

BIOMASS AND PRODUCTIVITY DETERMINATION OF ALGAE IN THE GREAT LAKES

PREPARED FOR

Working Group II
of the Water Management
Steering Committee



Ontario

Ministry
of the
Environment

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IN THE GREAT LAKES**

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Working Group II
of the Water Management
Steering Committee

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NOTE

The receiving water assessment techniques presented in this report describe the methods commonly in use within the Ministry. Alternative techniques exist and/or will eventually be developed. Consultation with Ministry of the Environment staff is advisable to determine the suitability of the alternative techniques.

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SUMMARY

This report presents a brief overview of eutrophication - related algal biomass increases in the Great Lakes and their relationship to such water quality problems as: taste and odour and clogging of filters at water treatment plants, oxygen depletion, alteration of aquatic habitat, aesthetic impairment and toxicity.

The majority of the report deals with a review of methods available for the assessment of algal biomass and productivity, and, for illustration, relates methods employed by the MOE Great Lakes Surveys Unit in the nearshore of Lake Ontario. The application of data gathered by these methods in the categorization of lake trophic status is also briefly discussed.

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A. PROBLEMS RELATED TO ALGAL BIOMASS INCREASES

1. Cause

Point sources (municipal and industrial), diffuse sources (land runoff, groundwater), the atmosphere (wet and dry precipitation) and shoreline erosion all have the potential to accelerate the input of nutrients (e.g. nitrogen and phosphorus) to the Great Lakes (Andren *et al*, 1977; Chapra and Sonzogni, 1979). Cultural eutrophication, or the manifestations of an increase in the input rate of nutrients (notably phosphorus) from human activities, has been identified as a major causative factor in the algal (planktonic and attached) and macrophyte biomass increases in the lower Great Lakes. Of specific concern are loadings derived from municipal point sources (e.g. sewage treatment plants), which are believed to mainly contain biologically available phosphorus (Millard *et al.*, 1979). Cognizant of the relationship between phosphorus and eutrophication, the water management strategies encompassed in the Great Lakes Water Quality Agreement of 1978 included provisions for stringent control of phosphorus arising from human sources.

With the aid of empirical models presently being developed for the Great Lakes, it should be possible to predict algal biomass response to anticipated reductions in phosphorus inputs. Adequate ground truth information is essential, however, for successful calibration of these models.

2. Effects

The many objectionable effects of algae, particularly the blue-greens, can be traced to the following characteristics (Prescott, 1968):

1. Their rapid rate of multiplication under favourable conditions.

2. The pseudovacuoles of some blue-green algae, which cause them to float high in the water and to concentrate at the surface where they are destroyed by intense light.
3. The mucilaginous sheaths of some species (e.g. blue-greens), which cause them to aggregate and form floating mats.
4. The high protein content of some algae, responsible for offensive odors and tastes.
5. Their ability to produce and give off antibiotics and toxins.

Generally, a decrease in the N:P (nitrogen:phosphorus) ratio in the water favours development of vacuolate, nitrogen-fixing blue-green algae that are most objectionable from a water quality standpoint. Conversely, an increase in the N:P ratio, such as that anticipated from phosphorus control programs, is believed to shift the algal species composition to decreased numbers and more favourable forms (e.g. green algae).

Major problems associated with eutrophication and excessive algal biomass include:

(a) Taste and Odour Problems

When alive, abundant growths of certain algal species can impart disagreeable tastes and odours to waters used for domestic supply. Algae responsible for taste and odour problems are largely found in the blue-green, diatom and chrysophyte groups, although some species of green algae may also be involved. Examples from these groups are given in Table 1. Tastes associated with algae can be sweet or bitter, and produce a dry, oily or metallic sensation on the tongue. Odours depend both on the species and its abundance, and can be aromatic, grassy, musty, spicy, fishy, septic, or geranium-, cucumber-, muskmelon-, nasturtium-, or violet-like (Palmer, 1962). When highly proteinaceous algae die and decompose, they may produce an even more objectionable odour. For example, extensive growths of *Cladophora* and other large

filamentous algae detached from their substrate by wave action accumulate on shores and, through decomposition, generate septic odours.

(b) Clogging of Intake Filters

If present in sufficient numbers, species from any of the algal groups can cause economic loss by clogging screens and sand filters in water supply systems. Records of the Belleville Water Treatment Plant show that prior to 1978 and the implementation of phosphorus control in the Bay of Quinte, microstrainers were in use 4-5 months of the year, as opposed to only a few weeks during 1978 and 1979 when phytoplankton densities had declined by 50-60 % (Nicholls, 1980).

In some instances, the algal groups involved may be the same as those mentioned for taste-and odour-producing algae, although the species involved need not necessarily be the same as those listed in Table 1. Of note are the diatoms (Bacillariophyceae), whose resistant siliceous walls can form layers over sand filters, thus facilitating the collection of organic matter and the development of a thick mat of filamentous algae (Aufwuchs) which can completely obstruct the filter (Prescott, 1968).

c) Oxygen Depletion

Highly productive areas such as the central basin of Lake Erie and Adolphus Reach of the Bay of Quinte in Lake Ontario are both highly productive and experience hypolimnetic oxygen depletion during stratification in the summer (Charlton, 1979; Great Lakes Water Quality Board, 1979). Dense algal populations can exert a substantial oxygen demand on the water column, due to algal cellular respiration or decomposition of detrital material (e.g. dead algal cells).

These are not the sole determinants of hypolimnetic oxygen depletion in eutrophic lakes, since that condition is also dependent on other variables such as lake depth and shape, water level, flushing rate and the thickness of the epilimnion and hypolimnion. Lake productivity and other possible

factors have also been linked with metalimnetic oxygen minima in Lake Ontario (Boyd, 1980). This condition was evident in the generally more productive nearshore waters (Stadelman *et al.*, 1974) and was most pronounced in the highly productive northeast corner of the lake near the St. Lawrence River outlet (Glooschenko *et al.*, 1974).

(d) Alteration of Aquatic Habitat

Since embayments and littoral zones receive the bulk of point and diffuse source inputs (land runoff, groundwater, shoreline erosion), these areas respond initially and may undergo significant changes in their environmental conditions, and, as a result, in their biota.

As lakes become more productive and their hypolimnia undergo periods of oxygen reduction and increases in the metabolic products of microbial decomposition, the number of organisms adapted to these conditions decreases dramatically. For example, the littoral zone normally consists of a rich benthic fauna with high oxygen demands, whereas the profundal zone is more homogeneous and becomes more so (as may the littoral) as productivity increases and species diversity decreases.

However, since predation as well as competitive pressures for available nutrients are considerably reduced, the biomass of surviving species may increase. This was the case in the western basin of Lake Erie, in which the less tolerant insect larvae were eliminated (e.g. *Hexagenia*) or reduced in numbers (eg: Chironomidae), with a concomitant increase in more tolerant forms (e.g. Tubificidae). A similar change was noted in the Bay of Quinte; Johnson and Owen, 1971).

Fish composition also changes dramatically during eutrophication: from salmonid and coregonid species with quite stringent oxygen (high) and thermal (low) requirements towards more tolerant warmwater species (eg: cyprinids). Other changes associated with eutrophication, such as changes in the littoral zone, food sources and increased organic deposition, may also affect the reproductive success of fish populations by reducing survival of the young.

(e) Aesthetic Impairment

Increased primary production due to cultural eutrophication may eventually lead to adverse effects on the aesthetics of lakes which are extensively used for swimming, boating, fishing, etc. This is particularly evident in the nearshore of Lakes Erie and Ontario, where extensive growths of the attached green alga *Cladophora* (Shear and Konasewich, 1975) or of macrophytes are a continuing problem. Also, a number of species (see Table 1) of free-floating algae accumulate at times to form loose, visible aggregations called "blooms" which may cover large areas and reduce water clarity, produce unpleasant odours, interfere with reaeration, release toxins and/or deplete oxygen as a result of cellular respiration or decay of the bloom.

(f) Algal Toxicity

Exotoxins and/or endotoxins produced by some species of freshwater algae have been implicated in adverse effects on both man and animals. As shown by Table 1, these algae are primarily blue-greens, but representatives of the green and dinoflagellate algae have also been shown to contain toxins (Prescott, 1968).

While algal toxins have been shown to be lethal to some fish species in laboratory tests and acute and often fatal poisonings of birds and domestic animals have been recorded after their drinking from water supplies (e.g. ponds) containing dense algal blooms, such toxicity problems are not expected in the Great Lakes nearshore, with the possible exception of confined embayments.

TABLE 1. Algal Species Related To Water Quality Problems In Freshwater Lakes.

(adapted from Palmer, 1962a, Prescott, 1963b and Taylor *et al.*, 1981c)

T & O = Taste- and odour-producing alga
 FC = Filter-clogging alga
 AN = Aesthetic nuisance alga
 T = Toxic alga

Algal Species	Associated Problems	Source
Blue-Green Algae (Cyanophyceae):		
Anabaena circinalis	T & O, T	a,b
Anabaena flos-aquae	FC, T	a,b
Anabaena inaequalis	T	b
Anabaena lemmermanni	T	a,c
Anabaena limnetica	T	b
Anabaena Nadsonii	T	b
Anabaena planctonica	T & O, AN	a,c
Anabaena Scheremetievi	T	b
Anabaenopsis sp.	T & O	c
Anacystis cyanea	T & O	a
Anacystis dimidiata (Chroococcus turgidus)	FC	a
Aphanizomenon flos-aquae	T & O, T, AN	a,b,c
Aphanothece nidulans	T	b
Aphanothece cyanea	T	b
Coelosphaerium Kuetzingianum	T & O, T, AN	c
Cylindrospermum musicola	T & O	a
Gloeotrichiae chinulata	FC, T	a,b
Gomphosphaeria lacustris, kuetzingianum type	T & O, T	a
Lyngbya Birgei	T	b
Lyngbya contorta	T	a,c

Table 1: cont'd

<i>Lyngbya limetica</i>	AN	c
<i>Microcystis aeruginosa</i>	T & O, T, AN	a,b,c
<i>Microcystis flos-aquae</i>	T	a,c
<i>Microcystis toxica</i>	T	a,b
<i>Nodularia spumigena</i>	T	a,b
<i>Oscillatoria amphibia</i>	FC	a,c
<i>Oscillatoria chalybya</i>	FC	a
<i>Oscillatoria curviceps</i>	T & O	a
<i>Oscillatoria ornata</i>	FC	a
<i>Oscillatoria princeps</i>	FC	a,c
<i>Oscillatoria pseudogeminata</i>	FC	a
<i>Oscillatoria rubescens</i>	FC	a
<i>Oscillatoria splendida</i>	FC	a
<i>Oscillatoria tenuis</i>	T & O	c
<i>Rivularia dura</i>	FC	a
<i>Rivularia haematites</i>	T & O	a
Green and Yellow-Green Algae (non-motile Chlorophyceae, etc):		
<i>Actinastrum</i> sp.	T & O	c
<i>Chara vulgaris</i>	T & O	a
<i>Chlorella</i> sp.	T & O, T	c,b
<i>Chlorella pyrenoidosa</i>	FC	a
<i>Cladophora aegagropila</i>	FC	a
<i>Cladophora insignis</i>	T & O	a
<i>Closterium moniliferum</i>	FC	a,c
<i>Cosmarium portianum</i>	T & O	a
<i>Dichotomosiphon tuberosus</i>	FC	a
<i>Dictyosphaerium ehrenbergianum</i>	T & O	a,c
<i>Dictyosphaerium pulchellum</i>	FC	a,c
<i>Gloeocystis planctonica</i>	T & O	a,c
<i>Hydrodictyon reticulatum</i>	T & O, FC	a
<i>Mougeotia sphaerocarpa</i>	FC	a

Table 1 : cont'd

Nitella gracilis	T & O	a
Palmella mucosa	FC	a
Pediastrum tetras	T & O	a,c
Scenedesmus sp.	T	b
Scenedesmus abundans	T & O	a,c
Spirogyra majuscula	T & O	a
Spirogyra porticalis	FC	a
Staurastrum paradoxum	T & O	a,c
Tribonema bombycinum	FC	a
Ulothrix sp.	T & O	a
Ulothrix variabilis	FC	a
Zygnema insigne	FC	a
Diatoms (Bacillariophyceae):		
Asterionella formosa (A. gracillima)	T & O, FC, AN	a,c
Cyclotella compta	T & O	a ,c
Cyclotella meneghiniana	FC	a,c
Cymbella ventricosa	FC	a,c
Diatoma vulgare	T & O, FC	a,c
Fragilaria construens	T & O	a,c
Fragilaria crotonensis	FC	a,c
Melosira sp.	T & O	c
Melosira granulata	FC	a,c
Melosira varians	FC	a
Meridion sp.	T & O	c
Navicula graciloides	FC	a
Navicula lanceolate	FC	a,c
Nitzschia palea	FC	a,c
Pleurosigma sp.	T & O	c
Stephanodiscus binderanus	FC	a
Stephanodiscus hantzschii	FC	a,c
Stephanodiscus niagarae	T & O	a,c
Synedra acus	FC	a,c

Table 1 : cont'd

Synedra acus var. radians (<i>S. delicatissima</i>)	FC	a,c
Synedra ulna	T & O	a,c
Synedra pulchella	FC	a,c
Tabellaria fenestrata	T & O, FC	a,c
Tabellaria flocculosa	FC	a,c
Pigmented Flagellates (motile Chlorophyceae, Chrysophyceae, Dinophyceae, Euglenophyceae):		
Ceratium hirundinella	T & O, FC	a,c
Chlamydomonas globosa	T & O	a,c
Chrysosphaerella longispina	T & O	a
Cryptomonas erosa	T & O	a,c
Dinobryon divergens	T & O	a,c
Dinobryon sertularia	FC	a,c
Euglena sanguinea	T & O	a
Eudorina sp.	T & O	c
Glenodinium palustre	T & O	a
Gonium sp.	T & O	c
Gonyaulaxca tenella	T	b
Gymnodinium brevis	T	b
Mallomonas caudata	T & O	a,c
Pandorina morum	T & O	a,c
Peridinium cinctum	t & O	ac
Peridinium wisconsinense	FC	a,c
Synura uvella	T & O	a,c
Trachelomonas crebea	FC	a,c
Uroglenopsis americana	T & O	a
Volvox aureus	T & O	a

B. ASSESSMENT OF THE PROBLEM

Below is a general outline of procedures employed by the Great Lakes Surveys Unit on Lake Ontario, as well as some brief comments on other possible methods. These procedures represent the application of general principles outlined in manuals or papers by Strickland (1960), Strickland and Parsons (1968), Tailing and Driver (1963), Vollenweider (1969) and Carnes and Millner (1980). Such references are highly recommended since it is recognized that our methods may not be applicable to inland lake studies without modification. In many instances, the equipment will not need to be as extensive or elaborate.

1. Assessment Techniques: Phytoplankton

Biomass is the weight of all living material per unit area (or volume) at a given point in time. In the case of phytoplankton, biomass is equivalent to standing crop. Since biomass reflects the availability of nutrients, (e.g. cultural eutrophication) seasonal population fluctuations, and cropping losses, its estimation is essential in any analysis of aquatic plant populations or of productivity dynamics.

Numerous methods have been used to evaluate the biomass and productivity of phytoplankton, including: enumeration, volume, wet (fresh) weight, carbon content, carbon fixation, photosynthetic pigments, energy as heat of combustion, ATP*, and the rates of exchange of oxygen and carbon dioxide.

(a) Sampling Regime (see Appendix for sampling apparatus)

Sampling should utilize depth-integrated methods to yield volume-weighted biomass estimates for the water column. In the nearshore, depth sampling along transects has been found useful in delineating inshore-offshore gradients which may be present, whereas station grids are preferable in harbours or embayments.

* Adenosine -5'-triphosphate

The approximate depth(s) of phytoplankton peak abundance at a station and hence, the sampling depths, are determined by initial depth profiling of chlorophyll a and turbidity using a flow-through equipped fluorometer and turbidimeter. The maximum sampling depth for productivity studies is determined with the aid of a visible light profile. In temperate waters, this is usually the compensation depth (where available light is 1% of the surface intensity), but if the mixed layer is much deeper than this, then biomass could be underestimated and productivity overestimated due to circulation of phytoplankton in deeper waters - hence the need for a preliminary chlorophyll a profile determination.

A sufficient volume of water is also taken from each depth to allow for analysis of the following physical and chemical parameters: temperature, dissolved oxygen, total and dissolved phosphorus, Kjeldahl nitrogen, ammonia, nitrite, nitrate, dissolved silica, chlorophylls a and b (corrected and uncorrected for degradation products), total and inorganic carbon, alkalinity and pH. (The latter three parameters are important for primary production determinations and calculations). Samples are also taken and preserved for later phytoplankton enumeration, speciation and biomass estimates. In addition, primary productivity is determined for each depth by use of the ¹⁴C - light and dark bottle method outlined later (see "Productivity Estimation").

Sampling frequency is dictated by local conditions. For example, during the summer, the Great Lakes nearshore is affected by frequent upwellings which can affect stratification and, hence, vertical phytoplankton distribution (Bowers, 1980). Therefore, intensive sampling (e.g. weekly or more frequently if possible) is essential, and should be done in conjunction with continuous monitoring of the thermal regime (using *in-situ* temperature recorders) in order to relate biomass changes to dynamics of the nearshore. However, during the winter, when ice conditions often preclude sampling by vessel, isothermal conditions permit more or less complete mixing of the nearshore (horizontally and vertically) so that samples obtained from water intakes are adequate for the assessment of nutrient and phytoplankton status. Smith and Shapiro (1981) have emphasized that models (e.g. chlorophyll total phosphorus) intended for predictive purposes be based on data collected during

summer months when algal biomass is most closely related to nutrient concentration.

(b) Biomass Estimation

- (i) Enumeration and Volume - Numbers are perhaps the best qualitative expression of phytoplankton populations (Vollenweider, 1969) and differentiation among species and of organisms from detrital particles. This is usually done on a preserved sample of known volume which has been concentrated by sedimentation (Utermohl, 1958) or on live samples concentrated by centrifugation or collected on a membrane filter (Holmes, 1962). Cell numbers have also been determined using electronic dimensional particle-counters (eg: Coulter counter).

However, numbers do not yield a true index of biomass because species vary greatly in cell or colony size and, hence, volume and weight. Electronic counters can also yield information about particle volumes but are unable to distinguish between organic detritus, inorganic material and plankton. Therefore, the preferred biomass estimate, and the one used by this Unit, is to multiply the number of individuals of each species by their average cell volume (determined from mean cell dimensions) to obtain the total volume of phytoplankton. This value can then be used to calculate the fresh weight biomass (see "Conversion Factors") per unit volume or surface area.

In addition to enumeration and volume, speciation of algae is essential to the determination of changes in the phytoplankton community, both spatially and temporally. Such changes may cause filter clogging or taste and odour problems at water intakes (Palmer, 1962).

- (ii) Photosynthetic Pigments - Owing to their importance in photosynthesis, measurements of algal pigments have

received considerable attention as an indicator of biomass. Chlorophyll a is the pigment of major interest, due to its ubiquity in all phytoplankton phyla and (usually) greater abundance than chlorophyll b or c.

For the range of chlorophyll a concentrations commonly encountered in the Great Lakes (1-20 µg/L), 0.5 to 2 litres of water is collected and filtered through membrane or glass fibre filters in the presence of magnesium carbonate (Vollenweider, 1969). Either acetone or methanol is then used to extract pigments from the cells ;in the dark and usually refrigerated) and the chlorophyll a, b or c concentration calculated by *in vitro* spectrophotometric analysis of the extract in the laboratory and the use of appropriate equations (see Talling, 1969; Westlake, 1969; Wetzel and Westlake, 1969). A number of procedures have been used when pigment extraction is incomplete with certain algal species. Some of these are: grinding, sonication and the use of alternate solvents such as dimethyl sulfoxide (Shoaf and Lium, 1976) and boiling methanol.

For *in vitro* analysis of samples from waters of low productivity (chlor. a 1 µg/L) as well as rapid *in vivo* (no extraction) field measurements, a more sensitive fluorometric method is available (Lorenzen, 1966; Tunzi *et al.*, 1974). During *in vivo* fluorescence measurements, the variability of the fluorescence yield coefficient of chlorophyll a with local species, light and nutrient conditions has meant that such data is in many instances not strictly comparable with that obtained from other study areas and/or from laboratory *in vitro* solvent extraction analyses. However, the addition of the photosynthetic electron transport blocking agent DCMU [3 -(3,4 dichlorophenyl) - 1,1 dimethylurea] to samples before measurement eliminates the variability in photochemical quenching and *in vivo* fluorescence yield, which then becomes a constant

function of cellular chlorophyll a (Slovacek and Hannan, 1977).

While cell pigment content varies appreciably (as do elemental constituents other than carbon) with species and environmental parameters, the ability to correct accurately for pigment degradation products (Lorenzen, 1967; Whitney and Daryl, 1979) in order to measure only the functional pigment content of phytoplankton (distinct from particulate detritus) permits effective analyses of composite phytoplankton population changes (Wetzel, 1975).

The Water Resources Branch presently uses field filtration of discrete water samples with acetone extraction. However, dimethyl sulfoxide extraction seems to be a promising alternative. Despite the occasional lack of correlation between in vivo chlorophyll a determinations by fluorometry (not DCMU - enhanced) and in vitro levels obtained by solvent extraction, the former is being used as a quick scan of phytoplankton biomass distribution in lakes (see "Sampling Regime").

- (iii) Weight (Biomass) - Fresh weight (the weight of organisms without any adherent water) is essentially equivalent to wet weight. However, because of the highly variable water content of nearly all organisms, wet weight determination by direct weighing of a centrifuged or filtered sample should be avoided as an estimate of biomass (Wetzel, 1975). Fresh weight can also be calculated from cell volume data (see "Enumeration and Volume" and "Conversion Factors").

Dry weight is widely used in production analysis, but varies with drying temperatures below 105°C due to retention of residual water.

Organic (ash-free) dry weight is usually obtained by determining the loss in weight after ignition at 550°C and is

the preferred biomass method for larger organisms, since ash content may comprise 50% or more of the dry weight of algae with massive inorganic skeletal structures (e.g. diatoms with siliceous walls) - (Vollenweider, 1969). However, there is an inherent problem in this method due to the difficulty in separating algae and other small microorganisms from detrital particulate organic matter.

- (iv) Cellular Constituents - The organic carbon content of plants (obtained by oxidising the organic plant material back to the carbon dioxide from which it originated in photosynthetic reduction) is one of their least variable constituents - the average carbon content of algae is 53 + 5% of the ash-free organic dry weight (Wetzel, 1975). However, extraneous organic detritus is still a problem with this estimate of biomass.

The measurement of ATP has been used as an indicator of both microbial and phytoplankton biomass in lakes since it is present in all living organisms. Its disappearance immediately after death and a fairly uniform conversion factor to cellular organic carbon (Holm - Hansen, 1970), make the measurement of ATP a useful adjunct to other determinations which are unable to distinguish between living and detrital biomass.

- (v) Conversion Factors - Several conversion factors have been used to obtain biomass estimates of one cellular component from another. Cellular volume can be converted to fresh weight, assuming a mean specific density of one for the phytoplankton (e.g. $\text{mm}^3/\text{L} = \text{mg}/\text{m}^3$).

Phytoplankton carbon content is usually estimated from average species content and extrapolated to natural populations by volume measurements since the carbon to volume relationship in algae has been found to be allometric (Mullin *et al.*, 1966); it can also be estimated from ATP

determinations (Holm- Hansen. 1970). Areal Standard Units (one A.S.U. = area of algal material equivalent to $400 \mu\text{m}^2$), whereby phytoplankton are counted and their optical, cross-sectional areas measured (Schenk and Thompson, 1965), are used by the Ontario Ministry of the Environment (MOE) in the monitoring of some Great Lakes water treatment plant intakes. A regression equation between this parameter and total cell volume has been derived by Nicholls (1980) which would allow for estimation of biomass from A.S.U.

The relationship between other components may be more complex than this or dependent on various external (environmental) conditions (e.g. carbon to nitrogen or nitrogen to phosphorus relationships in phytoplankton) and should be used with caution. For example, one of the chief aims of the MOE's nearshore surveillance program is to monitor the effectiveness of phosphorus removal programs by assessing the trophic status of the nearshore. At present, this is accomplished through the monitoring of phosphorus and chlorophyll a levels in the receiving water. This latter parameter may then be used to estimate biomass using a conversion factor. However, despite the accuracy and the rapidity of the actual chlorophyll a determination, the measure of biomass by this indirect method is rather imprecise since chlorophyll a in algal cells is dependent on:

- ▶ variations in the physiological condition of the population (senescent cells contain less pigment, while light-limited cells contain more).
- ▶ variations in the taxonomic composition of the phytoplankton community (absolute quantities of chlorophyll a and relative proportions of this pigment to other chlorophylls varies between species). incomplete pigment extraction for certain species.

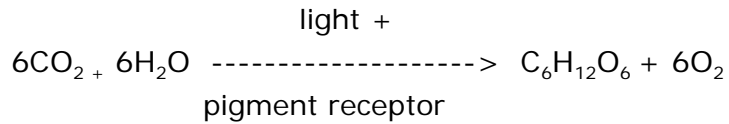
Hence, chlorophyll a levels and phytoplankton volume (i.e. biomass) may not correlate well. In addition, chlorophyll a content may not be indicative of the productivity of cells. However, this method of assessment is quick and if only one or two depths are sampled, a large area of the nearshore can be assessed.

(C) Productivity Estimation

It is important to know not only the biomass, but also the rate at which organic material is produced in a water body. This rate reflects not only the availability of nutrients and their interaction with environmental factors to determine the productivity of a given water body, but also determines to a large degree conditions of food availability, ionic composition, pH, oxygen levels, etc. that delimit the range and competitive abilities of aquatic fauna. Primary production is the quantity of new organic matter (biomass) created by photosynthesis (or chemosynthesis) or the stored energy which this material represents per unit area or volume. Primary productivity, then, is the production per unit area (or volume) per unit time and is usually an average of a number of measured instantaneous rates expressed on a daily or yearly basis, since natural phytoplankton communities are affected by many factors causing rapid, frequent, and irregular changes in the instantaneous rate. It is important, however, that the determinations be done with regard to the regeneration times (life cycles) of the phytoplankton.

Numerous techniques have been used to estimate primary production of natural phytoplankton communities, using either isolated or non-isolated populations. However, estimates of the production rates of phytoplankton based on changes in biomass are much more difficult than for aquatic macrophytes or attached algae (Vollenweider, 1969) owing to potentially substantial losses by zooplankton grazing, current transport, sedimentation or death and decomposition.

The following equation describes the overall reactions occurring during photosynthesis by photo-autotrophs:



Productivity can therefore be estimated from measurements of changes in oxygen, pH, total carbon or conductivity in the water over short time intervals. The more usual method, particularly with phytoplankton, is to measure changes in the rates of oxygen production or carbon dioxide uptake during photosynthesis by isolated samples of natural communities. These are incubated for brief intervals (1-4 h) in a depth profile at the points of collection (in- situ) or under simulated natural conditions aboard ship or in the laboratory. These changes may then be converted to the appropriate units of productivity (e.g. $\text{g C}\cdot\text{m}^{-3}\cdot\text{day}^{-1}$). Two methods have been most frequently used, both based on the light-and dark-bottle technique in which samples are incubated in clear and opaqued bottles. In the oxygen method, the initial oxygen content of water at each of the depths sampled (C_1) is determined. At the end of the incubation period, the oxygen concentration is again determined in both dark (C_2) and light (C_3) bottles.

Hence:

$$C_1 - C_2 = \text{respiration during the time interval,}$$

$$C_3 - C_1 = \text{net photosynthetic oxygen production during the time interval,}$$

$$C_3 - C_2 = \text{gross photosynthetic production of oxygen during the time interval.}$$

Although this method is relatively quick compared to the following one, it is relegated to relatively productive waters since the smallest change in oxygen measurable is about 20 $\mu\text{g/L}$ during the exposure period (approximately equivalent to 20 $\text{mg C}\cdot\text{m}^{-3}\cdot\text{day}^{-1}$). In addition, the presence of active zooplankton and bacteria can

result in errors due to their oxygen consumption. In the ^{14}C method, a known amount of $^{14}\text{CO}_2$ (usually as $\text{NaH}^{14}\text{CO}_3$) is added to water samples of known total $^{12}\text{CO}_2$ content and the samples incubated. At the end of the incubation period, the algae are collected on membrane filters and their activity determined with a geiger or, if fluor is used, a scintillation counter. Since the $^{14}\text{CO}_2$ assimilated by the phytoplankton is proportional to the $^{12}\text{CO}_2$ assimilated, then the total CO_2 assimilated by the cells can be calculated:

$$^{12}\text{CO}_2 \text{ assimilated} = \frac{^{14}\text{CO}_2 \text{ assimilated (light-dark bottle)}}{^{14}\text{CO}_2 \text{ added}} \times ^{12}\text{CO}_2 \text{ available} \times 1.06$$

This method is approximately 50 to 100 times more sensitive than the foregoing. However, it does not measure respiration directly and may underestimate primary production if appreciable amounts of ^{14}C organics are released from cells.

2. Assessment Techniques: Attached Algae (e.g. *Cladophora*)

(a) Sampling Regime

Biomass or standing crop data obtained from surveys should be expressed on an aerial basis. Productivity can be expressed in a variety of ways (e.g: per unit area or weight). Use of a sampling grid is necessary to adequately delineate the areal extent of the *Cladophora* problem.

Adequate background data should also be collected. This would include such parameters as nutrient levels, substrate characteristics, current velocity and direction, wave height, temperature and light profiles (visible as well as with specific filters). Sampling should be done at regular intervals during the growing period (May through September), the critical time being the early summer. Frequency of sampling should be site-specific and may need to be as often as once a week.

(b) Biomass Estimation

- (i) Weight/Cellular Constituents - Owing to the filamentous nature of *Cladophora*, the use of counting in conjunction with average cell volume for the determination of fresh weight biomass from total cellular volume is difficult, if not impossible. Therefore, the biomass of samples (scraped from a known area of substrate) should be determined on a biovolume, wet weight, dry weight, organic (ash-free) dry weight or organic carbon basis, the most common being dry weight.
- (ii) Photosynthetic Pigments - Determination of *in vivo* chlorophyll a of samples extracted with the appropriate solvent is likely the best method for these algae, and, with the use of the appropriate conversion factor, would provide a useful indication of algal health in conjunction with the dry weight biomass estimate.

(c) Productivity Estimation

Both the oxygen and ^{14}C techniques, as outlined previously for phytoplankton, have been used to determine the productivity of attached algae and submerged macrophytes. These studies have employed either isolated or non-isolated communities. For *Cladophora*, known areas can be enclosed by plexiglass incubation chambers which extend into the sediments and have ports to allow for sampling and injection (of ^{14}C). The ^{14}C method is preferred since it avoids the major problem with the oxygen light and dark method: consumption of oxygen by sedimental oxidative processes and by fauna. This problem can be circumvented if known quantities (weight) of *Cladophora* are scraped from the substrate and placed in incubation bottles (much like phytoplankton). However, because of the marked oxygen evolution by such samples (say, 1 g wet weight), the greater sensitivity of the ^{14}C method is not required, and the oxygen light and dark bottle method or even changes in the pH of the incubating medium can

be used.

Productivity estimates on non-isolated *Cladophora* communities, by measuring diurnal changes in oxygen, carbon dioxide or pH are feasible, and allow longer measurement periods, but necessitate determination of other parameters (exchange rates of CO₂ or O₂ between air and water).

C. Application of Data

A general categorization of the characteristics of lakes between the extremes of very oligotrophic and very eutrophic systems has been attempted many times, using a multitude of criteria and has fostered the development of a number of trophic status indices (e.g: Beeton and Edmondson, 1972; Michalski and Conroy, 1972; Carlson, 1977). The most realistic parameter for such a categorization is one that can be quantitatively determined directly, such as a rate of growth, and one that integrates the host of environmental parameters controlling the synthesis of the organic matter that enters the system (i.e. rates of autochthonous primary production). The general ranges of primary productivity of phytoplankton commonly associated with the trophic status of a lake are given in Table 2, along with several related characteristics. Although such groupings should only be used with caution, the general relationships predominate in a majority of inland waters (see Table 3 for comparable Great Lakes data adapted from Chapra and Sonzogni (1979) and Vollenweider *et al* (1974)).

TABLE 2: General Ranges of Primary Productivity of Phytoplankton and Related Characteristics of Lakes of Different Trophic Categories (from Wetzel, 1975).

Trophic Type	Mean Primary Productivity* mg C.m ⁻² .day ⁻¹	Phyto-plankton Density (cm ³ . m ⁻³)	Phyto-plankton Biomass (mg C.m ⁻³)	Chlorophyll <u>a</u> concentration (mg. m ⁻³)	Dominant Phytoplankton	Light Extinction Coefficients (n . m ⁻¹)	Total Organic Carbon (mg . L ⁻¹)	Total P (µg. L ⁻¹)	Total N (µg. L ⁻¹)	Total Inorganic Solids (mg. L ⁻¹)
Ultra-oligotrophic	<50	<1	<50	0.01-0.5	-	0.03-0.8	-	<1-5	<1-250	2-15
Oligotrophic	50-300	-	20-100	0.3-3	Chrysophyceae Cryptophyceae	0.05-1.0	<1-3	-	-	-
Oligo-mesotrophic	-	1-3	-	-	Dinophyceae, Bacillario-phyceae	-	-	5-10	250-600	10-200
Mesotrophic	250-1000	-	100-300	2-15	-	0.1-2.0	<1-5	-	-	-
Meso-eutrophic	-	3-5	-	-	-	-	-	10-30	500-1100	100-500
Eutrophic	>1000	-	>300	10-500	Bacillario-phyceae, Cyanophyceae	0.5-4.0	5-30	-	-	-
Hyper-eutrophic	-	>10	-	-	Chlorophyceae, Eugleno-phyceae	-	-	30->5000	500->15000	400-60000
Dystrophic	< 50-500	-	< 50-200	0.1-10	-	1.0-4.0	3-30	<1-10	<1-500	5-200

* Referring to approximately net primary productivity, such as measured by the ¹⁴C method.

TABLE 3. Comparison of Primary Productivity and Related Parameters for the Great Lakes(from: Vollenweider *et al*, 1974).

	Mean Daily Primary Productivity for year		Phytoplankton Biomass x 10 ³ mg.m ⁻³	Chlorophyll <u>a</u> mg m ⁻³	Total P* (mean) µg.L ⁻¹	Trophic Type
	mg C.m ⁻² .day ⁻¹	mg C.m ⁻³ .h ⁻¹				
Lake Ontario						
inshore	119-2003	2.9-25.0	0.8-7.4	2.7-12.0		secondary eutrophic**
offshore	58-1443	1.7-12.4	0.6-9.0	1.8-7.9	21.0	mesotrophic
Lake Erie						
Western basin	30-4760	4.8-146.9	0.8-13.2	3.3-19.3	39.1	hypereutrophic
Central basin	120-1690	5.5-21.4	0.6-6.0	2.5-9.2	19.4	secondary eutrophic
Eastern basin	140-1440	3.2-13.9	1.0-4.2	1.4-5.4	17.2	mesotrophic
Lake Huron						
Saginaw Bay	N.A.	4.1-127.2	1.6-17.3	9.5-27.4	30.9	hypereutrophic
offshore	147-698	2.2-9.9	0.3-1.8	1.4-2.2	5.5	oligotrophic
Lake Michigan						
inshore	67-1567	N.A.	N.A.	1.1-10.3	15.0-40.0	secondary eutrophic
offshore	67-1030	N.A.	N.A.	0.6-3.7	8.0	oligotrophic
Lake Superior	50-260	2.2-8.8	N.A.	0.4-9.7	4.6	oligotrophic

* from: Chapra and Sonzogni (1979)

** due to local pollution

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APPENDIX

Sampling Apparatus

a) Water Samplers

Sampling of the water column at discrete depths for physical, chemical and biological parameters can be accomplished with any of a number of commercially available samplers, as long, as they are of sufficient volume and easily deployed and maintained. On the Great Lakes, Nansen, Niskin, Kemmerer and Van Dorn bottles are commonly used and are available in a variety of sizes/volumes. The first two, because of their weight, require a winch. The Van Dorn sampler is particularly amenable to sampling of discrete water strata because of its horizontal orientation during sampling, whereas the other three samplers, because of their vertical orientation, will integrate a water stratum equal to their length. If a power source is available on the vessel, a deck-based or submersible pump system with attached hose allows rapid sampling of many depths at discrete intervals. In addition, the sample flow from this system can be split and connected to onboard measuring systems, such as fluorometers or turbidimeters, equipped with flow-through cells.

b) Instrumentation and Field Measurements

The ability to perform onboard measurements is dependent on vessel size and electrical power availability. Lack of the latter may make it difficult to utilize electrically-powered instruments. However, some instruments such as pH meters, turbidimeters, temperature/dissolved oxygen meters and solarimeters are available with built-in battery packs. This is particularly important for such parameters as pH, alkalinity and inorganic carbon which are preferably measured in the field (i.e. as soon as possible after collection) due to their changeable nature with time. Alkalinity and inorganic carbon can be determined titrimetrically onboard.

Also, if the oxygen light - and dark-bottle method is used for primary productivity determinations, oxygen should be determined in the field titrimetrically (by the Winkler method).

Primary productivity samples using the ^{14}C -bicarbonate tracer method should be filtered in the field, as should water samples taken for chlorophyll analysis. A filtration manifold with an electrical vacuum pump is helpful for this, particularly if the sample number is large. However, a hand-operated vacuum pump can be used instead.