May 9, 2013	1	17	0.3853
Family	May 9, 2013	2	17	0.1031
Family	May 9, 2013	3	18	0.3775
Family	May 23, 2013	1	14	0.9989
Family	May 23, 2013	2	14	0.8201
Family	May 23, 2013	3	12	0.7047
Family	June 27, 2013	1	15	0.0685
Family	June 27, 2013	2	17	0.0744
Family	June 27, 2013	3	18	0.2862
Order	March 17, 2013	1	16	0.7478
Order	May 9, 2013	1	14	0.3997
Order	May 9, 2013	2	13	0.0778
Order	May 9, 2013	3	15	0.3779
Order	May 23, 2013	1	12	0.9989
Order	May 23, 2013	2	12	0.8260
Order	May 23, 2013	3	10	0.7037
Order	June 27, 2013	1	12	0.0941
Order	June 27, 2013	2	15	0.0708
Order	June 27, 2013	3	14	0.2902
Class	March 17, 2013	1	11	0.7003
Class	May 9, 2013	1	9	0.4530
Class	May 9, 2013	2	8	0.1363
Class	May 9, 2013	3	10	0.3908
Class	May 23, 2013	1	9	0.9991
Class	May 23, 2013	2	7	0.8596
Class	May 23, 2013	3	7	0.7063
Class	June 27, 2013	1	8	0.1036
Class	June 27, 2013	2	9	0.1194
Class	June 27, 2013	3	10	0.2738
Phylum	March 17, 2013	1	8	0.6860
Phylum	May 9, 2013	1	8	0.4389
Phylum	May 9, 2013	2	7	0.1282
Phylum	May 9, 2013	3	7	0.4877
Phylum	May 23, 2013	1	7	0.9989

Table B-2. Continued				
Taxa Level	Date	Station	N samples	p-Value
Phylum	May 23, 2013	2	6	0.8290
Phylum	May 23, 2013	3	6	0.6415
Phylum	June 27, 2013	1	7	0.1580
Phylum	June 27, 2013	2	7	0.1612
Phylum	June 27, 2013	3	8	0.3469

Table B-3. Colony count per mL for St. Johns River at different taxonomic levels using microscope and FlowCAM®

Taxa Level	Date	Station	N samples	p-Value
Genus	November 11, 2012	Leo	31	0.5085
Genus	March 13, 2013	Creslm	30	0.5566
Genus	May 7, 2013	Leo	35	0.3719
Genus	July 9, 2013	Creslm	33	0.0584
Genus	July 9, 2013	Leo	28	0.0489*
Genus	July 22, 2013	Leo	25	0.1117
Genus	August 13, 2013	Leo	27	0.0573
Genus	August 27, 2013	Leo	22	0.4353
Genus	October 1, 2013	Pipe	30	0.1720
Family	November 11, 2012	Leo	24	0.5295
Family	March 13, 2013	Creslm	24	0.5873
Family	May 7, 2013	Leo	29	0.3856
Family	July 9, 2013	Creslm	28	0.0748
Family	July 9, 2013	Leo	22	0.0601
Family	July 22, 2013	Leo	21	0.1115
Family	August 13, 2013	Leo	20	0.1166
Family	August 27, 2013	Leo	17	0.4773
Family	October 1, 2013	Pipe	26	0.1447
Order	November 11, 2012	Leo	18	0.4906
Order	March 13, 2013	Creslm	17	0.5353
Order	May 7, 2013	Leo	21	0.3724
Order	July 9, 2013	Creslm	21	0.0751
Order	July 9, 2013	Leo	17	0.0550
Order	July 22, 2013	Leo	16	0.1151
Order	August 13, 2013	Leo	16	0.1254
Order	August 27, 2013	Leo	13	0.4597
Order	October 1, 2013	Pipe	20	0.1548
Class	November 11, 2012	Leo	11	0.4377
Class	March 13, 2013	Creslm	12	0.6267
Class	May 7, 2013	Leo	14	0.3681
Class	July 9, 2013	Creslm	15	0.0747
Class	July 9, 2013	Leo	11	0.0587
Class	July 22, 2013	Leo	11	0.0579
Class	August 13, 2013	Leo	11	0.1286
Class	August 27, 2013	Leo	8	0.5304
Class	October 1, 2013	Pipe	14	0.1417
Phylum	November 11, 2012	Leo	8	0.3376
Phylum	March 13, 2013	Creslm	9	0.6464
Phylum	May 7, 2013	Leo	10	0.4562
Phylum	July 9, 2013	Creslm	10	0.0807
Phylum	July 9, 2013	Leo	8	0.1689
Phylum	July 22, 2013	Leo	7	0.1654
Phylum	August 13, 2013	Leo	9	0.1300
Phylum	August 27, 2013	Leo	6	0.5245
Phylum	October 1, 2013	Pipe	10	0.1595

APPENDIX C
RESULTS OF PAIRED T-TEST FLUID-IMAGING FLOWCAM® ABD BIO-VOLUMES
COMPARED TO MICROSCOPE BIO-VOLUMES

Table C-1. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD bio-volumes in Lake Lochloosa

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	22	0.0127*
Genus	May 9, 2013	1	28	0.0127*
Genus	May 9, 2013	2	29	0.0690
Genus	May 9, 2013	3	30	0.0041*
Genus	May 23, 2013	1	26	0.2552
Genus	May 23, 2013	2	27	0.1010
Genus	May 23, 2013	3	25	0.1346
Genus	June 13, 2013	1	26	0.0943
Genus	June 13, 2013	2	30	0.0011*
Genus	June 13, 2013	3	29	0.0055*
Genus	June 27, 2013	1	31	<0.0001*
Genus	June 27, 2013	2	30	0.0045*
Genus	June 27, 2013	3	28	0.0664
Family	March 17, 2013	1	18	0.0235*
Family	May 9, 2013	1	23	0.0220*
Family	May 9, 2013	2	23	0.0557
Family	May 9, 2013	3	26	0.0043*
Family	May 23, 2013	1	21	0.2699
Family	May 23, 2013	2	23	0.1003
Family	May 23, 2013	3	21	0.1431
Family	June 13, 2013	1	21	0.0962
Family	June 13, 2013	2	23	0.0016*
Family	June 13, 2013	3	24	0.0063*
Family	June 27, 2013	1	25	0.0002*
Family	June 27, 2013	2	24	0.0061*
Family	June 27, 2013	3	22	0.0750
Order	March 17, 2013	1	13	0.0570
Order	May 9, 2013	1	17	0.0187*
Order	May 9, 2013	2	17	0.1469
Order	May 9, 2013	3	20	0.0232*
Order	May 23, 2013	1	15	0.1939
Order	May 23, 2013	2	18	0.1398
Order	May 23, 2013	3	16	0.1018
Order	June 13, 2013	1	16	0.0697
Order	June 13, 2013	2	18	0.0015*
Order	June 13, 2013	3	19	0.0142*
Order	June 27, 2013	1	19	0.0037*
Order	June 27, 2013	2	20	0.0060*
Order	June 27, 2013	3	17	0.1067

Table C-1. Continued

Taxa Level	Date	Station	N samples	p-Value
Class	March 17, 2013	1	11	0.1056
Class	May 9, 2013	1	11	0.0458*
Class	May 9, 2013	2	12	0.2081
Class	May 9, 2013	3	14	0.0586
Class	May 23, 2013	1	11	0.1816
Class	May 23, 2013	2	12	0.2068
Class	May 23, 2013	3	11	0.1883
Class	June 13, 2013	1	11	0.1699
Class	June 13, 2013	2	11	0.0209
Class	June 13, 2013	3	12	0.1185
Class	June 27, 2013	1	13	0.0221*
Class	June 27, 2013	2	13	0.0565
Class	June 27, 2013	3	11	0.2347
Phylum	March 17, 2013	1	9	0.2046
Phylum	May 9, 2013	1	7	0.1768
Phylum	May 9, 2013	2	9	0.3274
Phylum	May 9, 2013	3	9	0.1891
Phylum	May 23, 2013	1	9	0.2479
Phylum	May 23, 2013	2	8	0.3860
Phylum	May 23, 2013	3	7	0.3944
Phylum	June 13, 2013	1	8	0.1760
Phylum	June 13, 2013	2	8	0.1088
Phylum	June 13, 2013	3	9	0.2907
Phylum	June 27, 2013	1	9	0.0925
Phylum	June 27, 2013	2	9	0.1232
Phylum	June 27, 2013	3	9	0.3652

Table C-2. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD bio-volumes in Lake Santa Fe

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	18	0.4596
Genus	May 9, 2013	1	18	0.1178
Genus	May 9, 2013	2	17	0.1563
Genus	May 9, 2013	3	18	0.1852
Genus	May 23, 2013	1	14	0.5553
Genus	May 23, 2013	2	14	0.5554
Genus	May 23, 2013	3	12	0.4639
Genus	June 27, 2013	1	15	0.0239*
Genus	June 27, 2013	2	17	0.0243*
Genus	June 27, 2013	3	18	0.1039
Family	March 17, 2013	1	18	0.4596
Family	May 9, 2013	1	17	0.1527
Family	May 9, 2013	2	17	0.1563
Family	May 9, 2013	3	18	0.1852
Family	May 23, 2013	1	14	0.5553
Family	May 23, 2013	2	14	0.5554
Family	May 23, 2013	3	12	0.4639
Family	June 27, 2013	1	15	0.0239*
Family	June 27, 2013	2	17	0.0243*
Family	June 27, 2013	3	18	0.1039
Order	March 17, 2013	1	16	0.4414
Order	May 9, 2013	1	14	0.1760
Order	May 9, 2013	2	13	0.1310
Order	May 9, 2013	3	15	0.2134
Order	May 23, 2013	1	12	0.5111
Order	May 23, 2013	2	12	0.5536
Order	May 23, 2013	3	10	0.4804
Order	June 27, 2013	1	12	0.0339*
Order	June 27, 2013	2	15	0.0191*
Order	June 27, 2013	3	14	0.1612
Class	March 17, 2013	1	11	0.4152
Class	May 9, 2013	1	9	0.2362
Class	May 9, 2013	2	8	0.2063
Class	May 9, 2013	3	10	0.3412
Class	May 23, 2013	1	9	0.5666
Class	May 23, 2013	2	7	0.6485
Class	May 23, 2013	3	7	0.4395
Class	June 27, 2013	1	8	0.0276*
Class	June 27, 2013	2	9	0.0537
Class	June 27, 2013	3	10	0.1673
Phylum	March 17, 2013	1	8	0.3891
Phylum	May 9, 2013	1	8	0.2350
Phylum	May 9, 2013	2	7	0.2085

Table C-2. Continued

Taxa Level	Date	Station	N samples	p-Value
Phylum	May 9, 2013	3	7	0.4347
Phylum	May 23, 2013	1	7	0.4932
Phylum	May 23, 2013	2	6	0.5291
Phylum	May 23, 2013	3	6	0.2829
Phylum	June 27, 2013	1	7	0.0585
Phylum	June 27, 2013	2	7	0.0851
Phylum	June 27, 2013	3	8	0.2629

Table C-3. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD bio-volumes in St. Johns River

Taxa Level	Date	Station	N samples	p-Value
Genus	November 11, 2012	Leo	31	0.0215*
Genus	March 13, 2013	Creslm	30	0.0288*
Genus	May 7, 2013	Leo	35	0.0011*
Genus	July 9, 2013	Creslm	33	<0.0001*
Genus	July 9, 2013	Leo	28	0.0018*
Genus	July 22, 2013	Leo	25	0.0433*
Genus	August 13, 2013	Leo	27	0.0078*
Genus	August 27, 2013	Leo	22	0.0837
Genus	October 1, 2013	Pipe	30	0.0294*
Family	November 11, 2012	Leo	24	0.0388*
Family	March 13, 2013	Creslm	24	0.0447*
Family	May 7, 2013	Leo	29	0.0020*
Family	July 9, 2013	Creslm	28	<0.0001*
Family	July 9, 2013	Leo	22	0.0042*
Family	July 22, 2013	Leo	21	0.0440*
Family	August 13, 2013	Leo	20	0.0269*
Family	August 27, 2013	Leo	17	0.1103
Family	October 1, 2013	Pipe	26	0.0133*
Order	November 11, 2012	Leo	18	0.0350*
Order	March 13, 2013	Creslm	17	0.1152
Order	May 7, 2013	Leo	21	0.0074*
Order	July 9, 2013	Creslm	21	< 0.0001*
Order	July 9, 2013	Leo	17	0.0083*
Order	July 22, 2013	Leo	16	0.0576
Order	August 13, 2013	Leo	16	0.0576
Order	August 27, 2013	Leo	13	0.0992
Order	October 1, 2013	Pipe	20	0.0343*
Class	November 11, 2012	Leo	11	0.0500
Class	March 13, 2013	Creslm	12	0.1051
Class	May 7, 2013	Leo	14	0.0119*
Class	July 9, 2013	Creslm	15	0.0008*
Class	July 9, 2013	Leo	11	0.0231*
Class	July 22, 2013	Leo	11	0.0147*
Class	August 13, 2013	Leo	11	0.0490*
Class	August 27, 2013	Leo	8	0.1636
Class	October 1, 2013	Pipe	14	0.0270*
Phylum	November 11, 2012	Leo	8	0.0446*
Phylum	March 13, 2013	Creslm	9	0.2231
Phylum	May 7, 2013	Leo	10	0.0810
Phylum	July 9, 2013	Creslm	10	0.0060*
Phylum	July 9, 2013	Leo	8	0.0746
Phylum	July 22, 2013	Leo	7	0.0880
Phylum	August 13, 2013	Leo	9	0.0613

Table C-3. Continued				
Taxa Level	Date	Station	N samples	p-Value
Phylum	August 27, 2013	Leo	6	0.1717
Phylum	October 1, 2013	Pipe	10	0.0566

APPENDIX D
RESULTS OF PAIRED T-TEST FLUID-IMAGING FLOWCAM® ESD BIO-VOLUMES
COMPARED TO MICROSCOPE BIO-VOLUMES

Table D-1. Comparing the inverted microscope bio-volumes to the FlowCAM®'s ESD bio-volumes in Lake Lochloosa

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	22	0.0531
Genus	May 9, 2013	1	28	0.1117
Genus	May 9, 2013	2	29	0.2695
Genus	May 9, 2013	3	30	0.0299*
Genus	May 23, 2013	1	26	0.7902
Genus	May 23, 2013	2	27	0.3612
Genus	May 23, 2013	3	25	0.4865
Genus	June 13, 2013	1	26	0.4591
Genus	June 13, 2013	2	30	0.0165*
Genus	June 13, 2013	3	29	0.0694
Genus	June 27, 2013	1	31	0.0017*
Genus	June 27, 2013	2	30	0.0426*
Genus	June 27, 2013	3	28	0.3352
Family	March 17, 2013	1	18	0.0783
Family	May 9, 2013	1	23	0.1152
Family	May 9, 2013	2	23	0.2747
Family	May 9, 2013	3	26	0.0314*
Family	May 23, 2013	1	21	0.7916
Family	May 23, 2013	2	23	0.3582
Family	May 23, 2013	3	21	0.4902
Family	June 13, 2013	1	21	0.4674
Family	June 13, 2013	2	23	0.0207*
Family	June 13, 2013	3	24	0.0779
Family	June 27, 2013	1	25	0.0028*
Family	June 27, 2013	2	24	0.0473*
Family	June 27, 2013	3	22	0.3569
Order	March 17, 2013	1	13	0.1248
Order	May 9, 2013	1	17	0.0954
Order	May 9, 2013	2	17	0.3945
Order	May 9, 2013	3	20	0.0772
Order	May 23, 2013	1	15	0.7520
Order	May 23, 2013	2	18	0.3945
Order	May 23, 2013	3	16	0.4331
Order	June 13, 2013	1	16	0.4236
Order	June 13, 2013	2	18	0.0173*
Order	June 13, 2013	3	19	0.0966
Order	June 27, 2013	1	19	0.0163*
Order	June 27, 2013	2	20	0.0407*
Order	June 27, 2013	3	17	0.3957

Table D-1. Continued

Taxa Level	Date	Station	N samples	p-Value
Class	March 17, 2013	1	11	0.1931
Class	May 9, 2013	1	11	0.1529
Class	May 9, 2013	2	12	0.4567
Class	May 9, 2013	3	14	0.1511
Class	May 23, 2013	1	11	0.7889
Class	May 23, 2013	2	12	0.4867
Class	May 23, 2013	3	11	0.5023
Class	June 13, 2013	1	11	0.5355
Class	June 13, 2013	2	11	0.0601
Class	June 13, 2013	3	12	0.2837
Class	June 27, 2013	1	13	0.0539
Class	June 27, 2013	2	13	0.1536
Class	June 27, 2013	3	11	0.5170
Phylum	March 17, 2013	1	9	0.2923
Phylum	May 9, 2013	1	7	0.3104
Phylum	May 9, 2013	2	9	0.5480
Phylum	May 9, 2013	3	9	0.2951
Phylum	May 23, 2013	1	9	0.8081
Phylum	May 23, 2013	2	8	0.6096
Phylum	May 23, 2013	3	7	0.6588
Phylum	June 13, 2013	1	8	0.5220
Phylum	June 13, 2013	2	8	0.1963
Phylum	June 13, 2013	3	9	0.4528
Phylum	June 27, 2013	1	9	0.1645
Phylum	June 27, 2013	2	9	0.2433
Phylum	June 27, 2013	3	9	0.6131

Table D-2. Comparing the inverted microscope bio-volumes to the FlowCAM® ESD
bio-volumes in Lake Santa Fe

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	18	0.6276
Genus	May 9, 2013	1	18	0.3988
Genus	May 9, 2013	2	17	0.4543
Genus	May 9, 2013	3	18	0.4815
Genus	May 23, 2013	1	14	0.9916
Genus	May 23, 2013	2	14	0.9955
Genus	May 23, 2013	3	12	0.1580
Genus	June 27, 2013	1	15	0.0752
Genus	June 27, 2013	2	17	0.082
Genus	June 27, 2013	3	18	0.2459
Family	March 17, 2013	1	18	0.6276
Family	May 9, 2013	1	17	0.4348
Family	May 9, 2013	2	17	0.4543
Family	May 9, 2013	3	18	0.4816
Family	May 23, 2013	1	14	0.9916
Family	May 23, 2013	2	14	0.9955
Family	May 23, 2013	3	12	0.1580
Family	June 27, 2013	1	15	0.0762
Family	June 27, 2013	2	17	0.0802
Family	June 27, 2013	3	18	0.2459
Order	March 17, 2013	1	16	0.6174
Order	May 9, 2013	1	14	0.4445
Order	May 9, 2013	2	13	0.4223
Order	May 9, 2013	3	15	0.5102
Order	May 23, 2013	1	12	0.9904
Order	May 23, 2013	2	12	0.9956
Order	May 23, 2013	3	10	0.1908
Order	June 27, 2013	1	12	0.0903
Order	June 27, 2013	2	15	0.0672
Order	June 27, 2013	3	14	0.3101
Class	March 17, 2013	1	11	0.6163
Class	May 9, 2013	1	9	0.5290
Class	May 9, 2013	2	8	0.5059
Class	May 9, 2013	3	10	0.6331
Class	May 23, 2013	1	9	0.9923
Class	May 23, 2013	2	7	0.9966
Class	May 23, 2013	3	7	0.1945
Class	June 27, 2013	1	8	0.0716
Class	June 27, 2013	2	9	0.1445
Class	June 27, 2013	3	10	0.3628
Phylum	March 17, 2013	1	8	0.5949
Phylum	May 9, 2013	1	8	0.6329
Phylum	May 9, 2013	2	7	0.5020

Table D-2. Continued

Taxa Level	Date	Station	N samples	p-Value
Phylum	May 9, 2013	3	7	0.6821
Phylum	May 23, 2013	1	7	0.9911
Phylum	May 23, 2013	2	6	0.9966
Phylum	May 23, 2013	3	6	0.1141
Phylum	June 27, 2013	1	7	0.1216
Phylum	June 27, 2013	2	7	0.1787
Phylum	June 27, 2013	3	8	0.4483

Table D-3. Comparing the inverted microscope bio-volumes to the FlowCAM® ESD bio-volumes in St. Johns River

Taxa Level	Date	Station	N samples	p-Value
Genus	November 11, 2012	Leo	31	0.0893
Genus	March 13, 2013	Creslm	30	0.1724
Genus	May 7, 2013	Leo	35	0.0052*
Genus	July 9, 2013	Creslm	33	<0.0001*
Genus	July 9, 2013	Leo	28	0.0136*
Genus	July 22, 2013	Leo	25	0.1821
Genus	August 13, 2013	Leo	27	0.0346*
Genus	August 27, 2013	Leo	22	0.2928
Genus	October 1, 2013	Pipe	30	0.0775
Family	November 11, 2012	Leo	24	0.1188
Family	March 13, 2013	Creslm	24	0.2066
Family	May 7, 2013	Leo	29	0.0077*
Family	July 9, 2013	Creslm	28	0.0001*
Family	July 9, 2013	Leo	22	0.0182*
Family	July 22, 2013	Leo	21	0.1847
Family	August 13, 2013	Leo	20	0.0626
Family	August 27, 2013	Leo	17	0.3081
Family	October 1, 2013	Pipe	26	0.0607
Order	November 11, 2012	Leo	18	0.1040
Order	March 13, 2013	Creslm	17	0.3085
Order	May 7, 2013	Leo	21	0.0188*
Order	July 9, 2013	Creslm	21	0.0001*
Order	July 9, 2013	Leo	17	0.0268*
Order	July 22, 2013	Leo	16	0.1897
Order	August 13, 2013	Leo	16	0.1086
Order	August 27, 2013	Leo	13	0.2838
Order	October 1, 2013	Pipe	20	0.1036
Class	November 11, 2012	Leo	11	0.1024
Class	March 13, 2013	Creslm	12	0.2991
Class	May 7, 2013	Leo	14	0.0208*
Class	July 9, 2013	Creslm	15	0.0007*
Class	July 9, 2013	Leo	11	0.0342*
Class	July 22, 2013	Leo	11	0.0737
Class	August 13, 2013	Leo	11	0.0768
Class	August 27, 2013	Leo	8	0.2627
Class	October 1, 2013	Pipe	14	0.0914
Phylum	November 11, 2012	Leo	8	0.0874
Phylum	March 13, 2013	Creslm	9	0.4182
Phylum	May 7, 2013	Leo	10	0.1093
Phylum	July 9, 2013	Creslm	10	0.0059*
Phylum	July 9, 2013	Leo	8	0.1117
Phylum	July 22, 2013	Leo	7	0.1899
Phylum	August 13, 2013	Leo	9	0.0905

Table D-3. Continued				
Taxa Level	Date	Station	N samples	p-Value
Phylum	August 27, 2013	Leo	6	0.2685
Phylum	October 1, 2013	Pipe	10	0.1276

APPENDIX E
RESULTS OF PAIRED T-TEST FLUID-IMAGING FLOWCAM® ABD AND ESD BIO-
VOLUMES COMBINED COMPARED TO MICROSCOPE BIO-VOLUMES

Table E-1. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD
and ESD combined bio-volumes in Lake Lochloosa

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	22	0.0285*
Genus	May 9, 2013	1	28	0.0632
Genus	May 9, 2013	2	29	0.1874
Genus	May 9, 2013	3	30	0.0150*
Genus	May 23, 2013	1	26	0.5825
Genus	May 23, 2013	2	27	0.2126
Genus	May 23, 2013	3	25	0.2789
Genus	June 13, 2013	1	26	0.2678
Genus	June 13, 2013	2	30	0.0077*
Genus	June 13, 2013	3	29	0.0360*
Genus	June 27, 2013	1	31	0.0007*
Genus	June 27, 2013	2	30	0.0223*
Genus	June 27, 2013	3	28	0.2162
Family	March 17, 2013	1	18	0.0462*
Family	May 9, 2013	1	23	0.0651
Family	May 9, 2013	2	23	0.1910
Family	May 9, 2013	3	26	0.0161*
Family	May 23, 2013	1	21	0.5831
Family	May 23, 2013	2	23	0.2144
Family	May 23, 2013	3	21	0.2908
Family	June 13, 2013	1	21	0.2760
Family	June 13, 2013	2	23	0.0108*
Family	June 13, 2013	3	24	0.0437*
Family	June 27, 2013	1	25	0.0014*
Family	June 27, 2013	2	24	0.0281*
Family	June 27, 2013	3	22	0.2320
Order	March 17, 2013	1	13	0.0927
Order	May 9, 2013	1	17	0.0575
Order	May 9, 2013	2	17	0.3153
Order	May 9, 2013	3	20	0.0514
Order	May 23, 2013	1	15	0.5117
Order	May 23, 2013	2	18	0.2611
Order	May 23, 2013	3	16	0.2398
Order	June 13, 2013	1	16	0.2388
Order	June 13, 2013	2	18	0.0096*
Order	June 13, 2013	3	19	0.0667
Order	June 27, 2013	1	19	0.0116*
Order	June 27, 2013	2	20	0.0245*
Order	June 27, 2013	3	17	0.2806

Table E-1. Continued

Taxa Level	Date	Station	N samples	p-Value
Class	March 17, 2013	1	11	0.1522
Class	May 9, 2013	1	11	0.0998
Class	May 9, 2013	2	12	0.3756
Class	May 9, 2013	3	14	0.1108
Class	May 23, 2013	1	11	0.5852
Class	May 23, 2013	2	12	0.3547
Class	May 23, 2013	3	11	0.3246
Class	June 13, 2013	1	11	0.3559
Class	June 13, 2013	2	11	0.0444*
Class	June 13, 2013	3	12	0.2335
Class	June 27, 2013	1	13	0.0414*
Class	June 27, 2013	2	13	0.1189
Class	June 27, 2013	3	11	0.4116
Phylum	March 17, 2013	1	9	0.2664
Phylum	May 9, 2013	1	7	0.2540
Phylum	May 9, 2013	2	9	0.4801
Phylum	May 9, 2013	3	9	0.2523
Phylum	May 23, 2013	1	9	0.8269
Phylum	May 23, 2013	2	8	0.5109
Phylum	May 23, 2013	3	7	0.5201
Phylum	June 13, 2013	1	8	0.3601
Phylum	June 13, 2013	2	8	0.1667
Phylum	June 13, 2013	3	9	0.4105
Phylum	June 27, 2013	1	9	0.1336
Phylum	June 27, 2013	2	9	0.2036
Phylum	June 27, 2013	3	9	0.5268

Table E-2. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD and ESD combined bio-volumes in Lake Santa Fe

Taxa Level	Date	Station	N samples	p-Value
Genus	March 17, 2013	1	18	0.5270
Genus	May 9, 2013	1	18	0.2882
Genus	May 9, 2013	2	17	0.3778
Genus	May 9, 2013	3	18	0.3842
Genus	May 23, 2013	1	14	0.8465
Genus	May 23, 2013	2	14	0.8338
Genus	May 23, 2013	3	12	0.2564
Genus	June 27, 2013	1	15	0.0618
Genus	June 27, 2013	2	17	0.0664
Genus	June 27, 2013	3	18	0.1817
Family	March 17, 2013	1	18	0.5270
Family	May 9, 2013	1	17	0.3248
Family	May 9, 2013	2	17	0.3778
Family	May 9, 2013	3	18	0.3843
Family	May 23, 2013	1	14	0.8465
Family	May 23, 2013	2	14	0.8338
Family	May 23, 2013	3	12	0.2564
Family	June 27, 2013	1	15	0.0618
Family	June 27, 2013	2	17	0.0664
Family	June 27, 2013	3	18	0.1817
Order	March 17, 2013	1	16	0.5108
Order	May 9, 2013	1	14	0.3298
Order	May 9, 2013	2	13	0.3440
Order	May 9, 2013	3	15	0.4093
Order	May 23, 2013	1	12	0.8240
Order	May 23, 2013	2	12	0.8326
Order	May 23, 2013	3	10	0.2764
Order	June 27, 2013	1	12	0.0740
Order	June 27, 2013	2	15	0.0534
Order	June 27, 2013	3	14	0.2432
Class	March 17, 2013	1	11	0.5037
Class	May 9, 2013	1	9	0.4157
Class	May 9, 2013	2	8	0.4269
Class	May 9, 2013	3	10	0.5526
Class	May 23, 2013	1	9	0.8549
Class	May 23, 2013	2	7	0.8713
Class	May 23, 2013	3	7	0.2812
Class	June 27, 2013	1	8	0.0566
Class	June 27, 2013	2	9	0.1188
Class	June 27, 2013	3	10	0.2662
Phylum	March 17, 2013	1	8	0.4847
Phylum	May 9, 2013	1	8	0.4167
Phylum	May 9, 2013	2	7	0.4321

Table E-2. Continued

Taxa Level	Date	Station	N samples	p-Value
Phylum	May 9, 2013	3	7	0.6173
Phylum	May 23, 2013	1	7	0.8340
Phylum	May 23, 2013	2	6	0.6320
Phylum	May 23, 2013	3	6	0.1796
Phylum	June 27, 2013	1	7	0.1030
Phylum	June 27, 2013	2	7	0.1509
Phylum	June 27, 2013	3	8	0.3789

Table E-3. Comparing the inverted microscope bio-volumes to the FlowCAM® ABD and ESD combined bio-volumes in St. Johns River

Taxa Level	Date	Station	N samples	p-Value
Genus	November 11, 2012	Leo	31	0.0598
Genus	March 13, 2013	Creslm	30	0.1069
Genus	May 7, 2013	Leo	35	0.0032*
Genus	July 9, 2013	Creslm	33	<0.0001*
Genus	July 9, 2013	Leo	28	0.0063*
Genus	July 22, 2013	Leo	25	0.1161
Genus	August 13, 2013	Leo	27	0.0208*
Genus	August 27, 2013	Leo	22	0.2373
Genus	October 1, 2013	Pipe	30	0.0488*
Family	November 11, 2012	Leo	24	0.0842
Family	March 13, 2013	Creslm	24	0.1392
Family	May 7, 2013	Leo	29	0.0049*
Family	July 9, 2013	Creslm	28	<0.0001*
Family	July 9, 2013	Leo	22	0.0084*
Family	July 22, 2013	Leo	21	0.1192
Family	August 13, 2013	Leo	20	0.0427
Family	August 27, 2013	Leo	17	0.2531
Family	October 1, 2013	Pipe	26	0.0360*
Order	November 11, 2012	Leo	18	0.0785
Order	March 13, 2013	Creslm	17	0.2410
Order	May 7, 2013	Leo	21	0.0142*
Order	July 9, 2013	Creslm	21	<0.0001*
Order	July 9, 2013	Leo	17	0.0159*
Order	July 22, 2013	Leo	16	0.1310
Order	August 13, 2013	Leo	16	0.0848
Order	August 27, 2013	Leo	13	0.2319
Order	October 1, 2013	Pipe	20	0.0706
Class	November 11, 2012	Leo	11	0.0823
Class	March 13, 2013	Creslm	12	0.2242
Class	May 7, 2013	Leo	14	0.0165*
Class	July 9, 2013	Creslm	15	0.0007*
Class	July 9, 2013	Leo	11	0.0260*
Class	July 22, 2013	Leo	11	0.0400*
Class	August 13, 2013	Leo	11	0.0585
Class	August 27, 2013	Leo	8	0.2142
Class	October 1, 2013	Pipe	14	0.0569
Phylum	November 11, 2012	Leo	8	0.0757
Phylum	March 13, 2013	Creslm	9	0.3605
Phylum	May 7, 2013	Leo	10	0.0998
Phylum	July 9, 2013	Creslm	10	0.0061*
Phylum	July 9, 2013	Leo	8	0.0913
Phylum	July 22, 2013	Leo	7	0.1644
Phylum	August 13, 2013	Leo	9	0.0753

Table E-3. Continued				
Taxa Level	Date	Station	N samples	p-Value
Phylum	August 27, 2013	Leo	6	0.2288
Phylum	October 1, 2013	Pipe	10	0.0903

APPENDIX F
SUMMARY ANALYSES AUTOMATICALLY EXPORTED FROM THE FLUID-IMAGING
FLOWCAM® FOR ONE LAKE LOCHLOOSA SAMPLE

	Genus	Particles / ml	Summary Stats	Mean	Min	Max	StdDev	CV%
3/17/13	Cosmarium	43	Area (ABD)	239.72	176.77	402.28	108.74	45.36
3/17/13	Cosmarium	43	Aspect Ratio	0.6	0.37	0.73	0.16	26.28
3/17/13	Cosmarium	43	Ch1 Peak	0	0	0	0	0
3/17/13	Cosmarium	43	Ch2 Peak	0	0	0	0	0
3/17/13	Cosmarium	43	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Cosmarium	43	Circle Fit	0.52	0.21	0.74	0.23	44.42
3/17/13	Cosmarium	43	Circularity	0.65	0.51	0.85	0.15	22.5
3/17/13	Cosmarium	43	Circularity (Hu)	0.83	0.63	0.94	0.14	16.59
3/17/13	Cosmarium	43	Compactness	1.59	1.18	1.97	0.34	21.08
3/17/13	Cosmarium	43	Diameter (ABD)	17.18	15	22.63	3.65	21.25
3/17/13	Cosmarium	43	Diameter (ESD)	19.75	16.25	27.91	5.47	27.68
3/17/13	Cosmarium	43	Geodesic Aspect Ratio	0.51	0.25	1	0.34	67.07
3/17/13	Cosmarium	43	Geodesic Length	27.52	15.13	39.05	10.17	36.94
3/17/13	Cosmarium	43	Geodesic Thickness	11.66	7.97	15.13	3.03	25.96
3/17/13	Cosmarium	43	Intensity	115.47	88.36	161.66	32.01	27.72
3/17/13	Cosmarium	43	Length	23.85	18.29	36.46	8.47	35.51
3/17/13	Cosmarium	43	Perimeter	78.36	60.52	103.54	18.36	23.43
3/17/13	Cosmarium	43	Roughness	1.21	1.06	1.53	0.21	17.45
3/17/13	Cosmarium	43	Transparency	0.12	0.06	0.19	0.06	46.01
3/17/13	Cosmarium	43	Volume (ABD)	2944.36	1767.91	6069.58	2088	70.92
3/17/13	Cosmarium	43	Volume (ESD)	4793.52	2245.41	11386.87	4402.57	91.84
3/17/13	Cosmarium	43	Width	13.4	12.78	13.85	0.5	3.76
3/17/13	Staurastrum	183	Area (ABD)	598.98	272.9	2270.8	476.94	79.63
3/17/13	Staurastrum	183	Aspect Ratio	0.73	0.42	0.95	0.16	21.39
3/17/13	Staurastrum	183	Ch1 Peak	0	0	0	0	0
3/17/13	Staurastrum	183	Ch2 Peak	0	0	0	0	0
3/17/13	Staurastrum	183	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Staurastrum	183	Circle Fit	0.3	0	0.49	0.13	45.56
3/17/13	Staurastrum	183	Circularity	0.24	0.14	0.37	0.06	25.91
3/17/13	Staurastrum	183	Circularity (Hu)	0.72	0.5	0.83	0.09	12.77
3/17/13	Staurastrum	183	Compactness	4.42	2.74	7.2	1.22	27.6
3/17/13	Staurastrum	183	Diameter (ABD)	26.35	18.64	53.77	8.53	32.36
3/17/13	Staurastrum	183	Diameter (ESD)	36.08	23.84	75.14	12.23	33.9
3/17/13	Staurastrum	183	Geodesic Aspect Ratio	0.09	0.05	0.16	0.03	31.39

3/17/13	Staurastrum	183	Geodesic Length	89.57	49.93	216.31	39.91	44.55
3/17/13	Staurastrum	183	Geodesic Thickness	7.58	4.92	11.57	1.61	21.23
3/17/13	Staurastrum	183	Intensity	123.22	91.91	146.39	16.71	13.56
3/17/13	Staurastrum	183	Length	41.26	26.78	93.25	15.81	38.33
3/17/13	Staurastrum	183	Perimeter	194.31	115.37	455.76	81.78	42.09
3/17/13	Staurastrum	183	Roughness	1.68	1.28	1.99	0.19	11.38
3/17/13	Staurastrum	183	Transparency	0.27	0.22	0.35	0.03	11.75
3/17/13	Staurastrum	183	Volume (ABD)	13039.84	3391.26	81401.3	18561.16	142.34
3/17/13	Staurastrum	183	Volume (ESD)	34366.31	7097.58	2.22E+05	50779.33	147.76
3/17/13	Staurastrum	183	Width	29.23	20.17	52.37	7.23	24.74
3/17/13	pennate diatoms	280	Area (ABD)	130.49	81.03	250.67	45.62	34.96
3/17/13	pennate diatoms	280	Aspect Ratio	0.21	0.1	0.6	0.11	54.47
3/17/13	pennate diatoms	280	Ch1 Peak	0	0	0	0	0
3/17/13	pennate diatoms	280	Ch2 Peak	0	0	0	0	0
3/17/13	pennate diatoms	280	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	pennate diatoms	280	Circle Fit	0.07	0	0.45	0.12	178.19
3/17/13	pennate diatoms	280	Circularity	0.19	0.06	0.41	0.09	49.51
3/17/13	pennate diatoms	280	Circularity (Hu)	0.32	0.14	0.72	0.15	47.49
3/17/13	pennate diatoms	280	Compactness	7	2.45	17.42	4.11	58.71
3/17/13	pennate diatoms	280	Diameter (ABD)	12.71	10.16	17.87	2.16	17.02
3/17/13	pennate diatoms	280	Diameter (ESD)	27.54	15.51	47.12	9.4	34.14
3/17/13	pennate diatoms	280	Geodesic Aspect Ratio	0.07	0.02	0.18	0.04	57.32
3/17/13	pennate diatoms	280	Geodesic Length	58.7	26.63	110.1	22.72	38.72
3/17/13	pennate diatoms	280	Geodesic Thickness	3.46	2.02	5.63	1.01	29.29
3/17/13	pennate diatoms	280	Intensity	156.51	126.13	169.87	10.33	6.6
3/17/13	pennate diatoms	280	Length	41.53	18.78	73.81	15.54	37.42
3/17/13	pennate diatoms	280	Perimeter	124.31	62.89	224.63	44.17	35.53
3/17/13	pennate diatoms	280	Roughness	1.39	1.14	1.78	0.19	14.05
3/17/13	pennate diatoms	280	Transparency	0.5	0.28	0.7	0.12	23.89
3/17/13	pennate diatoms	280	Volume (ABD)	1169.16	548.69	2985.57	626.46	53.58
3/17/13	pennate diatoms	280	Volume (ESD)	14802.03	1953.28	54766.88	14372.29	97.1
3/17/13	pennate diatoms	280	Width	6.71	3.63	10.26	1.58	23.6
3/17/13	Microcystis	852	Area (ABD)	718.47	80.78	4070.17	774.04	107.73
3/17/13	Microcystis	852	Aspect Ratio	0.67	0.33	0.98	0.16	23.61
3/17/13	Microcystis	852	Ch1 Peak	0	0	0	0	0
3/17/13	Microcystis	852	Ch2 Peak	0	0	0	0	0
3/17/13	Microcystis	852	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Microcystis	852	Circle Fit	0.44	0	0.83	0.22	50.37
3/17/13	Microcystis	852	Circularity	0.35	0.04	0.68	0.13	38.31
3/17/13	Microcystis	852	Circularity (Hu)	0.8	0.36	0.99	0.14	18.18
3/17/13	Microcystis	852	Compactness	3.66	1.48	26.61	3.2	87.47
3/17/13	Microcystis	852	Diameter (ABD)	26.8	10.14	71.99	14.11	52.63

3/17/13	Microcystis	852	Diameter (ESD)	33.04	12.31	78.86	16.58	50.19
3/17/13	Microcystis	852	Geodesic Aspect Ratio	0.16	0.01	0.46	0.09	53.96
3/17/13	Microcystis	852	Geodesic Length	78.06	17.38	232.04	51.21	65.6
3/17/13	Microcystis	852	Geodesic Thickness	9.69	2.65	28.68	4.82	49.7
3/17/13	Microcystis	852	Intensity	130.84	84.22	166.06	18.7	14.29
3/17/13	Microcystis	852	Length	38.96	14.03	91.58	19.93	51.16
3/17/13	Microcystis	852	Perimeter	175.51	50.71	475.08	106.4	60.62
3/17/13	Microcystis	852	Roughness	1.7	1.14	3.44	0.48	27.97
3/17/13	Microcystis	852	Transparency	0.19	0.06	0.46	0.07	38.14
3/17/13	Microcystis	852	Volume (ABD)	19798	546.15	1.95E+05	32817.46	165.76
3/17/13	Microcystis	852	Volume (ESD)	34780.59	977.7	2.57E+05	49411.45	142.07
3/17/13	Microcystis	852	Width	25.77	6.34	65.2	13.66	53
3/17/13	Chroococcus	237	Area (ABD)	387.04	82.28	955.13	288.5	74.54
3/17/13	Chroococcus	237	Aspect Ratio	0.71	0.35	0.95	0.18	25.33
3/17/13	Chroococcus	237	Ch1 Peak	0	0	0	0	0
3/17/13	Chroococcus	237	Ch2 Peak	0	0	0	0	0
3/17/13	Chroococcus	237	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Chroococcus	237	Circle Fit	0.4	0	0.79	0.23	56.58
3/17/13	Chroococcus	237	Circularity	0.32	0.05	0.55	0.14	44.11
3/17/13	Chroococcus	237	Circularity (Hu)	0.76	0.43	0.97	0.16	21.46
3/17/13	Chroococcus	237	Compactness	4.42	1.81	21.15	4.16	94.01
3/17/13	Chroococcus	237	Diameter (ABD)	20.8	10.24	34.87	7.93	38.14
3/17/13	Chroococcus	237	Diameter (ESD)	27.79	13.26	57.49	12.21	43.95
3/17/13	Chroococcus	237	Geodesic Aspect Ratio	0.14	0.02	0.29	0.08	54.86
3/17/13	Chroococcus	237	Geodesic Length	70.39	26	272.18	54.4	77.28
3/17/13	Chroococcus	237	Geodesic Thickness	7.3	3.36	15.3	3.07	42.03
3/17/13	Chroococcus	237	Intensity	143.49	95.04	169.93	18.4	12.82
3/17/13	Chroococcus	237	Length	32.5	15.95	75.57	14.74	45.35
3/17/13	Chroococcus	237	Perimeter	155.4	61.83	552.82	108.08	69.55
3/17/13	Chroococcus	237	Roughness	1.77	1.25	3.31	0.48	27.18
3/17/13	Chroococcus	237	Transparency	0.23	0.1	0.45	0.09	38.24
3/17/13	Chroococcus	237	Volume (ABD)	6807.61	561.46	22205.4	7262.3	106.68
3/17/13	Chroococcus	237	Volume (ESD)	18309.43	1221.6	99489.14	25363.62	138.53
3/17/13	Chroococcus	237	Width	21.45	9.59	39.92	8.67	40.42
3/17/13	Unknown	270	Area (ABD)	145.29	78.78	292.47	55.22	38.01
3/17/13	Unknown	270	Aspect Ratio	0.68	0.5	0.98	0.14	20.48
3/17/13	Unknown	270	Ch1 Peak	0	0	0	0	0
3/17/13	Unknown	270	Ch2 Peak	0	0	0	0	0
3/17/13	Unknown	270	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Unknown	270	Circle Fit	0.66	0.36	0.87	0.14	21.81
3/17/13	Unknown	270	Circularity	0.71	0.27	0.89	0.14	20.23

3/17/13	Unknown	270	Circularity (Hu)	0.88	0.57	0.98	0.09	10.5
3/17/13	Unknown	270	Compactness	1.51	1.13	3.72	0.52	34.31
3/17/13	Unknown	270	Diameter					
3/17/13	Unknown	270	(ABD)	13.39	10.02	19.3	2.47	18.42
3/17/13	Unknown	270	Diameter					
3/17/13	Unknown	270	(ESD)	15.09	11.06	21.53	2.67	17.67
3/17/13	Unknown	270	Geodesic					
3/17/13	Unknown	270	Aspect Ratio	0.62	0.1	1	0.29	47.09
3/17/13	Unknown	270	Geodesic					
3/17/13	Unknown	270	Length	19.7	10.64	35.89	6.81	34.57
3/17/13	Unknown	270	Geodesic					
3/17/13	Unknown	270	Thickness	10.58	3.74	16.16	2.8	26.5
3/17/13	Unknown	270	Intensity	98.99	73.46	136.56	17.67	17.85
3/17/13	Unknown	270	Length	18.1	12.8	24.5	3.36	18.57
3/17/13	Unknown	270	Perimeter	60.55	42.56	87.36	11.63	19.2
3/17/13	Unknown	270	Roughness	1.13	1.05	1.42	0.08	7.2
3/17/13	Unknown	270	Transparency	0.11	0.04	0.33	0.06	50.03
3/17/13	Unknown	270	Volume (ABD)	1383.32	526.01	3762.49	810.47	58.59
3/17/13	Unknown	270	Volume (ESD)	1967.99	707.46	5222.57	1088.62	55.32
3/17/13	Unknown	270	Width	11.56	7.33	19.71	2.57	22.26
3/17/13	Planktolyngbya	367	Area (ABD)	400.59	85.8	1140.28	255.52	63.79
3/17/13	Planktolyngbya	367	Aspect Ratio	0.66	0.19	0.97	0.24	35.68
3/17/13	Planktolyngbya	367	Ch1 Peak	0	0	0	0	0
3/17/13	Planktolyngbya	367	Ch2 Peak	0	0	0	0	0
3/17/13	Planktolyngbya	367	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Planktolyngbya	367	Circle Fit	0.4	0	0.87	0.27	66.54
3/17/13	Planktolyngbya	367	Circularity	0.37	0.1	0.8	0.18	48.7
3/17/13	Planktolyngbya	367	Circularity (Hu)	0.73	0.29	1	0.24	32.69
3/17/13	Planktolyngbya	367	Compactness	3.52	1.25	9.77	2.09	59.45
3/17/13	Planktolyngbya	367	Diameter					
3/17/13	Planktolyngbya	367	(ABD)	21.53	10.45	38.1	6.91	32.09
3/17/13	Planktolyngbya	367	Diameter					
3/17/13	Planktolyngbya	367	(ESD)	27.53	14.68	45.28	6.94	25.22
3/17/13	Planktolyngbya	367	Geodesic					
3/17/13	Planktolyngbya	367	Aspect Ratio	0.2	0.03	1	0.2	99.66
3/17/13	Planktolyngbya	367	Geodesic					
3/17/13	Planktolyngbya	367	Length	59.08	23.68	111.44	23.34	39.5
3/17/13	Planktolyngbya	367	Geodesic					
3/17/13	Planktolyngbya	367	Thickness	9.43	3.18	25.56	5.58	59.2
3/17/13	Planktolyngbya	367	Intensity	161.77	143.91	175.92	8.69	5.37
3/17/13	Planktolyngbya	367	Length	32.36	19.78	53.32	7.35	22.71
3/17/13	Planktolyngbya	367	Perimeter	137.01	63.14	230.66	44.83	32.72
3/17/13	Planktolyngbya	367	Roughness	1.72	1.16	2.62	0.36	20.74
3/17/13	Planktolyngbya	367	Transparency	0.23	0.07	0.47	0.1	44.93
3/17/13	Planktolyngbya	367	Volume (ABD)	6875.33	597.82	28965.31	6606.84	96.09
3/17/13	Planktolyngbya	367	Volume (ESD)	13052.27	1656.49	48619.24	10476.41	80.27
3/17/13	Planktolyngbya	367	Width	21.29	5.76	34.62	8.17	38.4
3/17/13	Detritus	8108	Area (ABD)	524.29	78.53	7533.33	906.36	172.87
3/17/13	Detritus	8108	Aspect Ratio	0.64	0.07	0.99	0.19	28.9
3/17/13	Detritus	8108	Ch1 Peak	0	0	0	0	0

3/17/13	Detritus	8108	Ch2 Peak	0	0	0	0	0
3/17/13	Detritus	8108	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Detritus	8108	Circle Fit	0.4	0	0.9	0.22	55.38
3/17/13	Detritus	8108	Circularity	0.35	0.04	0.94	0.19	53.99
3/17/13	Detritus	8108	Circularity (Hu)	0.74	0.06	1	0.19	25.3
3/17/13	Detritus	8108	Compactness	4.07	1.07	26.1	2.97	72.84
3/17/13	Detritus	8108	Diameter (ABD)	21.36	10	97.94	14.54	68.09
3/17/13	Detritus	8108	Diameter (ESD)	28.5	10.85	141.79	18.57	65.15
3/17/13	Detritus	8108	Geodesic Aspect Ratio	0.18	0.01	1	0.19	101.82
3/17/13	Detritus	8108	Geodesic Length	68.61	10.28	544.98	59.1	86.14
3/17/13	Detritus	8108	Geodesic Thickness	7.95	1.57	41.67	4.74	59.59
3/17/13	Detritus	8108	Intensity	143.64	59.82	202.44	19.66	13.69
3/17/13	Detritus	8108	Length	34.45	12.01	195.66	22.99	66.72
3/17/13	Detritus	8108	Perimeter	153.13	41.12	1122.86	121.85	79.58
3/17/13	Detritus	8108	Roughness	1.78	1.03	5.7	0.53	29.79
3/17/13	Detritus	8108	Transparency	0.24	0.04	0.72	0.11	45.64
3/17/13	Detritus	8108	Volume (ABD)	15836.7	523.51	4.92E+05	47863.64	302.23
3/17/13	Detritus	8108	Volume (ESD)	34743.07	669.18	1.49E+06	1.05E+05	301.41
3/17/13	Detritus	8108	Width	20.91	4.19	114.54	14.43	68.99
3/17/13	Limnothrix	205	Area (ABD)	217.63	82.28	785.08	197.88	90.92
3/17/13	Limnothrix	205	Aspect Ratio	0.21	0.08	0.76	0.16	73.29
3/17/13	Limnothrix	205	Ch1 Peak	0	0	0	0	0
3/17/13	Limnothrix	205	Ch2 Peak	0	0	0	0	0
3/17/13	Limnothrix	205	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Limnothrix	205	Circle Fit	0.05	0	0.45	0.12	245.47
3/17/13	Limnothrix	205	Circularity	0.23	0.08	0.53	0.12	51.43
3/17/13	Limnothrix	205	Circularity (Hu)	0.27	0.09	0.69	0.13	49.62
3/17/13	Limnothrix	205	Compactness	5.77	1.89	12.64	3.23	55.89
3/17/13	Limnothrix	205	Diameter (ABD)	15.57	10.24	31.62	6.05	38.87
3/17/13	Limnothrix	205	Diameter (ESD)	32.73	13.52	80.67	16.42	50.16
3/17/13	Limnothrix	205	Geodesic Aspect Ratio	0.09	0.03	0.27	0.06	66.11
3/17/13	Limnothrix	205	Geodesic Length	65.84	22.07	190.59	43.28	65.72
3/17/13	Limnothrix	205	Geodesic Thickness	4.34	2.84	6.03	1.07	24.65
3/17/13	Limnothrix	205	Intensity	161.87	145.99	173.42	8.76	5.41
3/17/13	Limnothrix	205	Length	47.39	17.47	106.43	21.36	45.07
3/17/13	Limnothrix	205	Perimeter	140.37	56.16	391.3	86.64	61.72
3/17/13	Limnothrix	205	Roughness	1.26	1.08	1.73	0.19	15.24
3/17/13	Limnothrix	205	Transparency	0.49	0.22	0.66	0.11	22.44
3/17/13	Limnothrix	205	Volume (ABD)	3006.09	561.46	16547.43	4505.38	149.87
3/17/13	Limnothrix	205	Volume (ESD)	34586.39	1294.36	2.75E+05	63264.94	182.92

3/17/13	Limnothrix	205	Width	10.3	3.57	43.55	11.79	114.49
3/17/13	Closterium	54	Area (ABD)	271.56	94.65	528.72	162.63	59.89
3/17/13	Closterium	54	Aspect Ratio	0.25	0.16	0.32	0.06	25.44
3/17/13	Closterium	54	Ch1 Peak	0	0	0	0	0
3/17/13	Closterium	54	Ch2 Peak	0	0	0	0	0
3/17/13	Closterium	54	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Closterium	54	Circle Fit	0.05	0	0.26	0.12	223.61
3/17/13	Closterium	54	Circularity	0.21	0.12	0.44	0.13	64.49
3/17/13	Closterium	54	Circularity (Hu)	0.37	0.24	0.54	0.12	31.12
3/17/13	Closterium	54	Compactness	6.06	2.28	8.55	2.47	40.74
3/17/13	Closterium	54	Diameter					
3/17/13	Closterium	54	(ABD)	17.93	10.98	25.95	5.49	30.62
3/17/13	Closterium	54	Diameter					
3/17/13	Closterium	54	(ESD)	32.64	15.38	49.22	12.9	39.51
3/17/13	Closterium	54	Geodesic					
3/17/13	Closterium	54	Aspect Ratio	0.08	0.04	0.2	0.07	82.26
3/17/13	Closterium	54	Geodesic					
3/17/13	Closterium	54	Length	76.92	26.63	128.58	36.49	47.45
3/17/13	Closterium	54	Geodesic					
3/17/13	Closterium	54	Thickness	4.67	3.67	5.37	0.76	16.27
3/17/13	Closterium	54	Intensity	150.27	130.8	164.81	12.51	8.33
3/17/13	Closterium	54	Length	47.59	21.75	71.95	19.43	40.83
3/17/13	Closterium	54	Perimeter	163.18	64	267.53	72.87	44.65
3/17/13	Closterium	54	Roughness	1.58	1.18	2.12	0.43	27.06
3/17/13	Closterium	54	Transparency	0.43	0.29	0.51	0.09	20.52
3/17/13	Closterium	54	Volume (ABD)	3718.61	692.65	9145.46	3262.74	87.74
3/17/13	Closterium	54	Volume (ESD)	25017.08	1903.76	62447.15	24183.34	96.67
3/17/13	Closterium	54	Width	11.77	6.59	16.48	3.57	30.36
3/17/13	Scenedesmus	97	Area (ABD)	113.11	82.03	179.98	29.24	25.85
3/17/13	Scenedesmus	97	Aspect Ratio	0.64	0.4	0.83	0.15	24.26
3/17/13	Scenedesmus	97	Ch1 Peak	0	0	0	0	0
3/17/13	Scenedesmus	97	Ch2 Peak	0	0	0	0	0
3/17/13	Scenedesmus	97	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Scenedesmus	97	Circle Fit	0.6	0.18	0.77	0.18	30.64
3/17/13	Scenedesmus	97	Circularity	0.59	0.37	0.8	0.13	22.81
3/17/13	Scenedesmus	97	Circularity (Hu)	0.84	0.56	0.95	0.13	15.24
3/17/13	Scenedesmus	97	Compactness	1.78	1.24	2.72	0.43	24.32
3/17/13	Scenedesmus	97	Diameter					
3/17/13	Scenedesmus	97	(ABD)	11.92	10.22	15.14	1.46	12.24
3/17/13	Scenedesmus	97	Diameter					
3/17/13	Scenedesmus	97	(ESD)	14.17	12.75	16.62	1.46	10.29
3/17/13	Scenedesmus	97	Geodesic					
3/17/13	Scenedesmus	97	Aspect Ratio	0.44	0.16	1	0.32	72.6
3/17/13	Scenedesmus	97	Geodesic					
3/17/13	Scenedesmus	97	Length	21.68	12.91	32.93	5.66	26.1
3/17/13	Scenedesmus	97	Geodesic					
3/17/13	Scenedesmus	97	Thickness	8.23	5.15	15.46	3.52	42.78
3/17/13	Scenedesmus	97	Intensity	120.96	99.4	141.48	14.16	11.7
3/17/13	Scenedesmus	97	Length	16.95	13.94	21.3	2.39	14.08
3/17/13	Scenedesmus	97	Perimeter	59.82	51.64	76.16	7.24	12.11

3/17/13	Scenedesmus	97	Roughness	1.18	1.07	1.32	0.08	6.35
3/17/13	Scenedesmus	97	Transparency	0.16	0.09	0.26	0.06	36.03
3/17/13	Scenedesmus	97	Volume (ABD)	924.24	558.89	1816.28	378.3	40.93
3/17/13	Scenedesmus	97	Volume (ESD)	1531.72	1085.2	2402.96	501.81	32.76
3/17/13	Scenedesmus	97	Width	10.26	8.88	12.4	1.46	14.26
3/17/13	Animal	22	Area (ABD)	291.16	221.23	361.09	98.9	33.97
3/17/13	Animal	22	Aspect Ratio	0.77	0.76	0.78	0.01	1.56
3/17/13	Animal	22	Ch1 Peak	0	0	0	0	0
3/17/13	Animal	22	Ch2 Peak	0	0	0	0	0
3/17/13	Animal	22	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Animal	22	Circle Fit	0.67	0.58	0.76	0.12	18.1
3/17/13	Animal	22	Circularity	0.52	0.43	0.62	0.13	25.56
3/17/13	Animal	22	Circularity (Hu)	0.92	0.89	0.96	0.05	5.43
3/17/13	Animal	22	Compactness	1.98	1.63	2.34	0.51	25.56
3/17/13	Animal	22	Diameter (ABD)	19.11	16.78	21.44	3.29	17.24
3/17/13	Animal	22	Diameter (ESD)	21.89	20.37	23.41	2.15	9.82
3/17/13	Animal	22	Geodesic Aspect Ratio	0.28	0.19	0.36	0.12	43.34
3/17/13	Animal	22	Geodesic Length	37.02	35.17	38.88	2.63	7.09
3/17/13	Animal	22	Geodesic Thickness	10.17	7.52	12.82	3.74	36.82
3/17/13	Animal	22	Intensity	116.59	111.86	121.32	6.69	5.73
3/17/13	Animal	22	Length	24.53	22.58	26.49	2.77	11.28
3/17/13	Animal	22	Perimeter	94.39	92.81	95.97	2.23	2.37
3/17/13	Animal	22	Roughness	1.26	1.21	1.32	0.08	6.5
3/17/13	Animal	22	Transparency	0.13	0.08	0.18	0.07	50.07
3/17/13	Animal	22	Volume (ABD)	3818.56	2475.39	5161.73	1899.53	49.74
3/17/13	Animal	22	Volume (ESD)	5569.38	4424.05	6714.71	1619.74	29.08
3/17/13	Animal	22	Width	18.53	17.46	19.6	1.51	8.16
3/17/13	Ankistrodesmus	22	Area (ABD)	231.84	139.91	323.76	130	56.07
3/17/13	Ankistrodesmus	22	Aspect Ratio	0.65	0.54	0.77	0.17	25.6
3/17/13	Ankistrodesmus	22	Ch1 Peak	0	0	0	0	0
3/17/13	Ankistrodesmus	22	Ch2 Peak	0	0	0	0	0
3/17/13	Ankistrodesmus	22	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Ankistrodesmus	22	Circle Fit	0.24	0.05	0.42	0.26	110.7
3/17/13	Ankistrodesmus	22	Circularity	0.26	0.16	0.36	0.15	56.39
3/17/13	Ankistrodesmus	22	Circularity (Hu)	0.66	0.49	0.84	0.25	37.8
3/17/13	Ankistrodesmus	22	Compactness	4.56	2.74	6.37	2.57	56.39
3/17/13	Ankistrodesmus	22	Diameter (ABD)	16.83	13.35	20.3	4.92	29.24
3/17/13	Ankistrodesmus	22	Diameter (ESD)	23.99	16.18	31.8	11.04	46.03
3/17/13	Ankistrodesmus	22	Geodesic Aspect Ratio	0.11	0.06	0.15	0.07	66.69
3/17/13	Ankistrodesmus	22	Geodesic Length	60.91	35.71	86.11	35.64	58.51

			Geodesic					
3/17/13	Ankistrodesmus	22	Thickness	5.16	4.79	5.53	0.52	10.15
3/17/13	Ankistrodesmus	22	Intensity	151.12	149.45	152.79	2.36	1.56
3/17/13	Ankistrodesmus	22	Length	29.51	18.22	40.79	15.96	54.1
3/17/13	Ankistrodesmus	22	Perimeter	132.14	82.48	181.8	70.23	53.15
3/17/13	Ankistrodesmus	22	Roughness	1.6	1.44	1.77	0.23	14.33
3/17/13	Ankistrodesmus	22	Transparency	0.27	0.18	0.36	0.13	49.11
3/17/13	Ankistrodesmus	22	Volume (ABD)	2813.65	1244.95	4382.34	2218.47	78.85
3/17/13	Ankistrodesmus	22	Volume (ESD)	9526.04	2218.25	16833.83	10334.77	108.49
3/17/13	Ankistrodesmus	22	Width	17.86	14.05	21.67	5.39	30.18
3/17/13	Dictyosphaerium	11	Area (ABD)	140.7	140.7	140.7	0	0
3/17/13	Dictyosphaerium	11	Aspect Ratio	0.49	0.49	0.49	0	0
3/17/13	Dictyosphaerium	11	Ch1 Peak	0	0	0	0	0
3/17/13	Dictyosphaerium	11	Ch2 Peak	0	0	0	0	0
3/17/13	Dictyosphaerium	11	Ch2/Ch1 Ratio	0	0	0	0	0
3/17/13	Dictyosphaerium	11	Circle Fit	0.12	0.12	0.12	0	0
3/17/13	Dictyosphaerium	11	Circularity	0.16	0.16	0.16	0	0
3/17/13	Dictyosphaerium	11	Circularity (Hu)	0.43	0.43	0.43	0	0
3/17/13	Dictyosphaerium	11	Compactness	6.42	6.42	6.42	0	0
3/17/13	Dictyosphaerium	11	Diameter (ABD)	13.38	13.38	13.38	0	0
3/17/13	Dictyosphaerium	11	Diameter (ESD)	20.65	20.65	20.65	0	0
3/17/13	Dictyosphaerium	11	Geodesic Aspect Ratio	0.06	0.06	0.06	0	0
3/17/13	Dictyosphaerium	11	Geodesic Length	59.96	59.96	59.96	0	0
3/17/13	Dictyosphaerium	11	Geodesic Thickness	3.31	3.31	3.31	0	0
3/17/13	Dictyosphaerium	11	Intensity	151.47	151.47	151.47	0	0
3/17/13	Dictyosphaerium	11	Length	25.35	25.35	25.35	0	0
3/17/13	Dictyosphaerium	11	Perimeter	126.54	126.54	126.54	0	0
3/17/13	Dictyosphaerium	11	Roughness	1.77	1.77	1.77	0	0
3/17/13	Dictyosphaerium	11	Transparency	0.35	0.35	0.35	0	0
3/17/13	Dictyosphaerium	11	Volume (ABD)	1255.49	1255.49	1255.49	0	0
3/17/13	Dictyosphaerium	11	Volume (ESD)	4610.88	4610.88	4610.88	0	0
3/17/13	Dictyosphaerium	11	Width	13.85	13.85	13.85	0	0

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BIOGRAPHICAL SKETCH

Dawn Davis grew up in south Florida in a small town, Wauchula, and attended the University of Florida for her undergraduate degree in biology. Dawn's intentions were to become a dentist and then specialize in orthodontics. Dawn had the opportunity to shadow her dentists for many years and also volunteered in many laboratories at the Fisheries and Aquatic Sciences Program. She graduated in 2012 from the University of Florida majoring in biology with an emphasis on pre-profession and minoring in fisheries and aquatic sciences. Thereafter, Dawn started her master's degree with Dr. Daniel E. Canfield, Jr. at the University of Florida Program of Fisheries and Aquatic Sciences. In her spare time, Dawn enjoys being creative and crafty, playing all kinds of sports and spending her days in outdoor adventures.