3 DESIGNING AN OBSERVING SYSTEM FOR EARLY DETECTION OF HARMFUL ALGAL BLOOMS

Bengt Karlson[1], Clarissa R. Anderson[2], Kathryn J. Coyne[3], Kevin G. Sellner[4], and Donald M. Anderson[5]

[1]Swedish Meteorological and Hydrological Institute, Gothenberg, Sweden
[2]University of California, Santa Cruz, Santa Cruz, CA USA
[3]University of Delaware, Lewes, DE USA
[4]Chesapeake Research Consortium, Edgewater, MD USA
[5]Woods Hole Oceanographic Institution, Woods Hole, MA USA

3.1 Introduction 89
3.2 Designing an observation system 90
3.3 Background information 90
3.3.1 Characterizing the physical and chemical environment 91
3.3.2 Characterizing phytoplankton community composition 91
3.4 Identifying existing infrastructure 93
3.5 Sampling methods 93
3.5.1 Sampling from shore or from vessels 93
3.5.1.1 Fixation procedures for plankton samples 96
3.5.2 Water transparency – Turbidity tubes and Secchi discs 97
3.5.3 Chlorophyll-*a* and other photosynthetic pigment 98
3.5.3.1 *In vivo* and *in situ* chlorophyll fluorescence 98
3.5.4 Automated water sampling 99
3.5.5 Sampling using fixed platforms 100
3.5.6 Ships of opportunity and Ferrybox systems 101
3.5.7 Flow-through systems on land or on buoys 102
3.5.8 In-water optical instrumentation for detecting HABs 102
3.6 Identification and enumeration of HAB organisms 102
3.6.1 Essential information for identification phytoplankton 103
3.6.1.1 Books for identification of harmful algae and phytoplankton 103
3.6.1.2 Web sites with information on harmful algae and phytoplankton 104
3.6.2 Light microscopy 104
3.6.3 Fluorescence microscopy 105
3.6.4 Electron microscopy 105
3.6.5 Imaging flow cytometry 106
3.6.6 Molecular techniques 107
3.7 Satellite remote sensing 108
3.8 Transport and delivery of harmful algal blooms 108
3.8.1 Empirical and numerical models 108
3.8.2 High-resolution circulation models to resolve flow near intakes 109
3.8.3 An example of a regional HAB forecast system 110
3.9 Distributing warnings and information 110
3.10 Data storage and distribution 112
3.11 Facilities, equipment, and personnel 112
3.12 Summary 114
3.13 References 115

3.1 INTRODUCTION

Harmful algal blooms (HABs) are a serious and growing threat to many desalination plants. It is therefore important to limit the impact from HABs by preventing blooms from reaching seawater reverse osmosis (SWRO) plants in the first place, while also mitigating their effects through pretreatment and other actions within the plant once intake has occurred.

In this chapter, traditional and emerging technologies in the field of HAB detection and monitoring are summarized. Also advice on designing "observing systems" for early detection or characterization of algal blooms is provided. These systems will vary dramatically in terms of the number of parameters to be measured, the number of stations, frequency of sampling and instruments used - all determined by desalination plant budgets and personnel skills, the nature of the HAB threat for a given plant or region, and other such considerations. An observing system might be as simple as visual observations of the color or nature of the intake water, or as complex as a moored array of autonomous sensors outside the plant, or weekly surveys from small vessels to determine what algal species and blooms are in the intake area or surrounding waters, and thus likely to impact the plant.

There are a number of factors that complicate the design of an observing system. One is the diversity of HAB species. Potentially harmful phytoplankton are found in many groups (mainly eukaryotes) such as dinoflagellates, raphidophytes, diatoms, euglenophytes, cryptophytes, haptophytes, pelagophytes, and chlorophytes (see Chapter 1), but prokaryotes, (cyanobacteria) are also a concern. While dinoflagellates comprise the majority of toxic HAB species in the marine environment where desalination plants are located, many of the toxic species that pose a threat to drinking water supply in fresh- or brackish-water systems are cyanobacteria.

A second factor is that phytoplankton distribution in the sea is not uniform vertically or horizontally in space or in time. This is termed "patchiness" and results from the interaction between physical and biological processes. Examples are presented later in this chapter. The simultaneous use of multiple monitoring methods is therefore often necessary to characterize the species composition and extent of blooms, but even then, a full picture of the distribution of a HAB may not be achievable.

3.2 DESIGNING AN OBSERVATION SYSTEM

In the context of providing observations of the water and plankton that can guide desalination operations and plant siting, a HAB observing system can be very informative in many locations. The main goal of such a system is to provide information for actions (rapid response) to avoid or minimize operational disruptions and damage to desalination plants. Prior to the design and construction of a plant, a HAB observing system can be used to gather information on the nature and function of the regional oceanographic system, its role in HAB occurrence, and the historical patterns and extent of HAB events. This can be used to provide input on where to place, and how to design, water intake systems, as well as highlighting the types of pretreatment equipment that might be needed in order to minimize damage from HABs during operation. Figures 3.1 – 3.3 show some features to be considered in this regard.

3.3 BACKGROUND INFORMATION

To design an observation system, it is necessary to gather background information on the occurrence of harmful algae in relation to local physical and chemical conditions. Existing information should be used when possible (likely from monitoring programs run by government, industry, or academic institutions), but often a pilot study may be required. A physical oceanographic model describing regional hydrodynamics surrounding the desalination facility would be a valued asset to any observing system. These are often developed by universities and other academic institutes, as well as government agencies, and are sometimes utilized in studies of brine dispersion and recirculation during plant design. Models in HAB monitoring and management are discussed in section 3.8.1.

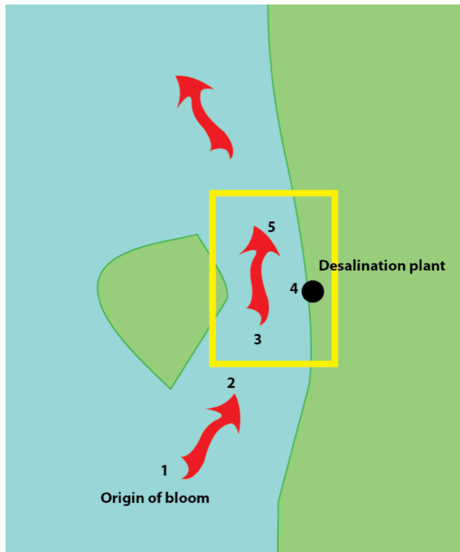

Figure 3.1. A schematic drawing illustrating the effect of local currents on the selection of sampling locations. In the yellow rectangle, tides dominate the currents on a 24-hour cycle. Water from locations 3, 4, and 5 all pass location 4. The northward current dominates over longer time scales, and since blooms develop in the South, observations at station 1 may be of great importance.

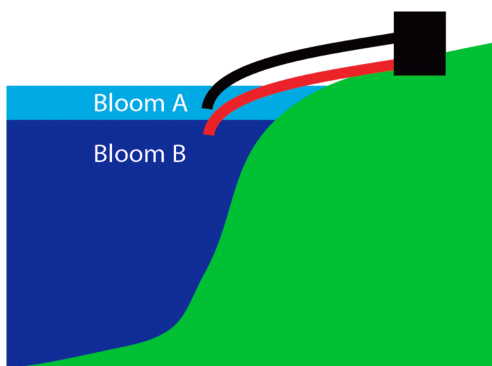

Figure 3.2. This schematic shows a strongly stratified water column. Water intakes at two different depths are depicted for the desalination plant. Bloom A in near surface water is reaching the plant through the black intake while subsurface blooms (bloom B) can be taken in through the red, deeper intake.

3.3.1 Characterizing the physical and chemical environment

The geographic position and depth of the water inlet of a desalination plant is one factor that will influence the design of the observing system. Local and larger scale current conditions as well as seasonal fluctuations in water column stratification are important physical parameters to assess or monitor. Questions about the physical and chemical environment that should be considered during the design of an observing system include: do the HABs develop locally or do currents transport them to the area (Figure 3.1)? What are the dominant sources of nutrients available for local algal growth – pollution discharges from nearby population centers for example, or natural sources through the circulation of water masses? And how dynamic is the hydrographic system outside the plant – are water masses and their associated blooms moving rapidly along the coast, or is it a more gradual and constant flow? These and other example questions that need to be answered before an observing system is designed are listed in Table 3.1.

Distributions of currents and circulation patterns in the area of the facility should be determined, and if possible, models used to estimate particle delivery to the plant's intake under normal weather patterns and during/following major meteorological events. Bottom sediment type and depths relative to the facility intake location should also be known to minimize bottom-derived sediment intake but also to assess the potential for blooms derived from resuspension of HAB cysts or spores up-current of the plant.

3.3.2 Characterizing phytoplankton community composition

The phytoplankton community in a given region often consists of hundreds of different species, with that community composition changing through time. Only a fraction of these species are potentially harmful. When a HAB organism reaches high biomass levels and becomes the main species present, it can cause problems due to that biomass, but for some species, a relatively low number of cells can still present operational concerns for a desalination plant if toxins deleterious to health are produced. The amount of toxin that might be present in blooms of different sizes is discussed in Chapter 1, and Chapter 10 evaluates the risk associated with the small amounts of residual toxins that might be present after desalination has occurred. HAB

Table 3.1. Questions about the physical and chemical environment that should be considered when designing a HAB observation system.

Questions	**Data needed**
What distance may HABs be transported during a tidal cycle?	*Local and regional data on current speed and direction at depths where HABs occur.*
Are there currents transporting HABs to the location of the water intake of the desalination plant?	*Data from in situ instruments, e.g. an ADCP (Acoustic Doppler Current Profiler). Simulations from a physical oceanographic model developed and verified for the area are also useful.*
Is the water stratified during the whole year or part of the year?	*Depth profiles of salinity and temperature together with measurements of chlorophyll fluorescence, a proxy for phytoplankton biomass.* Measurements are usually made from research vessels using a CTD, an instrument used to determine depth profiles of conductivity and temperature together with other parameters. Conductivity and temperature are used to calculate salinity (calculated using the practical salinity scale). Also moored depth profiling platforms are available providing information on subsurface algal blooms in near real time.
Are there short-term events that may favor HAB-development?	*Background data on air temperature, precipitation, river flow, wind speed and direction, cloud cover.* A meteorological and hydrological institute may provide the data.
Are there nutrients supporting HAB growth available during the whole year or part of the year?	*Data on concentrations of inorganic nutrients, i.e. phosphate, silicate, nitrate and ammonium from the surface mixed layer.* Water sampling and chemical analysis in a laboratory on ship or on land should be carried out by laboratories specialized in saline samples. The samples do not preserve well and should be analyzed within a few hours after collection or frozen for later analysis. Also, riverine input of nutrients is important.

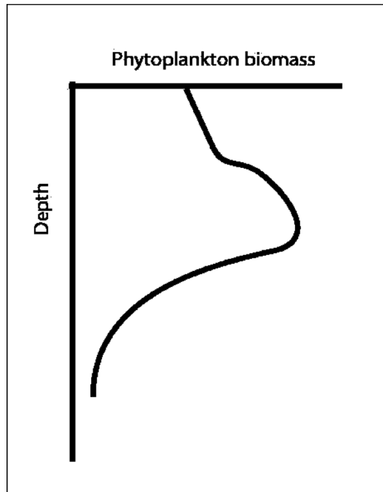

Figure 3.3. Typical vertical distribution of algae in the water column. The graph illustrates a common vertical distribution with a sub-surface maximum of chlorophyll or phytoplankton biomass (often 10-20 m deep), which may move vertically to the surface and back (termed migration), depending on the day-night cycle. Water sampling and automated observations should account for this heterogeneity.

toxins are sufficiently well removed that toxins in treated water should not be a concern, but they are an operational issue that should be identified and monitored when such a threat is present.

Since many of the non-harmful and harmful algal species are similar in appearance (morphology) it is necessary to have trained personnel identifying the organisms. Semi-automated systems exist, as described below, but staff or outside experts with knowledge of the instruments and phytoplankton taxonomy are needed to set up the systems and evaluate the results. Other information on the local phytoplankton community can often be obtained from existing monitoring programs in a region, perhaps conducted by a state, province, or municipality. Some of the questions and required data relative to phytoplankton population dynamics are listed in Table 3.2.

3.4 IDENTIFYING EXISTING INFRASTRUCTURE

In many cases, existing sampling infrastructure may be used when setting up a HAB observation system. An oceanographic laboratory with facilities for working with phytoplankton nearby the sampling sites is ideal. If there are already on-going marine monitoring programs, they can be adapted to undertake HAB work. Ships of opportunity, e.g. ferries, with a stable timetable, may be used for automated sampling. It is useful to investigate if there are buoys or permanent structures (e.g. pilings) in the area that can be used for mounting automated sensors and water sampling devices. Although existing buoys may not be available for mounting sensors and water sampling devices, colocation of HAB buoys is useful to avoid problems with fishing and traffic of merchant vessels. These approaches are described in more detail below.

3.5 SAMPLING METHODS

3.5.1 Sampling from shore or from vessels

The most basic sampling procedure for observing algal bloom species that cause problems for desalinations plants is to collect a water sample and analyze it with a microscope. This can complement online, continuous analyses, such as chlorophyll fluorescence, discussed below. The recommended frequency for sampling is once per week, as phytoplankton can grow and accumulate very rapidly (some can double their cell concentrations in a day or less). If resources are limited, bi-weekly sampling can provide reasonable protection. The number of sampling locations and the depth of sampling will depend on the local conditions and available funding and staff. One approach is to sample at the seawater intake, but that gives little advance notice or information about the geographic extent of the bloom. At the other extreme, ship-based surveys can be conducted, covering an area several km or more from the intake. During a HAB event, increased sampling frequency and spatial coverage can be very informative, as it can reveal the spatial extent of a bloom, its vertical distribution, and other factors that can help the plant anticipate future impacts and potential treatments.

Nearshore samples can be taken from land, but the sampling point must be before the waves break. A jetty, dock, or other extended feature can be used for that purpose. Both dedicated ships, i.e. research vessels, and other boats may be used. Ships should be of a size suitable for

work in rough weather and have room for both the crew and technical personnel. For this type of nearshore sampling, the ships should be fitted with CTD (conductivity, temperature, depth profilers) or other such devices to measure water column structure. The CTD should include a sensor for chlorophyll fluorescence if possible (see Table 3.1). A laboratory area on the ship is important for filtering of samples and other activities.

Table 3.2. Questions regarding HAB species and bloom dynamics that need to be addressed when designing an observation system. Some of the information is likely available in other institutions and should be explored prior to initiating the observing system.

Question	Data needed
Which HAB species occur in the area? What is the temporal and spatial distribution of HAB species? Do HABs develop upstream of the desalination plant? During what time of year do the HABs occur? What is the background composition of the phytoplankton community in the area?	*Abundance and distribution of phytoplankton, in general, and of HAB species, in particular. Frequent (e.g. weekly) water sampling and microscopy-based analyses of the samples by skilled personnel. In addition, automated analyses using imaging flow cytometry and/or genetic methods can be useful. Surveys including water sampling at multiple locations are needed to document the spatial distribution of HAB species. Data on current speed and direction support the design of the surveys.*
What are the ecological and bloom dynamics of the local HAB species? Can the HAB species regulate their position in the water column? What is the growth rate of the HAB species?	*Long-term observations and experimental work are needed to characterize the ecology of HAB species.* This may be outside the scope of the observation system, but some observations (e.g., vertical swimming behavior) are relatively simple to make, and are important for minimizing HAB intake.
Do the HAB-species produce resting stages? What is the distribution of these?	*Resting stages (cysts or spores) should be documented from observations or the scientific literature. If a common HAB-organism in the region produces resting stages, a distributional survey may be useful, as this can guide understanding of the timing and location of blooms.*
Do the HAB species produce toxins that may cause health problems for humans?	*Toxins produced by the species in the area.* Field samples of phytoplankton should be analyzed for toxin content using methods described in Chapter 2. Once the local HAB species are identified, known toxin profiles are likely available in the scientific literature.
Are there local nutrients to support HABs?	*Concentrations of inorganic nutrients* (see Table 1). This information can help explain the frequency and size of HABs in the area.

Water samples should be collected for laboratory analysis of phytoplankton and chlorophyll - *a*, and if possible also for inorganic nutrients, oxygen, and other parameters. In waters beyond the intake area, the focus should be on the surface (0 - 1m), mixed layer, as described in Chapter 1. Fixed depth sampling (e.g. 0, 10, 20 m, and a near sea floor sample,) can be informative, but this will depend on the local conditions, and whether there is a need for that degree of vertical resolution. Otherwise, a surface sample is all that is needed. If a more comprehensive measurement is needed that accounts for vertically migrating cells, water samples can be collected from individual depths using Niskin bottles, and these can then be pooled for later phytoplankton analyses, with one count to characterize the entire water column or mixed layer. Integrated hose sampling (see below) is another useful approach that is ideal for keeping the number of samples low while sampling the surface mixed layer. Where possible, the depth of the maximum chlorophyll fluorescence (typically determined with a vertical profiling instrument) should be sampled directly for phytoplankton analysis.

Water sampling devices are needed, regardless of the platform from which the samples are taken. Bucket samples at the very surface of the water can be used, but also can sometimes be misleading, so ideally, a surface sample should be collected 1 m or so below the actual surface using a Niskin-style bottle (Figure 3.4). There are simpler sampling devices like the Ruttner sampler and advanced types like the GoFlo bottles. The Niskin and GoFlo samplers may be mounted on special racks called rosette samplers to facilitate sampling at multiple depths on a single cast (i.e. a lowering of the bottle and associated instruments on a cable) from the ship. These bottles are cocked open during descent, and are commonly released by either a weight that is dropped down the line or wire once the desired depth is reached, or by a computer when the bottles are mounted on a rosette (Figure 3.4).

Figure 3.4. Water sampling devices. Left: Individual Niskin-type bottles mounted in a rosette for sampling at multiple depths on a single cast; right: a water sampling device of the Ruttner type. Photo: B. Karlson.

Since phytoplankton are often not distributed uniformly in the water column (see Chapter 1), hoses can be used to sample the mixed layer at the surface of the water column, e.g. 0-10 m. A 10 m-long segment of hose or silicone tubing (Figure 3.5) can be lowered through the water with a weight on the bottom end. Sometimes a valve can be attached to the top end. When no valve is present, a string or a line attached to the weight can then be pulled to the surface, being careful that water is not lost during this process. By removing the bottom end

of the hose from the water first, no sample is lost, and the contents can then be emptied into a bucket. If a valve is used, it needs to be closed once the hose has been lowered, as this will help to retain the sample on retrieval. If need be, segments of hose can be added or subtracted to give the appropriate depth for sampling.

Figure 3.5. A hose used for phytoplankton sampling. The valves are open when lowering the hose into the water. The top valve is closed before lifting the tube out of the water. Photo: B. Karlson.

Figure 3.6 A plankton net, used to collect large amounts of biomass. Note that these types of samples are not quantitative.

Plankton nets (Figure 3.6) can also be used to sample the phytoplankton community, but this type of collection is not quantitative. It is a good way to collect a large amount of biomass to see if HAB species are present, even if they are at low cell concentrations. It could be used to collect sufficient material for toxin analysis, for example. For general surveys, plankton nets with 20 - 25 µm mesh diameter are commonly employed. If smaller species are to be monitored, a mesh size from 5 to 10 µm should be used to ensure that both nanoplankton (2-20 µm) and microplankton (20-200 µm) are sampled. Picoplankton (0.2 – 2 µm) are too small to be collected by a net. The net is towed vertically up and down through the water column and the material contained in the end container is poured into a sampling bottle. The net can also be trailed alongside or behind a slowly moving boat to collect surface plankton. The planktonic material collected in the cup at the bottom of the net should not be preserved, and should be kept in the cold and dark until analyzed. This material can be concentrated further using a filter and extracted for toxin analysis. Examination of a live sample facilitates identification of species, since it is often easier to make a species identification on a cell that is swimming or that has its normal pigmentation.

Equipment such as water samplers, hoses, and sample bottles should be rinsed in fresh water and dried before storage for future use. Drying should be rapid to prevent unwanted algal growth within the hoses and tubes. Plankton nets should be rinsed thoroughly with fresh water to remove all plankton cells that may be attached to the net. At the same time, the nets should be checked to ensure there are no tears or holes. Plankton nets should be hung up to dry in an area protected from direct sunlight, and sharp or pointed objects. Plankton nets should be washed regularly in soapy water, at least once a year. They should be soaked for one day and then rinsed in abundant fresh water. After rinsing, they should be placed in fresh water for one day and then dried and stored.

3.5.1.1 Fixation procedures for plankton samples

There are a number of different fixation methods for phytoplankton, but for routine monitoring programs, Lugol's is the preferred preservative. The recipe for the acidic form is given here, but note that if the fluorescent dye called Calcofluor is to be used to delineate the thecal plates of some dinoflagellates (a very useful tool for species identification: Fritz and Treimer 1985; Edler and Elbrächter 2010; Andersen 2010), neutral Lugol's is needed.

Acid Lugol's is made by dissolving 100 g potassium iodide (KI) in 1L of distilled water, then 50 g crystalline iodine (I_2) is dissolved in this solution followed by the addition of 100 mL of glacial acetic acid. This produces about a 3% solution. For neutral Lugol's, the acid is omitted. Lugol's should be stored in the dark or in a brown bottle as the iodine is light sensitive and will degrade. It should also be stored with a tight fitting lid and kept away from live sample areas (e.g. the general culture environment).

For cultures, add 1 drop of 3 % Lugol's solution to 1 mL of a culture, and for field samples, 10 drops per 200 mL of sample or until the color of weak tea. Overuse of Lugol's will cause some delicate flagellate species to over stain, lose flagella, or break up entirely.

3.5.2 Water transparency – Turbidity tubes and Secchi discs

An algal bloom with a high biomass decreases water transparency. Perhaps the simplest way to measure turbidity is using a turbidity tube. This is a tall glass or plastic cylinder with a white or black and white disc at the bottom, and is useful when a desalination plant does not have easy access to a dock or small boat, or if waters are shallow. The tubes are available commercially or can be easily constructed from common laboratory supplies. Water is poured into the tube until the disc at the bottom is no longer visible. For turbidity tubes which have a turbidity scale marked on the side, read the number on the nearest line to the water level. This is the turbidity of the water. If the tube does not have a scale marked, measure the distance from the bottom of the tube to the water level with a tape measure and look up or calculate the turbidity o f the water sample using the instructions provided with the tube.

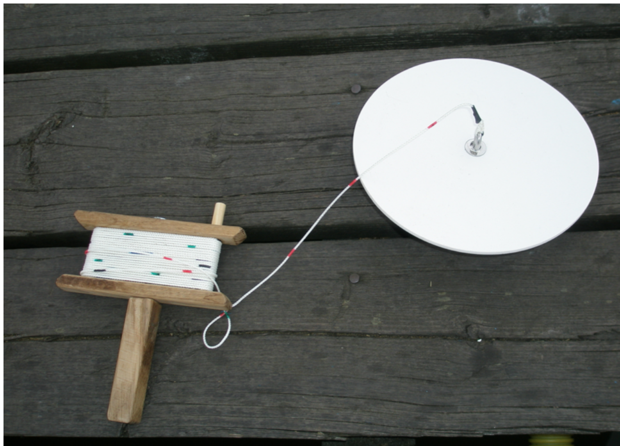

Figure 3.7. A Secchi disc is used to measure water transparency. Photo: B. Karlson.

A related way to measure water transparency and detect blooms is to use a Secchi disc (Figure 3.7). This can be purchased or made by hand. A weighted, white, circular disc, usually 30 cm in diameter, is lowered from a boat or a dock using a thin rope with markings every meter or every half meter until the disc is not visible. Then the disc is raised until it is barely visible. The distance from the sea surface to the disc is called the Secchi depth. It is important to measure the Secchi depth on the side of the boat or the dock that is in the shadow or has the least sun glint. From small boats it is recommended to use an aquascope to minimize effects of reflections. Some scientists working in freshwater prefer the disc be divided into quarters painted alternately black and white. This is not a standard Secchi disc and should be avoided, at least in the sea. It is important to carry out the Secchi depth measurements in a consistent way, e.g. carrying out the measurements at certain time of day, and to collect water samples and net samples at the same time. The amount of suspended particles from sediments influences water transparency making the intrepretation of the Secchi depth difficult during and after high wind events and close to river mouths. The Secchi depth is related to the attenuation coefficient which may be calculated if the light field is measured at several depths in the water column. This may be carried out e.g., using a light meter mounted on a CTD. A reference light meter mounted in air is also needed.

3.5.3 Chlorophyll-*a* and other photosynthetic pigment

Chlorophyll-*a* (chl *a*) is the main photosynthetic pigment in most algae, and therefore it can be used as a proxy for phytoplankton biomass. Since chl *a* content is not a constant fraction of phytoplankton biomass, this proxy must be used with caution. Light history and nutrient conditions and other factors may influence the chl *a* content of microalgae.

Chl *a* is often estimated using water sampling and subsequent filtering and extraction of the pigment, which is measured using a spectrophotometer or a laboratory fluorometer. The most common way to separate phytoplankton cells from seawater is to filter the seawater sample to concentrate all the particles. The filters are then soaked in a solvent (typically 90% acetone) that will extract the pigments from the cells. This extract can then be measured in a fluorometer (to detect chlorophyll fluorescence) or a spectrophotometer (to detect light absorbance by chlorophyll). Details of the fluorometric method can be found in Welschmeyer (1994).

A more exact method for use on water samples is High Performance Liquid Chromatography (HPLC), which separates the different photosynthetic pigments before they are quantified. HPLC is considered by many to be the new standard for chl *a* analysis. HPLC also gives information on pigments such as chl b, chl c_1, c_2, c_3, carotenoids and other accessory pigments. Some of these pigments are specific for certain phytoplankton groups, e.g. peridinin for most dinoflagellates. Thus HPLC analysis gives what is called chemotaxonomic information on the phytoplankton community. It is, however, unlikely that a desalination plant will have an HPLC available for this type of measurement, so the fluorometric method (above) or *in vivo* fluorescence (below) are recommended.

3.5.3.1 In vivo *and* in situ *chlorophyll fluorescence*

The chlorophyll in live phytoplankton produces red fluorescence when exposed to light, (e.g. sunlight or the blue excitation light in fluorometers). Fluorometers mounted on CTDs and other *in situ* instruments that are lowered through the water column are often called *in situ* fluorometers. These may also be mounted on oceanographic buoys or in Ferrybox systems on ships of opportunity. The fluorescence is calibrated against measurements of chl *a* in samples from the same location, making the fluorescence an easy-to-measure proxy for phytoplankton biomass. Indeed, many desalination plants have fluorometers mounted within their plants, measuring online chlorophyll fluorescence continually. Although this gives no information about the species of algae or the other pigments that are present, it does give an approximate indication of algal biomass. The same is true for *in situ* fluorescence measurements.

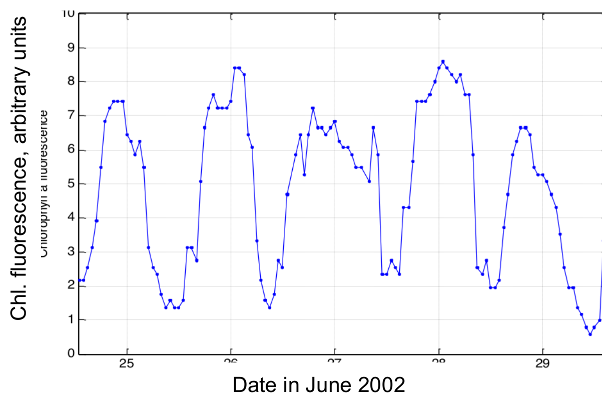

Figure 3.8. Variability of in *vivo* chl *a* fluorescence measured at approximately 2 m depth using the oceanographic buoy Läsö E. in the Kattegat, near the North Sea in 2002. The night to day ratio is about 2-3. Figure: B. Karlson.

Some limitations of the approach should be noted, however. First – the relationship between chl *a* and fluorescence is not constant across all phytoplankton species, nutritional conditions, and times of sampling. Some cells are large, and some small, and thus chl *a* will vary accordingly. Likewise, cells that are nutrient- or light-limited can have lower chl *a* content than the same cells under more

favorable conditions. Furthermore, chl *a* fluorescence is influenced by the light history of the organisms. The nighttime (nocturnal) to daytime ratio of chl *a* fluorescence of the same phytoplankton community may vary by a factor of 2-3. In Figure 3.8, data on hourly measurements of chl *a* fluorescence at approximately 2 m depth in the Kattegat, adjacent to the North Sea, are presented. Note the low daytime values and the high nocturnal values. It is likely that the same phytoplankton community was present day and night. Since nocturnal data are the most consistent, it is recommended to use only the night time chl *a* fluorescence data for near surface sensors.

Despite all of these limitations and caveats, when *in vivo* fluorescence measurements are high in an area relative to past measurements, this is indicative of a major algal bloom, and thus can be used to guide pretreatment options. Additional information on the identification and abundance of the algal species causing the fluorescence would be even more informative. Methods to obtain that type of data manually or using autonomous instruments are given elsewhere in this chapter.

Figure 3.9. An automated water-sampling device, which is part of a Ferrybox-system in the Baltic Sea. Photo: B. Karlson.

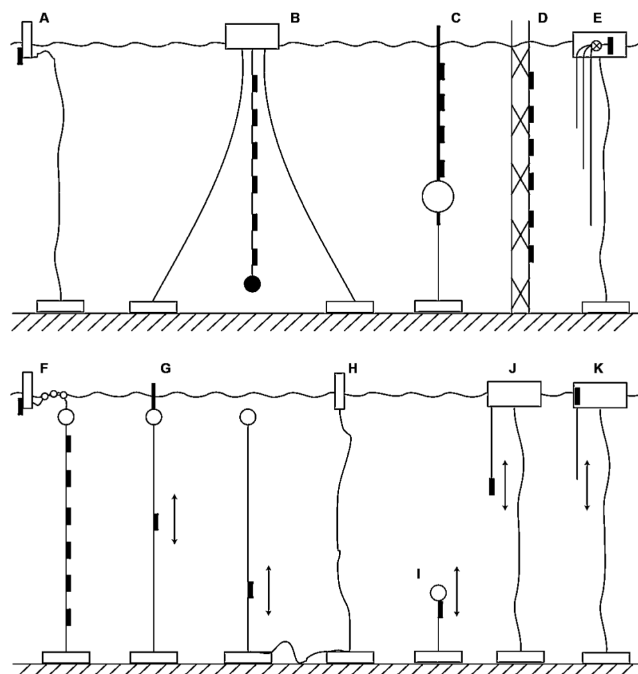

Figure 3.10. Mooring designs. The black rectangles represent sensors. In the depth profiling designs (G-K) and the one with a pump (E) only one sensor package is needed to cover a large depth interval.

3.5.4 Automated water sampling

To achieve cost efficient observations of HAB-organisms, automated sampling may be used. There are commercially available, refrigerated water sampling devices that hold 24 one-liter samples (Figure 3.9). These types of samplers are used in water treatment facilities and also in Ferrybox systems on ships and in flow through systems on land. It is useful to have two water sampling devices at each location; one is used for live and the other for preserved samples. For the latter, preservatives such as Lugols or formalin are put in the bottles such that the organisms are instantly preserved when the sample is added. If Lugol's iodine solution is used as preservative, the sampling device will turn brownish. Sampling may be programmed for certain hours or locations. Another option for automated water sampling is *in situ* systems. At present there are few of these systems available commercially. Samples are collected in plastic bags prefilled with preservative. These devices are used, for example, in a monitoring program using oceanographic buoys in the United Kingdom.

3.5.5 Sampling using fixed platforms

To achieve high-frequency sampling at static locations, oceanographic buoys and other fixed platforms such as pilings, oil platforms, and wind turbines may be used for mounting sensors and automated water-sampling devices. Figure 3.10 shows a schematic of the different types of mooring designs that could be considered. A disadvantage in mounting automated sampling devices on buoys is that the samples need to be brought to the laboratory for analyses. This is useful for research, but may be less useful for near real-time observing systems. If sufficient power is available, e.g. in cabled ocean observatories, advanced instruments such as Imaging FlowCytobot (Sosik and Olson 2007 and Olson and Sosik 2007) or the FlowCam and other automated laboratories may be used to obtain data in near real time. These are discussed in section 3.6.6. Buoys may be serviced at sea, but it is often cost effective to carry out service and calibrations on land and to have two systems, one in operation and one being serviced or ready to be deployed. In Figure 3.11, examples of fixed platforms are presented, and Figure 3.12 shows instruments that can do vertical profiling.

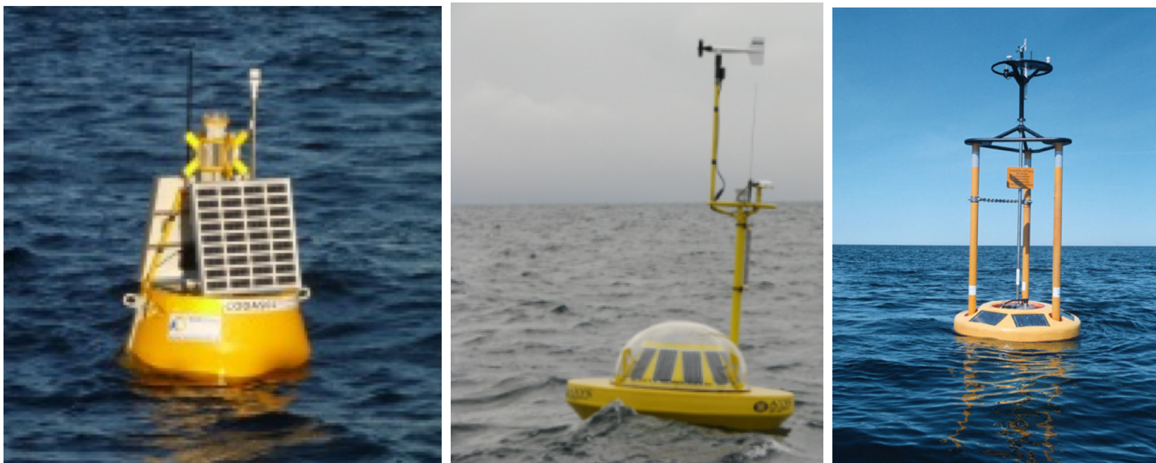

Figure 3.11. Examples of instrumented oceanographic buoys operated by the Swedish Meteorological and Hydrological Institute. The most important parts, i.e. the underwater sensors, are not shown. Left: the coastal Koster fjord buoy in the Skagerrak, Sweden designed by Techworks Marine Ltd. Ireland, middle: the offshore Huvudskär buoy in the Baltic Sea designed by Axys Tecnologies Inc. Canada, and right: the offshore Läsö buoy in the Kattegat, between Sweden and Denmark, designed by Fugro-Oceanor, Norway. Photos: Fredrik Waldh, Henrik Lindh/Per Olsson and Bengt Karlson.

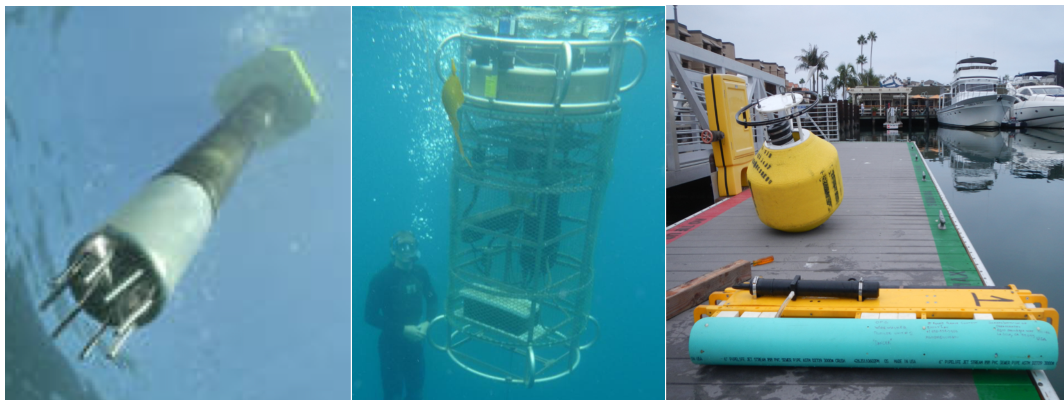

Figure 3.12. Examples of automated depth-profiling instrument platforms. All can be fitted with sensors useful for HAB observations. Left: the ArvorC from NKG, France, (http://www.nke-instrumentation.fr, middle, the Thetis from Wetlabs Inc., USA (http://www.wetlabs.com) and right: the Wirewalker. Photo: R. Kudela).

3.5.6 Ships of opportunity and Ferrybox systems

Ships of opportunity and Ferrybox systems are cost-efficient platforms for collecting information on near-surface HABs, but require a ship owner's willingness to provide free access to their vessel to install and service the instruments. The systems are often mounted on ferries, but are also found on other merchant and research vessels. Ships that have stable timetables and cross an area-of-interest frequently (e.g. every other day) are the most suitable. The owner of the ship should allow two holes (or through-hull fittings) in the ship at 3-4 m depth, one for a seawater inlet and the other for seawater discharge. Inside the ship, a small, but dedicated area for the Ferrybox system is needed to mount a pump that will not damage delicate phytoplankton, a de-bubbling device, an automated cleaning system, sensors, and water sampling devices. Data on chlorophyll fluorescence, turbidity, salinity, temperature, dissolved oxygen, and phycocyanin, are collected continuously every few hundred meters (Figure 3.13). Water samples are collected and archived when the ship passes predefined locations. Ideally, an Internet connection makes it possible to send data in near-real time. Sending data while the ship is in harbor and within reach of low cost wireless communication may be sufficient. A service team collects water samples while the boat is docked in the harbor and transports samples to a laboratory for analysis. During the visit to the ship, sensors are cleaned and other maintenance is conducted. Karlson et al. (2016) describes a Ferrybox system in some detail (Figure 3.14.)

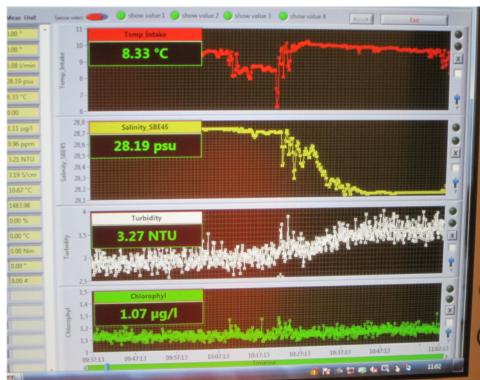

Figure 3.13. Screen shot of a Ferrybox system in operation. Continuous values of temperature, salinity, turbidity and chlorophyll fluorescence are shown. Photo: D.M. Anderson.

Figure 3.14. Components of a Ferrybox system: Left: sensors for conductivity, oxygen, chlorophyll fluorescence (proxy for phytoplankton biomass), phycocyanin fluorescence (proxy for cyanobacteria biomass), and turbidity Photos: Bengt Karlson; right: an example of a fully configured, commercially available Ferrybox system: http://www.4h-jena.de/.

3.5.7 Flow-through systems on land or on buoys

Systems very similar to Ferrybox systems may also be mounted on land or in large buoys. It would be highly informative if this type of sensor and automated sampling device were mounted in the water flow that leads to a desalination plant. The system should be indoors or at least in an environment protected from rain and sea spray. Other options are to mount the system on the dock or on a pier accessible to a service team that would collect samples and maintain the system. It is important to avoid locations where sediments are suspended regularly, e.g. through ship traffic. Seawater should be pumped to the sensors using pumps that do not damage the phytoplankton, particularly if archived samples are to be collected for microscopic species analysis; large peristaltic pumps are commonly used for this. Flow-through systems on land are also useful when connected to imaging flow cytometry instrumentation for automated identification and enumeration of HAB organisms, especially if both power and a fast internet connection are available (further discussion below).

3.5.8 In-water optical instrumentation for detecting HABs

Submersible optical instruments can be deployed on a range of platforms from moorings to autonomous underwater vehicles (AUVs) and provide information on the in situ constituents, from phytoplankton biomass to detrital particles. Obtaining HAB-specific signals is more difficult but could be accomplished with multivariate approaches that combine inherent (e.g. phytoplankton absorption and backscatter) and apparent (e.g. diffuse attenuation coefficient, remote-sensing reflectance) optical properties with information from other sensors, such as temperature, salinity, and nutrients. As with any moored instrument, however, the constant threat of biofouling requires continual maintenance and creative solutions to clearing organic matter that will attach to any sampling device.

3.6 IDENTIFICATION AND ENUMERATION OF HAB ORGANISMS

There are multiple reasons to monitor the species of algae that are in the intake waters for a desalination plant. Knowledge of which species are present makes it possible to anticipate pretreatment or operational strategies. Some species are toxic, so it is important to identify them and to ensure that membranes and other removal processes are functioning properly. To document the safety of the desalinated drinking water, it may be necessary to make toxin measurements in the intake and drinking water when major blooms of toxic HABs are detected. Non-toxic species also need to be documented, as they can cause clogging or fouling at low and high cell densities; some species are prolific producers of organic materials, and others are not. It is thus important to know not only what species and cell concentrations are present in intake waters, but also to have records of what happened in the plant in the past when that species was present. Detailed record keeping of species, concentrations, impacts and treatment strategies should thus be a standard operating procedure. Unfortunately, this is not currently done at many desalination plants, as there is seldom appropriate expertise for identifying and counting algal species, and often, there is no appreciation of the significance and utility of such information at the managerial level. The following sections provide information on this important aspect of algal monitoring.

To identify and to determine the abundance of phytoplankton, cells are typically counted under a microscope or using automated cell counters, described below. It is fairly easy to count cells (see Appendix 4), though identification to the species level can be challenging. As described below, there are a number of online resources, and many HAB or phytoplankton experts throughout the world who can help. The unit for abundance is usually cells per liter (cells/L) or cells per milliliter (cells/mL). To determine the abundance of harmful organisms and the biodiversity of samples, organisms should be identified to the species level if possible.

This is often done for the most abundant 5 or 10 taxa, with other less plentiful species noted, but not counted. Microscopy is the classic method and includes light microscopy, fluorescence microscopy and electron microscopy. The latter is necessary if the identification of smaller cells is needed or if it is otherwise difficult to identify an organism at the species-level. Microscopy and molecular methods for quantitative phytoplankton analyses are described in a UNESCO – IOC Handbook (Karlson et al. 2010).

3.6.1 Essential information for identification phytoplankton

It would be quite useful for a plant operator to have knowledge of the species that are in the intake waters or those surrounding the plant. To be able to correctly identify organisms that cause problems for desalination plants, it is necessary to have access to personnel who know how to identify those organisms. The IOC Harmful Algal Bloom Centre, University of Copenhagen, Denmark, arranges courses in microalgae identification that can train staff. In some cases, local universities can arrange courses on the topic. Identification guides and taxonomic keys are available as books but also web sites provide useful information. A list of these is provided below. Older books may be difficult to find and are not listed.

3.6.1.1 Books for identification of harmful algae and phytoplankton

Bérard-Therriault, L., Poulin, M., Bossé, L. 1999. *Guide d'identification du phytoplancton marin de l'estuaire et du golfe du Saint-Laurent incluant également certains protozoaires.* Canadian Special Publication of Fisheries and Aquatic Sciences No. 128. 387 pp.

Fukuyo, Y., Takano, H., Chikara, M., Matsuoka, K. 1990. *Red Tide Organisms in Japan: An Illustrated Taxonomic Guide*. Uchida Rokakuho Co. Ltd., Tokyo, Japan, 407 pp.

Hallegraeff, G. M. 1991, *Aquaculturists' Guide to Harmful Australian Microalgae.* Fishing Industry Training Board of Tasmania Inc., CSIRO Division of Fisheries, Hobart, Tasmania, Australia, ISBN 0-643-05184-8, 111 pp.

Hoppenrath, M., Elbrachter, M., Drebes, g. 2009. *Marine Phytoplankton. Selected Microphytoplankton Species from the North Sea Around Helgoland and Sylt.*, Kleine Senckenberg-Reihe 49, Stuttgart, Germany, ISBN 978-3-510-61392-2, 264 pp.

Horner, R.A. 2002. *A Taxonomic Guide to Some Common Marine Phytoplankton*, Biopress Limited, Bristol, England, ISBN 0-948737-65-4, 195 pp.

Lassus, P. Chomérat, N., Hess, P., and Nézan, E. 2016. *Toxic and Harmful Microalgae of the World Ocean*. International Society for the Study of Harmful Algae/ Intergovernmental Oceanographic Commission of UNESCO, Denmark (2016). IOC manuals and Guides 68. (Bilingual English/French)

Al-Kandari, M., Al-Yamani, F.Y., Al-Rifaie, K. 2009. *Marine Phytoplankton Atlas of Kuwait's Waters*. Kuwait Institute for Scientific Research, Safat, Kuwait, ISBN 99906-41-24-2, 351 pp. http://www.issha.org/Welcome-to-ISSHA/Web-shop/Toxic-and-Harmful-Microalgae-of-the-World-Ocean

Kraberg, A., Baumann, M., Dürselen, C.D. 2010. *Coastal Phytoplankton: Photo Guide for Northern European Seas*. Verlag Dr. Friedrich Pfeil, München, Germany, ISBN 978-3-89937-113-0, 204 pp.

Larsen, J., Moestrup O. 1989. *Guide to Toxic and Potentially Toxic Marine Algae*. The Fish Inspection Service, Ministry of Fisheries, Copenhagen. 61 pp.

Omura, T., Iwataki, M., Borja, V.M., Takayama, H., Fukyo, Y. 2012. *Marine Phytoplankton of the Western Pacific*. Kouseisha Kouseikaku, Tokyo, 160 pp.

Thomsen, H. A. 1992. *Plankton i de indre danske farvande*. Havforskning fra Miljøstyrelsen, Nr. 11. Copenhagen. http://www.mst.dk/Publikationer/Publikationer/1992/11/87-7810-034-8.htm

Throndsen, J., Hasle, G.R. and Tangen, K. (2007). *Phytoplankton of Norwegian Coastal Waters*, Almater Forlag, 341 pp.

Tomas, C. (Editor) 1997. *Identifying Marine Phytoplankton*. Academic Press, San Diego. 858 pages.

3.6.1.2 Web sites with information on harmful algae and phytoplankton

IOC-UNESCO Taxonomic Reference List of Harmful Micro Algae:

http://www.marinespecies.org/hab

AlgaeBase: http://algaebase.org

World Register of Marine Species (the parts on algae are based on AlgaeBase):

http://marinespecies.org

Nordic Microalgae: http://nordicmicroalgae.org/

Center of Excellence for Dinophyte Taxonomy: http://www.dinophyta.org/

Phytoplankton guide (Louisiana Universities Marine Consortium):

http://phytoplanktonguide.lumcon.edu/

Phyto'pedia: http://www.eos.ubc.ca/research/phytoplankton/

PlanktonNet: http://planktonnet.awi.de/

3.6.2 Light microscopy

The recommended method for enumerating and identifying most HAB organisms is the Utermöhl sedimentation chamber technique (Figure 3.15). This is also the most common method for quantitative analysis of phytoplankton. Step-by-step instructions on how to use the Utermöhl method are found in Edler and Elbrächter (2010). A description is also found in Appendix 4, along with a description of the use of an alternative counting chamber called the Sedgewick Rafter slide. With the Utermöhl method, organisms are concentrated through sedimentation. An inverted microscope with high quality optics is necessary to carry out the analyses (Figure 3.15). Qualified training in phytoplankton identification, taxonomy and systematics is very important to ensure high quality results. A disadvantage with the method is that organisms smaller than about 5-10 µm cannot readily be identified to the species level. Another problem is that a relatively small volume (10-20 mL) is most often analyzed, although 50-100 mL chambers are also routinely used. This means that rare species may be overlooked. This is unlikely to be a problem for monitoring blooms affecting desalination plants, as it will be blooms at high cell

Figure 3.15. The Utermöhl method for settling and counting algae. Left: concentration of samples using sedimentation chambers; right: an inverted microscope. Photos: Å. Edler, B. Karlson.

concentrations that are of primary concern. A useful tool when counting phytoplankton samples and working with the resulting data is the free software Plankton Toolbox available at http://nordicmicroalgae.org/tools (Karlson et al. 2015).

Although it is possible to do basic cell identification and enumeration using simple microscopes, this process will be easier if a microscope fitted with contrast enhancement equipment is available, either phase contrast or differential interference contrast (DIC, often termed Nomarski). Epifluorescence is also useful. Objectives should include 4-5x, 10x, 20x, 40x, and for high-magnification work, 100x. With oculars of 10x this results in a magnification of 40-1000x.

3.6.3 Fluorescence microscopy

Fluorescence microscopy is a useful tool to enumerate organisms that dominate algal blooms, making it possible to differentiate particles that are algae from other organisms or detritus that lack photosynthetic pigments. Figure 3.16 shows the natural colors of autofluorescence in a sample. Fluorescence microscopy is also used with samples treated with chemicals that are used in a diagnostic fashion, e.g. fluorescent RNA-probes (described below), calcofluor to stain the cell wall features of the many dinoflagellates with cellulose cell walls, and DAPI (4',6-diamidino-2-phenylindole) to reveal cell nuclei.

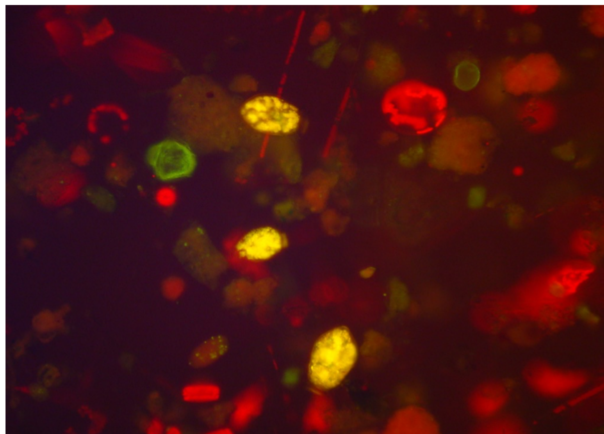

Figure 3.16. Autofluorescence in phytoplankton sample. The red cells contain chlorophyll and the yellow cells phycoerythrin. The green cell is non-photosynthetic. Photo: B. Karlson.

Samples for fluorescence microscopy are most often concentrated by filtering and then examined with inverted or conventional microscopes equipped with specific excitation lamps and filters. A combination of staining with calcofluor and light microscopy with the Utermöhl method is presented in Edler and Elbrächter (2010). Analysis of autotrophic picoplankton (0.2-2 µm) is often carried out using fluorescence microscopy. Autofluorescence from phycobilins in *Synechococcus*-type cyanobacteria facilitates analysis.

3.6.4 Electron microscopy

Electron microscopy is a costly and time-consuming method. It is used to identify many small phytoplankton organisms to the species level, or to visualize small, distinctive features on larger cells (Figure 3.17). Scanning electron microscopy (SEM) and transmission electron microscopy (TEM) should only be used as an occasional complement to light microscopy and imaging flow cytometry.

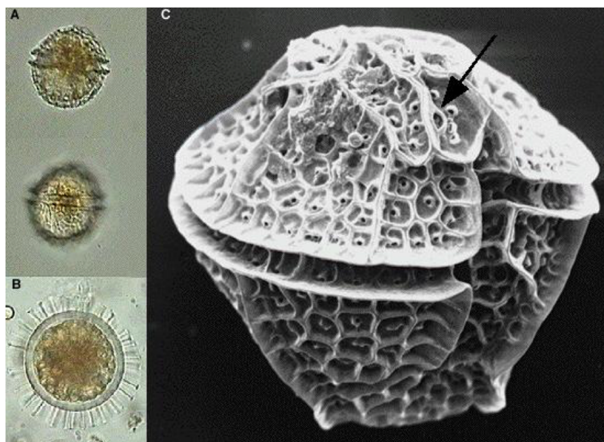

Figure 3.17. Right: a scanning electron micrograph of *Protoceratium reticulatum*. The arrow shows the ventral pore used to help identify this species. To the left, the same species as photographed using light microscopy. A the motile, vegetative stage cell; B) the resting cyst. Photos: M. Kuylenstierna.

3.6.5 Imaging flow cytometry

Flow cytometers are particle counters that were originally developed for counting and differentiating blood cells. Most models use one or a few lasers for creating light that is used for exciting fluorescent particles. In addition, light scattering properties of particles are used to differentiate cells. For phytoplankton research, they were first mainly used for pico- and nanoplankton (0.2-2 and 2-20 µm respectively) and the fluorescent and scattering properties of the algae were used to differentiate the algae to a very rough group level. Later, imaging flow cytometers were developed, now available as *in situ* instruments (Sosik and Olson 2007; Olson and Sosik 2007). In the latter instruments, fluorescence of chlorophyll is commonly used to trigger a camera and all particles that fluoresce, i.e., algal cells, are documented in a digital image. Automated image analysis is used to identify the organisms and to measure size, etc. The software must first be developed ('trained') by experts on the local phytoplankton community, but thereafter the instrument can run autonomously, taking

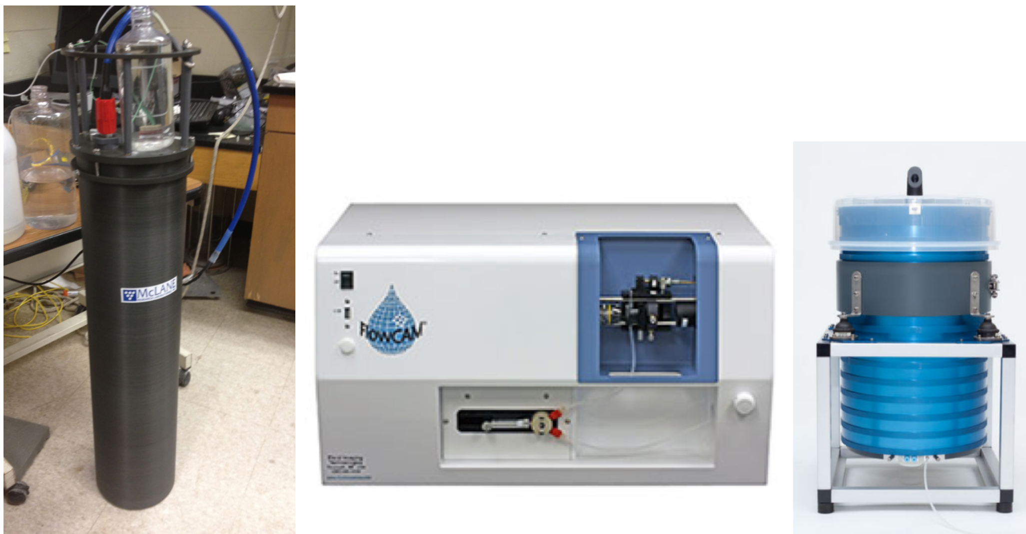

Figure 3.18. Imaging flow cytometers. Left to right: Imaging FlowCytobot (IFCB) from McLane Laboratories, Inc., the FlowCam from FluidImaging Inc., and the CytoSense from CytoBuoy.

thousands of images every minute, and classifying the organisms to genus and even to the species level. When new species are observed, new training sets are developed and added to the software. There are currently at least three imaging flow cytometers available commercially that are useful for phytoplankton analyses (Figure 3.18). Some are available both as desktop and as *in situ* instruments deployable on oceanographic buoys. These sensors could be placed online within a desalination plant to record the abundance and identity of algal species in the intake waters, providing a high-frequency, high-resolution record of the species that can be compared to the plant's operational data to identify problem species, and to guide pretreatment strategies. An example of the output from the Imaging FlowCytobot (IFCB) is shown in Figure 3.19.

Imaging flow cytometers:

FlowCam: http://www.fluidimaging.com/products-particle-vision-pv-series.htm
CytoSense: http://www.cytobuoy.com/
Imaging FlowCytobot: (IFCB): http://www.mclanelabs.com/master_page/product-type/samplers/imaging-flowcytobot

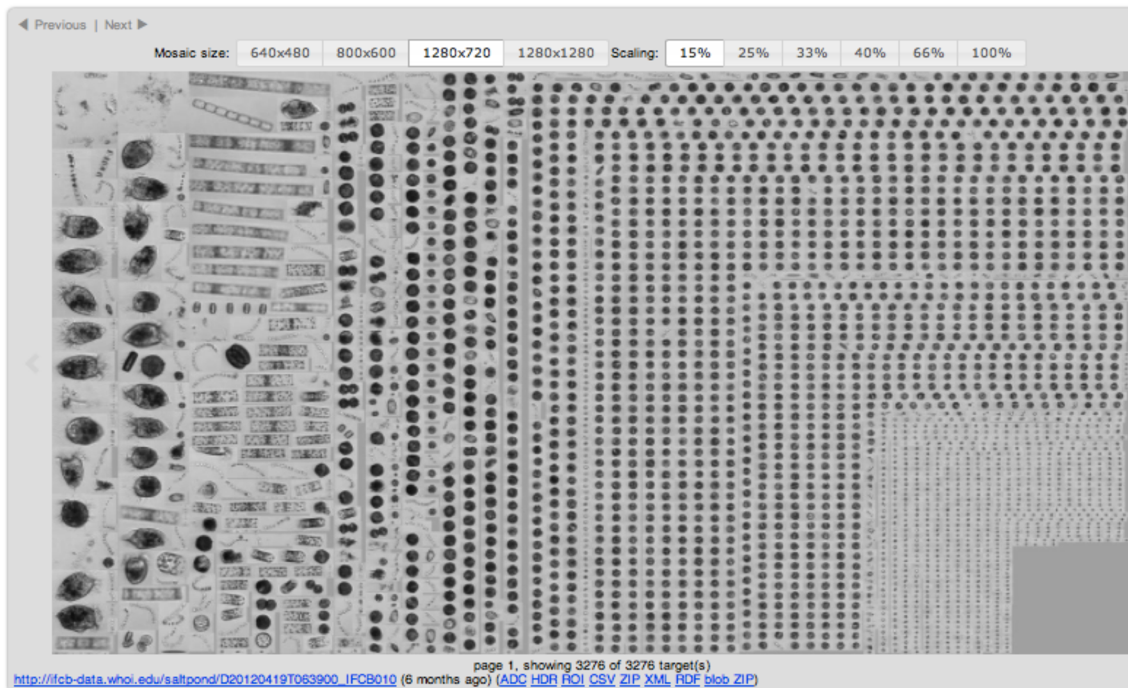

Figure 3.19. Example output from the autonomous IFCB. From left to right, the large cells on the left are microzooplankton grazers, the rectangular cells are diatoms and the round cells are the toxic HAB dinoflagellate *Alexandrium fundyense*. Photo: M. Brosnahan.

3.6.6 Molecular techniques

Advancements in molecular technology offer efficient and powerful alternatives to microscopic methods for detection and enumeration of HAB species in natural assemblages (reviewed in Sellner et al. 2003; Kudela et al. 2010). The application of molecular methods for HAB monitoring is particularly valuable for species that cannot be reliably identified by light microscopy due to small size and/or the lack of distinguishing morphological features (e.g. Coyne et al. 2001), or for sensitive detection of invasive species (e.g. Cary et al. 2014). In addition, fragile HAB species that disintegrate or distort in the presence of chemical fixatives may be underestimated by microscopy, but can be detected and accurately quantified using molecular methods (Doll et al. 2014).

It is unlikely that a desalination plant would have the capabilities to undertake most of the molecular techniques discussed here, but the methods are presented to demonstrate what can be done by outside laboratories, and to help explain the technology that is being incorporated into some of the new sensors and instruments that may be used by desalination plants at some point in the future.

Molecular approaches for detection of harmful algae are based on species-specific differences in nucleic acid (DNA or RNA) sequences. These methods often target the small or large subunit of the ribosomal RNA (rRNA) genes, which are present in high copy number (tens to tens of thousands) in the genomes of all organisms. The rRNA gene sequences contain conserved and variable regions, allowing development of molecular assays for different taxonomic levels of distinction, ranging from strains and species to genera and classes of phytoplankton, or encompassing the entire prokaryotic or eukaryotic communities. The design and *in silico* validation of molecular assays is facilitated by the vast amount of rRNA

sequence information currently available in public databases from a broad range of species (Hugerth et al. 2014).

While most molecular techniques still require collection of water for laboratory analysis, autonomous platforms such as the Environmental Sample Processor (ESP; Figure 3.20) and the Autonomous Microbial Genosensor have been designed to conduct automated molecular assays for rapid *in situ* detection of HAB species (Scholin et al. 2009; Scholin 2010; Yamahara et al. 2015). Two-way wireless communication allows data access and remote control of sampling times. These devices collect particulates by filtration for molecular detection of a range of microbial species, including HABs and pathogens, as well as antibody-based detection of HAB toxins in an enzyme-linked immunosorbent assay (ELISA) format (Doucette et al. 2009). Initially designed to be moored at a fixed location changes in technology have allowed ESP deployment in the deep sea (Ussler et al. 2013) and on gliders (Seegers et al. 2015) for additional flexibility. One important limitation of the ESP technology is that it detects "target" organisms, i.e. those for which molecular probes have been developed and which are incorporated into the ESP assay system. This means that it could be useful in detecting known toxic HAB species, for example, but not for identifying new or unknown species.

Figure 3.20. The Environmental Sample Processor (green canister), an autonomous "laboratory in a can" that can detect and enumerate HAB species and measure toxins. Photo: Woods Hole Oceanographic Institution.

3.7 SATELLITE REMOTE SENSING

In the context of desalination and the development of observing capabilities for HABs, satellite remote sensing has great potential to be of use to operators. A comprehensive overview is provided in Chapter 4.

3.8 TRANSPORT AND DELIVERY OF HARMFUL ALGAL BLOOMS

Desalination plants would benefit greatly from forecasts of algal bloom transport and landfall, but such capabilities typically require numerical models of coastal hydrography. These are typically far beyond the technical or financial resources of many individual plants, but regional approaches to this type of technology are being explored, and thus the fundamentals of such systems are described here.

3.8.1 Empirical and numerical models

Technological advances have expanded capabilities for research and monitoring of HABs, but the blooms will always be under sampled because of the large space and time scales over which they occur. As a result, models are being used to help extrapolate and interpret these sparse observations. These include empirical and numerical models. An example of an innovative and useful empirical model is that of Raine et al. (2010) who developed a model for predicting *Dinophysis* blooms on the southwestern coast of Ireland based on the wind index as a proxy for wind-driven exchange of water and HAB probability onto the shelf. This empirical approach was successful because these blooms occur during summer when offshore water is advected by onshore winds into the highly-stratified nearshore environment (downwelling – see Chapter 1). The model has improved understanding of the dynamics of diarrhetic shellfish poison (DSP) intoxications that greatly impact the shellfish in Bantry Bay.

Hydrodynamic circulation models are commonly used to track and visualize bloom formation and duration as well as to understand the physical processes controlling phytoplankton bloom dynamics. Models with varying levels of sophistication have been developed. Some are purely three-dimensional physical models capable of resolving hydrography, and into which HAB cells can be introduced as passive particles. In the event that a particular HAB can be identified through detection methods discussed above, algorithms can be used to predict its trajectory and map the bloom using Lagrangian Particle Transport (LPT). This can be coupled to either a 2D or 3D circulation model. LPT is widely used in oil spill tracking and studies of fish or shellfish larval transport and is now seeing growing popularity for HAB risk management. LPT can be a powerful tool for estimating the timing and spatial impact of HAB landfall because many blooms originate offshore and are moved into the regions where most intakes to desalination plants would be located (either surface or subsurface). This is the approach used in a HAB forecasting system developed for *Karenia brevis* blooms in the Gulf of Mexico in which HAB forecasts are made twice weekly during bloom events (Stumpf et al. 2009). Blooms are detected using SeaWiFS and MODIS imagery, and bloom transport is then predicted using hydrographic modeling with passive particle transport. Vélo-Suarez et al. (2010) determined the physical processes responsible for the demise of a *Dinophysis acuminata* bloom, illustrating the importance of retention-dispersion patterns driven by the physics of the bay. Another study used a combination of models to track particles and identify the dominant sites of discharge along the Saudi Coast of the Red Sea (Zhan et al. 2015). This had direct implications to blooms as well as sediment transport. In each of these cases, particle transport models provided crucial spatial information about bloom or sediment transport that could potentially serve as an early warning to desalination plants.

The next step in sophistication and complexity is to couple a detailed biological submodel (one that incorporates cyst germination, cell growth, nutrient uptake, mortality and swimming behavior) to a hydrographic model, as has been done for *Alexandrium* dynamics in the Gulf of Maine region in the US (McGillicuddy et al. 2005; He et al. 2008). This level of modeling is species-specific, and not generally appropriate for desalination plants, however, at least at this point in time.

An alternative but practical approach to biophysical modeling blends empirical and dynamic methods to leverage the power of both simple and complex modeling approaches. In California, where seawater desalination plants already exist and are being revitalized or built for the first time, there is a great need to understand toxin production and transport of HABs nearshore. To this end, a HAB forecasting system has been developed to predict domoic acid-producing *Pseudo-nitzschia* blooms from empirical HAB models that are computed routinely in near real-time from satellite ocean color parameters and physical output (salinity and temperature) in a physical oceanographic model (Anderson et al. under review).

3.8.2 High-resolution circulation models to resolve flow near intakes

High-resolution hydrodynamic models for studying physical features ~10-50 km in size, such as eddies, plumes and fronts, are becoming more accessible as technological advances reduce computing times and costs. Many of these models were initially developed to study patterns of fish larval recruitment or runoff close to shore. Models with a degree of physical complexity are useful for determining the initiation and termination of eddies and their movement near intake systems. Additional complexity in the form of coupled biological/ecosystem models provides specific guidance on where nutrient loading is highest in order to better inform the placement of intakes, initial design strategies or early warning of

bloom conditions. Such an approach was demonstrated for the Karkheh Reservoir in Iran (Afshar et al. 2012).

3.8.3 An example of a regional HAB forecast system

Efforts are underway to combine the technologies described above into forecast systems that would be of value to multiple desalination plants within large regions. In a pilot project that is underway at this time, coordinated by the Middle East Desalination Research Center (MEDRC), an observation and forecast system is being developed that can provide plant operators with a broader view of the environment around their plants, allowing them to anticipate algal blooms that are approaching, thereby allowing more adaptive management and informed decision-making (D.M. Anderson, K. Price, unpub. data). This capability is being developed through a pilot-project early warning system that involves: 1) development of satellite remote sensing indices for HABs along the coasts of Oman and the United Arab Emirates; 2) refinement and expansion of a high-resolution numerical model of regional hydrography and circulation; 3) combination of satellite bloom data with the hydrographic model to predict the transport of blooms to the area of desalination intakes; and 4) development of a web-based portal to provide data and forecasts to plant operators and other users. The remote sensing and modeling approach to be taken here is similar to that used in the US to forecast HABS in the Gulf of Mexico and in Lake Erie (Stumpf et al. 2009; Wynne et al. 2011).

For this project, an Arabian Gulf - Sea of Oman atmosphere-ocean forecast system was developed. The system includes 1) a high-resolution weather forecast model (WRF) and 2) the Arabian Gulf - Sea of Oman hydrographic model (named AGSO-FVCOM). WRF is driven by a global weather forecast model. AGSO-FVCOM is a regional ocean model with a computational domain that has been expanded to cover the entire Arabian Gulf - Sea of Oman region.

A Lagrangian tracking model has also been implemented for forecasting HAB trajectories. An example of the forecasting approach and the types of data that can be generated is given in Figure 3.21. Satellite imagery provided by ROPME revealed an algal bloom event in the vicinity of the Barka desalination plant north of Muscat, Oman on November 1, 2015. The imagery was further processed and digitized so that it could be represented as a field of particles. The AGSO-FCVOM model was then run in particle tracking mode to forecast where the bloom might move over the next several days. The model run started on November 1, and produced a series of images (Figure 3.21) extending through 4 November, suggesting that the bloom patches were moving away from the Barka site. The forecasting model successfully predicted the general offshore movement of the bloom patches, as confirmed in subsequent satellite imagery.

This is an example of the manner in which the combination of remote sensing and numerical modeling can be used to forecast algal blooms that might affect desalination plants. The regional pilot project will end in 2016, but may be extended. The concept can be readily applied to many other parts of the world, particularly those where multiple desalination plants are located in relatively close proximity to each other.

3.9 DISTRIBUTING WARNINGS AND INFORMATION

Many plants will operate bloom observation systems independently, and thus may retain the data for their own internal use. Ideally, however, the information should also be broadly distributed since HABs may cover large sea areas, cross national borders, and affect neighboring desalination plants. All data collectors will benefit from receiving data produced by other data collectors. An e-mail listserv or regional HAB web page would facilitate such

information transfer and dialogue. The web site could also contain information of an educational nature, so that the public and others can find information (such as the causes of HABs and summaries of the effectiveness of toxin removal during desalination) to alleviate their concerns. It is of course important to avoid false positives, i.e. warnings that are wrong. An example may be that a HAB is observed near the location of a desalination plant and a warning is issued within the region. However, the HAB is transported away due to shifts in currents. False positives are inevitable and information to operators and the public describing this trajectory and likely diversion would be needed. In addition to updated information on the common web site, yearly reports on the HAB-events in the area should be produced and archived, as such historical information will be very valuable through time. Figure 3.22 shows a schematic of this data collection, analysis, and information flow.

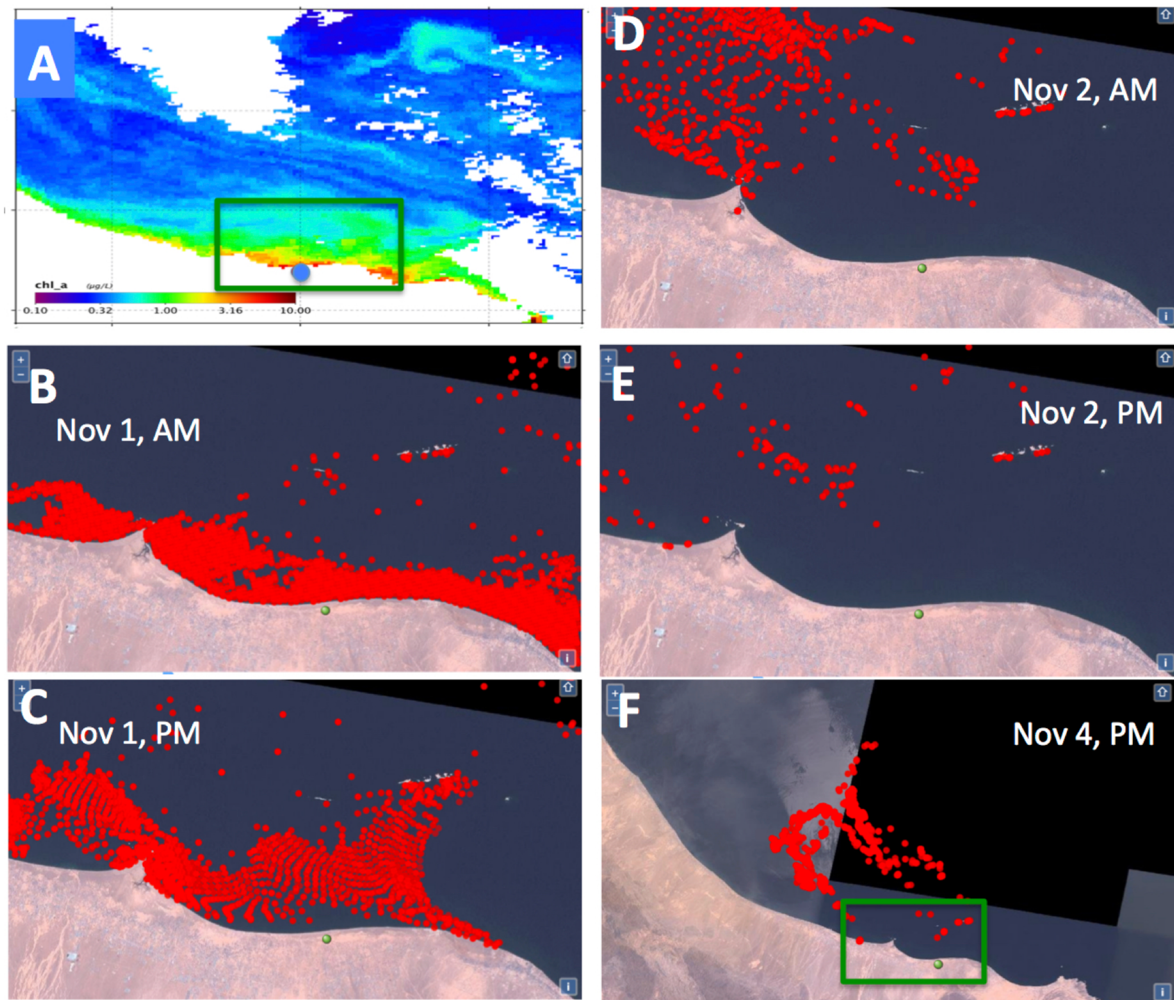

Figure 3.21. Real-time demonstration of the particle tracking module of the AGSO-FVCOM forecast system described above. A dense algal bloom was detected in satellite imagery (A) along the Oman coast near the Barka desalination plant (blue circle) on November 1, 2015 (chlorophyll depicted, with highest concentrations in red and yellow); B: The densest portion of the bloom was digitized and converted to passive particles (red circles) for November 1 AM. The forecast model was run with real-time and forecast weather and current patterns, showing the projected position of the bloom on November 1 PM (C), November 2 AM and PM (D and E), and November 4 (F) as the bloom was dispersed and transported away. Note that the scale of panels A and F are different from those for B-E. The green boxes in A and F depict the areas imaged in B-E. Source: D. M. Anderson, R. Kudela, and R. Ji, unpub. data.

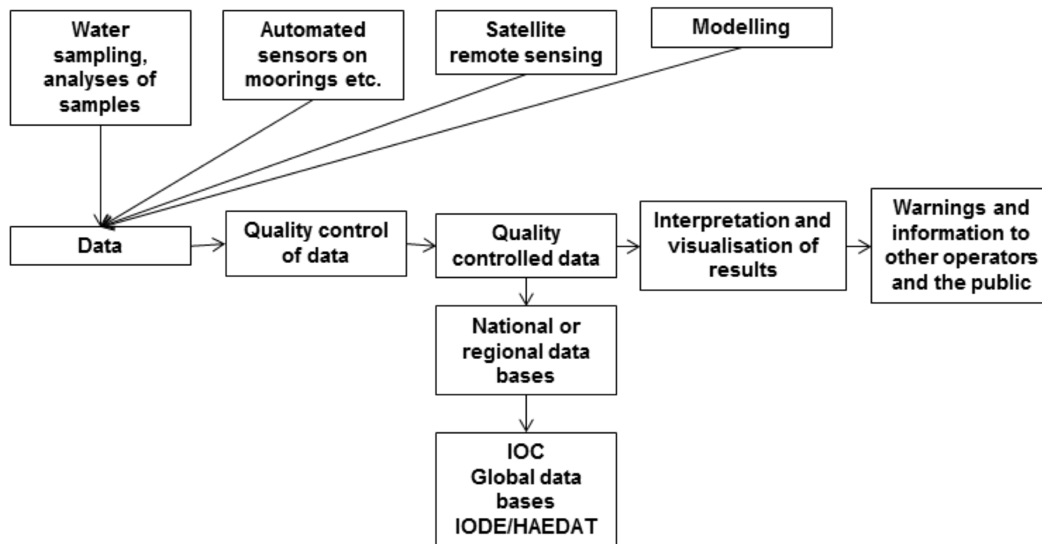

Figure 3.22. Illustration of flow of information from data sources to end users, national and international databases.

3.10 DATA STORAGE AND DISTRIBUTION

All data produced should be stored and their quality controlled. Ideally, the data should be made available freely to the scientific community. The UNESCO-IOC-Global Ocean Observing System (GOOS) and its regional organizations provide an umbrella-organization for this. IOC is the Intergovernmental Oceanographic Commission; it supports the Harmful Algal Event Database (http://haedat.iode.org) and the International Oceanographic Data and Information Exchange (IODE, http://iode.org). ICES (www.ices.dk) provides a system for handling quantitative physical, chemical, and biological data (including phytoplankton) for the North Atlantic. Similar systems exist or are being set up for other areas.

3.11 FACILITIES, EQUIPMENT, AND PERSONNEL

Facilities and equipment needed for a HAB-observation system are listed in Table 3.3, with some suggested priorities. However, facilities and priorities will vary dramatically among plants, depending upon available resources and the magnitude of the perceived HAB threat.

The number of personnel involved in HAB observing efforts will vary dramatically among desalination plants, as this once again will be determined by the nature of the HAB threat as well as the funding resources available. In some cases, existing staff can be assigned additional duties to undertake the sampling and analyses after adequate training. In others, a single individual might be assigned HAB monitoring responsibilities, with the mandate to draw upon additional plant personnel when more hands are needed. Local or international HAB experts (e.g. taxonomy, bloom dynamics, HAB observing systems, remote sensing, toxin analysis) should be identified and their contact details kept in a suitable archive so they can be of assistance in training or during outbreaks.

Table 3.3. An overview of facilities and equipment needed for a HAB observing system. A simple list of priorities is included. Many smaller standard items such as filtering equipment and plankton nets are not listed.

Facility/equipment	Priority 1-3, 1 being highest	Comment
Laboratory	1	
Small research vessel to sample beyond the intake waters	1	This could be chartered as needed.
CTD or multi-parameter sonde with *in situ* fluorometer and turbidity sensors	1	
Microscopes and appropriate cell counting chambers and slides	1	
Manuals and guides for identifying algal species, especially HAB species	1	Many of these are available online, but hard copies near the microscope are still useful.
Water sampling equipment for intake water and broader-scale surveys	1	
Computers and software for data storage, analysis and visualization.	1	These are needed for general data collection and analysis, but could also be used to download and analyze satellite imagery.
Laboratory supplies and simple toxin test kits	1	This would be for toxin screening only. Samples with positive results should be sent to expert laboratories for confirmation. The need for this capability would depend upon the potential for toxic HABs in an area.
Routine access to satellite imagery and trajectory models	1	Could develop in-house remote-sensing analysis capability, or could outsource. HAB forecast models would be very useful, but need regional cooperation.
Flow through system with sensors and water sampling devices, e.g. Ferrybox (for use on research vessels or ships of opportunity).	2	Can provide a steady flow of data over large areas and time if a repeatable transect can be monitored, as with ferry routes.
Oceanographic buoys	2	Buoys situated within and outside the intake area would be highly informative with the appropriate sensors, but costly to maintain.
Imaging Flow Cytometer	2	This would be very useful for continuous, autonomous monitoring of the seawater intake waters, but it is expensive.

Table 3.3. *(Continued)*

Facility/equipment	Priority 1-3, 1 being highest	Comment
Equipment and supplies (or service) for analyzing TEP and other organic molecules (Appendix 3)	2	These are useful measurements, especially when paired with phytoplankton counts in the intake water – helps to specify the species and cell concentrations that disrupt operations or that require specific pretreatments.
Equipment (or a service) for analyses of algal toxins (LC-MS)	3	This should be a low priority unless there is a significant risk for biotoxin contamination of water used for human consumption. These types of analyses are typically outsourced.
Equipment (or a service) for analyses of inorganic nutrients	3	This provides useful information relevant to HAB bloom growth and persistence, but typically are outsourced.
Electron microscope	3	This is a low priority and should be available at local universities.

3.12 SUMMARY

There are numerous techniques to detect HABs in order to react in time to minimize their effects on desalination plants. To accomplish this goal, local and regional monitoring of phytoplankton composition and biomass are needed. To carry out such monitoring, a combination of methods is recommended. A comprehensive monitoring program might include:

1. Long-term funding and staffing commitments
2. Appropriate facilities and equipment in the form of laboratory, shiptime, sampling equipment, microscope, analysis instruments
3. Frequent water sampling near or at the desalination plant. This could include:
    a. Manual sampling in water flowing into the plant or sampling from ships and boats in nearby waters.
    b. Automated sampling in water flowing into a plant or on fixed platforms or ships of opportunity outside the intake.
    c. Analysis of the composition and biomass of the phytoplankton by microscopy and/or imaging flow cytometry. Molecular techniques or SEM may be considered to complement the other methods when high-level species identification is needed.
    d. Continuous online analysis of chlorophyll-*a*, as a proxy for phytoplankton biomass
4. Automated measurements of in-water bio-optical and physical parameters, e.g. chlorophyll fluorescence, temperature and salinity, from fixed platforms such as moorings or by using Ferrybox systems. Biofouling of sensors must be considered when deciding frequency of service.
5. The use of ocean color data from satellite remote sensing. Algorithms for chlorophyll-*a*, a proxy for phytoplankton biomass, are useful and there are also algorithms for detecting certain phytoplankton groups in high biomass algal blooms.
6. The use of physical oceanographic models, verified for the geographic area in focus, to produce forecasts regarding advection of HABs.

7. Quality control and storage of data as well as distribution of data to regional and global data centers.
8. Interpretation of data and visualization of results.
9. Information officers who distribute warnings and information.

If funding is limited, regular water sampling using water bottles and plankton nets with subsequent microscope analysis of the HAB-species should be conducted. In addition, water transparency or turbidity should be measured using a Secchi disk or turbidity tube. This probably requires a part-time technician and linkages to a phytoplankton identification expert. However, such a low cost approach would miss many blooms and be very dependent on the availability of the key staff. In practice such a system could benefit from a larger regional program that detects and forecasts HABs within a region (using remote sensing and numerical modeling, for example), with minimal staffing at each desalination plant to respond to advancing HABs.

3.13 REFERENCES

Afshar, A., Saadatpour, M., and Marino, M. A. 2012. Development of a complex system dynamic eutrophication model: application to Karkheh reservoir. *Environmental Engineering Science* 29, 373–385.

Andersen, P. 2010. Filtering – calcofluor staining – quantitative epifluorescence microscopy for phytoplankton analysis. in: Microscopic and molecular methods for quantitative phytoplankton analysis, Eds. Karlson, B., Cusack, C. and Bresnan, E. *Intergovernmental Oceanographic Commission Manual and Guides* 55, 31-35.

Anderson, C. R., Kudela, R. M., Kahru, M., Chao, Y., Rosenfeld, L. K., Bahr, F. L., Anderson, D. M., and Norris, T. A. Accepted for publication. Towards an operational HAB forecast system for California. *Harmful Algae*.

Bosch van Drakestein, F. G. 2014. Modeling of tides and spreading of saline water in the Arabian Gulf and in the coastal area of Abu Dhabi with a flexible mesh model. Master thesis 67 pp.

Cary, S. C., Coyne, K. J., Rueckert, A., Wood, S. A., Kelly, S., Gemmill, C. E. C., Vieglais, C., and Hicks, B. J. 2014. Development and validation of a quantitative PCR assay for the early detection and monitoring of the invasive diatom *Didymosphenia geminata*. *Harmful Algae* 36, 63–70.

Coyne, K. J., Hutchins, D. A., Hare, C. E., and Cary, S. C. 2001. Assessing temporal and spatial variability in *Pfiesteria piscicida* distributions using molecular probing techniques. *Aquatic Microbial Ecology* 24, 275–285.

Doll, C., Main, C. R., Bianco, C., Coyne, K. J., and Greenfield, D. I. 2014. Comparison of sandwich hybridization assay and quantitative PCR for the quantification of live and preserved cultures of *Heterosigma akashiwo* (Raphidophyceae). *Limnology and Oceanography - Methods* 12, 232–245.

Doucette, G. J., Mikulski, C. M., Jones, K. L., King, K. L., Greenfield, D. I., Marin, R., Jensen, S., Roman, B., Elliott, C. T., and Scholin, C. A. 2009. Remote, subsurface detection of the algal toxin domoic acid onboard the Environmental Sample Processor: assay development and field trials. *Harmful Algae* 8, 880–888.

Edler, L. and Elbrächter, M. 2010. The Utermöhl method for quantitative phytoplankton analysis. in: Microscopic and molecular methods for quantitative phytoplankton analysis, Eds. Karlson, B., Cusack, C., and Bresnan, E. *Intergovernmental Oceanographic Commission Manual and Guides* 55, 13-20.

Fritz, L. and Triemer, R. E. 1985. A rapid simple technique utilizing Calcofluor White M2R for the visualization of dinoflagellate thecal plates. *Journal of Phycology* 21, 662-664.

He, R., McGillicuddy, D. J., Keafer, B. A., and Anderson, D. M. 2008. Historic 2005 toxic bloom of *Alexandrium fundyense* in the western Gulf of Maine: 2. Coupled biophysical numerical modeling. *Journal of Geophysical Research: Oceans* 113, 1978–2012.

Hugerth, L. W., Muller, E. E., Hu, Y. O., Lebrun, L. A., Roume, H., Lundin, D., Wilmes, P., and Andersson, A. F. 2014. Systematic design of 18S rRNA gene primers for determining eukaryotic diversity in microbial consortia. *PLoS One* 9: e95567.

Karlson, B., Cusack, C., and Bresnan, E. (editors) 2010. Microscopic and molecular methods for quantitative phytoplankton analysis. Paris, UNESCO. *IOC Manuals and Guides* 55, 110 pages.
http://hab.ioc-unesco.org/index.php?option=com_oe&task=viewDocumentRecord&docID=5440

Karlson, B., Godhe, A., Cusack, C., and Bresnan, B. 2010. Overview of methods for quantitative phytoplankton analysis in: Microscopic and molecular methods for quantitative phytoplankton analysis, Eds. Karlson, B., Cusack, C., and Bresnan, E. *Intergovernmental Oceanographic Commission Manual and Guides* 55, pp 5-12.

Karlson, B., Andreasson, A., Johansen, M., Mohlin, M., Skjevik, A-T., and Strömberg, P. 2015. Plankton Toolbox – open source software making it easier to work with plankton data, A. Lincoln MacKenzie [Ed]. Marine and Freshwater Harmful Algae 2014. *Proceedings of the 16th International Conference on Harmful Algae.* Cawthron Institute, Nelson, New Zealand and the International Society for the Study of Harmful Algae (ISSHA), pp 194-197.

Karlson, B., Andersson, L. S., Kaitala, S., Kronsell, J., Mohlin, M., Seppälä, J., and Wranne, A. W. 2016. Oceanographic variability in near surface waters of the Baltic Sea and the Kattegat – a comparison of Ferrybox data vs. monitoring data from research vessels. *Journal of Marine Systems* 162, 98–111.

Kudela, R. M., Howard, M., Jenkins, B. D., Miller, P. E., and Smith, G., J. 2010. Using the molecular toolbox to compare harmful algal blooms in upwelling systems. *Progress in Oceanography* 85, 108–121.

McGillicuddy, Jr., D. J., Anderson, D. M., Lynch, D. R., and Townsend, D. W. 2005. Mechanisms regulating large-scale seasonal fluctuations in Alexandrium fundyense populations in the Gulf of Maine: Results from a physical-biological model. *Deep-Sea Res. II* 52, 2698–2714.

Olson, R. J., and Sosik, H. M. 2007. A submersible imaging in flow instrument to analyze nano and microplankton: Imaging FlowCytobot. *Limnology and Oceanography - Methods* 5, 195–203.

Raine, R., McDermott, G., Silke, J., Lyons, K., and Nolan, G. 2010. A simple short range model for the prediction of harmful algal events in the bays of southwestern Ireland. *Journal of Marine Systems* 83, 150-157.

Scholin, C. A. 2010. What are“ ecogenomic sensors?” A review and thoughts for the future. *Ocean Science* 6, 51–60.

Scholin, C., Doucette, G., Jensen, S., Roman, B., Pargett, D., Marin III, R., Preston, C., Jones, W., Feldman, J., and Everlove, C. 2009. Remote detection of marine microbes, small invertebrates, harmful algae, and biotoxins using the environmental sample processor (ESP). *Oceanography* 22, 158-167.

Seegers, B. N., Birch, J. M., Marin, R., Scholin, C. A., Caron, D. A., Seubert, E. L., Howard, M. D., Robertson, G. L., and Jones, B. H. 2015. Subsurface seeding of surface harmful algal blooms observed through the integration of autonomous gliders, moored environmental sample processors, and satellite remote sensing in southern California. *Limnology and Oceanography* 60, 754–764.

Sellner, K. G., Doucette, G. J., and Kirkpatrick, G. J. 2003. Harmful algal blooms: causes, impacts and detection. *Journal of Industrial Microbiology and Biotechnology* 30, 383–406.

Sosik, H. M. and Olson, R. J. 2007. Automated taxonomic classification of phytoplankton sampled with imaging in flow cytometry. *Limnology and Oceanography - Methods* 5, 204–216.

Stumpf, R. P., Tomlinson, M. C., Calkins, J. A., Kirkpatrick, B., Nierenberg, K., Currier, R., and Wynne, T. T. 2009. Skill assessment for an operational algal bloom forecast system. *Journal of Marine Systems* 76, 151-161.

Ussler, W., Preston, C., Tavormina, P., Pargett, D., Jensen, S., Roman, B., Marin, R., Shah, S. R., Girguis, P. R., Birch, J. M., Orphan, V., and Scholin, C. 2013. Autonomous application of Quantitative PCR in the deep sea: In situ surveys of aerobic methanotrophs using the deep-sea Environmental Sample Processor. *Environmental Science and Technology* 47, 9339-9346.

Velo-Suárez, L., Reguera, B., González-Gil, S., Lunven, M., Lazure, P., Nézan, E., and Gentien, P. 2010. Application of a 3D Lagrangian model to explain the decline of a *Dinophysis* acuminata bloom in the Bay of Biscay. *Journal of Marine Systems* 83, 242–252.

Welschmeyer, N. A. 1994. Fluorometric analysis of chlorophyll a in the presence of chlorophyll b and pheopigments. *Limnology and Oceanography* 39(8), 1985-1992.

Wynne, T. T., Stumpf, R. P., Tomlinson, M. C., Schwab, D. J., Watabayashi, G. Y., and Christensen, J. D. 2011. Estimating cyanobacterial bloom transport by coupling remotely sensed imagery and a hydrodynamic model. *Ecological Applications* 2, 2709-2721.

Yamahara, K. M., Demir-Hilton, E., Preston, C. M., Marin, R., Pargett, D., Roman, B., Jensen, S., Birch, J. M., Boehm, A. B., and Scholin, C. A. 2015. Simultaneous monitoring of faecal indicators and harmful algae using an in-situ autonomous sensor. *Letters in Applied Microbiology* 61, 130–138.

Zhan, P., Yao, F., Kartadikaria, A. R., Viswanadhapalli, Y., Gopalakrishnan, G., and Hoteit, I. 2015. Far-field ocean conditions and concentrate discharges modeling along the Saudi Coast of the Red Sea. In: *Intakes and Outfalls for Seawater Reverse-Osmosis Desalination Facilities*, Springer. pp. 501–520.