

PILOT STUDY:

**MONITORING SUMMER METAPHYTON
GROWTH ALONG THE SOUTH AND
SOUTHEAST SHORE OF LAKE TAHOE**



FINAL REPORT

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Final Report

Pilot Study: Monitoring Summer Metaphyton Growth along the South and
Southeast shore of Lake Tahoe

Submitted to:
Nevada Division of State Lands

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Executive Summary

In the summers of 2015-2017, a pilot study was done by the UC Davis Tahoe Environmental Research Center (TERC) to develop and test methods that could be used to monitor metaphyton algae in the nearshore of Lake Tahoe. Metaphyton is algae which is neither strictly attached to a substrate (like periphyton) nor truly planktonic (like phytoplankton). In Lake Tahoe, the metaphyton is typically composed of aggregations of green filamentous algae ranging from small clumps of algae rolling between sand ripples on the shallow lake bottom, to larger clouds of algae hovering above, or resting on the bottom. Undesirable levels of algae can develop resulting in clouding of the nearshore waters and its deposition along the shore. Once on the shore, it forms accumulations that eventually decompose, but in the process, it can be both extremely unsightly and malodorous. Anecdotal evidence suggests that levels of metaphyton may have increased in recent years; however, little data has been collected. There currently is no consistent methodology or data collection effort on the occurrence and amounts of metaphyton in the nearshore of Lake Tahoe.

The emphasis in this study was on testing of field and lab methods that would be appropriate for monitoring metaphyton at select sites around the lake. The Lake Tahoe Nearshore Evaluation and Monitoring Framework report (Heyvaert et al., 2013) identified a need to know more about metaphyton and recommended that a few metaphyton monitoring sites should be included as part of periphyton monitoring, especially along the south shore region during summer. For sites such as popular beaches where shoreline users may consider metaphyton undesirable, such monitoring would provide baseline data to begin to assess trends. Such methods would also provide a means to study the potential causes of heavy metaphyton growth (i.e. nutrient loading, lake temperature, Asian clam presence and lake level fluctuation). For regional or lake-wide assessments of metaphyton, and for assessing the contributions of large patches of metaphyton along shore, additional methods such as aerial imaging will likely be needed. An assessment of regional metaphyton monitoring methods is planned in a follow-up study for NDSL to be done in the summers of 2018-2019. Methods found to be effective for metaphyton monitoring in the current study could be used in concert with other methods for regional and lake-wide metaphyton monitoring.

Monitoring methods were evaluated at three sites along the south and southeast shore of Lake Tahoe where metaphyton had been observed in the past: (Regan Beach, El Dorado Beach and Round Hill Pines Beach). The methods used to quantify the metaphyton included: (1) estimates of metaphyton percent coverage in the field; (2) estimates of metaphyton percent coverage from photos taken on site; (3) 60°C dry weight; (4) Ash Free Dry Weight (AFDW); and (5) Chlorophyll *a* concentration. In addition, measurements were made of certain physical features of the sites (i.e. water temperature, depth, distance from shore, thickness of algae along the bottom) and predominant algal types in the metaphyton were identified.

Of the metaphyton monitoring methods investigated, metaphyton percent cover estimated in the field, metaphyton percent cover estimated from photos and AFDW appeared to be the best choices for Lake Tahoe metaphyton monitoring. Visual estimates of metaphyton percent cover in the field were made using a quadrat (a plot used to isolate a standard area for measurement; a 0.25 m² or 1 m² square was used in this study). This proved to be a relatively rapid method to assess the level of metaphyton. The primary time involved with this method is in establishing sampling sites (i.e. marking out sites based on grids or randomization), recording GPS locations. Actual measures of percent cover in the field take only a few minutes per quadrat, additional time is required for photography (several minutes) and biomass sampling (5-10 minutes) when done. When data were compared among individual quadrats, field estimates of percent cover generally showed similar patterns and good linear associations with estimates of cover based on photos. Field estimates of percent cover often, but not always, showed good associations with AFDW biomass.

In this pilot study, we sampled up to 7 quadrats along the shoreline and 9-16 quadrats offshore at sites. Based on the variation in metaphyton percent cover observed in the study, particularly along the shoreline, the number of replicate quadrats required to estimate the mean level relatively accurately needs to be at similar levels or higher. For instance, for a moderately variable set of samples (range 5.4-22.4% cover, mean 15% cover, Std. Dev. = 6.3% cover, n=7) from El Dorado beach on 10/7/15, 9 replicate quadrats were estimated to be required to estimate the mean with 95% confidence interval of 10% cover. For highly variable shoreline metaphyton, i.e. Regan Beach 8/20/15 (range 6-70% cover, mean 36.8% cover Std. Dev. = 28.2%, n=6) 16 replicate quadrats would be needed to estimate the mean with 30% confidence interval.) Sampling with just a few quadrats may not provide an estimate of the mean with enough accuracy to make meaningful comparisons between sites and between years.

Percent coverage estimated from photos taken of metaphyton in quadrats also is a potentially useful method to assess levels of metaphyton. A method to determine percent cover from photos using Photoshop software is presented in this report. Percent cover was determined by counting the number of pixels associated with algae in photos and dividing this number by the total number of pixels enclosed in the quadrat in the photo. The method works well but requires greater overall time (about 10-15 minutes per photo analyzed) than time required to make estimates of coverage visually in the field. It is possible that if this method was adopted, a more automated image interrogation workflow could be developed. The photographic method requires that clear photos of metaphyton in quadrat be made. This is not always easy, as wind can produce ripples and waves that make photography from above the water surface very difficult. Poor water clarity can also limit the ability to obtain good photos underwater. The benefits of the photographic method are that it should provide a repeatable measure of percent cover when algae is accurately identified in processing of photos and the photo provides a permanent record of algal cover at the site. Both the field and photo methods rely on skill of the technician in identifying algae on the bottom.

The levels of percent cover often differed slightly between field estimates and estimates from photos. For instance percent cover estimates for Regan Beach on 8/20/15 along the shoreline were 50%, 70% and 15% estimated in the field and 60%, 78% and 1% based on the estimate from the photo. There were several incidences where the value from the photo was measured as 0% while the field estimate of percent cover was greater than 0%. Possible explanations for these differences were that small amounts of metaphyton were not readily visible in the photos or that the visual estimates for low levels of metaphyton overestimated the amount present. When metaphyton coverage was very high, the field estimates of percent cover were often lower than estimates obtained from photos. Differences between field estimates of cover and estimates from the photos could be reduced with additional training of field researchers in discerning various levels of percent cover. Field guides showing different levels of percent cover on cards or photos might be helpful for researchers to refer to in the field.

AFDW appeared to be a better method than dry weight or chlorophyll *a* for estimation of metaphyton biomass. Part of the reason for this was that AFDW appeared more suitable when substantial sand was present in samples. In this study, we found Lake Tahoe metaphyton can have variable and sometimes large amounts of sand associated with it. This sand contributes to the weight of the sample and as a result, dry weight is not a reliable indicator of algal biomass in the metaphyton. Metaphyton chlorophyll *a* content may also be impacted by amounts of sand in the samples. This is because chlorophyll *a* measurements are made on small subsamples of the metaphyton and differences in the proportion of sand can result in different estimates of chlorophyll *a*. Chlorophyll *a* further may not be ideal for algal biomass determination in metaphyton since the algae composing the metaphyton potentially can be a combination of various types of algae, and algae in different states of health and degradation – all of which may result in variable chlorophyll *a* content. AFDW measures the loss in weight samples dried at 60°C after combustion of the organic matter at high temperature (500°C). If other obvious organic matter (i.e. large pieces of plants and wood are removed) the AFDW should be primarily due to the metaphyton. Variability in sand content should have less of an impact on AFDW than for chlorophyll *a* since a much larger subsample size is used for AFDW. A particular benefit of the AFDW method is that it may not be necessary to separate sand from algal material.¹ AFDW measurements provide rapid estimates of the organic matter present in the samples and can easily be done in the lab over a 24-hour period.

Comparing associations of measures of percent coverage with measures of metaphyton biomass at the sites, AFDW showed the most frequent strong associations with field estimates of percent cover (*r* values were ≥ 0.80 in 7 of 10 groups of data) and estimates of percent cover calculated from photos (*r* values were ≥ 0.80 in 8 of 9 groups of data). In cases where the *r*-value was < 0.80

¹ (Separation of sand from algae in the lab was tried in association with dry weight, chlorophyll *a* or AFDW measurements, however the process proved time consuming and effective separation was not always achieved -it proved not a very efficient method for inclusion in routine monitoring).

for the percent cover vs. AFDW association, these occurred for offshore sites where percent cover was relatively low. Chlorophyll *a* had the least frequent strong associations with field estimates of percent cover (r values were ≥ 0.80 in only 6 of 10 groups of data) and estimates of percent cover calculated from photos (r values were ≥ 0.80 in only 5 of 9 groups of data). Comparing associations of measures of metaphyton biomass at the sites, AFDW showed strong associations with both 60°C dry weight and Chlorophyll *a* in all groups of data analyzed (r values were ≥ 0.80 in 10 of 10 groups of data) for both associations. 60°C dry weight and Chlorophyll *a* however, did not show strong associations in all groups (r values were ≥ 0.80 in 7 of 10 groups of data).

Monitoring in this pilot study showed that metaphyton was present offshore during all three summers during 2015-2017 at Regan and El Dorado beaches. This was interesting because metaphyton deposition onshore did not appear to occur in all three years. In 2015 and 2016 when lake level was low, near the natural rim elevation of 6223 ft., moderate amounts (means of ~15-60% cover) of metaphyton were measured along the shoreline at Regan and El Dorado Beaches. However, during the summer of 2017 when lake level was very high (over 6228 ft.) no metaphyton was measured along the shore at El Dorado beach and at Regan there was no beach, only rock lined shoreline, with no deposition of metaphyton there.

Monitoring results and field observations made in this study suggest nearshore slope or gradient and lake level may play a role in the degree to which metaphyton accumulates along the shoreline and is deposited on the beach. Lowered lake levels and minimal slope to the shoreline, favor accumulation of metaphyton right along the shoreline at Regan and El Dorado beaches. In 2015, the lake level was very low, 6222.61 ft. when test monitoring began on 8/6/15 and continued to drop throughout the summer. The lowering lake level left an expansive flat area of exposed lakebed adjacent to El Dorado and Regan beaches. The gradient was so slight that the water depth offshore at Regan and El Dorado was only about 30cm (about a foot deep) at a distance of 137m (150yds.) from shore. Accumulations of metaphyton were observed between sand ripples in the shallow region offshore. When wind and wave energy was sufficient, metaphyton moved along the bottom, including towards the shore at times. With little slope to impede movement of the metaphyton, it accumulated in the shallow areas right along the shoreline at El Dorado and Regan Beaches. There, waves could deposit it onshore or it could be left exposed as the lake level receded. However, in situations where there was a steeper slope along the shoreline, such as in 2017 along El Dorado beach, metaphyton was not observed onshore. At El Dorado Beach in 2017, metaphyton accumulated in a distinct patch offshore near where the steeply sloped beach changes to relatively flat bottom underwater. However, no metaphyton was observed onshore. Bottom topography nearshore may play a role in where metaphyton accumulates in the nearshore.

Members of the public using areas such as Regan Beach and El Dorado beach during low lake level years (such as 2015) have a vastly expanded shoreline to explore. When metaphyton is present in the nearshore in such years, there may be more opportunity to observe it, both as accumulations deposited onshore and as green clumps of algae scattered along the bottom in shallow waters offshore, visible while wading.

As part of the test monitoring, measurements of lake temperature were collected during sampling. Water temperature showed patterns at some sites. At El Dorado Beach during the August and October monitoring, the water temperature tended to decrease away from shore (i.e. the shoreline water temperature in August was about 22°C while at a distance of 137m the water temperature was about 16°C; in October the shoreline water temperature was 21.5°C and offshore was about 15°C.) At Regan Beach in August water temperature was as high as 28°C right at the shoreline and was about 18°C 137 m offshore while in September the temperatures were fairly similar along shore 13-15°C along the shore and 13°C offshore. At Round Hill Pines beach, the water temperatures tended to be similar nearshore and offshore near the surface (i.e. between 17-18°C in August and 14-15°C in September. Increased water temperature right along the shore at El Dorado and Regan beaches in the summer may have had an impact on the biology of the metaphyton. For instance, increases in temperature in shallow water along shore could increase rates of growth of some types of algae tolerant of increased temperatures and potentially increase degradation rates of algae and other organic material. The temperature measurements were only made during the time of sampling in the pilot study. More information on temperature patterns over a 24-hour period would be needed to better assess temperature impacts on metaphyton at the sites.

The monitoring in the study showed that there can be quite a bit of variability in levels of metaphyton along the shoreline. Monitoring evaluated in this pilot study may not be able to characterize fully contributions of larger patches in a region and the behavior of such patches - such as whether they are moving, increasing in size or decreasing. For regional or lake-wide assessments of metaphyton, and for assessing the contributions of large patches of metaphyton along shore, additional methods such as aerial imaging will likely be needed. In 2017, we worked with researchers from the UC Davis Department of Land, Air and Water Resources to undertake some trial imaging from the air (helicopter piloted by Mike Bruno, and UAV flights) with observations of metaphyton in the nearshore at Regan and El Dorado Beaches. Multispectral images were obtained. Regional distribution of larger metaphyton patches were apparent in the photos, thus showing the potential of the technique. Development of aerial methods for determining regional metaphyton distribution, along with semi-automated image processing workflows could be valuable approach in the future. Further assessment of aerial metaphyton monitoring methods is planned in the follow-up study for NDSL to be done in the summers of 2018-2019.

Finally, we took an initial look at some of the observational data on algae recorded by the public in the TERC Citizen Science 2 smartphone App (<https://citizensciencetahoe.org>). We wished to examine the extent to which the public might be voluntarily recording observations of metaphyton that could be used as part of a monitoring program. Seven of fifty-four observations made along the south shore between Baldwin Beach and Zephyr Cove appeared to relate to metaphyton. Of the seven observations, five were made in the El Dorado Beach area in 2016. El Dorado beach is a popular south shore beach, and based on the App results one in which algae along the shoreline is being noted by the public.

Introduction

In the summer of 2015, a pilot study was commenced by UC Davis TERC to develop and test methods that could be used to estimate the distribution and biomass of metaphyton. Anecdotal evidence suggests that levels of metaphyton may have increased in recent years, however, little data have been collected. The Lake Tahoe Nearshore Evaluation and Monitoring Framework report (Heyvaert et al., 2013) identified a need to know more about metaphyton and recommended that a few monitoring sites should be included as part of this framework, especially along the south shore region during summer. There currently is no consistent methodology or data collection effort on the occurrence and amounts of metaphyton in the nearshore of Lake Tahoe. Such information is essential to the determination of basic status and trends, which are core metrics for much of the long-term monitoring occurring throughout the Tahoe basin. Metaphyton is readily apparent to shore zone users during the summer when beaches are heavily used. Pilot study methodology testing and monitoring was done in the summer of 2015 with additional observations and testing in the summers of 2016 and 2017. This report presents the results of these initial pilot studies and monitoring.

Metaphyton is algae which is neither strictly attached to a substrate (like periphyton) nor truly planktonic (like phytoplankton). Wetzel (1975) indicates, “The metaphyton commonly originates from true phytoplankton populations that aggregate among macrophytes and debris of the littoral zone as a result of wind-induced water movements. In other situations, the metaphytonic algae derive from fragmentation of dense epipelagic and epiphytic algal populations. A surprisingly large number of descriptions exist of clustering of metaphytonic algae and macrophytes into “lake balls,” densely packed aggregations of algae or plant parts, or both. These balls are formed by the alternating rolling movements of wave action in the littoral zone (Nakazawa, 1973).”

In Lake Tahoe, the metaphyton is typically composed of aggregations of green filamentous algae ranging from small clumps of algae rolling between sand ripples on the shallow lake bottom, to larger clouds of algae hovering above, or resting on the bottom. Metaphyton can accumulate along portions of the south and southeast shore beaches of Lake Tahoe in the summer and fall. The bright green metaphyton can be quite apparent and visually unappealing to users of the Lake Tahoe shorezone. Occasionally thick blooms of metaphyton have been observed. For example, in 2008 thick metaphyton was observed in Marla Bay along the southeast shore (e.g. see Tahoe Daily Tribune, 2008), and high levels were also observed at some other south shore locations. Metaphyton can accumulate near the shoreline and eventually wash up along the shore to create foul-smelling accumulations of decaying algae (such conditions occurred at Regan Beach in the summer of 2014).

Information collected on Lake Tahoe metaphyton has been relatively limited. Annual visual observations of metaphyton have been made by TERC staff at one or more south shore locations each summer since 2008. Metaphyton presence, observable from piers or along the shoreline, appeared to vary summer-to-summer. In most years, however, observations were made only once in late summer, so it is possible heavier growth may have occurred before or after observations were made. Limited quantitative estimates of metaphyton biomass (as chlorophyll *a*) were made in 2008 and 2009. The results were highly variable ranging from 212 to 0.63 mg/m² (Schladow et al., 2012). Preliminary analysis of metaphyton collected from various south shore areas by the UC Davis Tahoe Environmental Research Center (TERC) prior to the present study indicated it often consisted of one or more types of filamentous (stringy) green algae (i.e. *Spirogyra*, *Mougeotia*, *Zygnema* spp.). Metaphyton has been observed along beaches, and it has also been observed in areas where Asian clams are present (Wittmann et al., 2011; Forrest et al., 2012). Asian clams excrete nutrients which may potentially stimulate algal growth (Wittmann et al., 2011). TERC has also received anecdotal accounts from some long-time users of the nearshore along the south portion of the lake that indicate the levels of metaphyton have increased in the last couple of decades.

The goal of work done for this pilot study was to develop and test methods that could be used to monitor metaphyton algae in the nearshore of Lake Tahoe. The emphasis in this study was on testing field and lab methods appropriate to track levels of metaphyton at select sites around the lake. Along popular beaches, the bright green clumps of metaphyton may be considered aesthetically undesirable by some beachgoers. Monitoring would provide baseline data to begin to assess trends. Such methods could also provide a means to study the potential causes of heavy metaphyton growth. For instance, they could be used to examine linkages of metaphyton to other factors in the nearshore such as nutrient loading, lake temperature, Asian clam presence and lake level fluctuation. For regional or lake-wide assessments of metaphyton, and for assessing the contributions of large patches of metaphyton along shore, additional methods such as aerial imaging will likely be needed. Such methods will be evaluated in a future study to be done in 2018-2019 for NDSL. Ultimately, the methods found to be effective for assessing metaphyton levels and distribution could be employed as part of a regular, long-term monitoring effort to document metaphyton status and trends. This information would assist basin agencies in future management decisions for the nearshore.

The results of pilot study monitoring are presented in the following report together with recommendations for future monitoring.

Methods

Station Site Selection

Stations were chosen at Regan Beach, El Dorado Beach and Round Hill Pines Beach aligned with obvious landmark trees or buildings in the backshore. Starting points for sampling along the shoreline (usually the starting point for center transects) were points with representative levels of metaphyton for the shoreline and lined up with the landmark in the backshore. GPS coordinates were taken of the landmark in the backshore and the starting point for sampling. Distances from the landmark to the starting point were also measured. Marking of the sampling sites took under an hour and was easier in shallow conditions where it was possible to wade compared with deeper conditions which required swimming. Several different sampling designs were used that are described below.

Center Transect Line with Randomized Replicates to Either Side

This method was used in August and September of 2015. A center transect line was established perpendicular to shore, lined up with a landmark in the backshore on the day of sample collections. Metaphyton sampling points were established in a stratified approach to include samples right at the shoreline; samples relatively close to shore (either 3, 10 or 20 yards away) and then at regular distances offshore (typically every 50 yards extending a maximum of 150 yards from shore). Sampling points at randomized distances ≤ 50 yards to the right and left of the center transect line points provided additional replicate samples at set distances from shore.

Center Transect Line with Regular-spaced Replicates to Either Side of the Centerline

This method was used Oct. 7, 2015 at El Dorado Beach for percent cover and photos only collected by one person in the field. A center transect line was established perpendicular to shore, lined up with a landmark in the backshore on the day of sample collections. Metaphyton sampling points were established right at the shoreline then at 25, 50, 100, 150 yards from shore. Samples to the right "R" and left "L" of the center transect points included: shoreline (50 yd. L, 25 yd. L, 25 yd. R, 50 yd. R, 54 yd. R-this was patch of heavy metaphyton we wanted to quantify, 75 yd. R, 100 yd. R), 25yd offshore (25 yd. L, 25 yd. R), 50 yd. offshore (25 yd. L, 25 yd. R), 100yd offshore (50 yd. L, 25 yd. L, 25 yd. R, 50 yd. R), 150 yd. offshore (50 yd. L, 25 yd. L, 25 yd. R, 50 yd. R).

Center Transect Line Only with Sampling Points at Set Distances from Shore

This method was used on August 19, 2016 by one person sampling along a transect starting at the water's edge, lined up with the landmark in the backshore at Regan and El Dorado Beaches. This was used to estimate percent cover in the field rapidly at the following distances away from shore: Regan Beach (2.3, 3.3, 16.7, 33.3, 50, 66.7, and 83.3 yds.), El Dorado Beach (1.7, 3.3, 16.7, 33.3, 50, 66.7, 117 yds.).

Center Transect Line with Regular-spaced Stations to Either Side of the Centerline (Grid)

This method was used in Aug. 2017 at Regan Beach. A center transect line was established perpendicular to shore, lined up with a landmark in the backshore on the day of sample collections. Metaphyton sampling points were established at 15, 65, 115, 165 yards from shore. Samples percent cover was measured at approximately 50 yds. right and 50 yds. left of the center transect.

Estimates of Metaphyton Percent Cover

Percent cover provides one means of assessing the visual or aesthetic impact of metaphyton at a site. The more coverage with algae over the bottom, the worse the level of metaphyton appears. The percent coverage is different from estimators of biomass such as AFDW or Chlorophyll *a*, which provide a number for biomass present. Percent cover may not always relate to biomass as factors such as whether the cover was over exposed sediments, was of different thicknesses or included algae in various states of health may affect the biomass for a certain percent coverage of the bottom.

Field Estimates of Metaphyton Percent Cover

Field estimates of percent cover were made at the sites, by aligning a 0.25 m² or 1m² quadrat at the designated sampling point (using a standardized method of lining a set corner of the quadrat or middle point of a side of the quadrat with the sampling point on the measuring tape). Percent cover of metaphyton inside the quadrat was visually estimated.

In the October 2015 and August 2016 samplings a 0.25 m² (0.5m x 0.5m) quadrat with string subdividing the quadrat into 10 cm x 10 cm boxes, was used. Having the 10cm x 10cm boxes outlined with string allowed for better estimates of percent coverage. Caution was needed however to avoid carryover of algae on the string from one site to another.

In August 2017, the quadrat size was increased to 1m². This was done to check if having a larger quadrat improved the field estimates of cover noticeably by encompassing a larger area. The inside of the 1 m² quadrat was divided into four 0.25 m² quadrats. The percent cover in each of the 0.25 m² quadrats was determined, and a 1 m² average was calculated.

Estimates of Percent Metaphyton Cover from Photos

This method entailed collection of several field photos of algal coverage within quadrats, then analysis of the percent metaphyton coverage within the quadrat by determining the number of pixels inside the quadrat representing metaphyton relative to the remaining number of pixels free of metaphyton. Photoshop was used to delineate metaphyton-covered areas in the photo and determine percent area covered.

Photography of Metaphyton

Photos of metaphyton were taken using a GoPro 3 Plus camera. For the first set of photos in the field, the GoPro camera was mounted on a frame above the 0.25 m² quadrat (such that the entire quadrat would be visible within the camera view). The photographer ultimately found it was easier to handhold the camera above the quadrat. A polarizing filter was used for photographs taken above water and removed for underwater photos. The narrowest field of view on the GoPro camera was used to minimize distortion. However some “barrel distortion” still occurred. This distortion appeared to be minor, but ultimately camera and lens settings should be used which result in no distortion of the photos. A series of 2-5 or more photos were taken. A white tile with a site number was placed adjacent to the quadrat to allow identification of the site during analysis of the photos. For monitoring done at El Dorado beach Oct. 7, 2015 and in 2016, and 2017 photos of quadrats were taken with a Panasonic Lumix TS6 digital camera.

Estimation of metaphyton percent cover from quadrat photos using Photoshop

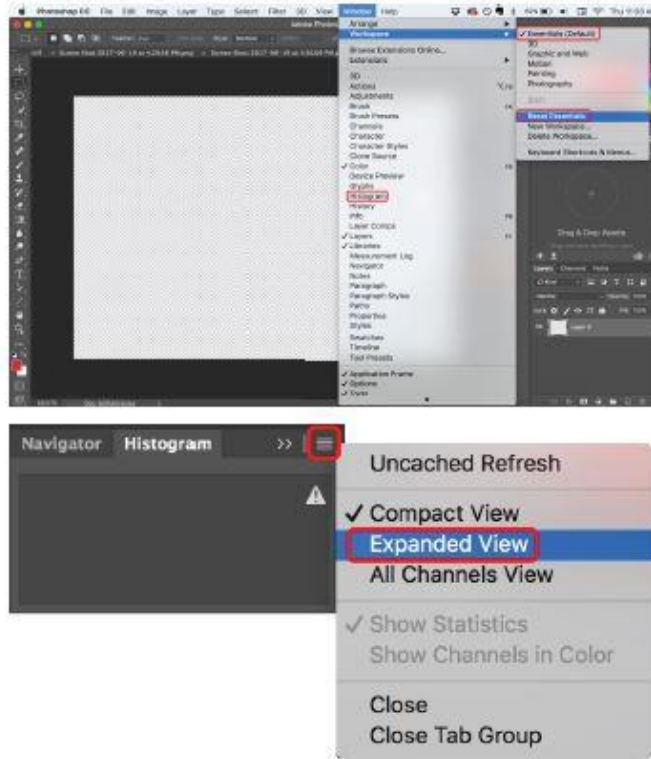
Photoshop was used to delineate metaphyton-covered areas in the photo and determine percent area covered. Tremendous appreciation goes out to intern Riley Rettig who refined the method for determining percent cover from photos and wrote it up for this report. She also analyzed percent cover in all the photos. The method for determination of percent cover is described below.

Finding Percent Coverage Using Photoshop

Image Processing in Photoshop can be used to make more accurate and consistent estimates of percent coverage of algae than visual estimates. These directions will guide you through the process for finding pixel coverage of algae blooms and comparing it to the total number of pixels in a plot in order to find percent coverage. In order for results to be consistent over time it is best to follow the procedures consistently.

Set-Up:

When you first open photoshop at the top of the screen go to Window > Workspace and ensure that Essentials is checked and press on the Reset Essentials. This will give us all the tools we need except one, the histogram. The histogram will be used to find the number of pixels selected in a photo, add this tool to your workspace by going to Window and clicking on Histogram. A new tool will pop up on the right side of the screen and at the top right of this tool window will be a button with four lines, click this and turn the histogram into expanded view. Now you're ready to start analyzing.



Step 1


With your picture open, under tools on the left hand side of the screen use Quick Selection (located under the magic wand tab when held down(W)) to select the area inside the plot (or select entire image if finding the coverage of the entire photo)

Note: Tools mentioned with a letter in parenthesis afterwards is a shortcut that can be used to speed up work. Example: Magic Wand (W) can be accessed by simply pressing W.

Step 2

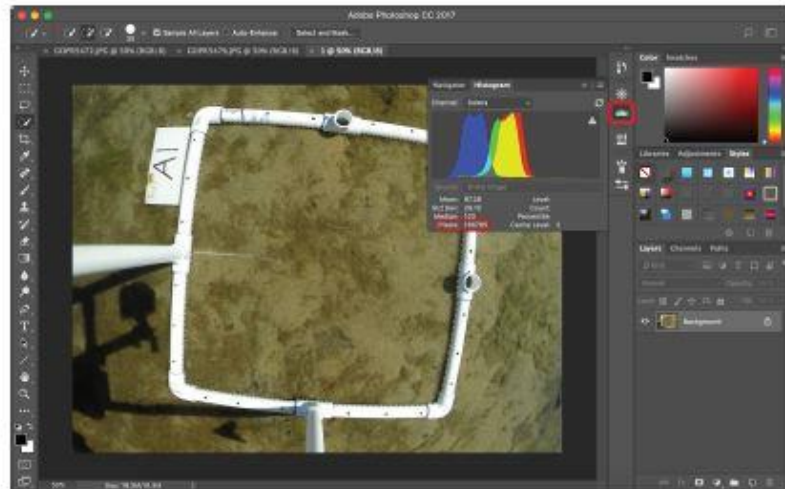
In the histogram window there will be a pixel reading at the bottom left. Record this number in the google Sheets template under Total Pixels.

Step 3

Switch to the lasso tool  (L) and deselect the area by clicking anywhere outside of it.

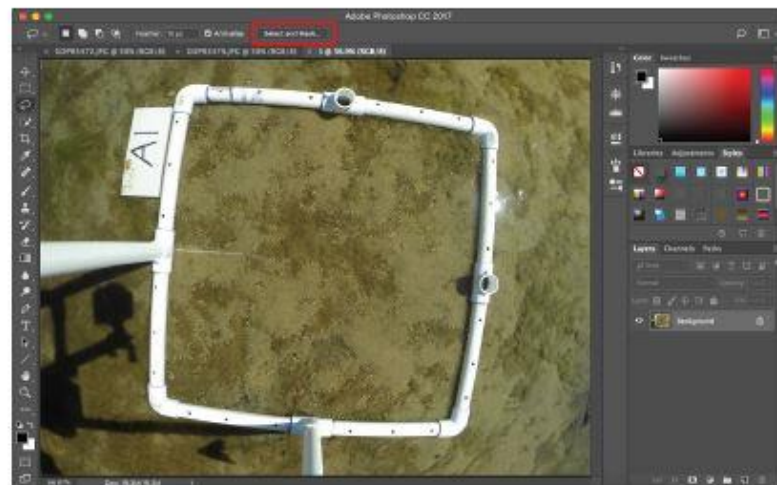
Step 4

Now use the lasso tool to select patches of algae. The edges do not have to be exact, but try to get the right shape of every patch. In order to select more than one bloom, hold down the shift key, then you can select more area without the first one disappearing.



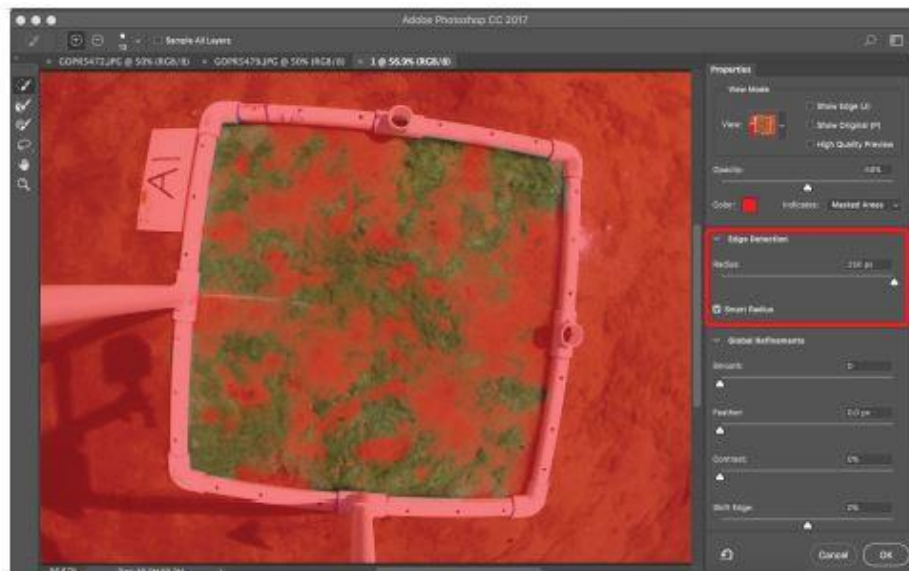
Step 5

After all algae in the measured area is selected, press on the button at the top of the screen that says "Select and Mask"



Step 6

In the screen that pops up find the tab the says “Edge Detection”. Under this click the Smart Radius checkbox and then turn the radius slider to the very right. Press okay at the bottom right of the window.

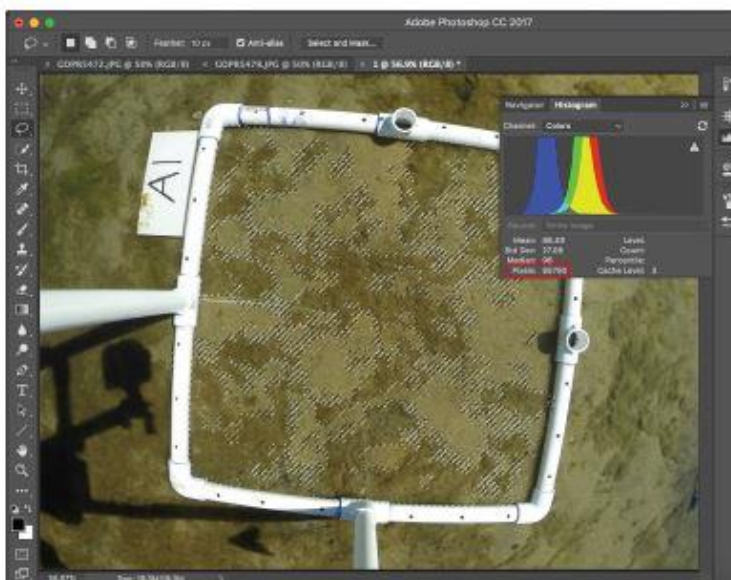


Step 7

Inspect the selected area and use the lasso tool while holding the alt/option key to deselect anything that the computer might have picked up. Sometimes clams, shadows or sediment will be selected, but this should not be included as part of the percent coverage of the algae.

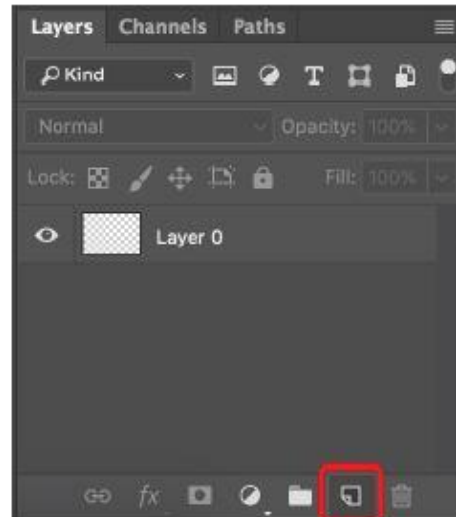
Step 8

After the selection looks good use the histogram again to find the number of pixels selected. Record this number under Covered Pixels and the sheet will automatically find the percent coverage by dividing the covered pixels by the total pixels.



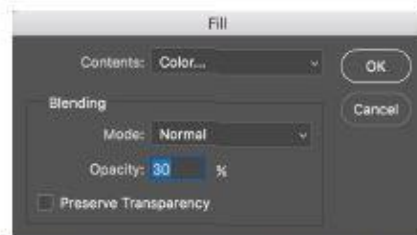
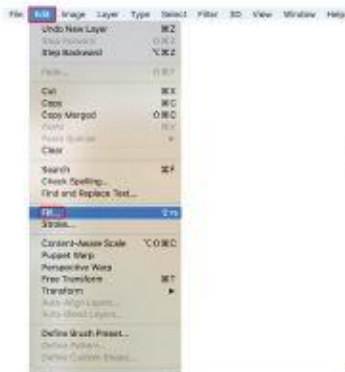
Step 9

In order to highlight the area selected with color, first make a new layer. In the bottom right there is a window with a tab labeled Layers. Click the new layer button at the bottom of this tab.



Step 10

At the top of your screen click Edit > Fill. A window should pop up, change the settings so that the color is red and then set the transparency to 30% and press okay.



Methods for Estimation of Metaphyton Biomass: 60°C Dry Weight, AFDW, Chlorophyll *a*

Sample collection for quantifiable estimates of metaphyton biomass

All metaphyton floating above or resting on the sediments within the quadrat was collected for determination of biomass. Onshore samples were collected by picking the filamentous algae off the sand with tweezers and transferring to a labeled Ziploc plastic bag. For metaphyton collection underwater, an aquarium fish net (17x25 cm opening, mesh size approximately 0.5 mm) was used to scoop metaphyton and transfer to a labeled plastic bag. The net was moved along the bottom in a regular pattern, as rapidly as possible to minimize drift of algae away from the net, and minimize loss due to algae moving outside of the quadrat due to currents. SCUBA was required for sampling biomass in 2017 in water depths of 4 feet or more. In sampling of both metaphyton exposed on shore and underwater, there was often a lot of sand mixed in with the algal filaments. The use of a net was suitable for most situations, however, sampling quantitatively in very thick metaphyton patches was not possible with the net, as too much material was stirred up and drifted in all directions.

Partitioning of samples into algae and sand fractions

Following sampling, the samples were returned to the lab at Incline Village, NV. One of the challenging aspects of processing metaphyton samples is that the sample can consist of a mix of algal, plant, and woody material and sand. Since we are interested only in the metaphyton algae biomass, we needed to separate the non-metaphyton material from the metaphyton. Removal of this material resulted in a more homogenous sample from which representative subsamples could be collected for dry weight, AFDW and chlorophyll *a*.

Sand and algal material in samples were separated to the extent possible. This was done either in the original collection plastic bag or in a shallow pan. The sample was mixed or swirled and the lighter algal material tended to remain suspended in the water while heavier material accumulated in the bottom of the bag or bottom of the pan. The algal material was separated off into another bag using a turkey baster, the sample swirled again and additional algae collected. When most of the algae had been separated off, large pieces of wood, plant or other debris was removed from the remaining material in the bottom of the bag and saved for weight measurement. The sand remaining in the bag or pan was collected for analysis. For smaller samples, algae and sand separation was done with aid of a dissecting scope. For most samples fairly good separation of algae and sand was achieved. However, for some samples, it was difficult to achieve good separation and both samples contained substantial algae and sand. This process proved to be time-consuming.

Both the algae fraction and sand fractions were removed from the water and dried to a damp consistency. The total damp weight of the algae sample was measured. Then small subsamples for 60°C Dry Weight, AFDW and chlorophyll *a* analysis were collected rapidly and damp weight

determined. Similarly, the sand portion of the sample was dried to a damp consistency then subsamples for DW, AFDW and chlorophyll *a* rapidly taken and damp weight determined. Samples for 60°C Dry Weight and AFDW were weighed in a pre-tared aluminum tin pan (pre-combusted at 500° C) then placed in a drying oven overnight at 60°C. Subsamples for chlorophyll *a* were weighed on a pre-tared half piece of a 4.75 cm dia. GF/C filter. The filter and sample were then immediately frozen. This process resulted in partitioning of the sample into algal and sand portions and these two fractions of the sample were analyzed separately for dry weight, chlorophyll *a* and AFDW.

60°C Dry Weight

Damp samples for dry weight were weighed in a pre-tared, pre-combusted aluminum tin to give a Sample Wet Weight (SSW) then dried overnight at a temperature of 60°C, allowed to cool in a desiccator, then weighed to determine 60°C dry weight (SDW 60°).

$$60^{\circ}\text{C Dry weight (g/m}^2) = (\text{TSWW/SWW}) * (\text{SDW}60^{\circ}) / 0.00053$$

["TWW" is Total Wet Weight of metaphyton sample collected; "SWW" is subsample wet weight; "SDW60°" is sample 60°C dry weight; 0.00053 m² is area sampled; all weights in grams].

Ash Free Dry Weight (AFDW)

After determination of 60°C dry weight, samples were combusted at 500°C for one hour. The loss in weight at this high temperature was assumed to be primarily due to combustion of organic material present in the sample. AFDW was calculated as:

$$\text{AFDW (g/m}^2) = (\text{TWW/SW}_{\text{afdw}}) * (\text{SDW}_{\text{afdw}}60^{\circ} - \text{SCW}_{\text{afdw}}500^{\circ}) / 5.3 \times 10^{-4}$$

Where:

"TWW" is Total Wet Weight (g) of metaphyton field sample collected (all weights in grams)

"SW_{afdw}" is AFDW subsample wet weight (g)

"SDW_{afdw} 60°" is sample 60°C dry weight (g)

"SCW_{afdw} 500°" is weight (g) of subsample after combusting at 500°C for 1 hour

5.3x10⁻⁴ m² is area sampled

Chlorophyll *a*

Sub-samples for metaphyton chlorophyll *a* were frozen immediately after measuring a damp weight. The analysis for chlorophyll *a* involved boiling the metaphyton sub-sample in 8-10 ml of 100% methanol for 2-3 minutes while grinding with a glass rod. Samples were then centrifuged to remove suspended particulate material from solution. The solution was then decanted, diluted with 100% methanol (usually a 1:2 dilution of the sample in methanol) to allow reading in a 4 cm glass spectrophotometric cell and the absorbance of the solution measured using a Shimadzu UV160U dual beam spectrophotometer at 750, 666, 653 nm. A 100% methanol solution was used as the reference blank. Non-chlorophyll *a* turbidity was determined

at 750 nm. The equation of Iwamura et al. (1970) was used to calculate the amount of chlorophyll *a* in each sample.

$$\text{Chlorophyll } a = (17.12 \times \text{Abs}_{666} - 8.68 \times \text{Abs}_{653}) \times ((\text{Vol}_{\text{me}} \times \text{TWW}) / (4 \times \text{SWW}_{\text{Chla}} \times 0.25))$$

Where:

“Abs₆₆₆” and “Abs₆₅₃” are absorbance readings at 666nm and 653nm respectively

“Vol_{me}” is volume of methanol (ml)

“TWW” is Total Wet Weight (g) of metaphyton field sample collected (all weights in grams)

“SWW_{Chla}” is Chlorophyll *a* subsample wet weight (g)

0.25 m² is area sampled

It should be noted, work on this project was put on hold for an extended period after the 2015 summer monitoring was completed while an agreement was worked out between UC Davis and Nevada State Lands on legal language in the final contract. Due to this break in project work, chlorophyll *a* samples from 2015 were stored for over a year frozen before analysis. We believe the long hold time did not substantially affect the 2015 chlorophyll *a* data or conclusions drawn from it. Evidence for this is the similar levels for chlorophyll *a* relative to AFDW for a limited number of samples collected in 2015 (chlorophyll *a* analyzed over year later) and 2017 (chlorophyll *a* analyzed only 22 days after collection). Chlorophyll *a* and AFDW levels for samples from Regan Beach in 2015 were 4.39 mg/m² and 3.18 g/m² for a sample collected on 8/20/15 (Appendix Table 1.c) and 4.73 mg/m² and 2.31 g/m² for a sample on 9/21/15 (Appendix Table 1d). Chlorophyll *a* and AFDW were in a similar range for a sample collected 8/9/17 when the chlorophyll *a* analysis was done within 22 days of collection (i.e. 4.64 mg/m² and 2.75 g/m²) (Appendix Table 1f). If degradation of chlorophyll *a* was substantial for the 2015 samples, it might be expected that the chlorophyll *a* levels would have proportionally less relative to AFDW (organic matter) in the 2015 samples compared to the 2017 sample.

Predominant algal species

Small subsamples of metaphyton from the overall sample were removed and analyzed under a microscope, generally under 10X magnification. The sample was examined and the predominant two or three algae species present were identified.

GPS measurements

GPS measurements were collected for the first set of sampling transects, with some additional measurements made on later dates. Many of the GPS measurements were made using an I-Phone 6S using an App called “Motion X GPS”. The accuracy of the I-Phone with this App ranges from 5-50 ft. At El Dorado beach, GPS measurements were made with an early I-Phone model (likely I-Phone 3). Information on the accuracy of this device was not available. GPS measurements for backshore landmarks were also made using a Magellan SporTrak Color GPS Mapping Receiver. Limited additional GPS readings are included in Table 1. The start location for transects on different dates shifted offshore or inshore as lake level fluctuated.

Results

Transect Line and Sampling Site Set-up during 2015-2017

Set-up of transect lines and marking of sites generally took under an hour on sampling days. In 2015 and 2016 set-up of sampling sites at El Dorado Beach and Regan Beach was relatively easy in the shallow lake conditions. Sites were easily wade-able 137m offshore; most sites were marked with flagging. At Round Hill Pines in 2015 in Sept. the deepest site was 1.5m and transect set-up was also relatively easy. In 2017, with high lake levels, set-up of sampling sites was more difficult. Two researchers (one snorkeling, one diving) worked together, measured out distances with a tape and marked sites with floats. Lining up sites, and working in the water was required much swimming. Care was exercised not to stir up metaphyton from the bottom.

Locations of sampling transects: GPS coordinates

The locations of the first set of transects used in the study are shown in Table 1. Subsequent sampling transects were established at the shoreline aligned with backshore markers. Figures 1-3 below show the approximate locations of the transects on Google Earth Images. GPS data was not collected on all dates. The start location for transects on different dates shifted offshore or inshore as lake level fluctuated. This shift was quite large at Regan and El Dorado beaches in 2015 when the slope of the shoreline to water's edge was very slight. For instance, the start point at water's edge for the Regan center transect was about 50 yards further lakeward in on 9/21/15 compared with the location on 8/20/15. At El Dorado beach, the start point was about 100 yards further lakeward on 10/7/15 compared to 8/6/15. At Round Hill Pines in contrast, the starting point only shifted about a yard further lakeward on 9/24/15 compared to 8/11/15 due to a relatively steep sloping shoreline.

Table 1. Coordinates of center transect lines and landmarks.

Station	Date	Center Transect Location	Latitude; Longitude
El Dorado Beach	8/6/15	Shoreline (Transect Start)	38.9453; -119.9758 ¹
El Dorado Beach	8/6/15	150 yd. (Transect End)	38.9467; -119.9764 ¹
El. Dorado Beach		Landmark Tree	38.9450; -119.9758 ^{1,4}
Round Hill Pines	8/11/15	Shoreline (Transect Start)	38.9893; -119.9532 ³
Round Hill Pines	8/11/15	50 yd. (Transect End)	38.9893; -119.9537 ²
Round Hill Pines		Landmark Tree	38.9892; -119.9528 ⁴
Regan Beach	8/20/15	Shoreline (Transect Start)	38.9455; -119.9838 ²
Regan Beach	8/20/15	150 yd. (Transect End)	38.9467; -119.9841 ²
Shore Landmark		Landmark Tree	38.9443; -119.9838 ^{2,4}
Regan Beach	9/21/15	Shoreline (Transect Start)	38.9459; -119.9839 ³
Regan Beach	9/21/15	150 yd. (Transect End)	38.9471; -119.9841 ²

- 1- I-Phone 3
- 2- I-Phone 6s with Motion X GPS app
- 3- Estimated using distance from sampling station with measured GPS
- 4- Magellan SporTrak Color GPS Mapping Receiver.

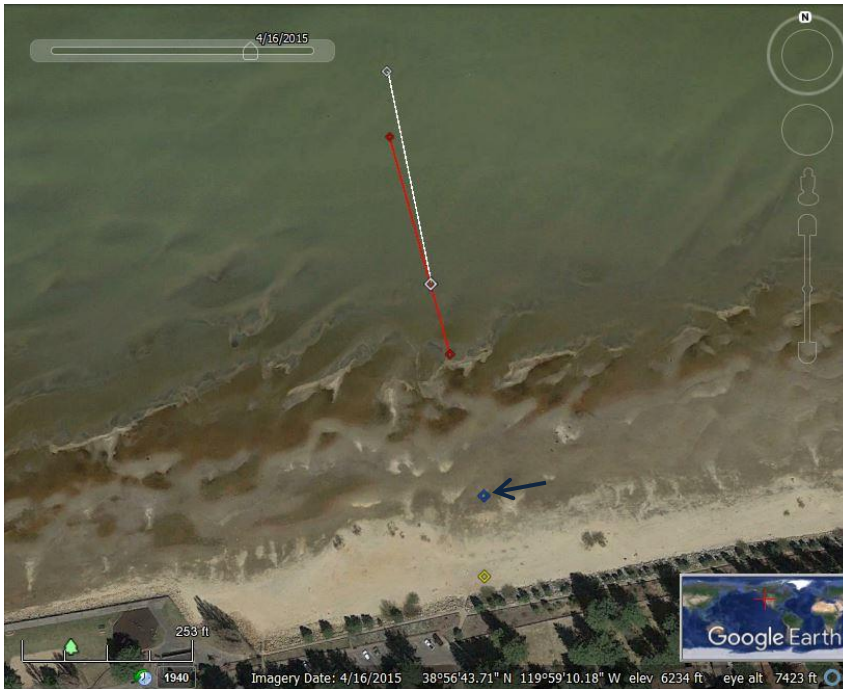


Figure 1. Google Earth image of Regan Beach area. Approximate locations of sampling transects for 8/20/15 (red line) 9/21/15 (white line) are indicated, blue arrow shows approximate location for transect start 8/19/16. This image taken 4/16/15, Google Earth.

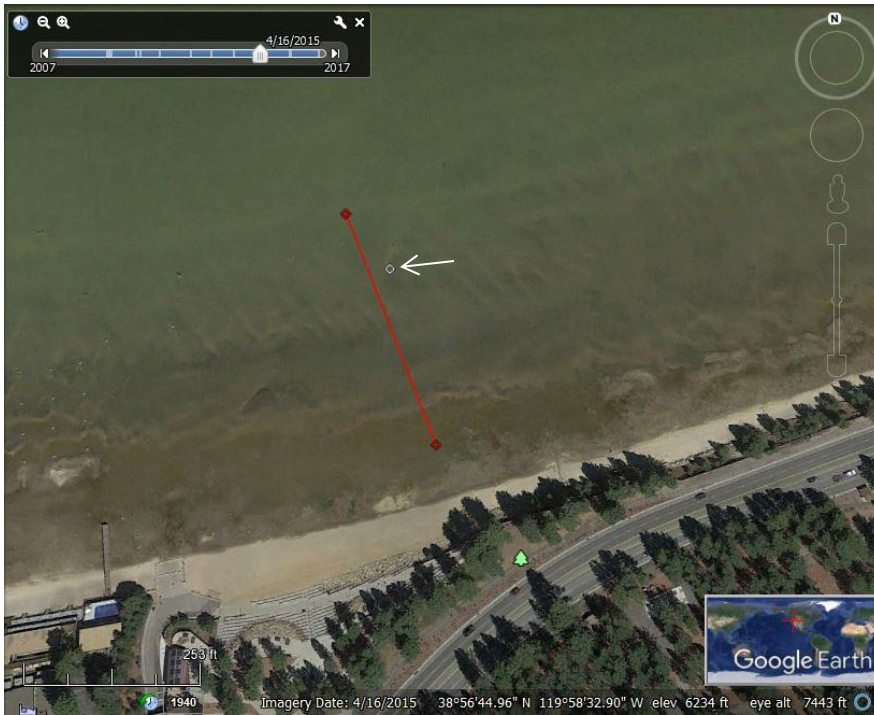


Figure 2. Google Earth image of El Dorado Beach area. Approximate location of sampling transect 8/6/15 (red line) is shown, white arrow points to approximate start location of 10/7/15 transect. This image taken 4/16/15, Google Earth.

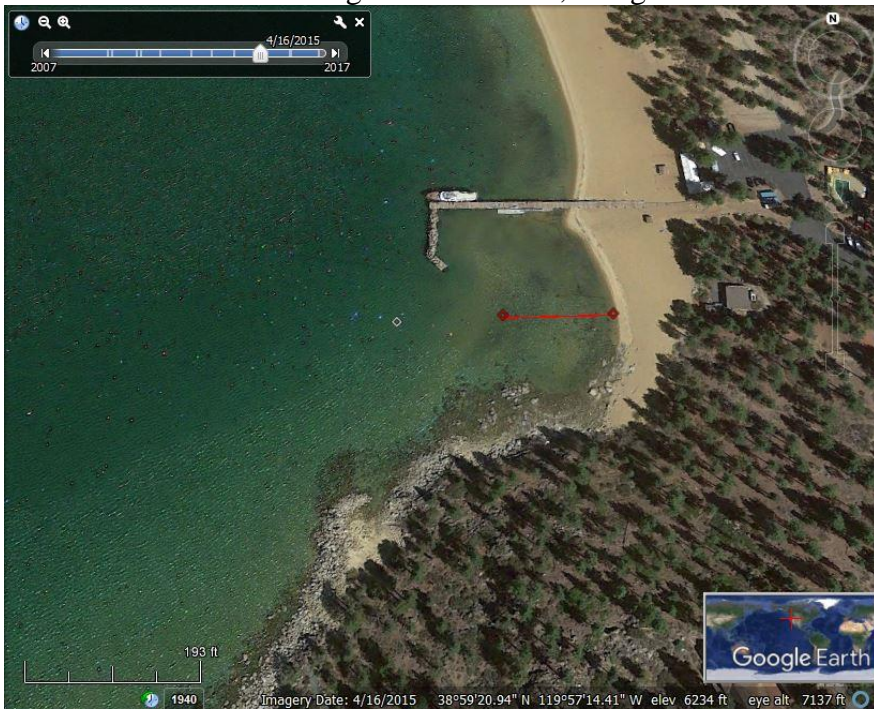


Figure 3. Google Earth image of Round Hill Pines area. Approximate location of sampling transect 8/11/15 (red line) is shown. This image taken 4/29/14, Google Earth.

In the summer of 2015, when much of the monitoring was done, there was an expansive area of exposed lakebed in the nearshore near Regan and El Dorado Beaches. The lake was also very shallow for a substantial distance offshore. As the lake receded during the summer of 2015, the sampling transects used for the second round of monitoring were located further offshore, however, there was some overlap in the transects. At Round Hill Pines Beach, the slope of the nearshore area is much steeper than for El Dorado and Regan, as a result there were small differences in the starting point for the transects there.

Site Physical Measurements and Predominant Algal Types

Summer 2015 Site Physical Measurements

Much of the pilot study monitoring was done during the summer of 2015. This was a period when the lake level was extremely low (below the natural rim elevation of 6223 ft.) due to a four-year drought. The lake surface elevation was 6222.61 ft. when test monitoring began on 8/6/15 and continued to drop throughout the summer. The lowering lake level left an expansive flat area of exposed lakebed adjacent to El Dorado and Regan beaches along the south shore. As part of the test monitoring, measurements of water depth, lake temperature and identification of predominant algae types were collected. Tables 2 to 7 below summarize the 2015 physical measurements and predominant metaphyton algal types at the sites. Photos of the sites are also presented in Figures 4 to 15.



Figure 4. El Dorado beach metaphyton along shore 8/6/15.

Figure 5. El Dorado beach metaphyton 8/6/15.

Table 2. El Dorado Beach 8/6/15 site physical measurements and predominant metaphyton algal types.

El Dorado Beach 8/6/15	Left Sampling Point					Center Transect				Right Sampling Point				
Distance Offshore in (m) /and (yd.)	Distance Left (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Distance Right (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)
Shoreline	19.2	0	NA	0.8	<i>S, z, m</i>	0	NA	<0.5	<i>S, z</i>	8.2	1.5	NA	1.0	<i>S, z</i>
18.3m / 20yd	35.7	2	22.0	1.0	<i>S, z, m</i>	5	22.0	1.0	<i>S, z, m</i>	31.1	4	22.0	1.0	<i>S</i>
45.7 m/ 50yd	40.2	12	18.0	NA		12	17.0	1.0	<i>S, z</i>	24.7	12	18.0	NA	
91.4 m/ 100yd	2.7	22	17.0	1.0	<i>S, z, m</i>	22	16.5	1.0	<i>S, z</i>	3.7	22	17.0	1.0	<i>S, z, m</i>
137.2 m/150yd	38.4	NA	16.0	1.0	<i>S, z, m</i>	31	17.0	1.0	<i>S, m</i>	10.1	31	16.0	1.0	<i>S</i>

Predominant algal types Capitalized, secondary algae in small letters (types included *S*= *Spirogyra*, *z*= *Zygnema*, *m*=*Mougeotia*); Lake surface elevation: 6222.61 ft.



Figure 6. El Dorado beach metaphyton along shore 10/7/15. **Figure 7.** El Dorado beach showing beach and exposed lakebed 10/7/15.

Table 3. El Dorado Beach 10/7/15 site physical measurements.

El Dorado Beach 10/7/15	Left Sampling Point			Left Sampling Point			Center Transect		Right Sampling Point		
Distance Offshore in (m) /and (yd.)	Distance Left (m)	Depth (cm)	T °C	Distance Left (m)	Depth (cm)	T °C	Depth (cm)	T °C	Distance Right (m)	Depth (cm)	T °C
Shoreline	45.7	0	NA	22.9	0	NA	0	NA	22.9	0	NA
22.9 m/ 25yd				22.9	5	21.5	7	21.5	22.9	5.5	21.0
45.7 m/ 50yd				22.9	11	17.5	12	19.0	22.9	11	20.0
91.4 m/ 100yd	45.7	20	15.0	22.9	19	15.0	20	15.0	22.9	19	16.0
137.2 m/ 150yd	45.7	32	15.0	21.9	32	14.0	30	14.0	22.9	27	NA

Lake surface elevation: 6221.87 ft.

El Dorado Beach 10/7/15	Right Sampling Point			Right Sampling Point			Right Sampling Point			Right Sampling Point		
Distance Offshore in (m) /and (yd.)	Distance Right (m)	Depth (cm)	T °C	Distance Right (m)	Depth (cm)	T °C	Distance Right (m)	Depth (cm)	T °C	Distance Right (m)	Depth (cm)	T °C
Shoreline	45.7	0	NA	49.6	1-2e	NA	68.6	1	NA	91.4	3	NA
22.9 m/ 25yd												
45.7 m/ 50yd												
91.4 m/ 100yd	45.7	20	16.5									
137.2 m/ 150yd	45.7	32	15.0									



Figure 8. Regan Beach metaphyton on shore 8/20/15.

Figure 9. Regan Beach shoreline and transect start 8/20/15.

Table 4. Regan Beach 8/20/15 site physical measurements and predominant metaphyton algal types.

Regan Beach 8/20/15	Left Sampling Point					Center Transect				Right Sampling Point				
Distance Offshore in (m) /and (yd.)	Distance Left (Yds.)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Distance Right (Yds.)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)
Shoreline	8.2	2.0	28.5	NA	<i>S,od</i>	0.0	NA	NA	<i>S</i>	15.5	0.0	NA	NA	<i>S,m,z,od</i>
9.1m/ 10yd	41.4	7.0	18.0	1.0	<i>S,m,z,od</i>	9.0	17.0	3.0	<i>S,z</i>	43.9	8.0	18.0	1.0	<i>S,z</i>
45.7 m/ 50yd	29.3	NA	NA	NA	<i>S,z</i>	14.0	NA	3.0	<i>S</i>	8.2	15.0	17.0	1.0	<i>S</i>
91.4 m/ 100yd	25.6	24.0	17.0	1.0	<i>S,z</i>	24.0	17.0	2.0	<i>S,z</i>	38.4	24.0	17.0	NA	<i>S,z,m</i>
137.2 m/150yd	12.8	30.0	18.0	2.5	<i>S,z</i>	30.0	17.5	2.5	<i>S,z</i>	13.7	31.0	18.0	2.5	<i>S</i>

Predominant algal types Capitalized, secondary algae in small letters (*S*= *Spirogyra*, *z*= *Zygnema*, *m*=*Mougeotia*, *od*=*organic detritus*). Lake surface elevation: 6222.47 ft.



Figure 10. Regan Beach 9/21/15.



Figure 11. Regan Beach looking offshore 9/21/15.

Table 5. Regan Beach 9/21/15 site physical measurements and predominant metaphyton algal types.

Regan Beach 9/21/15	Left Sampling Point					Center Transect				Right Sampling Point				
Distance Offshore in (m) /and (yd.)	Distance Left (Yds.)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Distance Right (Yds.)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)
Shoreline	23.8	0	NA	1.0	<i>S,z</i>	0.0	NA	1.0	<i>Z,od</i>	11.9	0.0	NA	<1.0	<i>Z,m</i>
9.1m/ 10yd	29.3	5.5	13.0	1.0	<i>S</i>	2	15.5	1.5	<i>Z,s</i>	28.3	0	NA	1.0	<i>Z,s</i>
45.7 m/ 50yd	19.2	9	11.0	1.0	<i>S,z</i>	7.5	11.0	1.0		11.0	8	11.5	0	
91.4 m/100yd	18.3	16.5	12.0	1.0	<i>S,z</i>	17	12.5	1.0	<i>Z,s</i>	28.3	15.5	12.5	1.0	<i>Z,s</i>
137.2 m/150yd	27.4	23	13.0	1.0	<i>S,z,m</i>	22	12.5	1.0	<i>S,z</i>	17.4	23	13.0	1.5	<i>S,z,m</i>

Predominant algal types Capitalized, secondary algae in small letters (*S*= *Spirogyra*, *z*= *Zygnema*, *m*=*Mougeotia*, *od*=*organic detritus*). Lake surface elevation: 6221.97 ft.

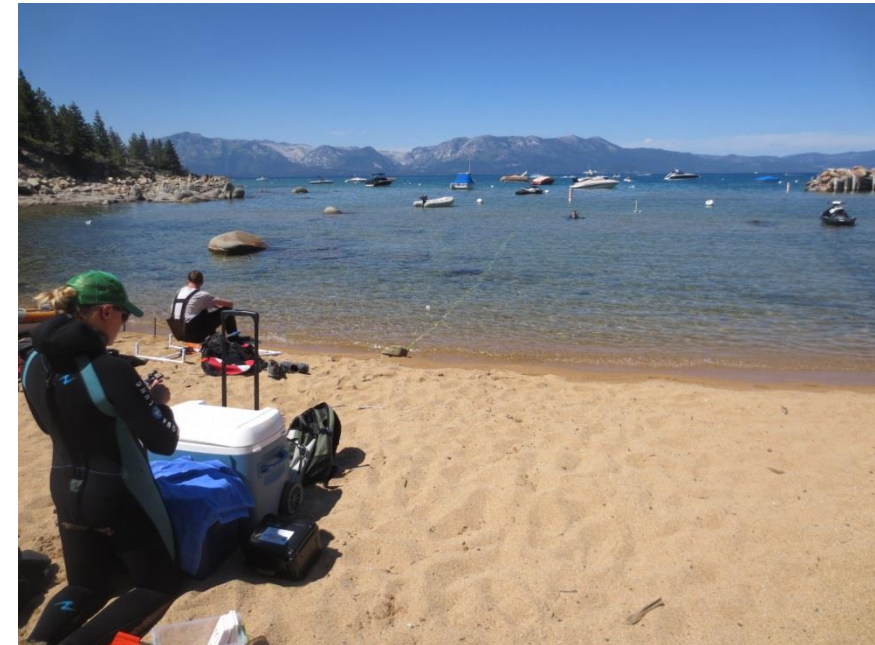


Figure 12. Round Hill Pines 8/11/15 no metaphyton on beach.

Figure 13. Round Hill Pines beach 8/11/15 and transect line.

Table 6. Round Hill Pines Beach 8/11/15 site physical measurements and predominant metaphyton algal types.

Round Hill Pines Beach 8/11/15	Left Sampling Point					Center Transect					Right Sampling Point				
Distance Offshore in (m) /and (yd.)	Distance Left (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Distance Right (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	
Shoreline	NA	0.0	NA	0		0.0	NA	0		NA	0.0	NA	0		
2.7m/3yd	10	46	17.5	3.0	Z,cy,g,s,m	49	18.0	4.0-5.0		20.1	48	18.0	1.0	Z, g,s,cy,m	
22.9m/25yd	5.5	66	17.0	1.0	Z, cy	52	17.0	2.0	Z, s, cy	10.1	69	18.5	1.0	Z	
45.7m/50 yd	21	100	17.0	1.0	Z, s, dia	97	18.0	1.0	Z, cy	NA ¹	NA ¹	NA ¹	NA ¹		

Predominant algal types Capitalized, secondary algae in small letters (Z=Zygnema, cy= cyanophytes, g= Gomphoneis, m= Mougeotia, s= Spirogyra; dia=misc. diatoms) Lake surface elevation: 6222.57 ft.



Figure 14. Round Hill Pines shoreline and transect 9/24/15.

Figure 15. Round Hill Pines shoreline and transect 9/24/15.

Table 7. Round Hill Pines Beach 9/24/15 site physical measurements and predominant metaphyton algal types.

Round Hill Pines Beach 9/24/15	Left Sampling Point					Center Transect				Right Sampling Point				
Distance Offshore in (m) /and (yd.)	Distance Left (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)	Distance Right (m)	Depth (cm)	T °C	Algal Layer Thick. (cm)	Main Algae Type(s)
Shoreline	16.5	0	NA	0		0.0	NA	0		17.4	0.0	NA	0	
2.7m/3yd	13.7	12	14.0	0.5	<i>Dia,cy,s,g</i>	16	14.0	<1		21	35	14.0	1.0	<i>Z,s,dia</i>
22.9m/25yd	7.3	53	14.0	0.5	<i>Z</i>	45	14.5	0.5	<i>Z,m,dia</i>	13.7	49	14.0	<1.0	<i>z,m,cy,dia</i>
45.7m/50 yd	6.4	80	14.5	0.5		76	14.5	0.5		16.5	66	15.0	0	
91.4m/100yd	7.3	152	15.5	NA	<i>Z</i>	168	15.0	1.0	<i>Z,cy</i>	18.3	213	15.0	0.5	<i>Z,s,m</i>

Predominant algal types Capitalized, secondary algae in small letters (*Z*=*Zygnema*, *cy*= cyanophytes, *g*= *Gomphoneis*, *m*= *Mougeotia*, *s*= *Spirogyra*; *dia*=misc. diatoms). Lake surface elevation:6221.95 ft.

The physical measurements taken as part of pilot monitoring in 2015 show dramatically both how shallow and how far out the shallow water extended off of Regan and El Dorado beaches. At El Dorado Beach, the water depth was 31cm (~ 1ft deep) at a distance of 137m (150 yards) away from the water's edge for both the August and October samplings. At Regan Beach, at 137m offshore, the water depth was about 30cm (<1 ft.) on Aug. 20 and even less, 23cm deep, on Sept. 21, 2015. The bottom slope from shoreline to well offshore at these two sites was minimal. As a demonstration of just how flat the shoreline was, when the wind blew onshore, the push of water moved the shoreline noticeably inshore (6.5 m on 8/6/15), almost as if a tide was "coming in."

At Round Hill Pines beach, there was a much steeper slope at the water's edge. On Aug. 11, 2015, the depth was near 0.5m within 3m of shore and near 1m deep 46m from shore. On Sept. 24, the depth ranged from 12-35cm within 3 m of shore and 66-80 cm within 46m of shore.

As part of the test monitoring, measurements of lake temperature were collected at the time of sampling. Water temperature showed patterns at some sites. At El Dorado Beach during the August and October monitoring, the water temperature tended to decrease away from shore (i.e. the shoreline water temperature in August was about 22°C while at a distance of 137m the water temperature was about 16°C; in October the shoreline water temperature was 21.5°C and offshore was about 15°C.) At Regan Beach in August water temperature was as high as 28°C right at the shoreline and was about 18°C 137 m offshore while in September the temperatures were fairly similar along shore 13-15°C along the shore and 13°C offshore. At Round Hill Pines beach, the water temperatures tended to be similar nearshore and offshore near the surface (i.e. between 17-18°C in August and 14-15°C in September. Increased water temperature right along the shore at El Dorado and Regan beaches in the summer may have had an impact on the biology of the metaphyton. For instance, increases in temperature in shallow water along shore could increase rates of growth of some types of algae tolerant of increased temperatures and potentially increase degradation rates of algae and other organic material. However, the temperature measurements were only made during the time of sampling in the pilot study. More information on temperature patterns over a 24-hour period would be needed to better assess temperature impacts on metaphyton at the sites.

The metaphyton tended to be more concentrated right along the shoreline (likely because of drifting or being blown inshore) at Regan and El Dorado Beaches in the summer of 2015. Metaphyton was widely dispersed in shallow water over the flat offshore area at El Dorado and Regan beaches in summer 2015. The metaphyton tended to be close to the bottom. The average thickness of the metaphyton layer was about 1 cm. As the lake receded, this algae was left deposited on top of the sand along the shoreline. In contrast at Round Hill Pines beach, there was only small amounts of metaphyton offshore, with some accumulation about 3 m offshore and there was no metaphyton deposited onshore.

Predominant Algal Types in Metaphyton at Sites Summer 2015

The predominant algae composing the metaphyton in summer of 2015 were green filamentous algae. At both El Dorado and Regan Beaches, the predominant algae was the green filamentous algae, *Spirogyra*. There was also some *Zygnema* and *Mougeotia* at El Dorado beach. At Regan beach, *Zygnema* was the other prevalent algae, with some *Mougeotia*. In contrast, the most prevalent algae in the metaphyton nearshore at Round Hill Pines was *Zygnema*.

Summer 2016 Site Physical Measurements

During 2016, we made limited observations of metaphyton levels at the three sites on a single date, August 19. Although 2016 was more of a normal precipitation year which caused the lake level to rise about 2 feet from the lowest level in 2015, the lake level was still relatively low (6223.37 ft.) in August. Site data collected at El Dorado beach is presented in Table 8 and a site photo is presented (Figure 16). At El Dorado beach, the water's edge was slightly upslope of a transition area where the sloping shore meets relatively flat shallow lakebed just offshore underwater. Within 4.6 m of shore, the water depth was only 12 cm. There was some accumulation of metaphyton right at the water's edge and also onshore at El Dorado beach. Data collected at Regan beach is presented in Table 9 and a site photo is presented in Figure 17. At Regan beach, there was much less exposed lakebed offshore compared to the previous year (the lake extended 119 m further inshore compared to late summer 2015). The water was relatively shallow off Regan, (at a distance of 76 m from shore, the depth was 21 cm). At Round Hill Pines physical measurements were not made in 2016, however a photo of the site on 8/19/16 is presented in Figure 18. Temperature measurements were made at only at Regan Beach on August 19, 2016. The shallow water was very warm ranging from 32°C in water 2 cm deep near the shoreline, to 26°C in water 23 cm deep, 76 m from shore.

Predominant Algal Types in Metaphyton at Sites Summer 2016

Metaphyton was present at all three sites in 2016. At El Dorado beach, the metaphyton was concentrated in the nearshore with metaphyton also deposited on the beach (Fig. 16). The algae right near shore was primarily *Cladophora* with some *Spirogyra*. At 107 m offshore at El Dorado, the algae was primarily *Spirogyra*, with some *Zygnema*. At Regan Beach, there was relatively light metaphyton in the nearshore and along the shoreline (Fig.17). The primary algae in the nearshore was *Zygnema*. Further offshore, (76 m away), the metaphyton consisted of a mix of *Zygnema* and *Cladophora*. At Round Hill Pines, there was noticeable small clumps of bright green metaphyton in the southeast cove area (Figure 18) in the sand ripples close to shore and several larger patches offshore. This algae was not collected for identification. No metaphyton was deposited on the beach at Round Hill Pines.



Figure 16 . El Dorado Beach 8/19/16.

Table 8. El Dorado Beach Site Data, 8/19/16 limited sampling, percent cover estimates and predominant algal species.

El Dorado Beach 8/19/16	Center Transect		
Distance Offshore in (m) /and (yd)	Depth (cm)	T °C	Main Algae Type(s)
Shoreline 1.5m/ 1.7yd	-	NA	Cl, s
Offshore			
4.6m/ 5yd	12	NA	
17.1m/ 18.7yd	21	NA	
32m/ 35yd	22	NA	
47.2m/ 51.7yd	26	NA	
62.5m/ 68.3yd	32	NA	
106.7m/ 116.7yd	41	NA	S,z,od

Predominant algal types Capitalized, secondary algae in small letters (types included Cl=*Cladophora*, s= *Spirogyra*, z= *Zygnema*, od=*organic debris*). Lake surface elevation: 6223.37 ft.



Figure 17. Regan Beach 8/19/16.

Table 9. Regan Beach Site Data, 8/19/16.

Regan Beach 8/19/16	Center Transect		
Distance Offshore in (m)/ and (yd)	Depth (cm)	T °C	Main Algae Type(s)
Shoreline 2.1m/ 2.3yd	0	NA	Z, od
3.0m/ 3.3yd	2	32	
Offshore			
15.2m/ 16.7yd	8	31	
30.5m/ 33.3yd	10	30	
45.7m/ 50yd	14	28	
61m/ 66.7yd	19	27.5	
76.2m/ 83.3yd	23	26	'cl,z,od

Predominant algal types Capitalized, secondary algae in small letters (types included cl=*Cladophora*, z= *Zygnema*, od=*organic debris*). Lake surface elevation: 6223.37 ft.



Figure 18. Round Hill Pines beach and nearshore 8/19/16. Note small clumps of green metaphyton are visible nearshore. There were also larger patches of metaphyton offshore. However, no algae was present onshore.

(NO PHYSICAL MEASUREMENTS AT ROUND HILL PINES BEACH 8/19/16)

Summer 2017 Site Physical Measurements

We made limited observations of metaphyton levels at Regan and El Dorado beaches in the summer of 2017. The winter of 2016-2017 was extremely wet with many large rain events and a large spring snowmelt. This resulted in a lake level rise of over six feet from minimum lake levels the previous fall to a nearly full lake in the summer of 2017. This provided an opportunity to test some of the metaphyton monitoring methods under high lake level conditions. Figures 19 to 21 show representative photos of the three sites in summer 2017.

A monitoring grid of 12 sampling points was established offshore at Regan Beach starting approximately 11 m away from shore extending out 148 m and 46 m on the sides on Aug. 9, 2017. This monitoring was coordinated with aerial imaging of the metaphyton from a helicopter. The lake at that time was at an elevation of 6228.83 ft. (the lake maximum height is 6229.1 ft.). At about 11 m offshore, the water depth was about 1.3 m with a large patch of metaphyton present. At about 57 m offshore, the depth was near 1.7 m. The sampling area was very heterogeneous and included a mix of metaphyton and underwater vegetation. The water temperature was warm, 24°C. Water clarity was very poor at Regan Beach, possibly due to wave activity stirring up organic particles from the bottom and from within the submerged vegetation.

Observations of metaphyton were also made at El Dorado beach on Sept. 10, 2017 coordinated with aerial imaging from the helicopter. The water's edge was located well up the sandy shoreline slope. The transition area from steeper shoreline flat lakebed was located about 18 m offshore at a depth of near 6223 ft. Just offshore of the transition area, a distinct patch of metaphyton paralleling the shore was observed. Algae from this patch did not move upslope and there was no metaphyton onshore. However, there were pieces of aquatic plants deposited on shore.

Predominant Algal Types in Metaphyton at Sites Summer 2017

Samples of metaphyton were collected at Regan Beach at two sites "B1" and "D3" and were identified as *Mougeotia*. Site "D3" also had some *Spirogyra* and *Zygnema*. The algae composing the metaphyton at El Dorado was a mix of unidentified thin filamentous green algae (possibly *Oedogonium* or *Mougeotia*) with some *Zygnema* and old algae filaments. Some of the filamentous algae also had diatoms attached. Algae from the nearshore edge also contained plant stems.

Overall Patterns for Predominant Metaphyton Algal Species 2015-2017

The predominant algal types composing the metaphyton at the sites showed some variation during the study. In 2015, the predominant algae was the green filamentous algae, *Spirogyra* at Regan and El Dorado beaches while at Round Hill Pines the predominant algae was *Zygnema*. In 2016 the predominant algae in the nearshore metaphyton at El Dorado beach was *Cladophora*, with some *Spirogyra* also present. At Regan beach the predominant algae was *Zygnema* in the nearshore. In 2017 the predominant algae at Regan beach was *an unidentified thin green filamentous algae* while the predominant algae at El Dorado was a mix of an unidentified thin filamentous algae (possibly *Oedogonium* or *Mougeotia*) with some *Zygnema* and old algae filaments. The predominant algae composing the metaphyton in summers of 2015-2017 were green filamentous algae.



Figure 19. Regan Beach and nearly full lake on August 9, 2017. Dark metaphyton nearshore patch can be seen beneath boats.



Figure 20. Round Hill Pines beach 8/4/17.



Figure 21. El Dorado Beach 9/9/17 no metaphyton was observed onshore, however, there were many pieces of aquatic plants.

Pilot Test Monitoring Results for Percent Cover Field Estimates, Percent Cover from Photo Estimates, 60°C Dry Weight, AFDW and Chlorophyll *a*

The results of pilot testing for field estimates of metaphyton percent cover, estimates of metaphyton percent cover from photos, and 60°C Dry weight, AFDW, and Chlorophyll *a* are summarized in Tables 10 to 18 below. The data are also summarized graphically in Figures 27 to 32. Additional data including sample replicates are presented in Appendix 1.

Field Estimates of Metaphyton Percent Cover

Field estimates of percent metaphyton cover in the quadrats were generally made right after photographs of metaphyton in the quadrats were taken. Either one or two observers made visual estimates of cover. These were subjective, best estimates, relating the amount of bottom within a quadrat covered with metaphyton, to the total area of the quadrat. The estimates generally took a few minutes to complete.

In 2015, El Dorado and Regan beach sites tended to show much greater levels of coverage along shore than offshore (Tables 10a, 12a, 13a, 15a; Figures 27a, 29a, 30a). At El Dorado Beach on Aug 6, percent coverage along the shoreline ranged from 30-75% while coverage ranged from zero to 5% in at distances from 18-137 m offshore using the 0.25m² quadrat. On 10/7/15 at El Dorado, metaphyton cover was measured at regular 22.9 m (50 yd.) intervals over a distance of 137 m (150 yd.) along the shoreline and found to range between 5-22%. One area of heavier algae coverage between the regular intervals was measured and found to have 72% cover. At Regan Beach on 8/20/15, percent metaphyton cover was found to range between 6-70% for shoreline sites and to range between 2-20% for offshore sites between 46 m and 137 m offshore. Metaphyton percent coverage along the shoreline at Regan ranged between 1-60% cover on 9/21/15 and offshore sites ranged between 0-10% coverage. Overall, this sampling showed algae tended to accumulate along the shoreline at El Dorado and Regan Beaches in 2015 and was more dispersed offshore.

In contrast, no metaphyton was observed along the shoreline at Round Hill Pines beach during the summer of 2015. Metaphyton levels offshore there ranged from <1% to 14% from 3-46 m offshore on 8/11/15, and from 0-7% from 3 to 91 m offshore on 9/24/15.

On 8/19/16, estimates of % cover were made along single transect lines at El Dorado beach (Table 16) and Regan beach (Table 17) At El Dorado beach, percent cover along the shoreline was about 20% while offshore between 5 and 107 m it ranged between 0-10%. At Regan beach shoreline metaphyton percent cover ranged from <1 to 56% while offshore percent cover ranged from 0-3% between 15 and 76 m offshore.

On 8/9/17, field estimates of percent cover were made at sampling sites evenly spaced on a 12 point grid offshore. For this monitoring a larger 1 m² quadrat was used. This quadrat was subdivided into four 0.25m² sub-areas. This sampling was designed to see if use of a larger

quadrat made a difference in estimates of percent cover. Observations were made by both a SCUBA diver and a researcher snorkeling.

The results of this monitoring are shown in Table 18. Water clarity was poor during this monitoring possibly due to resuspended metaphyton and other organic debris from the bottom during waves. For the 11m sites closest to shore, only the SCUBA diver was able to get close enough to the bottom and see clearly enough to make estimates of percent cover. The bottom was not visible to the snorkeler on the surface only 4-5 feet above. Coverage estimates ranged from 5 - 50% in a thick patch of metaphyton, which began about 11m from shore. Further offshore, clarity improved enough to allow both the diver and snorkeler both to make estimates of percent cover. Levels of percent coverage for the sites beyond the initial set of sampling points close to shore ranged between 2-11% for estimates from the SCUBA diver and between 2-5% for the snorkeler. Two monitoring stations had higher levels of coverage, one of these was a plant bed to which algae was attached (this was not included as metaphyton). The other site was one in which the quadrat was purposely placed to enclose a known amount of metaphyton for aerial imaging.

Both the 0.25 m² and 1 m² quadrats generally gave similar estimates of cover in offshore monitoring of metaphyton with the exception of one site where the larger quadrat accounted for an area with greater estimated coverage. Table 18 (snorkeler results), the levels of coverage for individual 0.25 m² quadrats were relatively similar to the 1 m² coverage at five of six sites. However, for one site, the use of the 0.25m² quadrat gave an estimate of 1% cover in 3 of 4 plots measured, and 16% in the other quadrat. The use of the larger quadrat averaged incorporated the high value into an average percentage cover of 5% at the site. The use of the larger quadrat was not tested along the shoreline where coverage can be quite variable. Trade-offs (with use of a 1m² quadrat compared with a 0.25m²) are: potentially a better accounting for the variation at a site with the larger quadrat: however, several minutes more of time are required in estimating coverage in each of the sub-quadrats; and additional time photographing each sub-quadrat with the larger quadrat. In addition, it is difficult to photograph the whole 1 m² quadrat in one photo. There is also potentially much additional time required for sampling biomass using the larger quadrat compared with the 0.25m² quadrat. Use of the 0.25m² quadrat may be most suitable for a routine monitoring program for Tahoe, since results were generally close to those made with the 1m² quadrat, but more rapidly made. However, additional evaluation may want to be done to see if there is benefit to using the larger quadrat along the shoreline where variation in coverage can be high. It is interesting to note that for studies assessing macroalgae coverage over coral reefs, a 0.25 m² quadrat was recommended as sufficient (Green et al., 2000).

There were slight differences in field estimates of percent cover made by the SCUBA diver and the snorkeler. In part, some of these differences may be attributable to the poor water clarity and difficulty of seeing metaphyton coverage from the surface of the water snorkeling compared with the better view by the SCUBA diver. In part, it may be due to subjective differences in estimates of coverage made by two different observers. In general though there were only slight

differences (3-6%) between estimates made by the diver and the snorkeler. When clarity is good, observations and photography while snorkeling can be done. If clarity is poor or water depth is such that it is not possible to distinguish the algae along the bottom, use of SCUBA will be necessary.

It was desirable to obtain some estimate of the relative accuracy with which mean percent cover could be determined with reasonable numbers of samples collected. Based on sampling in the pilot study, we would estimate a “reasonable number” of quadrats to be near 12 on the upper end to be manageable (percent cover sampling time of one hour or less). To determine the number of quadrats to sample to estimate different 95% confidence intervals for the means, we applied an equation from Green et al., 2000 (Eq. 1 below). This equation is used to estimate the number of samples needed for known confidence interval widths and a known estimate of variance determined from a pilot study.

$$\text{Eq. 1: } n = S^2 * t^2_{\alpha/2, (n-1)} / d^2.$$

Where: n = is the sample size; s^2 is the sample variance (calculated with $v = n-1$ degrees of freedom; $t^2_{\alpha/2, (n-1)}$ is the two-tailed critical value of Student's t with $v = n-1$ degrees of freedom and d is the half-width of the desired confidence interval.

Percent cover data from Regan and El Dorado beaches for shoreline and offshore samples was used to estimate the population variance S^2 and estimates of the number of samples required for different confidence intervals were determined. The results for these calculations are included in Appendix Table 2. For a moderately variable set of samples (range 5.4-22.4% cover, mean 15% cover, Std. Dev. = 6.3% cover, $n=7$) from El Dorado beach on 10/7/15, 9 replicate quadrats were estimated to be needed to estimate the mean with 95% confidence interval of 10% cover. For highly variable shoreline metaphyton, i.e. Regan Beach 8/20/15 (range 6-70% cover, mean 36.8% cover Std. Dev. = 28.2%, $n=6$) 10 replicate quadrats would be needed to estimate the mean with a large 40% confidence interval. In the pilot study, we sampled up to 7 quadrats along shore for shoreline monitoring and up to 12 quadrats offshore. Based on the heterogeneity in metaphyton observed in the study, particularly along the shoreline, it is recommended that the number of replicate quadrats sampled be at comparable or higher levels to estimate the mean metaphyton levels at sites. Sampling with just a few quadrats along the shoreline may not provide an estimate of the mean with enough accuracy to make meaningful comparisons between sites and between years.

For offshore data, the data ranges were generally smaller than for the shoreline sites. For instance, for El Dorado beach on 8/6/15, the range for percent cover values was very small (i.e. 0-5 %) with mean $2.6 \pm \text{std. dev. } 1.7\%$, 7 replicates would be needed to achieve a 95% confidence interval of $\leq 3\%$ and 13 replicates to achieve a 95% confidence interval of $\leq 2\%$. Offshore data from Regan beach on 8/20/15 with a range from 2-20% cover and mean of 10.22%

and std. dev. of 6.34% indicated 7 sample replicates would be needed to define a 95% confidence interval of 12% and 12 replicates for a 95% confidence interval of 8%.

Percent coverage proved to be a relatively rapid method to provide an assessment of the levels of metaphyton present. The extent to which the percent coverage related to other measures of biomass is examined below.

Estimates of Metaphyton Percent Cover from Photos

The results for percent cover determined using photos are presented in Tables 10a to 14a and graphically in Figures 27b to 31b. Photography of the metaphyton in the quadrats took about 5 minutes per site. Comparing percent cover data from field estimates with data from photos, some patterns were apparent. Estimates of percent cover from photos generally showed similar patterns among sample replicates as the field estimates, but the levels of percent cover often differed slightly. For instance percent cover estimates for Regan Beach on 8/20/15 for shoreline left, center and right sites were 50%, 70% and 15% estimated in the field and 60%, 78% and 1% based on the estimate from the photo. At low percent cover levels, there were several incidences where the value from the photo was indicated as 0% and the field estimate was a value greater than 0%. For instance note the large number of “0” values for percent cover determined from photos for Round Hill Pines beach on 9/24/15 (Figure 31.b.), while only one of the field estimates was 0 (Figure 31.a.). Possible explanations for these differences were that small amounts of metaphyton were not readily visible in the photos or that the visual estimates for low levels of metaphyton overestimated the amount present.

When metaphyton coverage was very high, the field estimates of percent cover were often lower than estimates obtained from photos (note the pattern Regan on 8/20/15 described above). Differences between field estimates of cover and estimates from the photos might be reduced with additional training of field researchers in discerning various levels of percent cover. Field guides showing different levels of percent cover on cards or photos might be helpful for researchers to refer to in the field.

Both field researchers and technicians working with photos must be skilled at identifying algae from other material covering the bottom. In the field, it is possible to examine the whole quadrat closely and account for algae that may be moving within and potentially in or out of the quadrat. A technician working with a photo does not have the option to closely inspect what may or may not be algae. Very low levels of algae may be missed in photos. On the other hand, as indicated above, photos may be beneficial in more accurately estimating high levels of percent cover.

The following conditions were found to increase the quality of photos for use in estimating amounts of metaphyton:

- Calm conditions – When nearshore water was very shallow such as in 2015, photography of metaphyton from above the lake surface was necessary. To see and photograph the

metaphyton without distortion from above the water, calm conditions were needed.² We typically did the metaphyton monitoring early in the morning to take advantage of calm conditions. When photographing underwater, problems associated with distortion were removed. However, movement of water in the nearshore associated with wind and waves can cause the metaphyton to move along the bottom in shallow water. This required rapid monitoring at sites if algae was moving into and out of the quadrats.

- Uniform shading of the quadrat - when photographing from above the water surface shading the whole area photographed reduced glare and reflections off the bottom.
- Clear water – when photographing underwater, clear water is obviously best. Working when the lake is calm and waves have not stirred up turbidity in the nearshore can help. Care should be taken not to stir up metaphyton along the bottom with fins or by walking along the bottom. If the lake is very turbid, SCUBA may be necessary to see the metaphyton near the bottom, photography of the quadrat may not be possible.

In 2017, underwater observations of cover and photography was done when the lake was approximately 6 feet deep. Underwater photography removed problems associated with waves and ripples. However, at some sites visibility of the quadrat along the bottom while snorkeling at the surface was much reduced due to poor water clarity. A Scuba diver was needed in such situations to estimate cover. Clear photos of the whole quadrat from above were not possible to obtain under conditions of poor water clarity.

60°C Dry Weight

All metaphyton enclosed within a 0.25 m² quadrat was collected for biomass estimates. Along the shoreline, these collections were made by picking the metaphyton off the surface of the beach using forceps. In the water, collections were made by scooping the algae in a small aquarium fish net (see figure 22). The metaphyton collected was returned to the lab and samples processed.

Tables 10.b to 14.b and Figures 27.c to 31.c show the results for 60°C dry weight. Dry weight showed similar patterns as visual estimates of percent cover at several of the sites. For instance note the similarity between patterns for field estimates of percent cover and Dry Weight for shoreline samples from: El Dorado Beach 8/6/15 (Figure 27), and Regan Beach on 8/20/15

²To allow viewing of quadrats in shallow water when the surface was rough we tried placing a large rectangular float with a clear plastic sheet on the water surface above the quadrat. We only had limited success using this as often either air bubbles were caught underneath the glass obscuring view of the bottom, or water splashed over the top of the plastic partially obscuring viewing. This viewer could be redesigned to improve use in the future by adding sides to prevent over-topping and minimizing surfaces which trap air underwater.

(Figure 29) and 9/21/15 (Figure 30). However, there was less similarity between coverage and dry weight for sites with low levels of percent cover (i.e. at Round Hill Pines on both dates and for many of the offshore sites at all sites, see Figures 28 and 31). The similarity between the patterns for percent cover and dry weight at certain sites may be due to the contributions the sand makes to the bulk of the algal mass, i.e. a large mass of sand mixed in with the algae increased the coverage of the material.



Figure 22. SCUBA diver collecting metaphyton within a quadrat at Regan Beach, 8/9/17. A small fishnet was used to scoop algae along the bottom.

The tendency of Lake Tahoe metaphyton to have much sand associated with it however, makes dry weight not a very reliable estimator of metaphyton biomass. There is typically variable amounts of sand mixed in with the algae (i.e. see Figure 23). This is true both for algae deposited along the shoreline and also often for clumps of algae rolling along the shallow lake bottom. We attempted to separate the sand fraction from the algae fractions in analyses, and then combine the individual results to determine an overall value (Dry weight, AFDW or chlorophyll *a*). Typically, it was not possible to completely separate sand and algae. Appendix Tables 1a-1e include the results for algae and sand fractions and the combined totals. The proportion of sand to algae is often varied among samples and sometimes sand constituted the major portion of the sample. For instance, sand in shoreline samples at El Dorado beach on 8/6/15 was 579.99, 661.02 and 927.92 g/m², these were 38%, 6% and 18% of the total sample dry weight respectively. Regan Beach shoreline metaphyton samples collected 8/20/15 had less sand

content but a greater proportion of the sample was sand (i.e. the sand dry weights for shoreline samples were 364.92, 349.40, 317.42 g/m², but these represented 87%, 61% and 91% of the total sample dry weights respectively). With a variable and sometimes large contribution of sand to the weight of the metaphyton, total sample dry weight is not a reliable indicator of metaphyton algae biomass. The process of separation of sand and algae is also time consuming and typically, not all the sand is separated from the algae. There can also be other material in the metaphyton, which is not algal biomass yet contributes to the weight. A measure more specific to algal biomass than dry weight is needed for Tahoe metaphyton.



Figure 23. Example of metaphyton samples from Regan Beach on shore, on 9/21/15, separated into sand and various organic and inorganic fractions. Separated algae fractions are on 3 pieces of white weighing paper at the top (Regan shoreline sites “Left” A-1, “Center” A-2, and “Right” A-3. Separated sand fractions for each site are below on foil (A-1) and two white weighing. Pieces of aquatic plants and clamshells were also separated from the metaphyton collected.

AFDW

Ash Free Dry Weight was also evaluated as potential measure of metaphyton algae biomass. This method provides a rapid estimate of the organic matter present in the sample based on the weight loss of dried material after combustion at very high temperature (500°C). When pieces of plants and woody debris are removed, the AFDW should be primarily due to the metaphyton algae. Some additional weight loss may be due to other organic debris, bacteria and loss of water remaining in the material after drying. A particular benefit of the method is that it may not be necessary to separate sand from algal material in the analysis, which may result in less lab processing time. AFDW provides an estimate of the organic content of the sample, and as long as the sample is well mixed, the content of sand should not affect the AFDW estimate.

The results for total sample AFDW are reported in Tables 10.b-14.b and Figures 27.d-31.d. Total sample AFDW ranged from 0 to a high of 35 g/m². The general patterns for AFDW were similar to percent cover in some but not all samplings. They were similar at Regan Beach on both sampling dates (8/20/15 and 9/21/15, see Figures 29 and 30), and at Round Hill Pines 9/24/15 (Figure 31). At El Dorado Beach on 8/6/15 (Figure 27) however, the patterns for shoreline AFDW and percent cover were slightly different. For percent cover the middle shoreline sample (30%) was lower than the two outer samples (70% and 75% cover). In contrast, for AFDW, the middle and left sites' AFDWs were similar (near 34 g/m²) and higher than the right side AFDW (near 25 g/m²). AFDW and percent cover do not always indicate the same metaphyton patterns.

The results for analysis of the individual fractions showed that the sand fraction typically had very little AFDW (see Appendix 1). For a few sites we did not analyze the AFDW associated with the sand fraction and the AFDW of the algae fraction alone was used to estimate the sample AFDW (these are the gray bars in the AFDW figures, i.e. see Figure 29 for Regan Beach 8/20/15. In a few samples in which the proportion of sand was very high, the contribution of AFDW with the sand was a significant contributor to total sample AFDW. For instance two of the Regan Beach shoreline samples collected 8/20/15 had sand content of 87% and 91% of the total dry weight. The AFDW associated with sand in these samples contributed 50% and 47% of the total sample AFDW. At Round Hill Pines 8/11/15, a few small sand samples also contributed substantially to the overall AFDW (these were the three samples in Figure 28.d with the largest AFDW, the sand contribution of AFDW was 51%, 87% and 46%. So occasionally, sand can contribute substantially to the AFDW.

AFDW provides a simple, rapid means to estimate metaphyton biomass. The AFDW method is easily done in the lab over a 24-hour period. The analysis can be done on a larger subsample (than for chlorophyll *a*), which is likely a better representatopn of total sample biomass. Although we separated algae and sand fractions in our testing we cannot see a major benefit in doing this in future monitoring, it should be possible to run AFDW on samples not separated into

the different fractions. This would speed the lab processing time greatly. AFDW appears to be a good method for estimates of metaphyton biomass.

Chlorophyll *a*

Chlorophyll *a* was measured in samples of metaphyton to see whether it might be a good estimator of metaphyton biomass. Chlorophyll *a* is an algal pigment, which is commonly used as a measure of algal biomass. Chlorophyll *a* is involved in energy capture from sunlight in the photosynthetic production of biomass within the algae. It is assumed that levels of chlorophyll *a* show a general relationship to algal biomass. However, the particular levels of chlorophyll *a* may depend on the types of algae present, the physiological states of the cells, and the amount of degradation of the cells.

In our analysis of metaphyton chlorophyll *a*, we were concerned that the large sand content in samples might result in variable estimates of total sample chlorophyll *a*. This is because chlorophyll *a* measurements are made on small subsamples of the metaphyton and differences in the proportion of sand can result in different estimates of chlorophyll *a*. The chlorophyll *a* results for sand and algae fractions are reported in Appendix A along with the total sample chlorophyll *a* levels. For several samples, we did not analyze the chlorophyll *a* associated with the sand fraction and the chlorophyll *a* of the algae fraction alone was used (these are represented by gray bars in the Figures (i.e. see figure 27.e El Dorado Chlorophyll *a* on 8/6/15).

Table's 10.c-14.c and Figures 27.e-31.e present the results for total chlorophyll *a*. Total sample chlorophyll *a* ranged from 0 to near 80 mg/m². The patterns for chlorophyll *a* seen in the figures were similar to patterns for AFDW at many of the sites and sampling dates (i.e. Regan Beach 8/20/15, 9/21/15 (Figures 29 and 30), and Round Hill Pines 8/11/15 and 9/24/15 (Figures 28 and 31). Both AFDW and chlorophyll *a* appeared to be good measures of biomass on those dates. However, El Dorado beach shoreline metaphyton on 8/6/15 (Figure 27) showed different patterns for AFDW and Chlorophyll *a*. Chlorophyll *a* was highest in the middle shoreline site (79 mg/m²) and less at the left and right sites (37, 30 mg/m²). While AFDW at the middle site was similar to the left shoreline site (i.e. near 34 g/m²) and slightly less for the shoreline right site (25 g/m²), AFDW chlorophyll *a* do not always indicate the same metaphyton patterns.

The results for analysis of the individual algae and sand fractions for chlorophyll *a* showed that the sand fraction typically had very little chlorophyll *a*. However, there were some exceptions; these were the same samples for which sand also contributed substantially to the AFDW. For instance, a Regan Beach shoreline sample collected 8/20/15 had sand content of 87% and the sand fraction contributed 42% to the overall chlorophyll *a*. At Round Hill Pines 8/11/15 three samples with sand content of 51%, 87% and 46%, had chlorophyll *a* contributions associated with the sand representing 35%, 67% and 39% to the total sample chlorophyll *a*. So occasionally, sand can contribute substantially to the chlorophyll *a*.

Replicate subsamples of chlorophyll *a* were analyzed in a limited number of samples (See Appendix 1). A moderate degree of variation was observed in the samples. The coefficient of variation (standard deviation / mean) ranged from 0 to 74% with an average of 20% for samples from all sites for which subsample replicates were measured. In a couple of samples, the chlorophyll *a* replicates were quite different even though they came from samples in which the sand had been removed mostly from the algae fraction. These were shoreline samples from Regan Beach on 8/20/15. The left shoreline sample (A-1) had chlorophyll *a* content of 4.11 and 9.41 g/m² and the center site (A-2) had chlorophyll content of 12.95 and 41.11 g/m². In contrast, AFDW replication for these samples was much better, AFDW replicates in the left (A-1) sample were 4.17 and 4.24 g/m² and in the center site (A-2) were 21.57 and 19.58 g/m². There is likely inherent variability in the metaphyton chlorophyll *a* samples, since the metaphyton material may be an aggregation of various types of algae and algae in different states of health and degradation. Examination of the metaphyton under the microscope bore this out. Often samples contained a mix of algae types. Some samples had algae that appeared quite healthy with bright green chloroplasts. In other samples, many of the cells were damaged and little chlorophyll was present. The color of the metaphyton observed at the sites ranged from various shades of green, to light brown to black decaying algae.

As an example of different chlorophyll *a* and AFDW associations, the relationship between chlorophyll *a* and AFDW at Regan Beach on 8/20/15 for all samples is plotted in Figure 24. A couple of samples A-1 and B-1 had notably lower chlorophyll *a* relative to AFDW compared with the other samples. The metaphyton at sites A-1 and B-1 also had a light brown color. Figure 25 shows the site A-1 algae, which appears brown and old. Examination of site A-1 and site B-1 algae under the microscope indicated the predominant algae was *Spirogyra* at both sites and there was also organic detritus noted in the samples. Site A-2 in contrast appeared to be mostly green (Figure 26). There was greater mean chlorophyll *a* in samples relative to AFDW compared with sites A-1 and B-1. The predominant algae was noted to be *Spirogyra* and organic detritus was not noted in the sample.

With the presence of algae in various states of health and mixed algal types in the metaphyton, there is likelihood that the relationship between chlorophyll *a* and metaphyton biomass is variable. Whether algae is fresh or old along a beach may not make too much difference visually (with regard to smell, presence of decaying algae is likely more disagreeable), but the overall amount of organic matter may be of primary importance. To characterize metaphyton in a range of conditions present along the beach or offshore, AFDW may provide a more representative estimate of organic material present than chlorophyll *a*.

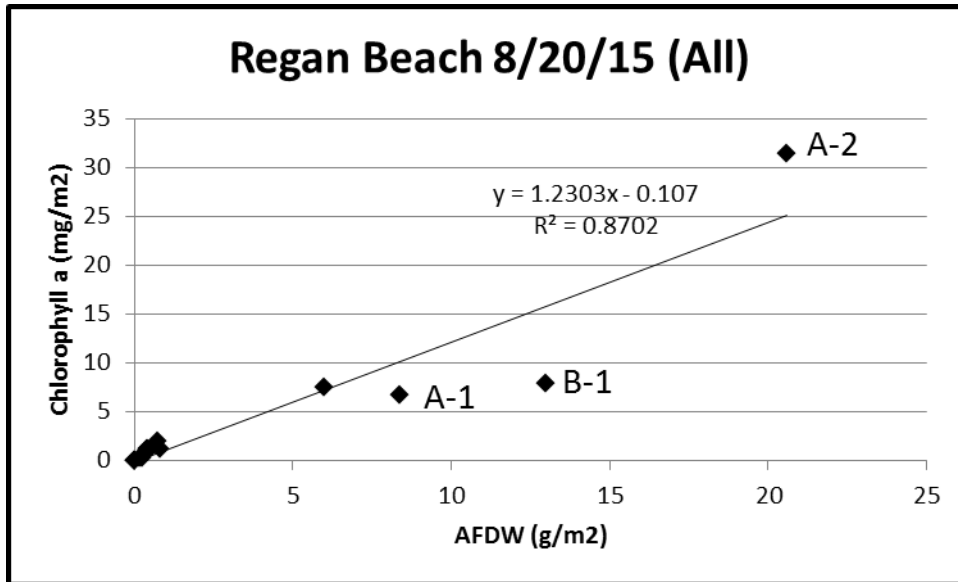


Figure 24. Association between AFDW and chlorophyll *a* for samples collected at Regan Beach on 8/20/15. The data point for site A-2 represents sample with green color and greater chlorophyll *a* relative to AFDW than site B-1 which was brown in color and likely had older degraded material, with lower chlorophyll *a* relative to AFDW (see photos below: Figures 25, 26).



Figure 25. Regan Beach 8/20/15 site B-1. The metaphyton algae had a brown appearance and possibly represented older or degrading algae. This site had low chlorophyll *a* relative to AFDW organic content (see Figure 24 above).



[Figure 26](#). Regan Beach 8/20/15 shoreline site A-2. The metaphyton algae was mostly various shades of green. The site had higher chlorophyll *a* relative to AFDW organic content (see Figure 24 above.)

Table 10.a. El Dorado Beach, 8/6/15, metaphyton percent cover estimated in the field “Field Est.” using a 0.25 m² quadrat and percent cover estimated from calculation of coverage from a photograph “Photo”.

El Dorado Beach 8/6/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d.(n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)
Shoreline	70%	30%	75%	58 ± 25% (3)	55%	42%	86%	61 ± 23% (3)
Offshore:								
18.3m / 20yd	1%	3%	5%	3 ± 2% (3)	0%	3%	1%	1 ± 2% (3)
45.7 m/ 50yd	0%	2%	0%	1 ± 1% (3)	0%	1%	0%	0 ± 1% (3)
91.4 m/ 100yd	2%	3%	4%	3 ± 1% (3)	1%	1%	3%	2 ± 1% (3)
137.2 m/150yd	3%	3%	5%	4 ± 1% (3)	2%	0%	8%	3 ± 4% (3)
Offshore Mean±s.d. (n)	2±1% (4)	3±0.1% (4)	4%±2% (4)	Overall Offshore 3 ± 2% (12)	1±1% (4)	1±1% (4)	3±4% (4)	Overall Offshore 2 ± 2% (12)

Table 10.b. El Dorado Beach 8/6/15, metaphyton 60°C Dry Weight and Ash Free Dry Weight (AFDW) in 0.25 m² quadrat.

El Dorado Beach 8/6/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d.	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d.(n)
Distance Offshore in (m) /and (yd)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)
Shoreline	1510.18	703.26	1134.73	1116.06±403.78 (3)	34.92	33.78	24.88	31.19±5.50 (3)
Offshore:								
18.3m / 20yd	2.13	18.29	12.17	10.86±8.16 (3)	0.06	0.27	0.15	0.16±0.11 (3)
45.7 m/ 50yd	0.00	2.60	0.00	0.87±1.50 (3)	0.00	0.13	0.00	0.04±0.08 (3)
91.4 m/ 100yd	1.73	2.50	5.49	3.24±1.99 (3)	0.07	0.13	0.26	0.15±0.10 (3)
137.2 m/150yd	10.43	1.29	24.7	12.14±11.80 (3)	0.13	0.07	0.62	0.27±0.30 (3)
Offshore Mean±s.d. (n)	3.57±4.66 (4)	6.17±8.10 (4)	10.59±10.64 (4)	Overall Offshore 6.78±7.99 (12)	0.06±0.05 (4)	0.15±0.08 (4)	0.26±0.26 (4)	Overall Offshore 0.16±0.17(12)

Table 10. c. El Dorado Beach 8/6/15, metaphyton Chlorophyll *a* in 0.25 m² quadrat.

El Dorado Beach 8/6/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)
Shoreline	37.07	78.96 a ¹	29.67	48.57±26.58 (3)
Offshore:				
18.3m / 20yd	0.11 a ²	0.63 a ³	0.36 a ⁴	0.37±0.26 (3)
45.7 m/ 50yd	0.00	0.34	0.00	0.11±0.20 (3)
91.4 m/ 100yd	0.19	0.35	0.62	0.39±0.22 (3)
137.2 m/150yd	0.19 a ⁵	0.17	1.54 a ⁶	0.63±0.79 (3)
Offshore Mean±s.d. (n)	0.12±0.09 (4)	0.37±0.19 (4)	0.63±0.66 (4)	0.38±0.42 (12)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis, (sand not analyzed): a¹-6%; a²-71%; a³-75%; a⁴-96%; a⁵-84%; a⁶-26%;

Table 11. a. Round Hill Pines 8/11/15, metaphyton percent cover in 0.25 m² quadrat, estimated in the field “Field Est.” and percent cover estimated from calculation of coverage from a photograph “Photo”.

Round Hill Pines Beach 8/11/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)
Shoreline	0%	0%	0%	0±0% (3)	0%	0%	0%	0±0% (3)
Offshore:								
2.7m/3yd	9%	5%	<1% ¹	5±4% (3)	NA	NA	NA	NA
22.9m/25yd	3%	6%	14%	8±6% (3)	NA	NA	NA	NA
45.7m/50 yd	3%	4%	NA	4±1% (2)	0.2%	0.5%	NA	0.4±0.2% (2)
Offshore Mean±s.d. (n)	5±4% (3)	5±1% (3)	7±10% (2)	6±4% (8)	0.2%	0.5%	NA	0.4±0.2% (2)

Notes: 1- for values “<” estimated as 1/2 value to calculate averages.

Table 11.b. Round Hill Pines 8/11/15, metaphyton 60°C Dry Weight and Ash Free Dry Weight in 0.25 m² quadrat.

Round Hill Pines Beach 8/11/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d.(n)
Distance Offshore in (m) /and (yd)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)
Shoreline	0	0	0	0± 0 (3)	0	0	0	0±0 (3)
Offshore:								
2.7m/3yd	301.51	0	338.80	213.44±185.78 (3)	2.04	0	1.17	1.07±1.02 (3)
22.9m/25yd	1.14	1.62	278.30	93.69±159.88 (3)	0.06	0.20	3.54	1.27±1.97 (3)
45.7m/50 yd	1.75	0.99	NA	1.37±0.54 (2)	0.09	0.05	NA	0.07±0.03 (2)
Offshore Mean±s.d.(n)	101.47±173.24(3)	0.88±0.82(3)	308.55±42.78 (2)	115.51±158.75 (8)	0.73±1.13 (3)	0.08±0.10 (3)	2.36±1.68 (2)	0.89±1.29 (8)

Table 11. c. Round Hill Pines Beach 8/11/15, metaphyton Chlorophyll *a* in 0.25 m² quadrat.

Round Hill Pines Beach 8/11/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)
Shoreline	0	0	0	0±0 (3)
Offshore:				
2.7m/3yd	1.39	0	0.73	0.70±0.69
22.9m/25yd	0.06 a ¹	0.31 a ²	3.73	1.37±2.05
45.7m/50 yd	0.09 a ³	0.05 a ⁴	NA	0.07±0.03
Offshore Mean±s.d. (n)	0.51±0.75 (3)	0.12±0.16 (3)	2.23±2.12 (2)	0.80±1.28 (8)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis, (sand not analyzed): a¹-83%; a²-49%; a³-23%; a⁴-80%;

Table 12. a. Regan Beach 8/20/15, metaphyton percent cover in 0.25 m² quadrat, estimated in the field “Field Est.” and percent cover estimated from calculation of coverage from a photograph “Photo”.

Regan Beach 8/20/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)
Shoreline	50%	70%	15%	45±28% (3)	60%	78%	1%	46±40% (3)
9.1m/ 10yd	65%	15%	6%	29±32% (3)	80%	6%	2%	29±44%
Shoreline Mean±s.d.(n)	58±11% (2)	43±39% (2)	11±6% (2)	37±28% (6)	70±14% (2)	42±51% (2)	2±1% (2)	38±39% (6)
Offshore:								
45.7 m/ 50yd	2%	5%	3%	3±2% (2)	0%	0%	0%	0±0%
91.4 m/ 100yd	10%	7%	15%	11±4% (3)	5%	3%	NA	4±1%
137.2 m/150yd	15%	15%	20%	17±3% (3)	4%	3%	11%	6±4%
Offshore Mean±s.d.(n)	9±7% (3)	9±5% (3)	13±9% (3)	10±6% (9)	3%±3%(3)	2±2% (3)	6±8% (3)	3±4% (8)

Table 12. b. Regan Beach 8/20/15, metaphyton 60°C Dry Weight and Ash Free Dry Weight in 0.25 m² quadrat.

Regan Beach 8/20/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)
Shoreline	421.24	576.97	348.92	449.04±116.54 (3)	8.37	20.58 a ¹	6.01	11.65±7.82 (3)
9.1m/ 10yd	384.14	19.24	5.48	136.29±214.76 (3)	12.98	0.83	0.16	4.66±7.22 (3)
Shoreline Mean±s.d.(n)	402.69±26.23 (2)	298.11±394.37 (2)	177.2±242.85 (2)	292.67±230.71 (6)	10.68±3.26 (2)	10.71±13.97 (2)	3.09±4.14 (2)	8.16±7.74 (6)
Offshore:								
45.7 m/ 50yd	0.01	3.10	0.36	1.16±1.69 (3)	0.01	0.03	0.01	0.02±0.01 (3)
91.4 m/ 100yd	13.40	6.50	8.54	9.48±3.55 (3)	0.42	0.24	0.46	0.37±0.12 (3)
137.2 m/150yd	4.86	7.76	11.11	7.91±3.13 (3)	0.42	0.43	0.75	0.53±0.19 (3)
Mean±s.d.(n)	6.09±6.78 (3)	5.79±2.41 (3)	6.67±5.61 (3)	6.18±4.58 (9)	0.28±0.24 (3)	0.23±0.20 (3)	0.41±0.37 (3)	0.31±0.25 (9)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis, (sand not analyzed: a¹-61%)

Table 12. c. Regan Beach 8/20/15, metaphyton Chlorophyll *a* in 0.25 m² quadrat.

Regan Beach 8/20/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)
Shoreline	6.76 a ¹	31.44	7.58	15.26±14.02 (3)
9.1m/ 10yd	7.88 a ²	1.21	0.28 a ³	3.12±4.15 (3)
Shoreline Mean±s.d. (n)	7.32±0.79 (2)	16.33±21.38 (2)	3.93±5.16 (2)	9.19±11.39 (6)
Offshore:				
45.7 m/ 50yd	0.01	0.04 a ⁴	0.02 a ⁵	0.02±0.02 (3)
91.4 m/ 100yd	0.94 a ⁶	0.32 a ⁷	1.08 a ⁸	0.78±0.40 (3)
137.2 m/150yd	1.24	1.21	1.99	1.48±0.44 (3)
Offshore Mean±s.d. (n)	0.73±0.64 (3)	0.52±0.61 (3)	1.03±0.99 (3)	0.76±0.70 (9)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis, sand not analyzed: a¹-87%; a²-80%; a³-76%; a⁴-99%; a⁵-97%; a⁶-66%; a⁷-50%; a⁸-25%;

Table 13. a. Regan Beach 9/21/15, metaphyton percent cover in 0.25 m² quadrat, estimated in the field “Field Est.” and percent cover estimated from calculation of coverage from a photograph “Photo”.

Regan Beach 9/21/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)
Shoreline	60%	15%	5%	27±29% (3)	80%	9%	1%	30±44% (3)
9.1m/ 10yd	1%	35%	20%	19±17% (3)	0%	37%	13%	17±19% (3)
Shoreline Mean±s.d. (n)	31±42% (2)	25±14% (2)	13±11% (2)	Overall 23±22% (6)	40±57% (2)	23±20% (2)	7±8% (2)	Overall 23±31% (6)
Offshore:								
45.7 m/ 50yd	1%	2%	0%	1±1% (3)	0%	0%	0%	0±0% (3)
91.4 m/100yd	2%	5%	10%	6±4% (3)	0%	1%	4%	2±2% (3)
137.2 m/150yd	2%	10%	7%	6±4% (3)	0%	1%	3%	1±2% (3)
Offshore Mean±s.d. (n)	2±1% (3)	6±4% (3)	6±5% (3)	4±4% (9)	0±0% (3)	1±1% (3)	2±2% (3)	1±2% (9)

Table 13.b. Regan Beach 9/21/15, metaphyton 60°C Dry Weight and Ash Free Dry Weight in 0.25 m² quadrat.

Regan Beach 9/21/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)
Shoreline	681.21	43.70	24.22	249.71±373.82 (3)	18.45	1.34	0.61	6.80±10.10 (3)
9.1m/ 10yd	0.27	217.27	68.52	95.35±110.96 (3)	0.01	7.32	2.64	3.32±3.70 (3)
Shoreline Mean±s.d. (n)	340.74±481.50 (2)	130.49±122.73 (2)	46.37±31.32 (2)	Overall 172.53±260.71 (6)	9.23±13.04 (2)	4.33±4.23 (2)	1.63±1.44 (2)	Overall 5.06±7.06 (6)
Offshore:								
45.7 m/ 50yd	0.01	0.59	0	0.20±0.34 (3)	0.01	0.03	0	0.01±0.02 (3)
91.4 m/100yd	0.67	2.89	15.96	6.51±8.26 (3)	0.02	0.16	0.72	0.30±0.37 (3)
137.2 m/150yd	1.16	2.92	8.72	4.27±3.96 (3)	0.04	0.12	0.52	0.23±0.26 (3)
Offshore Mean±s.d. (n)	0.61±0.58 (3)	2.13±1.34 (3)	8.23±7.99 (3)	Offshore Mean 3.66±5.35 (9)	0.02±0.02 (3)	0.10±0.07 (3)	0.41±0.37 (3)	0.18±0.26 (9)

Table 13. c. Regan Beach 9/21/15, metaphyton Chlorophyll *a* in 0.25 m² quadrat.

Regan Beach 9/21/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)
Shoreline	36.25 a ¹	1.43	0.56 a ²	12.75±20.36 (3)
9.1m/ 10yd	NES	11.00 a ⁸	4.73 a ³	7.87±4.43 (2)
Shoreline Mean±s.d. (n)	36.25 (1)	6.22±6.77 (2)	2.65±2.95 (2)	Overall 10.79±14.81 (5)
Offshore:				
45.7 m/ 50yd	NES	0.06	NA	0.06 (1)
91.4 m/100yd	0.05 a ⁴	0.25	1.15	0.48±0.59 (3)
137.2 m/150yd	0.04 a ⁵	0.26 a ⁶	1.38 a ⁷	0.56±0.72 (3)
Offshore Mean±s.d. (n)	0.05±0.01 (2)	0.19±0.11 (3)	1.27±0.16 (2)	0.46±0.56 (7)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis, sand not analyzed : a¹-76%; a²-89%; a³-59%; a⁴-56%; a⁵-72%; a⁶-11%; a⁷-1% ,a⁸-53%

Table 14. a. Round Hill Pines 9/24/15, metaphyton percent cover in 0.25 m² quadrat, estimated in the field “Field Est.” and percent cover estimated from calculation of coverage from a photograph “Photo”.

Round Hill Pines Beach 9/24/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)
Shoreline	0%	0%	0%	0±0% (3)				
Offshore:								
2.7m/3yd	3%	1%	7%	4±3% (3)	0%	0%	1%	0.3±0.6% (3)
22.9m/25yd	2%	2%	2%	2±0% (3)	1%	0%	0%	0.3±0.6% (3)
45.7m/50 yd	<1% ¹	1%	0%	1± 1 % (3)	0%	0%	0%	0.0±0.0% (3)
91.4m/100yd	<1% ¹	1%	2%	1± % (3)	0%	0%	NU	0.0±0.0% (3)
Offshore Mean±s.d. (n)	1.5±1.2% (4)	1.3±0.5% (4)	2.8±3.0% (4)	1.7±1.9% (12)	0.3±0.5% (4)	0±0% (4)	0.3±0.6% (3)	0.2±0.4% (11)

Notes: 1- for values “<” estimated as 1/2 value to calculate averages; “NU” indicates photo not used

Table 14. b. Round Hill Pines Beach 9/24/15, metaphyton 60°C Dry Weight and Ash Free Dry Weight in 0.25 m² quadrat.

Round Hill Pines Beach 9/24/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	DW 60°C (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)	AFDW (g/m ²)
Shoreline	0	0	0	0±0 (3)	0	0	0	0±0 (3)
Offshore:								
2.7m/3yd	0.63	0	9.66	3.43±5.40 (3)	0.08	0	0.47	0.18±0.25 (3)
22.9m/25yd	1.41	0.12	.30	0.61±0.70 (3)	0.02	0.01	0.04	0.02±0.02
45.7m/50 yd	0	0	0	0±0 (3)	NES	NES	0.00	0 (1)
91.4m/100yd	0.07	0.07	0.14	0.09±0.04 (3)	0	0	0.01	0±0.01
Offshore Mean±s.d. (n)	0.53±0.65 (4)	0.05±0.06 (4)	2.53±4.76 (4)	1.15±3.19 (12)	0.03±0.04(3)	0±0.01(3)	0.13±0.23(4)	0.08±0.17(10)

Notes: “NES” is not enough sample for analysis.

Table 14.c. Round Hill Pines 9/24/15, metaphyton Chlorophyll *a* in 0.25 m² quadrat.

Round Hill Pines Beach 9/24/15	Left Sampling Point	Center Transect	Right Sampling Point	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)	Chl <i>a</i> (mg/m ²)
Shoreline	0	0	0	0±0 (3)
2.7m/3yd	0.10 a ¹	0	0.61	0.24±0.33 (3)
22.9m/25yd	0.02 a ²	0.02	0.05	0.03±0.02 (3)
45.7m/50 yd	0	NES	0	0±0 (2)
91.4m/100yd	0.01	0.01	0.02	0.01±0.01 (3)
Offshore Mean±s.d. (n)	0.03±0.05 (4)	0.01±0.01 (3)	0.17±0.29 (4)	0.08±0.18 (11)

Notes- (a) Percent of Dry Wt. as sand removed prior to sample analysis: a¹-37%; a²-80%

Table 15.a. El Dorado Beach 10/7/15 metaphyton percent cover in 0.25 m² quadrat, estimated in the field “Field Est.” and estimated from a photograph “Photo”.

El Dorado Beach 10/7/15	Left 45.7m/50yd	Left 22.9m/25yd	Center Transect	Right 22.9m/25yd	Right 45.7m/50yd	Right ¹ 49.4 m/54yd	Right 68.6m/75yd	Right 91.4m/100yd	Mean±s.d. (n)
Distance Offshore in (m) /and (yd)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)	Field Est. (% Cover)
Shoreline	22.4%	19.5%	10.4%	5.4%	17.4%	72% ¹	19.7%	10.0%	15.0±6.3% ¹ (7)
22.9 m/ 25yd	nm	0.5%	0.3%	0%	nm	nm	nm	nm	0.27±0.003% (3)
45.7 m/ 50yd	nm	0.3%	0.2%	0.3%	nm	nm	nm	nm	0.27±0.001% (3)
91.4 m/ 100yd	0.2%	<0.2% ³	0.2%	0.2%	0%	nm	nm	nm	0.14±0.001% (5)
137.2 m/150yd	0.2%	0%	0.4%	0.2%	0.2%	nm	nm	nm	0.20±0.001% (5)
Offshore	0.2±0.0%	0.2±0.2%	0.3±0.1%	0.2±0.1%		nm	nm	nm	Overall Offshore
Mean±s.d. (n)	(2)	(4)	(4)	(4)					0.21±0.14% (16)

Table 15.a.cont’d.

El Dorado Beach 10/7/15	Left 45.7m/50yd	Left 22.9m/25yd	Center Transect	Right 22.9m/25yd	Right 45.7m/50yd	Right 49.4 m/54yd	Right 68.6m/75yd	Right 91.4m/100yd	Mean±s.d.
Distance Offshore in (m) /and (yd)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo Est. (% Cover)	Photo (% Cover)	Photo (% Cover)	Photo Est. (% Cover)	Photo (% Cover)
Shoreline	20.7%	28.6%	12.1%	4.2%	19.3%	93% ¹	23.1%	3.5%	15.9±9.6% ¹ (7)
22.9 m/ 25yd	nm	0%	0%	nm	nm	nm	nm	nm	0±0% (2)
45.7 m/ 50yd	nm	0.3%	0%	nm	nm	nm	nm	nm	0.15±0.21% (2)
91.4 m/ 100yd	nm	0%	nm	nm	nm	nm	nm	nm	0% (1)
137.2 m/150yd	nm	nm	nm	nm	nm	nm	nm	nm	nm
Offshore	nm	0.1±0.2%	0.0±0.0%	nm	nm	nm	nm	nm	Overall Offshore
Mean±s.d. (n)		(3)	(2)						0.06±0.13% (5)

Notes –1- Observations at “Shoreline 49.4m/ 54 yds. Right”, were made to provide example of high level of algae along transect not accounted for by sampling at regular distances, the data was not included with average since would bias the estimate; 2- A 0.25m² quadrat with 10cm X 10cm squares denoted by string was used to make small estimates of percent cover; 3- for values “<” used estimate of 1/2 value to calculate averages; “nm” not measured.

Table 16. El Dorado Beach Site Data, 8/19/16 percent cover estimates.

El Dorado Beach 8/19/16	Center Transect	
Distance Offshore in (m) /and (yd)	Depth (cm)	Visual Estimate % Cover
Shoreline 1.5m/ 1.7yd	-	~20%
Offshore		
4.6m/ 5yd	12	<1% ¹
17.1m/ 18.7yd	21	1-2%
32m/ 35yd	22	0%
47.2m/ 51.7yd	26	0%
62.5m/ 68.3yd	32	<1% ¹
106.7m/ 116.7yd	41	10%
Offshore Mean±s.d. (n)		2.1±3.9% (6)

Notes: 1- for values “<” estimated as 1/2 value to calculate averages

Table 17. Regan Beach Site Data, 8/19/16 percent cover estimates.

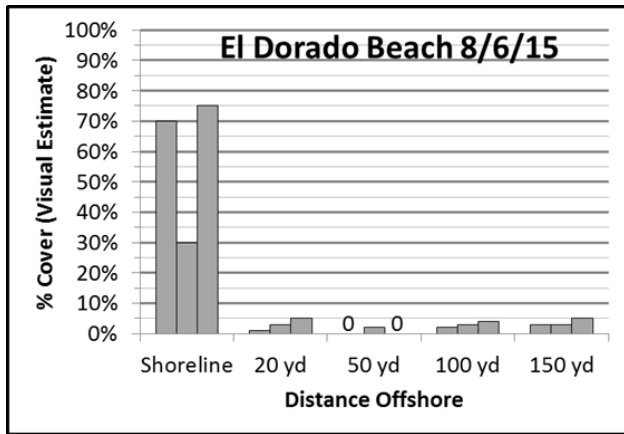
Regan Beach 8/19/16	Center Transect	
Distance Offshore in (m) /and (yd)	Depth (cm)	Visual Estimate % Cover
Shoreline 2.1m/ 2.3yd	0	~56%
3.0m/ 3.3yd	2	<1% ¹
Shoreline Mean±s.d. (n)		28±39% (2)
15.2m/ 16.7yd	8	1%
30.5m/ 33.3yd	10	<1% ¹
45.7m/ 50yd	14	2%
61m/ 66.7yd	19	3%
76.2m/ 83.3yd	23	0%
Shoreline Mean±s.d. (n)		1.3±1.2% (5)

Notes: 1- for values “<” estimated as 1/2 value to calculate averages

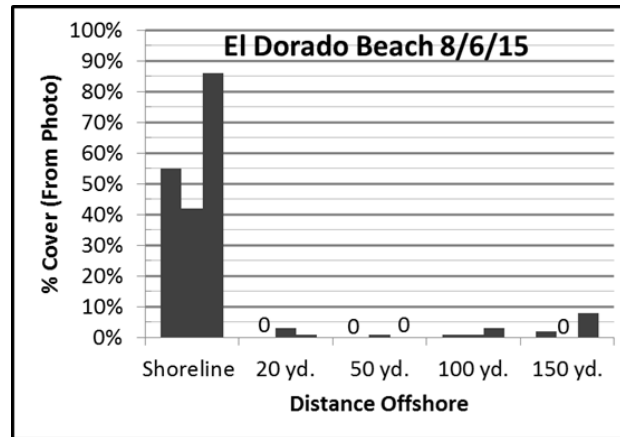
Table 18. Regan Beach metaphyton percent cover measurements 8/9/17. Sampling points were established on three transect lines separated by a distance of approximately 50 yards (45.7 meters) starting at a distance of 12 yds. (11m) away from shore. Observations were made by a diver using SCUBA (left set of observations in table) and a snorkeler on the surface, 4-5 feet above the bottom (right set of observations). A 1m² quadrat subdivided into four 0.25 m² quadrats was used to determine percent cover. The coverage in each 0.25m² quadrat was recorded and the mean of these gave the coverage for the 1m² quadrat. . Underwater visibility was very poor, particularly at the site closest to shore.

Regan Beach 8/9/17		46 m/50yd Left	Center Transect	46 m/50yd Right	46 m/50yd Left	Center Transect	46 m/50yd Right
		Observer 1 (w/SCUBA)	Observer 1 (w/SCUBA)	Observer 1 (w/SCUBA)	Observer2 (Snorkeling)	Observer 2 (Snorkeling)	Observer 2 (Snorkeling)
Distance from Shore		Visual Est. % Cover	Visual Est. % Cover	Visual Est. % Cover	Visual Est. % Cover	Visual Est. % Cover	Visual Est. % Cover
Shoreline					0%	0%	0%
11 m/ 12yd	4 X 0.25m ² Quadrats	All ~10% ¹	All~50% ¹	All<5% ¹	Poor Visib. ¹	Poor Visib. ¹	Poor Visib. ¹
11 m/ 12 yd	1m ² Quadrat Average	~10% ¹	~50% ¹	<5% ¹			
57 m/ 62yd	4 X 0.25m ² Quadrats	All ~10%	All <5%	All ~5%	NA	3%,2%, 2%,4%	5-7%,5-7%, 3-5%, 3-5%
57 m/ 62yd	1m ² Quadrat Average ± S.D.	~10%	<5%	5%	NA	3±1%	5±1%
102 m/ 112yd	4 X 0.25m ² Quadrats	80%,60%, 80%,80% ²	1%,1%, 1%, 5%	6%,13%, 10%, 13%	Algae over plants ²	1%,1%, 1%,5%	5%,6-7%, 3%,5-7%
102 m/ 112yd	1m ² Quadrat Average ± S.D.	75±10% ²	2±2%	11±3%		2±2%	5±2%
148 m/ 162 yd	4 X 0.25m ² Quadrats	5%,8%, 6%,10% ³	Total 70-80%	3%,2%, 13%,4 %	5-7%,3%, 4%,6% ³	Total ~70- 80%	1%,1%, 16%,1%
148 m/ 162 yd	1m ² Quadrat Average ± S.D.	7±2% ³	70-80% ⁴	6±5%	5±2% ³	70-80% ⁴	5±8%

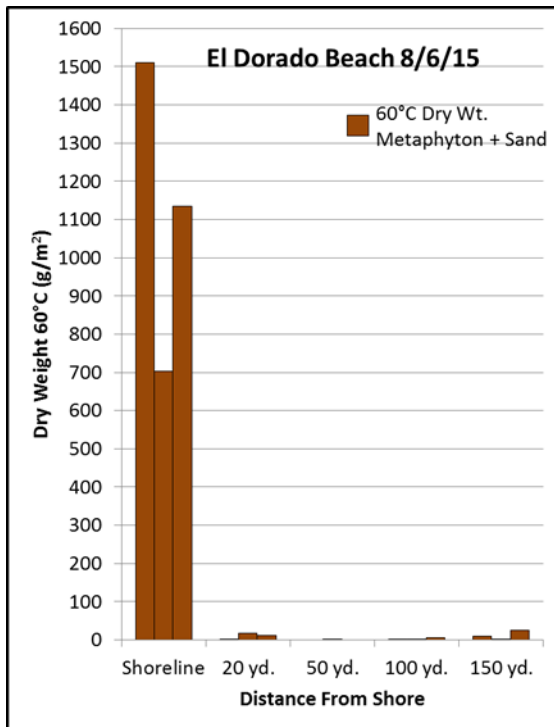
Notes: 1- Visibility underwater was very poor, snorkeler unable to see bottom to estimate coverage; 2- algae was associated with small aquatic plants, coverage estimate may include cover due to plants, snorkeler unable to discern cover from plants and metaphyton cover, do not include in site averages; 3- wind and water movement causing algae to move along bottom; 4- quadrat purposefully located over larger patch of metaphyton to see if the patch could be seen in aerial imaging, do not include in site average.



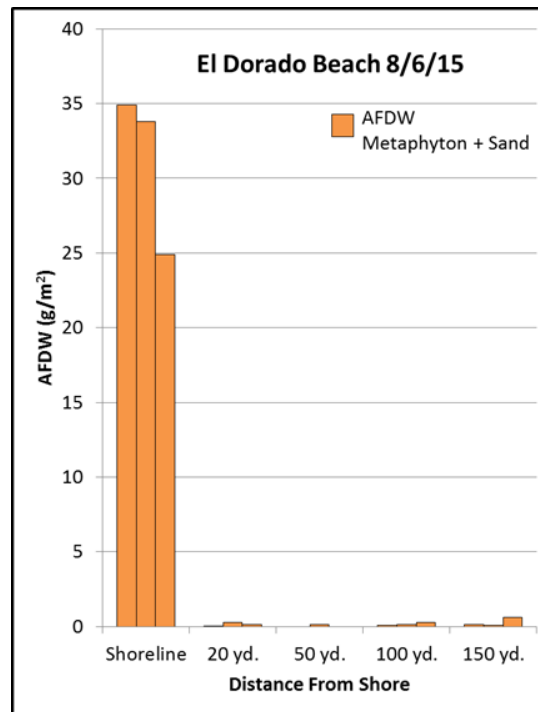
a. Metaphyton % Cover (Field Estimate)



b. Metaphyton % Cover (From Photo)

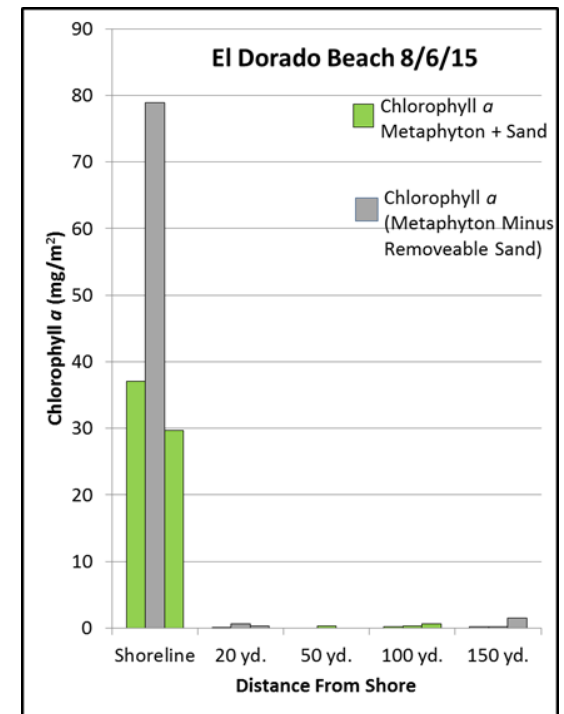


c. Metaphyton 60°C Dry Weight

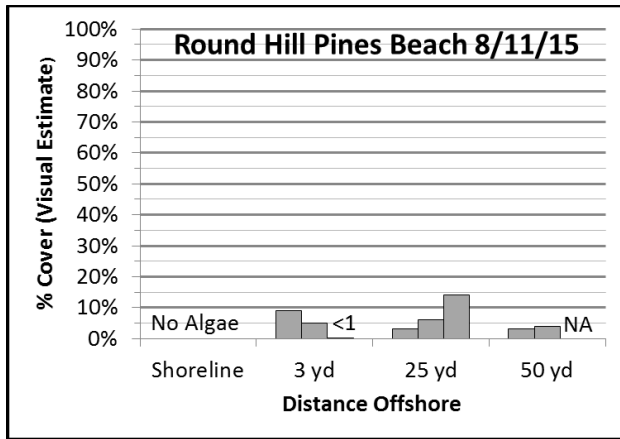


d. Metaphyton AFDW

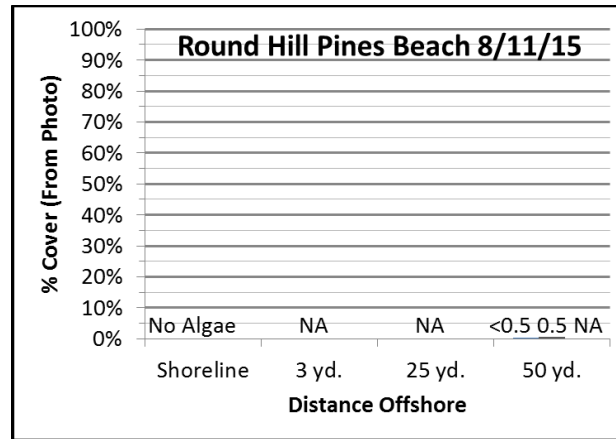
Figures 27a-e. El Dorado Beach 8/6/15 metaphyton monitoring results: (a) % cover, (field estimate); (b) % cover (from photo); (c) 60°C Dry Weight; (d) Ash Free Dry Weight (AFDW); Chlorophyll *a*. Bars represent left, center and right sample replicates at indicated distances offshore. Chlorophyll *a* minus sand is value for chlorophyll *a* of metaphyton after separating off removable sand.



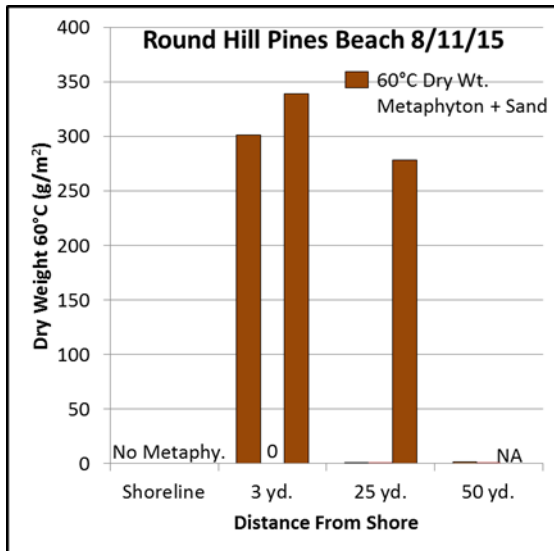
e. Metaphyton Chlorophyll *a*



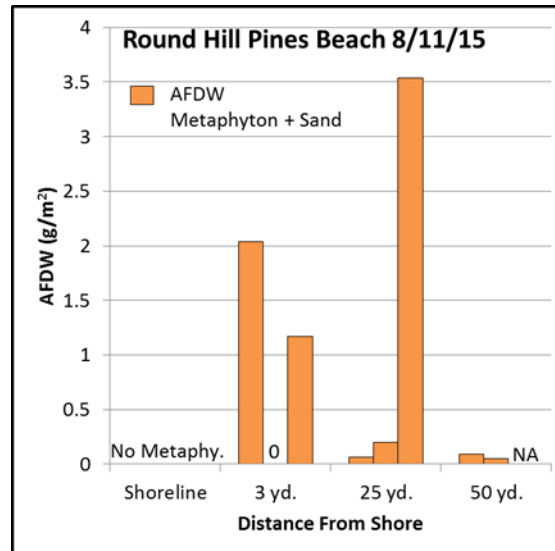
a. Metaphyton % Cover (Field Estimate)



b. Metaphyton % Cover (From Photo)

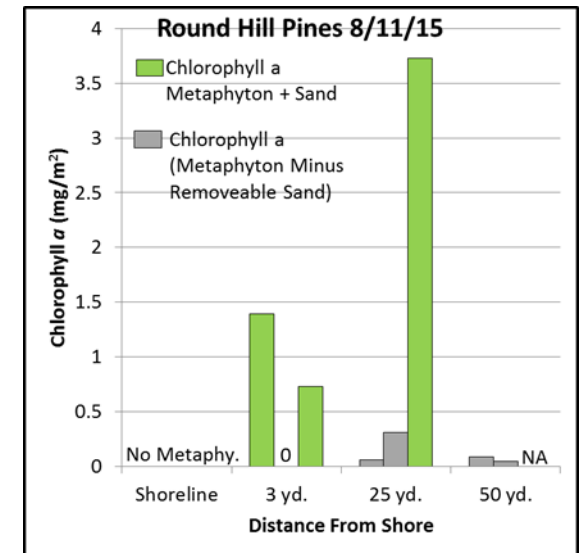


c. Metaphyton 60°C Dry Weight

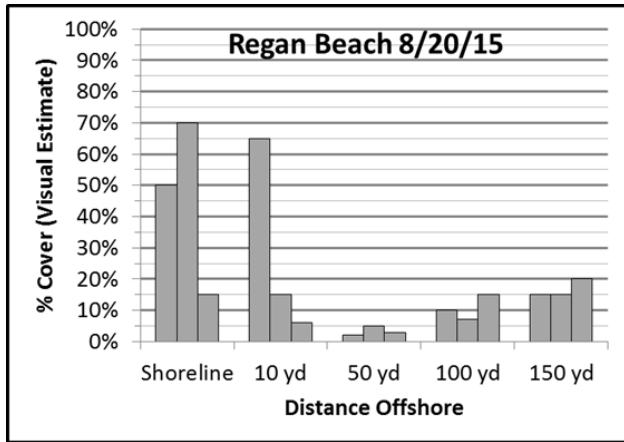


d. Metaphyton AFDW

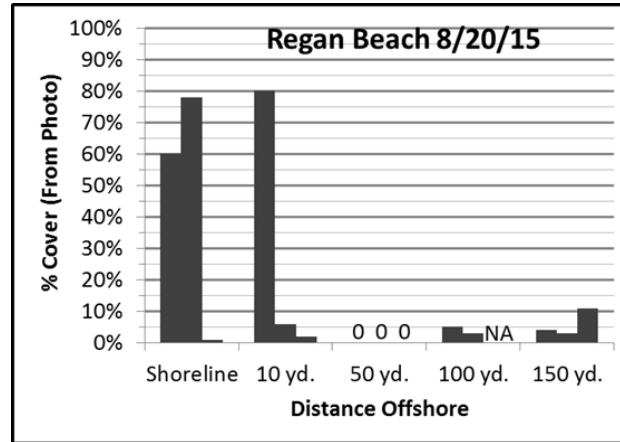
Figures 28a-e. Round Hill Pines Beach 8/11/15 metaphyton monitoring results: (a) % cover, (field estimate); (b) % cover (from photo); (c) 60°C Dry Weight; (d) Ash Free Dry Weight (AFDW); Chlorophyll *a*. Bars represent left, center and right sample replicates at indicated distances offshore. Chlorophyll *a* minus sand is value for chlorophyll *a* of metaphyton after separating off removable sand.



e. Metaphyton Chlorophyll *a*

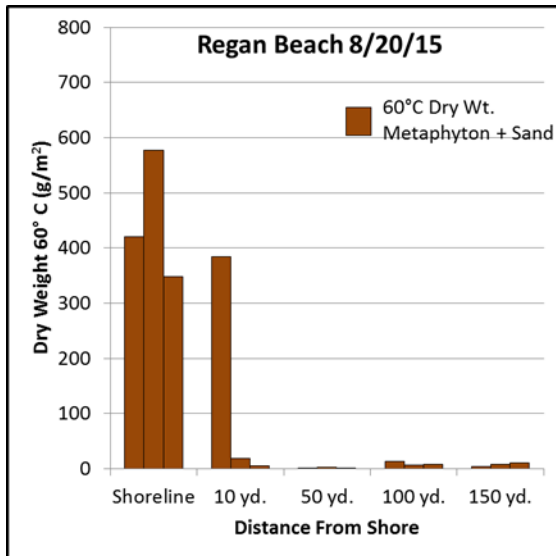


a. Metaphyton % Cover (Field Estimate)

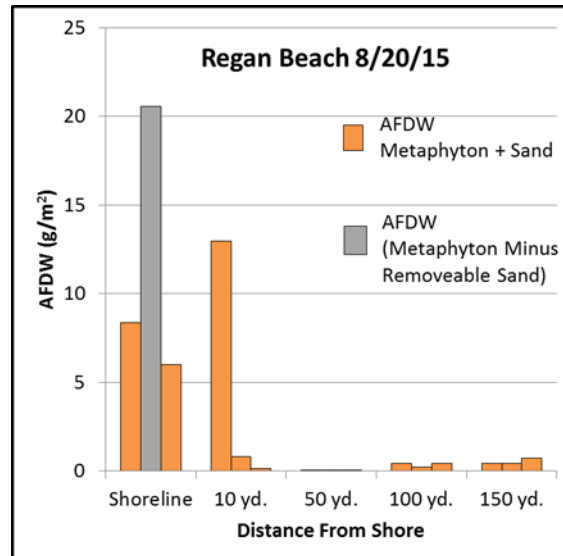


b. Metaphyton % Cover (From Photo)

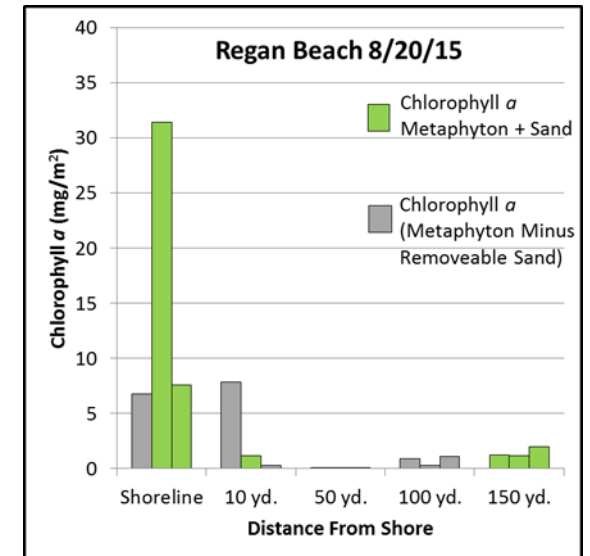
Figures 29a-e. Regan Beach 8/20/15 metaphyton monitoring results: (a) % cover, (field estimate); (b) % cover (from photo); (c) 60°C Dry Weight; (d) Ash Free Dry Weight (AFDW); Chlorophyll *a*. Bars represent left, center and right sample replicates at indicated distances offshore. AFDW or Chlorophyll *a* minus sand is value for metaphyton after separating off removable sand.



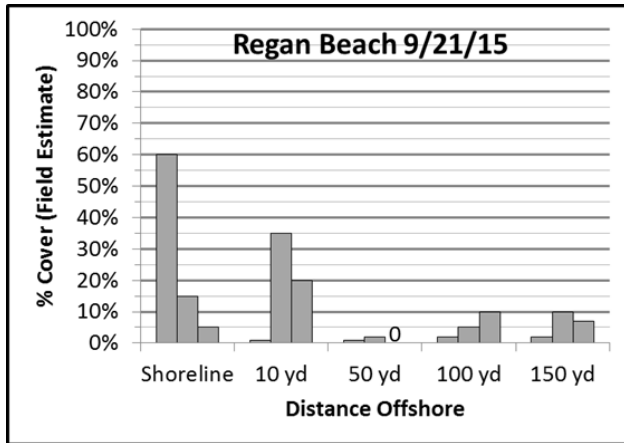
c. Metaphyton 60°C Dry Weight



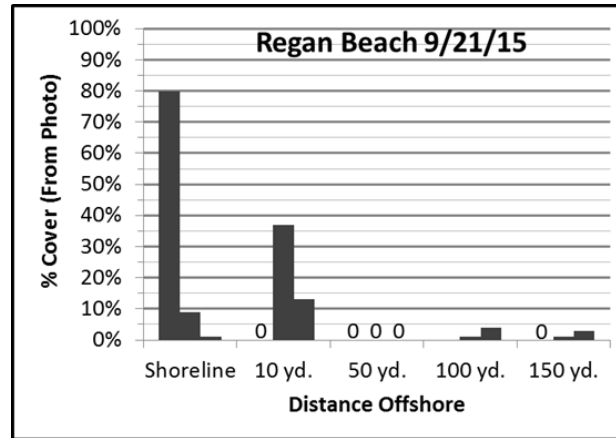
d. Metaphyton AFDW



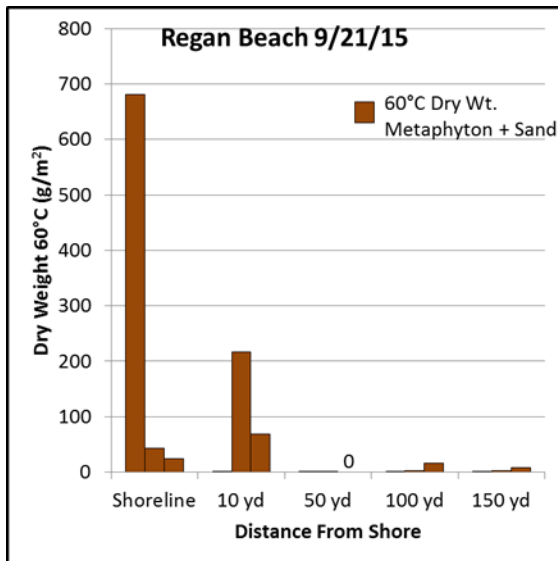
e. Metaphyton Chlorophyll *a*



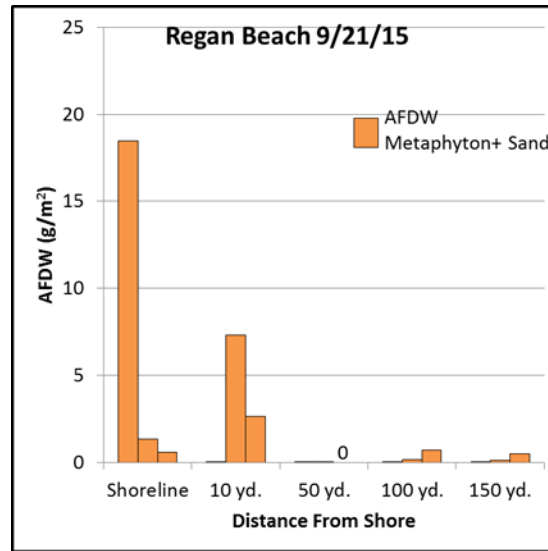
a. Metaphyton % Cover (Field Estimate)



b. Metaphyton % Cover (From Photo)

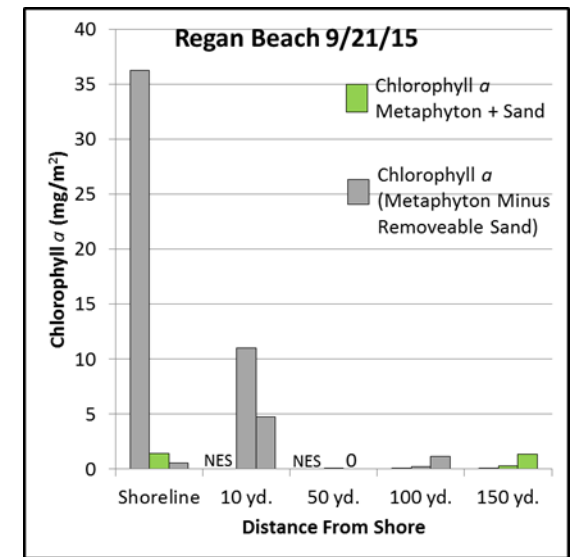


c. Metaphyton 60°C Dry Weight

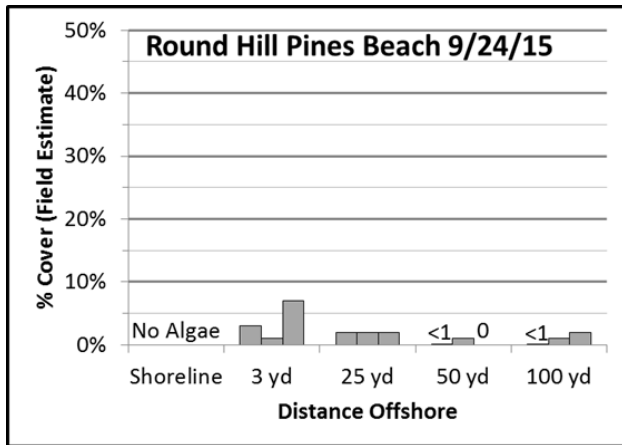


d. Metaphyton AFDW

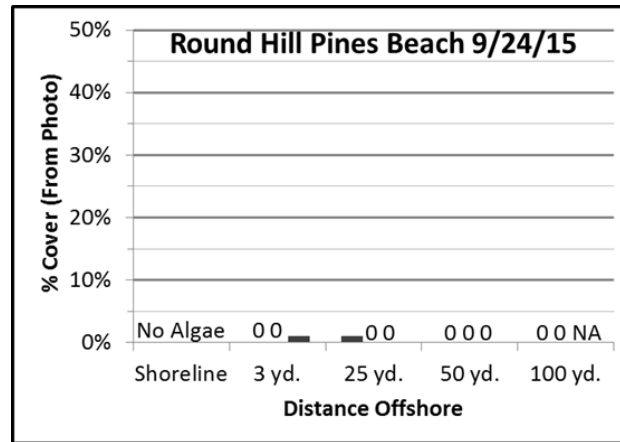
Figures 30a-e. Regan Beach 9/21/15 metaphyton monitoring results: (a) % cover, (field estimate); (b) % cover (from photo); (c) 60°C Dry Weight; (d) Ash Free Dry Weight (AFDW); Chlorophyll *a*. Bars represent left, center and right sample replicates at indicated distances offshore. Chlorophyll *a* minus sand is value for chlorophyll *a* of metaphyton after separating off removable sand.



e. Metaphyton Chlorophyll *a*

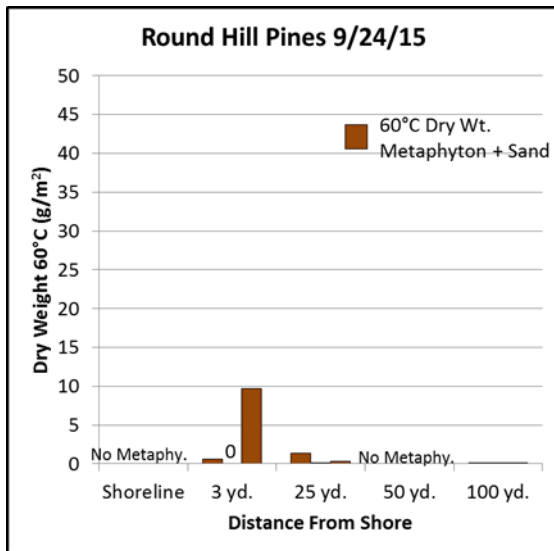


a. Metaphyton % Cover (Field Estimate)

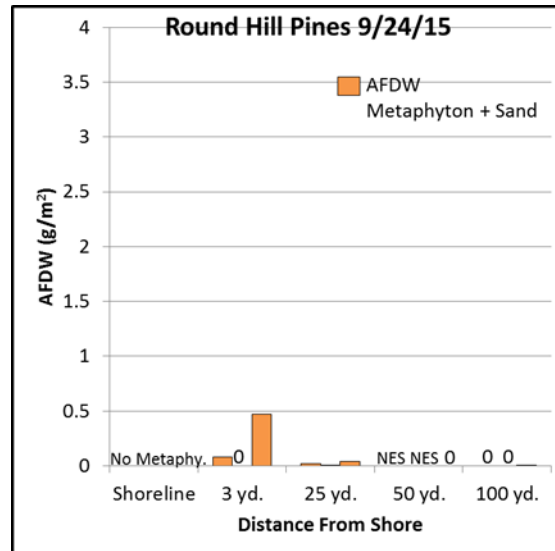


b. Metaphyton % Cover (From Photo)

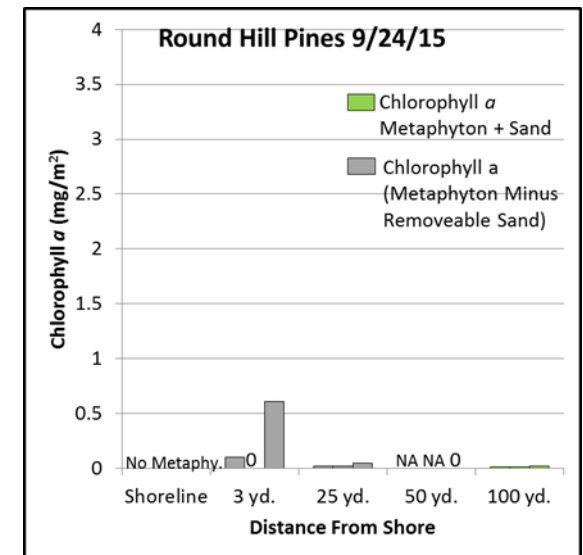
Figures 31a-e. Round Hill Pines Beach 9/24/15 metaphyton monitoring results: (a) % cover, (field estimate); (b) % cover (from photo); (c) 60°C Dry Weight; (d) Ash Free Dry Weight (AFDW); Chlorophyll *a*. Bars represent left, center and right sample replicates at indicated distances offshore. Chlorophyll *a* minus sand is value for chlorophyll *a* of metaphyton after separating off removable sand.



c. Metaphyton 60°C Dry Weight

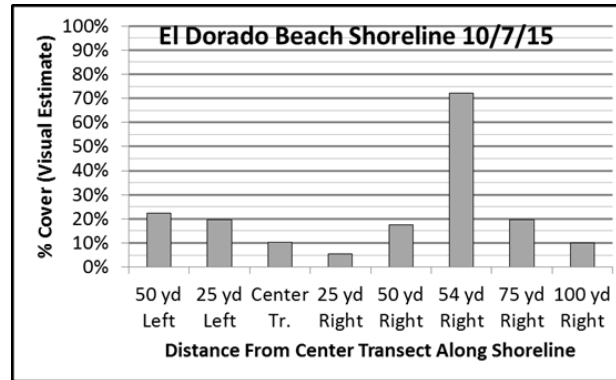


d. Metaphyton AFDW

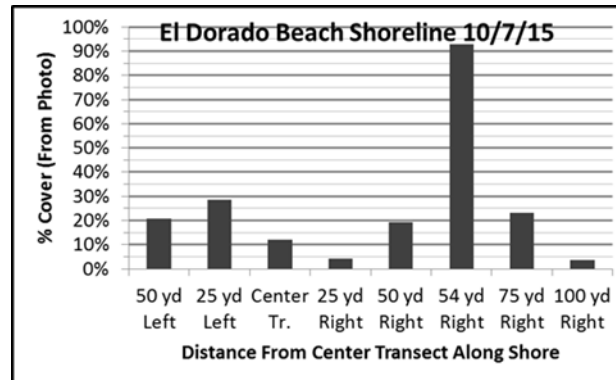


e. Metaphyton Chlorophyll *a*

Figures 32a-b. El Dorado Beach 10/7/15 metaphyton monitoring results, shoreline only: (a) % cover, (field estimate); (b) % cover (from photo). Offshore had very low (<1%) metaphyton % cover levels.



a. Metaphyton % Cover (Field Estimate)



b. Metaphyton % Cover (From Photo)

Associations Between Different Measures of Metaphyton Level

Correlation coefficients for data (percent cover from field estimates, percent cover from photos, 60°C Dry Weight, AFDW, Chlorophyll *a*), for each site on a date were compared to assess linear associations. The correlation coefficient *r* value was determined for all data from a site, for offshore data and for shoreline data (when enough data was available). The values for *r* are reported in Table 19. Strongest linear associations in the table (*r* values ≥ 0.80) are indicated in black, with *r* < 0.80 indicated in red.

For most sites, field estimates of percent cover and estimates of percent cover calculated from photos showed relatively strong associations. The *r* values were ≥ 0.80 in eight of 10 groups of data analyzed. The exceptions were for offshore data at a couple of sites (El Dorado 8/6/16 and Round Hill Pines 9/24/15). The offshore values for coverage at these sites were relatively low.

Comparing associations of measures of percent coverage with measures of metaphyton biomass at the sites, AFDW showed the most frequent strong associations with field estimates of percent cover (*r* values were ≥ 0.80 in 7 of 10 groups of data) and estimates of percent cover calculated from photos (*r* values were ≥ 0.80 in 8 of 9 groups of data). In cases where the *r* value was <0.80 for the percent cover vs. AFDW association, these occurred for offshore sites where percent cover was relatively low. Chlorophyll *a* had the least frequent strong associations with field estimates of percent cover (*r* values were ≥ 0.80 in only 6 of 10 groups of data) and estimates of percent cover calculated from photos (*r* values were ≥ 0.80 in only 5 of 9 groups of data).

Comparing associations of measures of metaphyton biomass at the sites, AFDW showed strong associations with both 60°C dry weight and Chlorophyll *a* in all groups of data analyzed (*r* values were ≥ 0.80 in 10 of 10 groups of data) for both associations. 60°C dry weight and Chlorophyll *a* however, did not show strong associations in all groups (*r* values were ≥ 0.80 in seven of 10 groups of data).

AFDW appears to be a good choice for estimates of metaphyton biomass based on the linear associations with other measures. Since AFDW is rapidly measured and does not require separation of sand and algae fractions, this measure of biomass would be most efficient to include in future monitoring. Either of the measures of percent cover appear to be useful to describe percent metaphyton cover. From a time standpoint, field estimates of percent cover would be the most efficient.

Table 19. Summary of correlation coefficient r values for linear associations between different measures of metaphyton presence at sites (n = number of samples). Strongest associations with $r \geq 0.80$ are shown in black, r values < 0.8 shown in red.

Site Date, Data Used	Measure of Metaphyton Amount	% Cover From Photo	60 °C Dry Wt.	AFDW	Chlorophyll <i>a</i>
El Dorado 8/6/15 All	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.972 (15)	0.977 (15) 0.928 (15)	0.884 (15) 0.882 (15) 0.940 (15)	0.659 (15) 0.722 (15) 0.732 (15) 0.920 (15)
El Dorado 8/6/15 Offshore only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.653 (12)	0.709 (12) 0.868 (12)	0.728 (12) 0.982 (12) 0.879 (12)	0.715 (12) 0.970 (12) 0.848 (12) 0.995 (12)
Round Hill Pines 8/11/15 Offshore Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	NA	0.327 (8) NA	0.782 (8) NA 0.814 (8)	0.839 (8) NA 0.683 (8) 0.975 (8)
Regan 8/20/15 All	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.983 (14)	0.886 (15) 0.862 (15)	0.936 (15) 0.916 (15) 0.951 (15)	0.801 (15) 0.751 (15) 0.853 (15) 0.933 (15)
Regan 8/20/15 Shoreline Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.990 (6)	0.840 (6) 0.641 (6)	0.920 (6) 0.869 (6) 0.916 (6)	0.719 (6) 0.641 (6) 0.803 (6) 0.913 (6)
Regan 8/20/15 Offshore Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.867 (8)	0.719 (9) 0.792 (8)	0.962 (9) 0.953 (8) 0.837 (9)	0.978 (9) 0.914 (8) 0.751 (9) 0.981 (9)
Regan 9/21/15 All	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.978 (15)	0.947 (15) 0.989 (15)	0.965 (15) 0.997 (15) 0.997 (15)	0.953 (12) 0.987 (12) 0.998 (12) 0.996 (12)
Regan 9/21/15 Shoreline Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.985 (6)	0.952 (6) 0.988 (6)	0.972 (6) 0.997 (6) 0.996 (6)	0.964 (5) 0.986 (5) 0.999 (5) 0.996 (5)
Regan 9/21/15 Offshore Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.803 (9)	0.769 (9) 0.979 (9)	0.753 (9) 0.994 (9) 0.987 (9)	0.648 (7) 0.948 (7) 0.880 (7) 0.945 (7)
Round Hill Pines 9/24/15 Offshore Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.688 (11)	0.915 (12) 0.763 (11)	0.941 (10) 0.654 (10) 0.986 (10)	0.935 (11) 0.653 (10) 0.986 (11) 0.9999 (10)
El Dorado 10/7/15 Shoreline Only	% Cover Field % Cover From Photo 60 °C Dry Wt. AFDW	0.990 (8)	NA NA	NA NA NA	NA NA NA NA

Summary of Metaphyton Levels Observed During the Study

Table 20 presents a summary of mean levels of percent cover (field and photo estimates), 60°C dry weight, AFDW and chlorophyll *a* observed for shoreline and offshore sites during 2015.

Table 21 presents a summary of percent cover field estimates made in 2016 and 2017.

One pattern that stood out was that there typically was much higher metaphyton along the shoreline than offshore at Regan and El Dorado Beaches in 2015 and 2016. AFDW mean± std. dev. (n) for shoreline samples from Regan in August and September 2015 were 8.16 ± 7.74 (6) and 5.06 ± 7.06 (6) g/m² respectively. Offshore mean AFDW amounts at Regan were 0.31 ± 0.21 (9) and 0.18 ± 0.26 (9) g/m² respectively. At El Dorado Beach mean AFDW for shoreline samples in August was 31.19 ± 5.50 (3) while the offshore amount was 0.16 ± 0.17 (12). Round Hill Pines did not have any metaphyton along the shoreline in 2015 and only relatively small amounts offshore (i.e. in August mean offshore AFDW was 0.89 ± 1.29 (8) and in September the mean offshore AFDW was 0.08 ± 0.17 (10)).

El Dorado Beach had the highest shoreline level of metaphyton in 2015 at 31.19 ± 5.50 g/m² (3). Regan Beach had slightly higher offshore levels of AFDW (0.31 ± 0.21 g/m² (n=9); 0.18 ± 0.26 g/m² (9) in 2015 compared with El Dorado Beach offshore AFDW 0.16 ± 0.17 g/m² (12). At all sites, levels of metaphyton were higher in August compared with Sept. and Oct. in 2015. Cooler water temperatures may have contributed to decreased metaphyton growth in Sept. and Oct. Water temperatures cooled by several degrees or more at Regan and Round Hill Pines in Sept. compared with August. However at El Dorado Beach the water temperatures at the time of sampling in Oct. were only slightly cooler in Oct. (21.5°C at shoreline and 15°C at 150 yds. offshore) than in August (22°C at shoreline and 16°C at 150 yds. offshore).

Metaphyton was present along the shoreline at Regan and El Dorado beaches also in 2016. At El Dorado Beach in August of 2016 the shoreline percent cover was estimated at about 20% while in 2015 it was $58 \pm 25\%$ (3). At Regan Beach the shoreline percent cover in 2016 was $28 \pm 39\%$ (2) while in 2015 it was $37 \pm 28\%$ (6). Since Regan and El Dorado were only sampled on one date in the summer of 2016, it is difficult to know if the samples were collected during the heaviest growth. Therefore, it is difficult to draw conclusions on whether metaphyton levels were greater in 2015 or 2016.

Metaphyton was present offshore in all three years 2015-2017 at Regan and El Dorado beaches. These years included a very dry year with extremely low lake levels in 2015 and a very wet year with very high lake levels in 2017. This was interesting because metaphyton deposition onshore did not appear to occur in all three years. In 2015 and 2016 when lake level was low, near the natural rim elevation of 6223 ft., moderate amounts (means of ~15-60% cover) of metaphyton were measured along the shoreline at Regan and El Dorado Beaches. However, during the summer of 2017 when lake level was very high (over 6228 ft.) no metaphyton was measured

along the shore at El Dorado beach and at Regan there was no beach, only rock lined shoreline, with no deposition of metaphyton there.

Table 20. Mean \pm Std. Dev. (n) for different measures of metaphyton presence along at the water's edge "Shoreline" and Offshore in 2015. For Regan Beach Shoreline included data collected 10 yds. offshore of the water's edge, water was extremely shallow in that zone.

	% Cover Field	% Cover From Photo	60°C DW	AFDW	Chlorophyll a
Regan 8/20/15 Shoreline +10yd	37 \pm 28% (6)	38 \pm 39% (6)	292.67 \pm 230.71 (6)	8.16 \pm 7.74 (6)	9.19 \pm 11.39 (6)
Regan 8/20/15 Offshore	10 \pm 6% (9)	3 \pm 4% (8)	6.18 \pm 4.48 (9)	0.31 \pm 0.21 (9)	0.76 \pm 0.70 (9)
Regan 9/21/15 Shoreline +10yd	23 \pm 22% (6)	23 \pm 31% (6)	172.53 \pm 260.71 (6)	5.06 \pm 7.06 (6)	10.79 \pm 14.81 (5)
Regan 9/21/15 Offshore	4 \pm 4% (9)	1 \pm 2% (9)	3.66 \pm 5.35 (9)	0.18 \pm 0.26 (9)	0.46 \pm 0.56 (7)
El Dorado 8/6/15 Shoreline	58 \pm 25% (3)	61 \pm 23% (3)	1116.06 \pm 403.78(3)	31.19 \pm 5.50 (3)	48.57 \pm 26.58 (3)
El Dorado 8/6/15 Offshore	3 \pm 2% (12)	2 \pm 2% (12)	6.78 \pm 7.99 (12)	0.16 \pm 0.17(12)	0.38 \pm 0.42 (12)
El Dorado 10/7/15 Shoreline	15.0 \pm 6.3% (7)	15.9 \pm 9.6% (7)	NA	NA	NA
El Dorado 10/7/15 Offshore	0.21 \pm 0.14% (16)	0.06 \pm 0.13% (5)	NA	NA	NA
Round Hill P. 8/11/15 Shoreline	0	0	0	0	0
Round Hill P. 8/11/15 Offshore	6 \pm 4% (8)	0.4 \pm 0.2% (2)	115.51 \pm 158.75 (8)	0.89 \pm 1.29 (8)	0.80 \pm 1.28 (8)
Round Hill P. 9/24/15 Shoreline	0	0	0	0	0
Round Hill P. 9/24/15 Offshore	1.7 \pm 1.9% (12)	0.2 \pm 0.4% (11)	1.15 \pm 3.19 (12)	0.08 \pm 0.17(10)	0.08 \pm 0.18 (11)

Table 21. Mean \pm Std. Dev. (n) for field estimate of metaphyton percent cover along the water’s edge “Shoreline” and Offshore in 2016 and 2017. For Regan Beach Shoreline included data collected 10yd offshore of the water’s edge, water was extremely shallow in that zone.

	% Cover Field	% Cover Field
Regan 8/19/16 Shoreline +3.3yd	28 \pm 39% (2)	
Regan 8/19/16 Offshore	1.3 \pm 1.2% (5)	
El Dorado 8/19/16 Shoreline	~20% (1)	
El Dorado 8/19/16 Offshore	2.1 \pm 3.9% (6)	
	Observer Snorkeling	Observer w/SCUBA
Regan 8/9/17 Shoreline	0	
Regan 8/9/17 Patch Nearest Shore (12yd)	NA- Poor Visibility	22 \pm 21% (12)
Regan 8/9/17 Offshore*	4.1 \pm 3.2% (24)	6.4 \pm 3.7% (28)

*Notes- Excludes quadrats with algae over aquatic plants, and deliberately placed quadrat containing 70-80% metaphyton.

Relationships Between Levels of Nearshore Algae, Algae Along the Shoreline and Bottom Topography

Measurements of metaphyton distribution made in this pilot study indicated metaphyton levels were highest along the shoreline or within a relatively short distance offshore of the shoreline at the Regan and El Dorado beach sites in 2015 and 2016 when lake level was low (see Figures 27, 29, 30 and Tables 10, 12, 13). The levels of metaphyton along the shoreline could be quite variable. Offshore to at least 137 m away, the metaphyton tended to be widely dispersed over the shallow nearshore sands, with no obvious patterns related to distance from shore. At Round Hill Pines, no metaphyton was observed along the shoreline, but some metaphyton was observed relatively close to the shore.

Monitoring results and field observations made in this study suggest nearshore slope or gradient and lake level may play a role in the degree to which metaphyton accumulates along the shoreline and is deposited on the beach. Lowered lake levels may favor accumulation of metaphyton in the shallow flat shoreline areas along Regan and El Dorado beaches. In 2015, the lake level was very low, 6222.61 ft. when test monitoring began on 8/6/15 and continued to drop throughout the summer. The lowering lake level left an expansive flat area of exposed lakebed adjacent to El Dorado and Regan beaches. The gradient was so slight that the water depth offshore at Regan and El Dorado was only about 30cm (about a foot deep) at a distance of 137m from shore. Accumulations of metaphyton often occur in between sand ripples in the shallow region offshore. When wind and wave energy is sufficient, metaphyton can be pushed toward the shore due to wind and currents. With little slope to impede movement of the metaphyton, it may accumulate in the shallow areas right along the shoreline at El Dorado and Regan Beaches. As the lake draws down during the summer, the metaphyton may be left exposed on the shoreline. Water in the very shallow areas nearshore also was warmer than in offshore areas, this may affect the metaphyton. More information on temperature patterns over a 24-hour period would be needed to better assess temperature impacts on metaphyton at the sites.

In summer of 2016, the water line at El Dorado Beach was just above the transition point where the more steeply sloped beach transitions to the flat lake bed (at about 6223 ft.). Within 4.6 m of shore, the water depth was only 12 cm. Metaphyton was observed at the water's edge and deposited onshore as a result of wave activity. Metaphyton may have moved towards shore along the flat bottom offshore, and been within a zone nearshore where wave activity mixed the water along the shoreline and deposited algae onshore. Figure 33 shows a photo of the steeply sloping beach at El Dorado Beach and the transition area to flatter lakebed sediments offshore during very low lake level in 2015. The transition point between the two occurs at about 6223 ft. Figure 34 shows the same area in the summer of 2016 when the lake level was 6223.37 ft., slightly above the transition area. Note the metaphyton in the nearshore water and deposited on the beach.



Figure 33. El Dorado Beach on 10/7/15 showing sloping beach and transition area near 6223 ft. to flatter exposed lakebed nearshore.



[Figure 34](#). El Dorado Beach 8/19/16. The same area as in Figure 33, but with lake elevation at 6223.37 ft. which was slightly above the transition area between sloping beach and flatter lakebed sediments offshore. Note the metaphyton in the nearshore water and deposited on the beach.

When there is a steeper sloping beach along the shoreline, the proximity of algae accumulated in the nearshore and the depth relative to the zone of wave activity may play a role in the amount of algae along the shore. At Round Hill Pines beach, for example, in August 2015, there was a relatively steep slope to the beach at the water's edge; 3 m away the water was about 0.5m. Some metaphyton was present at the base of the slope slightly offshore, but did not appear to move up the slope to be deposited onshore. As another example, at El Dorado beach in 2017, the water line was well up the steeply sloped beach and the transition point from steeply sloped beach to flatter lake bed was located about 15m offshore. Water depth at the transition point was nearly 2m deep. Metaphyton accumulated in a distinct patch offshore near where the shoreline slope becomes steeper but did not move upslope into the wave zone where it could be deposited on the beach.

We also observed metaphyton to accumulate in some pockets created by bars in the offshore area near Regan Beach. In the shallow offshore area near Regan Beach, there is interesting structure to the bottom created by many sand bars. When viewed from above, these bars have the same appearance as ripples on a sandy bottom but on a larger scale (Figure 35). When the water's edge is within this area of sand bars, pockets of water or small embayments are created. These may play a role in where metaphyton accumulates and is left on shore when the lake recedes. We did observe metaphyton accumulating in these pockets (Figure 36). It would be interesting

to further study the relationship of the bottom topography and water movements and water depth in this area to metaphyton.

With future monitoring it will be interesting to see if the patterns described above for metaphyton relative to lake low lake levels are consistently observed. It's possible deposition of metaphyton onshore may be more frequently observed during low lake level years as was observed in this study. As lake level lowers, there is a large expanse of shallow shoreline along the south shore. Metaphyton may drift inshore or be pushed inshore and left deposited onshore as the water recedes. The public using areas such as Regan Beach and El Dorado beach have much more shoreline to recreate on and due to the shallow nature of the offshore, offshore water in which to wade. They may view both increased levels of metaphyton deposited onshore and also metaphyton along the bottom in the shallow waters offshore.

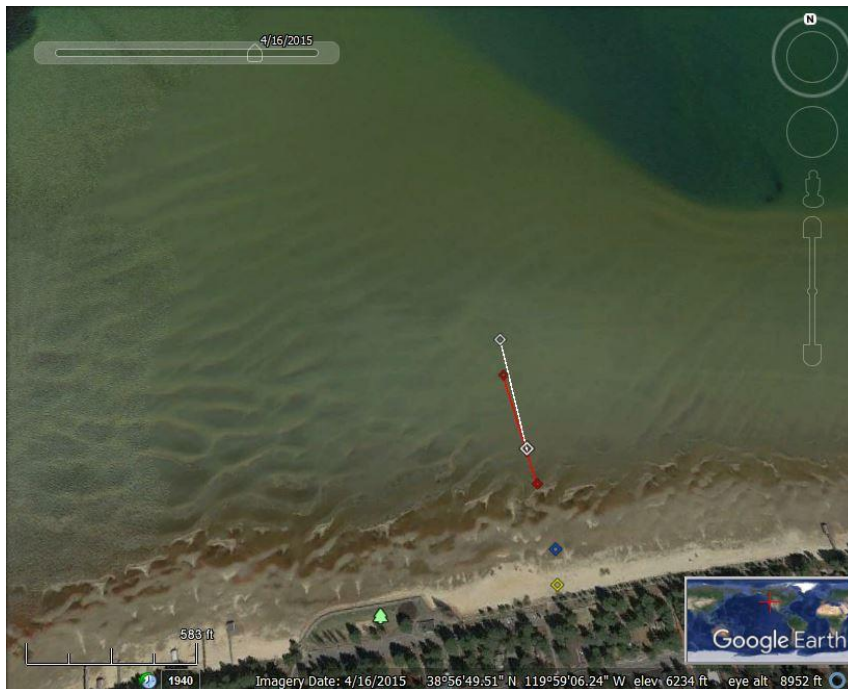


Figure 35. Google Earth view of Regan Beach area. Sampling area and sand bars along the shoreline and also out over the shallow shelf to the west (the bars almost appear like sand ripples underwater but on a much larger scale) are shown. This image taken 4/16/15, Google Earth.



Figure 36. Regan Beach 8/11/15 accumulations of metachyton in pockets created by sand bars.

Aerial Observations of Sites

The monitoring in the study showed that there can be considerable variability in levels of metachyton along the shoreline. Offshore, the metachyton tends to be quite dispersed, with some areas in which metachyton may form large patches. Monitoring as done in this pilot study provides an indication of levels of metachyton along the shoreline and offshore. However, it may not characterize fully contributions of larger patches in a region and the behavior of such patches - such as whether they are moving, increasing in size or decreasing. Tracking behavior of metachyton on a regional scale may help in better understanding factors driving metachyton distribution in the nearshore. It would be useful to incorporate some monitoring that allowed observations of metachyton on a regional scale. Observations from the air may provide such an opportunity. In 2017, we worked with researchers from the UC Davis Department of Land, Air and Water Resources (Dr. Yufang Jin, Andy Wong and Han Liu) to coordinate some imaging from the air (helicopter piloted by Mike Bruno and using a UAV) with observations of metachyton at Regan and El Dorado Beaches. Multispectral images were obtained. Figure 37 shows an example of a RGB image taken of Regan beach during monitoring on 8/9/17. Regional distribution of larger metachyton patches were apparent in the photos. Development of aerial methods for determining regional metachyton distribution would be valuable and potentially could be combined with monitoring methods described in this pilot study.



Figure 37. RGB image of Regan Beach monitoring site taken from helicopter on 8/9/17. A large patch of metaphyton with submerged vegetation was visible. Aerial imaging holds potential for assessing metaphyton distribution on a regional scale (Image courtesy of Yufang Jin, Andy Wong and Han Liu, UC Davis Division of Land, Air and Water Resources).

Relationships Between Algae Observations in TERC Citizens Science APP and Metaphyton Observed in Study

In 2015, TERC developed a mobile app “Tahoe Citizen Science 2” which can be used by the public to enter observations on various environmental conditions observed around Lake Tahoe. App users can record observations on such things as: presence of algae, water clarity, beach conditions, and species observed, as well as submit photos taken of what is seen. This information, together with GPS locations of where the observer was when recording information and the date and time are recorded in an “Observations” data archive. Users of the app and associated website can see summaries of observations and photos. For algae, a map showing a summary of points around the lake where algae observations were made, together with observations and photos associated with the data points. The website for the app is located at <https://citizensciencetahoe.org>.

We looked at algae observations recorded by users of the app for the south shore region (Baldwin Beach to Zephyr Cove) during the period July 2015 to November 2017 to see if there were any patterns for where observations of algae relative to the three study sites Regan Beach, El Dorado Beach and Round Hill Pines Beach. Of 340 observations lake-wide for the area, recorded in the Algae section of the app, about 54 were specific to the shoreline region of the

south shore. Of those, a portion had additional descriptive information on the algae or photos identifying the algae as metaphyton or periphyton (algae attached to rocks). Table 22 summarizes the occurrences of metaphyton and periphyton observations by year 2015-2017 and provides information on months in which observations were recorded.

There were seven algae reports likely describing metaphyton in the nearshore, and five of these were at or near the El Dorado Beach metaphyton monitoring site used in the pilot study. The other two were at Pope Beach west of the Tahoe Keys and at Zephyr Cove. The five citizen science app observations in the region of El Dorado beach were made in June and July of 2016. In our pilot study monitoring at El Dorado beach in August of 2016 (8/19/16), we found metaphyton percent coverage near the water's edge of about 20%, with metaphyton being deposited onshore (see photo in Figure 34). El Dorado beach is a popular south shore beach, and based on the app results one in which algae along the shoreline is being noted by the public. The other two sites with citizen reports of probable metaphyton are also popular beaches (Pope and Zephyr Cove). Interestingly there were no reports of metaphyton there in 2015 when we did much of our testing and found metaphyton along shore. The app had only recently been released then so may not have been in wide use. An absence of citizen observations of algae at El Dorado Beach in 2017 is consistent with our observations in Sept. 2017, which found no metaphyton along the shoreline.

No citizen monitoring app observations of algae were made at Regan Beach during 2015-2017. The only observation of algae in the region there was for algae on the rocks between Al Tahoe and the U. Truckee marsh in August of 2015. During pilot study monitoring, we did note metaphyton at Regan Beach in all three years. Therefore, metaphyton was present at Regan but was not reported by the public on the app during this period.

Similarly, no observations that likely were associated with metaphyton were reported by public using the Tahoe Citizen app at Round Hill Pines 2015-2017. There were a couple of reports algae there in May and June of 2016. It is possible these related to metaphyton or periphyton. Another report in June of 2016 at Round Hill Pines related to periphyton. We observed clumps of metaphyton offshore just offshore of the beach at Round Hill Pines in August 2016 and small patches of metaphyton near the shore in summer 2015.

Slimy algae on rocks (periphyton) was more frequently reported in the citizen app (14 of 54 measurements) than algae which likely was metaphyton (7 of 54 measurements) along the south shore region during 2015-2017. Most of the reports of algae on rocks were distributed between Elk Pt and Zephyr Cove along the south shore, there were a couple of reports of algae on rocks near Al Tahoe/ U. Truckee marsh and near Timber Cove. This is consistent with the composition of the shoreline areas. There is more rocky portions of shoreline in the Elk Pt. to Zephyr Cove region compared with sandy beach areas along much of the rest of the south shoreline. Again, most of the metaphyton measurements were made near El Dorado Beach.

Table 22. Summary of TERC Citizen Science Tahoe Mobile App 2 algae number of reports of probable metaphyton, algae attached to rocks (periphyton) and non-specific algae, along the south shore of Lake Tahoe from Baldwin Beach to Zephyr Cove, NV, July 2015 to Nov. 2017.

TERC Citizen Science Tahoe Mobile App 2 – Metaphyton and Periphyton (Attached Algae) Reports July 2015- November 2017	
	#
Shoreline Algae Observations (South Shore: Baldwin Beach to Zephyr Cove)	54
Offshore Algae Observations (South Shore: Baldwin Beach to Zephyr Cove)	6
Probable Shoreline Metaphyton Reports 2015-2017³	7
2015 Probable metaphyton reports (None)	0
2016 Probable metaphyton reports (4 El Dorado Beach., 1 west of Timber Cove, 1 Zephyr Cove)	6
2017 Probable metaphyton reports (1 Pope Beach)	1
Month Reported:	
June	2
July	4
Aug.	1
Shoreline Periphyton Reports 2015-2017⁴	14
2015 Periphyton reports (1 Al Tahoe; 2 Zephyr Cove)	3
2016 Periphyton reports (3 So. Elk Pt; 4 No. Elk Pt; 1 Round Hill Pines; 1 Zephyr Cove; 1 Timber Cove)	10
2017 Periphyton reports (1 near Elk Pt.)	1
Month Reported:	
Feb.	1
April	1
June	6
July	4
Aug.	1
Oct.	1
Non-specific Shoreline Algae Reports 2015-2017⁵	33

The citizen science app is providing useful information on what is occurring around the lake. Observations from the app showed algae, likely metaphyton, was present at El Dorado beach in June and July 2016. This information combined with our monitoring data in August 2016 suggests a good period for monitoring metaphyton there is during June to August. One possible suggestion for future additional useful information from the app would be to encourage further specification of whether algae observed was periphyton or metaphyton. There were a large number of algae reports for which it could not be determined if the algae observed was

³ Algae was indicated as probable metaphyton based on information provided including presence of mats in sandy areas, clumps of algae on sandy bottom and appearance of algae if photos provided.

⁴ Algae was indicated as periphyton (attached algae) based on information provided including slimy layer of algae on rocks, thick coating of algae on rocks and appearance of algae in photos.

⁵ Non-specific algae – there was no additional information provided that indicated algae was specifically periphyton or metaphyton.

periphyton or metaphyton. It will be interesting to see if what patterns further become apparent as more years of data are added to the archive.

Recommendations for Future Monitoring

Based on the work in this study we have the following recommendations for future monitoring of metaphyton:

- 1) We recommend 2-3 sites be selected for monitoring on an annual basis in the summer. Some of the characteristics that we suggest be considered in selection of sites include: (1) sites noted by the public and/or agencies to have had substantial metaphyton growth in the nearshore in recent years; (2) sites where metaphyton has been observed deposited along the shoreline; (3) sites with large numbers of Asian clams or nutrient inputs (monitoring could provide information on associations). Some suggested areas for future monitoring include: (i) sites included in this study: i.e., Regan beach, El Dorado Beach and Round Hill Pines beach; (ii) areas on the north shore which have been observed to develop metaphyton, such as near the mouths of Third and Incline Creeks.
- 2) We recommend the monitoring be done primarily during June-August. This recommendation is based on observations from this study and TERC's observations of metaphyton in several recent summers that suggests growth of metaphyton typically is elevated during this period. Two samplings per summer are suggested for each site to "capture" peak metaphyton growth.
- 3) Sampling points should be set-up on the date of sampling using a random sampling design or a grid pattern to avoid bias in sampling. The coordinates of each sampling point should be determined using an accurate GPS system. Based on the heterogeneity in metaphyton observed in the study, particularly along the shoreline, it is recommended that the number of replicate quadrat sample number be between 7-12 to estimate the mean metaphyton level with as high a level of accuracy as possible, with a reasonable (not overly time consuming) number of samples.
- 4) To monitor metaphyton levels at a few sites, we recommend using one or more of the methods found to be best for monitoring metaphyton in this pilot study: i.e., percent cover in the field using a 0.25 m² quadrat; percent cover calculated from photographs; and AFDW biomass. Field estimates of percent cover using a 0.25m² quadrat can be made rapidly (taking a few minutes at each site). Along with the field estimates of percent cover, photos of algae in quadrats should be taken to provide a photographic record of algae present with the option to later analyze the photos for percent cover. Collection of samples and analysis of AFDW provides an estimate of the amount of metaphyton biomass present to relate to the percent cover estimates. Estimates of the amount of biomass may also be important for assessing ecological questions about metaphyton linkages with other biological and chemical factors at sites. AFDW is a relatively simple lab procedure with an additional benefit that separation of sand from algae in the processing does not appear necessary. This saves time in processing. A

subsample of the algae collected for AFDW biomass can also be examined under the microscope for predominant algal species present.

- 5) It is recommended field technicians, practice identifying different levels of coverage to produce accurate field estimates of percent cover. Field guides that show examples of different levels of coverage would be useful as references in the field. For both field estimates of cover and photography, it is recommended that monitoring be done early in the morning when lake conditions tend to be calm for clear viewing of the metaphyton from above or below water.
- 6) We made estimates of cost for several basic approaches that could be used to monitoring metaphyton. The estimates of cost were based on assumptions that three sites would be monitored twice per summer, and that it would take two researchers a morning to sample a site, if samples were processed for AFDW an afternoon of processing of samples by the researchers would be required. The cost was based on hours of work and indirect costs.
 - (i) The most basic approach to monitoring would be to focus on visual estimates of percent cover, and concentrate those measurements right along the shoreline (area within about 10 yards of the waterline shoreward and lakeward). Since metaphyton tends to accumulate right along the shoreline, this would capture the area with the heaviest concentration of metaphyton, in particular in years when the lake level is relatively low. Costs associated with this level for three sites per summer would be \$3500 (not including reporting costs).
 - (ii) There are advantages to including measurements of percent cover for the shallow sandy offshore area (20-150 yards offshore) in addition to measurements along the shore; this is a second approach that should be considered. There was indication from this pilot study that when the lake level is high, metaphyton is still present in the nearshore, but may not be deposited along some beaches. Instead, it may be distributed both in small and large patches nearshore. It would be useful to know nearshore concentrations for comparisons year to year. In low lake level years, the metaphyton in the shallow water offshore can be quite apparent to waders, swimmers and boaters in the nearshore. The metaphyton in this offshore area also contributes metaphyton to the shoreline. It would be useful to estimate metaphyton levels in the nearshore each summer. Estimated combined cost for shoreline and offshore estimates of percent cover, would be approximately \$5000.
 - (iii) A third approach would be to add collection of samples of metaphyton from the shoreline quadrats for AFDW biomass assessment to visual estimates of percent cover. Estimated combined cost for visual estimates of percent cover and AFDW for the shoreline would be about \$6600 per summer.
 - (iv) A fourth approach would be to include both visual estimates of percent cover and estimates of AFDW biomass for the shoreline and shallow sandy offshore area. Estimated cost for this level would be about \$9000 per summer.

Costs for data analysis and reporting would need to be added to each individual cost.

- 7) The use of a combination of aerial imaging, with in-lake estimates of percent coverage and AFDW biomass may ultimately prove to be the best approach for assessing regional and lake-wide metaphyton patterns and levels. Aerial imaging, combined with in-lake monitoring, will be done in a follow-up study in 2018-2019 by TERC for the Nevada Division of State Lands. This study will look at regional distribution of metaphyton and potential associations of metaphyton with Asian clams and other nutrient sources.

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APPENDIX

Appendix 1.a. El Dorado Beach 8/6/15 Dry Weight, AFDW, and Chlorophyll *a* (coarse separation of algae and sand fractions done⁶). NA-Not Analysis.; NF-No sand fraction

El Dorado		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n	
8/6/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	Dry Wt.	929.81	913.52	947.23	930.19	16.86	3	633.57	650.94	698.53	661.02	33.63	3	931.99	911.07	940.09	927.72	14.97	3	
Shoreline	Sand	Dry Wt.	577.28	581.25	581.44	579.99	2.35	3	42.24			42.24	-	1	207.01			207.01	-	1	
Shoreline	A+S	Dry Wt.				1510.18	-					703.26	-					1134.73	-		
20 Yards	Algae	Dry Wt.	0.61			0.61	-	1	4.50			4.50	-	1	0.43			0.43	-	1	
20 Yards	Sand	Dry Wt.	1.52			1.52	-	1	13.79			13.79	-	1	11.74			11.74	-	1	
20 Yards	A+S	Dry Wt.				2.13	-					18.28	-					12.17	-		
50 Yards	Algae	Dry Wt.	0.00			0.00	-	1	2.60			2.60	-	1	0.00			0.00	-	1	
50 Yards	Sand	Dry Wt.	0.00			0.00	-	1	NF			NF	-	1	0.00			0.00	-	1	
50 Yards	A+S	Dry Wt.				0.00	-					2.60	-					0.00	-		
100 Yards	Algae	Dry Wt.	1.64			1.64	-	1	2.42			2.42	-	1	5.49			5.49	-	1	
100 Yards	Sand	Dry Wt.	0.09			0.09	-	1	0.08			0.08	-	1	NF			NF	-	1	
100 Yards	A+S	Dry Wt.				1.74	-					2.50	-					5.49	-		
150 Yards	Algae	Dry Wt.	1.66			1.66	-	1	0.93			0.93	-	1	18.20			18.20	-	1	
150 Yards	Sand	Dry Wt.	8.77			8.77	-	1	0.36			0.36	-	1	6.50			6.50	-	1	
150 Yards	A+S	Dry Wt.				10.43	-					1.29	-					24.70	-		
El Dorado		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n	
8/6/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	AFDW	30.82	29.13	26.18	28.71	2.35	3	34.60	33.77	32.14	33.51	1.25	3	22.38	23.68	23.69	23.25	0.75	3	
Shoreline	Sand	AFDW	3.42	3.30	11.90	6.21	4.93	3	0.28			0.28	-	1	1.63			1.63	-	1	
Shoreline	A+S	AFDW				34.92	-					33.78	-					24.88	-		
20 Yards	Algae	AFDW	0.05			0.05	-	1	0.24			0.24	-	1	0.12			0.12	-	1	
20 Yards	Sand	AFDW	0.01			0.01	-	1	0.03			0.03	-	1	0.03			0.03	-	1	
20 Yards	A+S	AFDW				0.06	-					0.27	-					0.15	-		
50 Yards	Algae	AFDW	0.00			0.00	-	1	0.13			0.13	-	1	0.00			0.00	-	1	
50 Yards	Sand	AFDW	0.00			0.00	-	1	NF			NF	-	0	0.00			0.00	-	1	
50 Yards	A+S	AFDW				0.00	-					0.13	-					0.00	-		
100 Yards	Algae	AFDW	0.07			0.07	-	1	0.13			0.13	-	1	0.26			0.26	-	1	
100 Yards	Sand	AFDW	0			0	-	1	0			0	-		NF			NF	-	0	
100 Yards	A+S	AFDW				.07	-					.13	-					0.26	-		
150 Yards	Algae	AFDW	0.10			0.10	-	1	0.07			0.07	-	1	0.60			0.60	-	1	
150 Yards	Sand	AFDW	0.03			0.03	-	1	0.00			0.00	-	1	0.02			0.02	-	1	
150 Yards	A+S	AFDW				0.13	-					0.07	-					0.62	-		
El Dorado			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n	
8/6/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
Shoreline	Algae	Chl. <i>a</i>	31.97	44.03	26.23	34.08	9.09	3	78.56	75.52	82.81	78.96	3.67	3	36.00	22.52	24.56	27.69	7.27	3	
Shoreline	Sand	Chl. <i>a</i>	3.20	2.39	3.38	2.99	0.53	3	NA			NA	-	0	1.98			1.98	-	1	
Shoreline	A+S	Chl. <i>a</i>				37.07	-					NA	-					29.67	-		
20 Yards	Algae	Chl. <i>a</i>	0.11			0.11	-	1	0.63			0.63	-	1	0.36			0.36	-	1	
20 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	NA			NA	-	0	NA			NA	-	0	
20 Yards	A+S	Chl. <i>a</i>				NA	-		NA			NA	-					NA	-		
50 Yards	Algae	Chl. <i>a</i>	0.00			0.00	-	1	0.34			0.34	-	1	0.00			0.00	-	1	
50 Yards	Sand	Chl. <i>a</i>	0.00			0.00	-	1	NF			NF	-		0.00			0.00	-	1	
50 Yards	A+S	Chl. <i>a</i>				0.00	-		0.34			0.34	-					0.00	-		

⁶ “Coarse separation of sand and algae” indicates separation of sand from algae was done to the extent possible with methods used, however often complete separation was not achieved.

El Dorado			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/6/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
100 Yards	Algae	Chl. <i>a</i>	0.19			0.19	-	1	0.35			0.35	-	1	0.62	0.62		0.62	0	2
100 Yards	Sand	Chl. <i>a</i>	0			0	-	1	0			0	-	1	NF			NF	-	
100 Yards	A+S	Chl. <i>a</i>				0.19	-		.35			.35	-					0.62	-	
150 Yards	Algae	Chl. <i>a</i>	0.18	0.20		0.19	0.01	2	0.17			0.17	-	1	1.53	1.55		1.54	0.01	2
150 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	0			0	-	1	NA			NA	-	0
150 Yards	A+S	Chl. <i>a</i>				NA	-					0.17	-					NA	-	

Appendix 1.b. Round Hill Pines Beach 8/11/15 Dry Weight, AFDW, and Chlorophyll *a* (coarse separation of algae and sand fractions done). NA-Not Analy.; NF-No sand fract.

Round H.		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/11/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	Dry Wt.	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	Sand	Dry Wt.	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	A+S	Dry Wt.				0	-					0	-					0	-	
3 Yards	Algae	Dry Wt.	3.94			3.94	-	1	0			0	-	1	0.34			0.34	-	1
3 Yards	Sand	Dry Wt.	297.57			297.57	-	1	0			0	-	1	338.46			338.46	-	1
3 Yards	A+S	Dry Wt.				301.51	-					0	-					338.80	-	
25 Yards	Algae	Dry Wt.	0.19			0.19	-	1	0.82			0.82	-	1	30.11			30.11	-	1
25 Yards	Sand	Dry Wt.	0.95			0.95	-	1	0.80			0.80	-	1	248.19			248.19	-	1
25 Yards	A+S	Dry Wt.				1.14	-					1.62	-					278.30	-	
50 Yards	Algae	Dry Wt.	1.34			1.34	-	1	0.20			0.20	-	1	NA			NA	-	0
50 Yards	Sand	Dry Wt.	0.41			0.41	-	1	0.79			0.79	-	1	NA			NA	-	0
50 Yards	A+S	Dry Wt.				1.75	-					0.99	-					NA	-	
Round H.		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/11/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	AFDW	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	Sand	AFDW	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	A+S	AFDW				0	-					0	-					0	-	
3 Yards	Algae	AFDW	1.01			1.01	-	1	0			0	-	1	0.15			0.15	-	1
3 Yards	Sand	AFDW	1.03			1.03	-	1	0			0	-	1	1.02			1.02	-	1
3 Yards	A+S	AFDW				2.04	-					0	-					1.17	-	
25 Yards	Algae	AFDW	0.04			0.04	-	1	0.18			0.18	-	1	1.63			1.63	-	1
25 Yards	Sand	AFDW	0.02			0.02	-	1	0.02			0.02	-	1	1.91			1.91	-	1
25 Yards	A+S	AFDW				0.06	-					0.20	-					3.54	-	
50 Yards	Algae	AFDW	0.08			0.08	-	1	0.04			0.04	-	1	NA			NA	-	0
50 Yards	Sand	AFDW	0.01			0.01	-	1	0.01			0.01	-	1	NA			NA	-	0
50 Yards	A+S	AFDW				0.09	-					0.05	-					NA	-	
Round H.			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/11/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
Shoreline	Algae	Chl. <i>a</i>	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	Sand	Chl. <i>a</i>	0			0	-	1	0			0	-	1	0			0	-	1
Shoreline	A+S	Chl. <i>a</i>				0	-					0	-					0	-	
3 Yards	Algae	Chl. <i>a</i>	0.77	1.02		0.90	0.18	2	0			0	-	1	0.22	0.25		0.24	0.02	2
3 Yards	Sand	Chl. <i>a</i>	0.39	0.58		0.49	0.13	2	0			0	-	1	0.43	0.54		0.49	0.08	2
3 Yards	A+S	Chl. <i>a</i>				1.39	-					0	-					0.73	-	
25 Yards	Algae	Chl. <i>a</i>	0.06	0.07		0.06	0.01	2	0.32	0.30		0.31	.01	2	2.13	2.41		2.27	0.20	2
25 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	NA			NA	-	0	1.52	1.40		1.46	0.09	2
25 Yards	A+S	Chl. <i>a</i>				NA	-					NA	-					3.73	-	
50 Yards	Algae	Chl. <i>a</i>	0.08	0.11		0.09	0.02	2	0.05	0.05		0.05	0	2	NA			NA	-	0
50 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	NA			NA	-	0	NA			NA	-	0
50 Yards	A+S	Chl. <i>a</i>				NA	-					NA	-						-	

Appendix 1.c. Regan Beach 8/20/15 Dry Weight, AFDW, and Chlorophyll *a* (coarse separation of algae and sand fractions done). NA-Not Analy.; NF-No sand fract.

Regan		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/20/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	Dry Wt.	55.87	56.77		56.32	0.64	2	225.54	229.64		227.57	2.88	2	31.80	31.20		31.50	0.42	2
Shoreline	Sand	Dry Wt.	364.92			364.92	-	1	349.40			349.40	-	1	317.42			317.42	-	1
Shoreline	A+S	Dry Wt.				421.24	-					576.97	-					348.92	-	
10 Yards	Algae	Dry Wt.	76.88			76.88	-	1	5.71			5.71	-	1	1.29			1.29	-	1
10 Yards	Sand	Dry Wt.	307.26			307.26	-	1	13.53			13.53	-	1	4.19			4.19	-	1
10 Yards	A+S	Dry Wt.				384.14	-					19.24	-					5.48	-	
50 Yards	Algae	Dry Wt.	0.01			0.01	-	1	0.02			0.02	-	1	0.01			0.01	-	1
50 Yards	Sand	Dry Wt.	NF			NF	-		3.08			3.08	-	1	0.35			0.35	-	1
50 Yards	A+S	Dry Wt.				0.01	-					3.10	-					0.36	-	
100 Yards	Algae	Dry Wt.	4.51			4.51	-	1	3.26			3.26	-	1	6.39			6.39	-	1
100 Yards	Sand	Dry Wt.	8.89			8.89	-	1	3.24			3.24	-	1	2.15			2.15	-	1
100 Yards	A+S	Dry Wt.				13.40	-					6.50	-					8.54	-	
150 Yards	Algae	Dry Wt.	4.86			4.86	-	1	7.76			7.76	-	1	11.11			11.11	-	1
150 Yards	Sand	Dry Wt.	NF			NF	-		NF			NF	-		NF			NF	-	
150 Yards	A+S	Dry Wt.				4.86	-					7.76	-					11.11	-	
Regan		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/20/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	AFDW	4.07	4.24		4.15	0.12	2	21.57	19.58		20.58	1.41	2	3.15	3.21		3.18	0.04	2
Shoreline	Sand	AFDW	4.22			4.22	-	1	NA			NA	-	0	2.83			2.83	-	1
Shoreline	A+S	AFDW				8.37	-					NA	-					6.01	-	
10 Yards	Algae	AFDW	9.90			9.90	-	1	0.57			0.57	-	1	0.13			0.13	-	1
10 Yards	Sand	AFDW	3.08			3.08	-	1	0.26			0.26	-	1	0.04			0.04	-	1
10 Yards	A+S	AFDW				12.98	-					0.83	-					0.17	-	
50 Yards	Algae	AFDW	0.01			0.01	-	1	0.01			0.01	-	1	0.01			0.01	-	1
50 Yards	Sand	AFDW	NF			NF	-		0.02			0.02	-	1	0			0	-	1
50 Yards	A+S	AFDW				0.01	-					0.03	-					0.01	-	
100 Yards	Algae	AFDW	0.35			0.35	-	1	0.21			0.21	-	1	0.44			0.44	-	1
100 Yards	Sand	AFDW	0.07			0.07	-	1	0.03			0.03	-	1	0.02			0.02	-	1
100 Yards	A+S	AFDW				0.42	-					0.24	-					0.46	-	
150 Yards	Algae	AFDW	0.42			0.42	-	1	0.43			0.43	-	1	0.75			0.75	-	1
150 Yards	Sand	AFDW	NF			NF	-		NF			NF	-		NF			NF	-	
150 Yards	A+S	AFDW				0.42	-					0.43	-					0.75	-	
Regan			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/20/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
Shoreline	Algae	Chl. <i>a</i>	4.11	9.41		6.76	3.75	2	12.95	41.11		27.03	19.91	2	4.20	4.57		4.39	0.26	2
Shoreline	Sand	Chl. <i>a</i>	NA			NA	-	0	4.41			4.41	-	1	3.19			3.19	-	1
Shoreline	A+S	Chl. <i>a</i>				NA	-					31.44	-					7.58	-	
10 Yards	Algae	Chl. <i>a</i>	7.88			7.88	-	1	1.12			1.12	-	1	0.28			0.28	-	1
10 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	0.10			0.10	-	1	NA			NA	-	0
10 Yards	A+S	Chl. <i>a</i>				NA	-					1.22	-					NA	-	
50 Yards	Algae	Chl. <i>a</i>	0.01			0.01	-	1	0.04			0.04	-	1	0.02			0.02	-	1
50 Yards	Sand	Chl. <i>a</i>	NF			NF	-		NA			NA	-	0	NA			NA	-	0
50 Yards	A+S	Chl. <i>a</i>				0.01	-					NA	-					NA	-	
100 Yards	Algae	Chl. <i>a</i>	0.94			0.94	-	1	0.32			0.32	-	1	1.08			1.08	-	1
100 Yards	Sand	Chl. <i>a</i>	NA			NA	-	0	NA			NA	-	0	NA			NA	-	0
100 Yards	A+S	Chl. <i>a</i>				NA	-					NA	-					NA	-	

Regan			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/20/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
150 Yards	Algae	Chl. <i>a</i>	1.24			1.24	-	1	1.21			1.21	-	1	1.99			1.99	-	1
150 Yards	Sand	Chl. <i>a</i>	NF			NF			NF			NF			NF			NF	-	
150 Yards	A+S	Chl. <i>a</i>				1.24	-					1.21	-					1.99	-	

Appendix 1.d. Regan Beach 9/21/15 Dry Weight, AFDW, and Chlorophyll *a* (coarse separation of algae and sand fractions done). NA-Not Analy.; NF-No sand fract.

Regan		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/21/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	Dry Wt.	162.18			162.18		1	8.99			8.99		1	2.75			2.75		1
Shoreline	Sand	Dry Wt.	519.03			519.03		1	34.71			34.71		1	21.47			21.47		1
Shoreline	A+S	Dry Wt.				681.21						43.70						24.22		
10 Yards	Algae	Dry Wt.	0.06			0.06		1	102.36			102.36		1	28.40			28.40		1
10 Yards	Sand	Dry Wt.	0.21			0.21		1	114.91			114.91		1	40.12			40.12		1
10 Yards	A+S	Dry Wt.				0.27						217.27						68.52		
50 Yards	Algae	Dry Wt.	0.01			0.01		1	0.59			0.59		1	0			0		1
50 Yards	Sand	Dry Wt.	NF			NF			NF			NF			NF			NF		
50 Yards	A+S	Dry Wt.				0.01						0.59						0		
100 Yards	Algae	Dry Wt.	0.29			0.29		1	2.89			2.89		1	15.96			15.96		1
100 Yards	Sand	Dry Wt.	0.38			0.38		1	NF			NF			NF			NF		
100 Yards	A+S	Dry Wt.				0.67						2.89						15.96		
150 Yards	Algae	Dry Wt.	0.32			0.32		1	2.59			2.59		1	8.60			8.60		1
150 Yards	Sand	Dry Wt.	0.84			0.84		1	0.33			0.33		1	0.11			0.11		1
150 Yards	A+S	Dry Wt.				1.16						2.92						8.72		
Regan		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/21/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	AFDW	13.89			13.89		1	1.06			1.06		1	0.25			0.25		1
Shoreline	Sand	AFDW	4.56			4.56		1	0.28			0.28		1	0.36			0.36		1
Shoreline	A+S	AFDW				18.45						1.34						.61		
10 Yards	Algae	AFDW	0			0		1	6.09			6.09		1	2.31			2.31		1
10 Yards	Sand	AFDW	0.01			0.01		1	1.23			1.23		1	0.33			0.33		1
10 Yards	A+S	AFDW				0.01						7.32						2.64		
50 Yards	Algae	AFDW	0.01			0.01		1	0.03			0.03		1	0			0		1
50 Yards	Sand	AFDW	NF			NF			NF			NF			NF			NF		
50 Yards	A+S	AFDW				0.01						0.03						0		
100 Yards	Algae	AFDW	0.02			0.02		1	0.16			0.16		1	0.72			0.72		1
100 Yards	Sand	AFDW	0			0		1	NF			NF		1	NF			NF		
100 Yards	A+S	AFDW				0.02						0.16						0.72		
150 Yards	Algae	AFDW	0.03			0.03		1	0.12			0.12		1	0.52			0.52		1
150 Yards	Sand	AFDW	0.01			0.01		1	0			0		1	0			0		1
150 Yards	A+S	AFDW				0.04						0.12						0.52		
Regan			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/21/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
Shoreline	Algae	Chl. <i>a</i>	36.25			36.25		1	1.20			1.20		1	0.56			0.56		1
Shoreline	Sand	Chl. <i>a</i>	NA			NA		0	0.23			0.23		1	NA			NA		0
Shoreline	A+S	Chl. <i>a</i>				1.43			1.43			1.43						NA		
10 Yards	Algae	Chl. <i>a</i>	NES			NES		0	11.00			11.00		1	4.73			4.73		1
10 Yards	Sand	Chl. <i>a</i>	NA			NA		0	NA			NA		0	NA			NA		0
10 Yards	A+S	Chl. <i>a</i>				NA						NA						NA		
50 Yards	Algae	Chl. <i>a</i>	NA			NA		0	0.06			0.06		1	NA			NA		1
50 Yards	Sand	Chl. <i>a</i>	NF			NF			NF			NF			NF			NF		
50 Yards	A+S	Chl. <i>a</i>				NA						0.06						NA		
100 Yards	Algae	Chl. <i>a</i>	0.05			0.05		1	0.25			0.25		1	1.15			1.15		1
100 Yards	Sand	Chl. <i>a</i>	NA			NA		0	NF			NF			NF			NF		
100 Yards	A+S	Chl. <i>a</i>				NA						0.25						1.15		

Regan			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/21/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
150 Yards	Algae	Chl. <i>a</i>	0.04			0.04		1	0.26			0.26		1	1.38			1.38		1
150 Yards	Sand	Chl. <i>a</i>	NA			NA		0	NA			NA		0	NA			NA		0
150 Yards	A+S	Chl. <i>a</i>				NA						NA								

Appendix 1.e. Round Hill Pines Beach 9/24/15 Dry Weight, AFDW, and Chlorophyll *a* (coarse separation of algae and sand fractions done). NA-Not Analy.; NF-No sand fract.

Round HP		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/24/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	Dry Wt.	0			0		1	0			0		1	0			0		1
Shoreline	Sand	Dry Wt.	0			0		1	0			0		1	0			0		1
Shoreline	A+S	Dry Wt.				0						0						0		
3 Yards	Algae	Dry Wt.	0.40			0.40		1	0			0		1	7.91			7.91		1
3 Yards	Sand	Dry Wt.	0.23			0.23		1	0			0		1	1.75			1.75		1
3 Yards	A+S	Dry Wt.				0.63						0						9.66		
25 Yards	Algae	Dry Wt.	0.27			0.27		1	0.12			0.12		1	0.30			0.30		1
25 Yards	Sand	Dry Wt.	1.14			1.14		1	NF			NF			NF			NF		
25 Yards	A+S	Dry Wt.				1.41						0.12						0.30		
50 Yards	Algae	Dry Wt.	NA			NA		0	NA			NA		0	0			0		1
50 Yards	Sand	Dry Wt.	NF			NF			NF			NF			NF			NF		
50 Yards	A+S	Dry Wt.				NA						NA						0		
100 Yards	Algae	Dry Wt.	0.07			0.07		1	0.07			0.07		1	0.14			0.14		1
100 Yards	Sand	Dry Wt.	NF			NF			NF			NF			NF			NF		
100 Yards	A+S	Dry Wt.				0.07						0.07						0.14		
Round HP		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/24/15			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
Shoreline	Algae	AFDW	0			0		1	0			0		1	0			0		1
Shoreline	Sand	AFDW	0			0		1	0			0		1	0			0		1
Shoreline	A+S	AFDW				0						0						0		
3 Yards	Algae	AFDW	0.08			0.08		1	0			0		1	0.46			0.46		1
3 Yards	Sand	AFDW	0			0		1	0			0		1	.01			.01		1
3 Yards	A+S	AFDW				0.08						0						0.47		
25 Yards	Algae	AFDW	0.01			0.01		1	0.01			0.01		1	0.04			0.04		1
25 Yards	Sand	AFDW	0.01			0.01		1	0			0		1	0			0		1
25 Yards	A+S	AFDW				0.02						0.01						0.04		
50 Yards	Algae	AFDW	NA			NA		0	NA			NA		0	0			0		1
50 Yards	Sand	AFDW	NF			NF			NF			NF			NF			NF		
50 Yards	A+S	AFDW				NA						NA						0		
100 Yards	Algae	AFDW	0			0		1	0			0		1	.01			.01		1
100 Yards	Sand	AFDW	NF			NF			NF			NF			NF			NF		
100 Yards	A+S	AFDW				0						0						0.01		
Round HP			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/24/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
Shoreline	Algae	Chl. <i>a</i>	0			0		1	0			0		1	0			0		1
Shoreline	Sand	Chl. <i>a</i>	0			0		1	0			0		1	0			0		1
Shoreline	A+S	Chl. <i>a</i>				0						0						0		
3 Yards	Algae	Chl. <i>a</i>	0.10			0.10		1	0			0		1	0.61			0.61		1
3 Yards	Sand	Chl. <i>a</i>	NA			NA		0	0			0		1	NA			NA		0
3 Yards	A+S	Chl. <i>a</i>				NA						0						NA		
25 Yards	Algae	Chl. <i>a</i>	0.02			0.02		1	0.02			0.02		1	0.05			0.05		1
25 Yards	Sand	Chl. <i>a</i>	NA			NA		0	NA			NA		0	NA			NA		0
25 Yards	A+S	Chl. <i>a</i>				NA						NA						NA		
50 Yards	Algae	Chl. <i>a</i>	NA			NA		1	NA			NA		0	0			0		1
50 Yards	Sand	Chl. <i>a</i>	NF			NF			NF			NF			NF			NF		

Round HP			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
9/24/15			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
50 Yards	A+S	Chl. <i>a</i>				NA						NA						0		
100 Yards	Algae	Chl. <i>a</i>	0.01			0.01		1	0.01			0.01		1	0.02			0.02		1
100 Yards	Sand	Chl. <i>a</i>	NF			NF			NF			NF			NF			NF		
100 Yards	A+S	Chl. <i>a</i>				0.01						0.01						0.02		

Appendix 1.f. Regan Beach 8/9/17 Dry Weight, AFDW, and Chlorophyll *a* (algae and sand fractions not separated).

Regan		Dry Wt.	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/9/17			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
62 Yards	A+S	Dry Wt.	0.17			0.17		1	NA			NA		0	NA			NA		0
112 Yards	A+S	Dry Wt.	NA			NA		0	NA			NA		0	14.91	16.46	14.90	15.42	0.90	3
Regan		AFDW	L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/9/17			g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²		g/m ²	g/m ²	g/m ²	g/m ²	g/m ²	
62 Yards	A+S	AFDW	0.05			0.05		1	0			0		1	NA			NA		0
112 Yards	A+S	AFDW	NA			NA		0	NA			NA		0	2.60	2.73	2.91	2.75	0.16	3
Regan			L-Rep.1	L-Rep.2	L-Rep.3	Mean	S. Dev.	n	Rep.1	Rep.2	Rep.3	Mean	S. Dev.	n	R-Rep.1	R-Rep.2	R-Rep.3	Mean	S. Dev.	n
8/9/17			mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²		mg/m ²	mg/m ²	mg/m ²	mg/m ²	mg/m ²	
62 Yards	A+S	Chl. <i>a</i>	0.09			0.09		1	0			0		1	NA			NA		0
112 Yards	A+S	Chl. <i>a</i>	NA			NA		0	NA			NA		0	4.96	4.34	4.63	4.64	0.31	3

Appendix Table 2. Pilot study values for visual estimates of percent cover, means, std. deviations, estimates of variance, and number of quadrats needed to estimate the mean percent cover with the indicated 95% confidence interval. “*d*” is the half-width for the confidence interval used in Equation 1 to estimate number of replicate quadrats needed.

Site	Pilot Study Visual Estimates of Percent Cover (%)	Pilot Study Mean % Cover ±S.D. (n)	Variance S ² (%) ²	95% Confidence Interval	<i>d</i>	Number of 0.25 m ² Quadrats Needed to Estimate the Mean
Shoreline Samples						
El Dorado 10/7/15	22.4, 19.7, 19.5, 17.4, 10.4, 10, 5.4 ⁷	14.97± 6.34 (n=7)	40.20	≤5%	≤2.5%	27
“	“	“	“	≤10%	≤5%	9
“	“	“	“	≤20%	≤10%	4
Regan 8/20/15	70, 65, 50, 15, 15, 6	36.83±25.17 (n=6)	794.167	≤20%	≤10%	33
“	“	“	“	≤30%	≤15%	16
“	“	“	“	≤40%	≤20%	10
Offshore Samples						
El Dorado 8/6/15	5, 5, 4, 3, 3, 3, 3, 2, 2, 1, 0, 0	2.6± 1.679 (n=12)	2.811	≤2%	≤1%	13
‘	‘	‘	‘	≤3%	≤1.5%	7
‘	‘	‘	‘	≤4%	≤2%	5
Regan 8/20/15	20, 15, 15, 15, 10, 7, 5, 3, 2	10.22±6.34 (9)	40.194	≤5%	≤2.5%	27
‘	‘	‘	‘	≤8%	≤4%	12
‘	‘	‘	‘	≤12%	≤6%	7

⁷ One site with very high percent cover was not included in measurements of the mean and S² since the sampling design to reduce bias (i.e. sampling every 25 yds. along a transect) was not used for that sample.

