

Final Report

**RECONNAISSANCE OF THE
BIOLOGICAL LIMNOLOGY OF
THE SALTON SEA**

Salton Sea Ecosystem Research Group
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EXECUTIVE SUMMARY

A three-year reconnaissance of the limnology of the Salton Sea was carried out during 1997-1999, with funding from EPA Grant R826552-01-0 for the 1999 field season and a write-up period. There were five objectives: a biotic inventory of the species found in the lake, a study of the dynamics of the benthos (or bottom dwelling organisms), a study of the dynamics of the plankton (or microscopic organisms living suspended in the water column), an analysis of physical and chemical conditions in the water column, and a survey for algal toxins. Thirty eight scientists, technicians, and graduate students from nine different institutions participated in this reconnaissance. Detailed findings have been presented in 26 Progress Reports submitted during the course of this work and in 24 manuscripts, listed at end of this report, that have been submitted or are in preparation for publication in the open scientific literature. A wide variety of methodologies have been used. For the main monitoring efforts, plankton and physical and chemical conditions of the water column were monitored at 2-5 week intervals over the whole three years, and the benthos was sampled at 2-month intervals during 1999.

The **biotic inventory** turned up ca. 300 species previously unreported for the Salton Sea, and many species that appear to be new to science. Most of this diversity lies with various types of microorganisms, as follows: cyanobacteria, 11 genera, possibly 2 new to science; cryptomonads, 8 genera, possibly 1 new to science; diatoms, 94 species, probably 3 new to science; dinoflagellates, 22 species, perhaps 5 new to science and one an important fish pathogen (*Amyloodinium*); ciliates, 141 species, including 1 (*Ambiphrya*) that sometimes covers the body surface of fish fry, and possibly 3 genera new to science; naked amebas, 45 species, probably 18 new to science; and one kinetoplastid flagellate (*Cryptobia*) parasitic on fish.

Metazoan invertebrates are much less diverse, but in addition to the dozen species previously known from the Sea, several new species were found. These included: 1 flatworm, 13 nematodes, 3 rotifers, 4 annelid worms, and 1 ostracod. Two species of monogenean fluke (*Gyrodactylus*) parasitic on fish were also found.

Principal invertebrates in the **benthos** (organisms living on or in the sediments) were the polychaetes *Neanthes succinea* (pileworm) and *Streblospio benedicti*, the oligochaetes *Thalassodrilides gurwitschi*, *T. belli*, and an enchytraeid, and, among crustaceans, the amphipods *Gammarus mucronatus* and *Corophium louisianum* and the barnacle *Balanus amphitrite*. *Neanthes succinea* was the dominant macroinvertebrate on the bottom at depths of 2-12 m. Its abundance declined greatly in offshore sediments at depths >2m during summer and fall, due to low oxygen levels at the sediment surface. It persisted year round on shoreline rocks, where densities of all invertebrate species and biomass of *Neanthes* increased from January to November. There is evidence of a decline in *Neanthes* abundance over the last several decades. In September and November 1999 it was two orders of magnitude less abundant than in September and November 1956.

The rocky shoreline had the highest numbers of benthic invertebrates per unit area. In that habitat maximum densities of *Neanthes* and the amphipods *Gammarus mucronatus* and *Corophium louisianum* exceeded previously reported values for those species from

other parts of their range. This demonstrates the high productivity of the Salton Sea, and the importance of the rocky shoreline habitat as a refuge for *Neanthes* and other food organisms for fish and birds during seasonal anoxia in deeper waters.

Phytoplankton, the microscopic algae found suspended or swimming in the water column, constitutes the base for the lake's food web. Three algal groups dominated the phytoplankton: dinoflagellates, diatoms and a raphidophyte. Total phytoplankton abundance was highest in late winter and lowest in late summer. Overturn events occurring in late summer or early fall were associated with low oxygen and high sulfide concentrations in the water, and phytoplankton abundance declined abruptly by an order of magnitude or more. After the crashes, phytoplankton density showed a gradual increase throughout the fall and winter, mostly involving diatoms and dinoflagellates. These late summer crashes in plankton did not occur in the 1950s. Species composition has also changed since the 1950s. We report a number of dinoflagellate and diatom species and one raphidophyte species that have not previously been documented, but some of which now dominate the phytoplankton.

Two species of phytoplankton were of special interest. The raphidophyte *Chattonella marina* was found in high abundance from April to November. Algae in this genus are known to be toxic and produce brevetoxins and superoxide radicals, both of which may be lethal to fish. Some fish kills in the Salton Sea may be due in part to this organism. Gills of fish collected during a *Chattonella* bloom showed gill damage.

Dense populations of the coccolithophore *Pleurochrysis pseudoroscoffensis* were found in surface films at several locations around the Salton Sea on various occasions between February and August. The cells of this species are covered with tiny, highly reflective, calcium carbonate plates (or coccoliths). Surface concentrations of this alga no doubt account for some of the swirls seen in satellite images of the Salton Sea.

Total **zooplankton** abundance was highest in summer following late winter/early spring phytoplankton blooms. At this time metazooplankton consisted mostly of the copepod, *Apocyclops dengizicus*, and the rotifer, *Brachionus rotundiformis*.

In August or September severe crashes in the metazooplankton populations occurred each year in mid-lake due to strong wind events that increased mixing and caused low oxygen and high sulfide concentrations throughout the water column. Larvae of the polychaete worm, *Neanthes succinea* and the barnacle, *Balanus amphitrite* were present mostly in late winter and spring. Their scarcity in summer is due in part to persistent anoxic bottom conditions that decrease adult populations and in part to predation by tilapia, an omnivorous fish that has become abundant in the lake since the 1960s.

Synchaeta spp., rotifers not previously reported from the Sea, were abundant in winter and spring and predation on these may have permitted the copepod to persist at low levels through the winter.

There have been two major changes in metazooplankton dynamics since 1954-1956 in addition to the appearance of two synchaetid rotifers in the fauna. First, there are now

much lower densities of barnacle and polychaete larvae in the fall, probably due to the invasion of the zooplanktivorous fish, tilapia. Second the precipitous crashes now seen in zooplankton densities, especially the copepod, in late summer-early fall did not occur in the 1950s possibly because fall overturn events did not result in as high sulfide levels then as they do now.

Physical and chemical variables in the lake were monitored as this information was essential to understanding the temporal and spatial dynamics of the benthic and planktonic populations.

The Salton Sea is a wind driven system, with predominant winds paralleling the long axis of the lake and being strongest in spring and weakest in summer and fall. The Sea presumably mixed daily or nearly daily between September and January. During this cooling period, moderate to high levels of dissolved oxygen (3-11 mg Γ^1) were found throughout the water column.

Mean water column temperature ranged from a minimum of 13-14 °C in early January to a maximum of 30-34 °C in July-September. During most of this warming period the Sea was thermally stratified but subject to periodic wind driven mixing events. During summer months, mid-lake surface waters were sometimes supersaturated with oxygen, and bottom waters were hypoxic or anoxic with sulfide concentrations sometimes > 5 mg Γ^1 .

Mixing events in late summer deoxygenated the entire water column for a period of days. Consumption of oxygen by sulfide oxidation likely was the principal mechanism for these deoxygenation events. Sulfide concentrations in surface waters were 0.5-1 mg Γ^1 approximately three days after one mixing event in mid-August 1999. Sulfide is highly toxic to most aerobic organisms, and these mixing events were associated with population crashes of phytoplankters and zooplankters and with large fish kills.

Conditions with respect to these variables have changed since the 1950s. The limnological studies carried out in 1954-1956 never found sulfide at as high concentrations in surface waters as we did. They also found that surface waters experienced both hypoxia *and* oxygen supersaturation less frequently than we did. In other words, oxygen levels in the 1950s showed less dramatic fluctuations than they do now.

High inputs of nutrients, nitrogen and phosphorus in particular, are responsible for the dense phytoplankton that characterizes this eutrophic lake. In the water column, the mean molar ratio of nitrogen to phosphorus averaged ca. 70, suggesting phosphorus is the nutrient limiting phytoplankton growth. Total nitrogen concentrations (mean = 5.9 mg N Γ^1) were due mostly to dissolved organic nitrogen (mean = 4.8 mg N Γ^1). Ammonia (mean = 0.9 mg N Γ^1), nitrate (mean = 0.08 mg N Γ^1), and nitrite (mean = 0.007 mg N Γ^1) were relatively scarce. Total phosphorus concentrations (0.16 mg P Γ^1) were primarily made up of the dissolved organic fraction (mean = 0.1 mg P Γ^1). Orthophosphate concentrations were generally low (mean = 0.02 mg P Γ^1). Particulate phosphorus (mean = 0.036 mg P Γ^1) exhibited seasonal and vertical variation that followed changes in phytoplankton abundance.

The penetration of light into the Sea determines to what depths photosynthesis can be carried out by algae, is regulated in part by the abundance of phytoplankton, and is very low. The depth to which a 20-cm diameter, flat, black and white Secchi disk remained visible from the edge of the boat, increased to a maximum of 170-190 cm in late summer or early fall and then decreased to a minimum of 60-80 cm in late winter or early spring. Radiometer readings showed a similar seasonal variation. Weak correlations between light penetration and chlorophyll a concentrations suggest that light penetration in the Salton Sea may be more strongly affected by dissolved organic matter, zooplankton, and inorganic particulate matter than is usually the case in lakes.

Algal toxins have been known to cause disease and mortality in fish, birds, mammals and even humans and have been postulated as possible causes of some bird die-offs at the Salton Sea, such as large eared grebe mortalities in 1992 and 1994. Most of the diverse phytoplankton samples we collected and assayed showed, at best, low level toxicity to brine shrimp and no toxicity to mice. One sample taken from a bloom of *Gymnodinium* spp. was highly active in the brine shrimp lethality assay. Although the ¹H NMR spectrum from this sample was extremely complex, the active material was lost during fractionation and we were not able to isolate a toxin. Given the rather small number of samples tested and the fact that several types of cyanobacteria, dinoflagellates, and raphidophytes known to occur in the Sea are capable of producing toxins in other environments, the role of algal toxins in this lake ecosystem remains an open question.

Dissolved organic material released by phytoplankton blooms can lower the surface tension of water and thereby facilitate penetration of avian plumage by lake water. We speculate that some past eared grebe die-offs may have resulted from hypothermia following waterlogging of plumage.

It is evident that large changes have taken place in the ecology of the Salton Sea since the 1950s and 1960s. This is not surprising given that since then salinity has increased by about 25 percent, tremendous amounts of additional nutrients have come into the lake, and tilapia invaded and became the most abundant fish in it. The separate influences of these three factors are not easily assessed. The system will change greatly once it becomes too saline for fish. But we can also expect that as salinity rises, other and unpredictable changes will take place in the lake long before the fish disappear, i.e. in the short term.

Our understanding of Salton Sea ecology, of its likely future, and of possible impacts of engineering projects aimed at restoration would be facilitated by new mechanisms for attracting and supporting research efforts that better build on existing information. Critical insights into what is going on at the Sea are not being taken advantage of. An **open-topic RFP and award process** would attract research proposals that themselves would be great eye-openers for the EPA, Salton Sea Authority, and Salton Sea Science Office.

INTRODUCTION

This final report summarizes the findings of a Reconnaissance of the Biological Limnology of the Salton Sea carried out under U.S. Environmental Protection Agency Grant No. R826552-01-0 to the Salton Sea Authority. Work was carried out by 38 scientists, graduate students and technicians at nine different U.S. and foreign institutions (see preceding page) and coordinated by the Salton Sea Ecosystem Research Group (SSERG) at San Diego State University. The reconnaissance represents the most detailed limnological investigation of the Salton Sea ever carried out. It also has documented the presence of hundreds of new species heretofore unknown from the Salton Sea, and many species completely new to science.

Details of methodologies and findings can be found in the 26 progress reports that were submitted during the course of our investigations and that are attached to this report as appendices.

Many of these progress reports are manuscripts that have been submitted for peer review and publication in scientific journals. In some cases they have already been published. Other manuscripts are still in preparation. A list of all submitted manuscripts and published papers follows is found at the end of this report. Eventually all findings of this reconnaissance will be published in the open scientific literature.

We do not in this final report attempt to reformat and re-present our findings in full. A great variety of topics and scientists are involved, and manuscripts are in various stages of review and revision. We also wish to make this report readable by the small (but select!) group of people who will read it -- to the very end, if they are wise. In this report, we identify location of detailed findings by referencing Progress Report numbers in parentheses following section headings, but otherwise do not reference specific manuscripts, whether published or unpublished.

This limnological reconnaissance has received strong support from the home institutions of the participating scientists and was actually initiated in 1996, two years prior to its funding by the EPA grant. The grant allowed great expansion of the reconnaissance and covered ca. 90 percent of our supply, equipment and travel costs. Most of the manpower has been provided on a *pro bono* basis, however. With a few exceptions, none of the collaborating scientists listed on the preceding page received any salary or stipend for their participation. The lead scientists and the key graduate student researchers have dedicated roughly three times more effort to the project than was funded by the EPA grant. Many participants are still involved in analyzing samples and data and writing up findings. This process will continue for some time.

REPRODUCTION AND DISTRIBUTION OF REPORT

This final report and attached progress reports present most of the information gathered on Salton Sea limnology by the Salton Sea Ecosystem Research Group and its collaborators during the period 1996-2001, including that gathered with funding from

sources other than the 1998-2000 EPA grant. It is not feasible to try to separate out information gathered under the EPA grant from that information gathered under other auspices.

Our early progress reports aimed at providing information as quickly as possible to those agencies carrying out the environmental impact analysis. Later progress reports included appendices containing our raw data files for particular variables and also containing results of our quality control assessments. This final report aims to inform EPA, the Salton Sea Authority, the USGS Salton Sea Science Office, and the community at large, in succinct fashion, of our key findings, without distinction as to funding source, from this 4-year project and their significance,

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The Progress Reports, however, are either preliminary reports or manuscripts in various stages or review and revision. They are not intended for wide public distribution and not appropriately cited in scientific or other reports. Persons wishing to use information presented in a Progress Report should consult with the senior author of that report or with Dr. D.M. Dexter or Dr. S.H. Hurlbert.

GENERAL METHODOLOGY

Our limnological reconnaissance had five main components: a biotic inventory of the species found in the lake, a study of the dynamics of the benthos (or bottom dwelling organisms), a study of the dynamics of the plankton (or microscopic organisms living suspended in the water column) an analysis of physical and chemical conditions in the water column, and a survey for algal toxins. We describe briefly the objectives and general methodological approaches for each of these components.

Biotic Inventory (Progress Reports 2, 4, 5, 6-15, 17, 21-24)

Aside from the birds and the fish, the biota of the Sea has been poorly studied over the years. In the Salton Sea, as in many aquatic ecosystems, the small organisms rule. They drive the system. They carry out most of the photosynthesis and most of the decomposition. Their metabolic and biogeochemical activities are a major determinant of water chemistry and water quality. They serve as the base of the food web that sustains the fish and bird populations at the Sea. Not much happens without their involvement. Some are pathogens or toxic.

During the 1996-98 phase of this reconnaissance, every careful look at samples of invertebrates and protists turned up forms not previously recorded from the Sea, or, in some cases, forms that were misidentified by previous workers. Yet nothing is more fundamental to an understanding of the biology of the Sea than knowing what species are present. Without that knowledge we cannot access the most pertinent scientific literature

and are poorly positioned to assess functional relationships, effects of future salinity increase, or impacts of engineering projects.

For this reason we proposed to carry out taxonomic inventories for several key groups of organisms in the Sea, giving special attention to groups that are abundant there but poorly known and difficult for non-specialists to identify with accuracy. These are mainly different sorts of microorganisms and small invertebrates. This sort of work was not requested in the Request for Proposal that was issued for this work. To their credit, the SSA and SSSS recognized the value of laying this foundation once it was pointed out in our proposal and were willing to fund the extensive participation of systematists in the project. Many national and state agencies that fund ecological and environmental studies are not so far-sighted.

The groups of organisms chosen and the specialists selected to inventory them were:

Cyanobacteria (blue-green 'algae')	- Anne Michelle Wood, Richard W. Castenholz
Cryptophyta (small flagellates)	- Paul Kugrens, Steven B. Barlow
Bacillariophyta (diatoms)	- Carina Lange, Mary Ann Tiffany
Dinophyta (dinoflagellates)	- Karen Steidinger
Ciliophora (ciliates)	- Eugene B. Small
Gymnamoebae (naked amebas)	- Andrew Rogerson
Protozoan fish parasites & symbionts	- Boris Kuperman
Platyhelminthes (flatworms)	- Yuri Mamkaev
Nematodes (roundworms)	- Richard M. Warwick

Other specialists aided with occasional identifications: P. Fong (Chlorophyta - green algae), R. Shiel (Rotifera - rotifers), M. Milligan (Oligochaeta - worms), L. Lovell (Polychaeta - worms), J. Thomas (Amphipoda - scuds), and R. Forester (Ostracoda - seed shrimp).

Methods for collecting, preserving, and identifying specimens were different for each group and will be found in the respective Progress Reports. A few of the specialists had funding that allowed them to visit the Sea and collect material themselves. All specialists also received fresh or preserved samples collected by SDSU scientists and forwarded to them by express mail.

Benthos Dynamics (Progress Reports 4, 13, 15)

Invertebrates associated with the lake bottom - especially amphipods, polychaetes, corixids, and barnacles -- provide food for fish and birds that forage on the bottom. The juveniles of these invertebrates are often planktonic and provide food for fish feeding in the water column and at the water surface. Because there are so few invertebrate species in the food chain, changes in the physical and chemical environment have the potential for destabilizing this assemblage and, as a consequence, adversely affecting fish and birds.

The objective of this component of the reconnaissance was to document the species composition, abundance, and seasonality of macroinvertebrates associated with specific benthic habitats. We sampled the sublittoral sediments of the Sea at depths of 2 to 12 m, and the shoreline habitats comprising barnacle shell substrates, barnacle-covered rocks, and algae-covered rocks. Additionally, the seasonal abundance of swarming heteronereids of *Neanthes succinea* was determined to quantify their availability to fish and birds foraging at night.

Benthic fauna of the soft offshore sediments

Benthic grab samples were taken at stations along 3 transects extending out from shore along the eastern side of the Sea. Sampling was carried out at two-month intervals starting in January 1999 at each of 6 stations along each transect at depths of 2, 4, 6, 8, 10, and 12m. We used a petite Ponar grab, which samples an area of 15 x 15 cm to a depth of 15 cm.

Sediment analysis

Sediment characteristics can be important factors affecting the distribution of infaunal invertebrates. Replicate samples of the top 15 cm of sediment were collected at each station in March and September. Total carbon, total nitrogen, and organic carbon content were determined. Particle size analysis was performed on sediments collected in March.

Macrozooplankton tows

On each sample date beginning at sunset, we conducted 3 replicate tows for macrozooplankton near Varner Harbor to determine the abundance and biomass of nektonic heteronereids of *Neanthes succinea*.

Flora and fauna associated with shoreline littoral substrates

Rocky Substrates:

This habitat consists mainly of rocks, dikes constructed for protection of marinas and shoreline, and precipitated calcium carbonate deposits. Quadrat sampling of epifaunal invertebrates on rocky substrates was carried out on the southern jetty at Red Hill Marina on two substrate types: rocky substrate dominated by the barnacle *Balanus amphitrite* and rocky substrate dominated by the chlorophytes *Chaetomorpha linum* Müller and *Enteromorpha intestinalis* (L.) Link. The locations of five quadrats on barnacle-covered rock were haphazardly selected along the exposed side of the jetty, while five quadrats on algae-covered rock were selected along both the exposed and protected sides of the jetty.

Barnacle shell substrates:

A major shoreline habitat all around the Salton Sea, especially at very shallow depths (less than 0.3 m), consists of intact and broken barnacle shells and associated unconsolidated sediments, ranging in particle size from coarse sand to gravel. Invertebrates present in barnacle shell substrates were sampled on open shoreline sites at Bombay Beach, Salt Creek, and State Recreation Area Headquarters using a stainless steel corer (area 0.01 m²) which penetrated the sediment to a depth of 10 cm.

Plankton Dynamics and Physical and Chemical Conditions (Progress Reports 1-3, 16-20, 24, 25, 26)

Monitoring of plankton populations and selected physical and chemical variables was carried out at three mid-lake stations beginning in January 1997, and at two near shore stations beginning in January 1999, and ending in January 2000. The first three stations were chosen to represent the main water mass of the Sea. The two additional ones were added to better sample the southern basin, monitor nearshore areas, and to document the influence of freshwater inflows. Monitoring was conducted at 2 to 5 week intervals, although in 1997 – 1998 some stations were not visited on all dates due to logistical problems or storms.

Measurements of temperature, dissolved oxygen, and specific conductance were made at each station from surface to bottom at 1 m. At each station phytoplankton, zooplankton, nitrogen, phosphorus, and silica concentrations, pH, oxidation-reduction potential, light attenuation, and Secchi depth were also sampled or measured. Specific conductance was measured starting in July 1998. Temperature and dissolved oxygen profiles were taken at five stations along each of two onshore-offshore transects on in June and August 1999 and in January 2000. We measured sulfide in the water column on two dates (31 July and 16 August 1999) at all five mid-lake sampling stations after sulfide was detected in the air during mixing events.

Survey for Algal Toxins (Progress Reports 2, 8, 16)

In other aquatic systems, algal toxins have been implicated in mortality events involving, fish, and mammals. Several species of phytoplankton found in the Salton Sea have the potential to form toxic blooms. These include dinoflagellates, raphidophytes, and prymnesiophytes. Some of these species have been implicated in mortality events in other systems while others are closely related to known toxin-producing species. In a 1994 preliminary study, a sample dominated by an unarmored gymnodinioid dinoflagellate with some *Gonyaulax grindleyi* also present showed high activity in a mouse bioassay indicating the presence of a toxin. Otherwise the toxicity of Salton Sea phytoplankton had not been previously investigated.

A survey of algal toxins in the Salton Sea was initiated in 1999 to determine if and when algal toxins are present in the lake, and to document the composition of the phytoplankton at those times. Five open water stations and three shoreline sites (North Shore, Bombay Beach and Red Hill) were visited biweekly from January to December 1999. Samples for taxonomic and toxicity analyses were collected when dense or unusual blooms were encountered or particular species were observed. Phytoplankton samples were collected and concentrated using several different methods. Concentrates were generally stored at -80°C until extraction. Concentrated algal samples were lyophilized and then extracted with methanol, water, and ethyl acetate. Each extract was assayed for lethality to brine shrimp (*Artemia salina*) and mice (*Mus musculus*). A nuclear magnetic resonance spectrum was recorded for each extract with a NMR spectrometer. Water samples for analysis of phytoplankton composition were taken prior to

concentration of algae, preserved with Lugol's solution, and counted using the Utermohl method.

BIOTIC INVENTORY

Results to date have been very interesting in that large numbers of species not previously reported from the Sea have been found. These include forms of interesting morphology, such as giant ciliates over 1 mm in length, forms that are potentially toxic to fish and other organisms, and forms that are completely new to science, including probably several new genera. In almost no case do we have a clear understanding of the importance of the individual species to the functioning of the Salton Sea ecosystem.

Prior to 1997 there were a few more than 70 species of invertebrates, protists, and cyanobacteria recorded from the Sea. The present effort has already added 300+ new forms to that list, though specific determinations have not been made yet for the majority of these. For this reason, we do not have a good idea of exactly how many species new to science are in the collections already made. Preliminary indications are that it will be at least two dozen. The painstaking work-up of these collections will continue well beyond the end of this reconnaissance project.

We have found several new species of metazoan invertebrates, but this on the whole is not a diverse assemblage. The protist assemblages, on the other hand, have very high diversity, higher than is typical of most lake and marine environments. Perhaps this diversity is related in part to scarcity of metazoan invertebrate species that in other aquatic environments are both competitors with and predators on the protists. Perhaps it is also related to the high concentrations of dissolved organic matter and of bacteria in the Sea. And certainly the salinity and ionic composition of the lake are similar enough to those of ocean water, that many marine forms find conditions in the Sea quite acceptable.

Findings are summarized below for the main groups this inventory has focused on.

Cyanobacteria (blue-green 'algae') (Progress Report 22)

Representatives of a total of 11 described genera (*Oscillatoria*, *Spirulina*, *Arthrospira*, *Geitlerinema*, *Lyngbya*, *Leptolyngbya*, *Calothrix*, *Rivularia*, *Synechococcus*, *Synechocystis*) were identified in the samples; additionally, the morphology of two cultured strains do not conform to any genus recognized by the standard bacteriological system. One of these is very unusual as it appears to contain chlorophyll *d* as its primary light harvesting pigment. Filamentous forms, particularly *Geitlerinema* and *Oscillatoria* were common in benthic and floating MICROBIAL mat material; different mat samples were dominated by different morphotypes of filamentous cyanobacteria and were usually composed of roughly equivalent biomass of cyanobacterial filaments and the chemolithotrophic sulfur oxidizing bacterium *Beggiatoa*. Unicellular and aggregating forms (mostly *Synechococcus* and *Synechocystis*) were common in the plankton. In contrast to most marine waters where the predominant strains of these genera utilize phycoerythrin as the dominant photosynthetic pigment, our samples included many

picocyanobacterial cells that synthesized phycocyanin as the dominant photosynthetic pigment.

Molecular phylogenetic analysis of a representative subset of filamentous cyanobacteria isolated from the sea highlighted the difficulties of evaluating cyanobacterial diversity from field samples alone. For example, three strains that would be classified as the same species using the current botanical system for cyanobacteria fell into two completely different major clades of cyanobacteria based on their 16s rDNA sequence.

The biotic inventory of cyanobacteria conducted in 1999 does not represent an exhaustive inventory of the cyanobacterial flora of the Salton Sea. Even so, it provides ample evidence that there is a diverse community of these prokaryotic primary producers in both the water column and benthos, and that the species present include some physiologically unusual organisms. The association of filamentous cyanobacteria with the bacterium *Beggiatoa*, which relies on reduced sulfur for energy, suggests that cyanobacterial communities are important primary producers under conditions of low oxygen concentration. Among taxa found in the sea, there is overlap at the generic level with five known toxin producing species; thus, cyanobacteria should also be considered as possible sources of toxins causing fish kills and avian mortality, particularly if there is any likelihood that animals will feed directly on the mats when they are growing on pilings, attached to the bottom, or floating in the water column.

Cryptophyta (small flagellates) (Progress Report 23)

Eight genera of cryptomonads from the Salton Sea are recorded for the first time. Comparative data from light and scanning electron microscopy were utilized to identify these genera and species. The genera included *Chroomonas*, *Falcomonas*, *Hemiselmis*, *Plagioselmis*, *Pyrenomonas/Rhodomonas*, *Storeatula*, *Teleaulax*, and the kathablepharid *Leucocryptos*. One putative genus remains unidentified and may represent a new taxon. Scanning electron micrography has not been conducted on this cryptomonad, but it has been isolated and is being maintained in culture. The genera and species identified from the Salton Sea are typical of marine rather than freshwater environments and may play an important role in primary productivity and as preferred food organisms for zooplankton.

Bacillariophyta (diatoms) (Progress Reports 7, 10)

In the plankton and benthos of the Salton Sea and its immediate tributaries, 94 diatom species were distinguished based on their morphological features at the light- and electron microscope level. Heretofore less than a dozen species of diatoms were known from the Sea. Four general categories of diatom assemblages related to their habitats can be distinguished: (1) A planktonic assemblage composed of *Chaetoceros muelleri* var. *subsalsum*, *Cyclotella choctawhatcheeana*, *Cyclotella* sp., *Cylindrotheca closterium*, *Pleurosigma ambrosianum*, *Thalassionema* sp); (2) a benthic assemblage with diatoms that live on the bottom (e.g. genera *Caloneis*, *Diploneis*, *Entomoneis*, *Gyrosigma*, *Plagiotropis*, *Pleurosigma*, *Surirella* and *Tryblionella*), or in algal mats (*Proschkinia bulnheimii*, several species of *Navicula* and *Seminavis gracilentia*); (3) an epiphytic community attached to the macroscopic green algae which grow on the rocks and other

hard surfaces near shore (e.g. *Achnanthes brevipes*, *Licmophora ehrenbergii*, *Tabularia parva*); and (4) a freshwater assemblage composed of species that get washed in by the rivers and other inflows discharging into the Sea (e.g. *Achnantheidium minutissimum*, *Cocconeis pediculus*, *Cyclotella atomus*, *C. scaldensis*, *Nitzschia elegantula*, *T. weissflogii*). The most striking feature of the phytoplankton is the abundance of species formerly known only from marine environments; this is not surprising given the high salinity and the peculiar history of the lake.

Flourishing populations of three sigmoid diatom species were observed in the Sea that were unusual in one way or another. The occurrence of *Gyrosigma balticum* (Ehrenberg) Rabenhorst so far inland is at least unusual if not unique. The ecology of *Gyrosigma wormleyi* (Sullivant) Boyer, previously typified as a freshwater species, must apparently be extended to include saline habitats. A new species new to science, *P. ambrosianum* is the dominant member of the plankton diatom assemblage during the winter months. We named the species after Ambrosia, believed to be one of the last fire-eaters of the Desert Cahuilla Indians who have lived in the Salton Basin for millennia.

Dinophyta (dinoflagellates) (Progress Reports 6, 10, 12, 14)

During much of the year, dinoflagellates dominate the phytoplankton and hence are major components of the ecosystem. This inventory found 22 dinoflagellate taxa, almost all previously unrecorded from the Salton Sea and perhaps a fourth of which are new to science. They include unarmored and armored, brackish water and marine, heterotrophic, mixotrophic, and auxotrophic species that have adapted to the high salinity of the Sea. Probably none of these species have been previously recorded from an inland saline lake. Unarmored dinoflagellates found included species in the genera *Amphidinium*, *Gymnodinium*, *Oxyrrhis*, *Gyrodinium* and *Woloszynskia*. Armored dinoflagellates included species in the genera *Prorocentrum*, *Alexandrium*, *Gonyaulax*, *Scrippsiella*, *Bysmatrum*, *Pentaparsodinium*, *Oblea*, *Heterocapsa*, *Kryptoperidinium*, *Protoperidinium*, and a new *Pfiesteria*-like species.

The nutrient dynamics of these dinoflagellate populations are complex, as some are completely autotrophic, some completely heterotrophic, utilizing particulate and dissolved organic matter, and some with both capabilities (mixotrophic). Some harbor bacteria on their surface. These may be either using released nutrients from the bacteria or feeding on the bacteria directly. Detailed autecological studies on nutrient dynamics would be needed to understand the physiology and ecology of these complex populations and how they influence the rest of the ecosystem. There are several dinoflagellates present that might form toxic blooms, e.g. *Gonyaulax grindleyi*, known to produce yessotoxin, *Alexandrium* sp. possibly producing saxitoxins and gonyautoxins, and some of the new *Gymnodinium* species that may produce bioactive compounds.

Hermesinium adriaticum is a rarely reported unicellular biflagellate organism with a solid, elaborate siliceous skeleton. Its classification is uncertain, but it has been considered by some as close to the dinoflagellates and by others has been put in the phylum Opalozoa. In the Salton Sea we found it in cyanobacterial mats, the plankton, and, in largest numbers in the sediments. Stages of the developing skeleton were studied with SEM and

the progression from small tetraaxial daughter skeletons to complete, asymmetrical adult skeletons was documented.

Ciliophora (ciliates) (Progress Report 17)

One hundred and forty-one species of benthic and planktonic ciliates representing nine classes of Ciliophora were collected from eleven shoreline sites around the perimeter of the lake area. An additional thirty-five species were found in June 1999 that were not during the January 1999 collection trip. Thus, the ciliates comprise one of the most diverse groups of organisms encountered in the Salton Sea. We have assigned one hundred and thirty eight of the one hundred and forty one observed species to genera. Three of the ciliates can only be assigned to family level to date and may represent new genera.

Gymnamoebae (naked amebas) (Progress Report 11)

This first examination of the naked amoeboid protozoans of the Salton Sea isolated 45 different morphospecies (considered to be species) of amoebae. Of the 45 taxa, around 18 (i.e. 40 percent) are probably new to science. Preliminary counts, based on enrichment cultivation methods, showed that amoebae in the water column ranged from 14,560 to 237,120 cells per liter. The ecological importance of the high densities and high diversity of Salton Sea amoebae is unknown. But it should be noted that several of the amoebae were actively grazing cyanobacterial and algal filaments and filaments of the bacterium *Beggiotoa*. Others were predominately associated with suspended particulates. Amoebae may be important in the cycling of carbon and nutrients in the Salton Sea.

Protozoan fish parasites & symbionts (Progress Reports 6, 9)

Parasitological examinations were made of 1,512 fish. Young tilapia, *Oreochromis mossambicus*, croaker, *Bairdiella icistia*, and mudsucker, *Gillichthys mirabilis*, were found infected by ectoparasites. Some persistent foci of fish infestation were found around the perimeter of the lake. The diversity of fish parasites was limited. We found three protozoan species, *Amyloodinium ocellatum* (Dinoflagellida), *Ambiphrya ameiuri* (Peritricha), *Cryptobia branchialis* (Bodonidae: Kinetoplastida), and two metazoans, the monogeneans *Gyrodactylus olsoni* and *Gyrodactylus imperialis*. These five species all have direct life cycles without involvement of intermediate hosts such as invertebrates or other fish. This type of development allows parasites to spread easily and widely. Endoparasites, which generally have more complex life cycles, were not detected at the Salton Sea. Two factors may explain this absence. First would be the limited number of potential intermediate host species. Second, the free-living larval stages of parasitic helminths with indirect life cycle are likely to have difficulty surviving in environments with such high salinities and temperatures.

Both *A. ocellatum* and *A. ameiuri* infected fish from spring through fall. The greatest infestations by *A. ocellatum* occurred in summer and the greatest ones by *A. ameiuri* in in spring and autumn. The seasonal dynamics of the *A. ocellatum* infestation of young

tilapia was documented. First appearing in May, it became maximal in June-August, decreased in October and was not detectable in November.

High parasite loads caused severe damage to such respiratory organs as gills and skin. They may depress respiration and osmoregulation and, in combination with other environmental stresses, cause fish suffocation and death. If a major cause of juvenile fish mortality, these parasites likely play a major role in determining fish population dynamics in the Salton Sea.

Nematodes (free-living roundworms) (Progress Report 21)

Nematodes occur in every conceivable habitat that can support life, and are the most numerous of all metazoans, yet had never been studied at the Salton Sea. Sampling of a variety of substrates in the Sea yielded thirteen nematode species. This doubles the number of species of multicellular invertebrates known to occur in the lake. All species were referable to known marine genera, and are regarded as having a marine coastal origin. The range of taxa present is representative of the full taxonomic spread found in marine coastal habitats. Evidently a widerange of marine nematode taxa are capable of adapting to the highly saline conditions. The broad spectrum of feeding types present suggests that nematodes play a variety of ecological roles within the lake. They are of major energetic importance in benthic systems, they form a significant part of the diet of many other animals, facilitate the mineralization of organic matter, and influence the physical stability of sediments and the exchange of materials between the sediment and water column.

Other invertebrates (Progress Reports 13, 15, 20)

A number of other invertebrates not previously known from the Sea were found and identified by specialists.

The turbellarian *Macrostomum pusillum*, is a small rhabdocoel belonging to the Family Macrostomidae, measures a few mm in length, and is the first flatworm (Phylum Platyhelminthes) to be recorded from the Sea. It has heretofore been known only from coastal marine environments of North America and Europe. At the Salton Sea, specimens were associated with algal mats and muddy substrates in shallow embayments within Varner Harbor and near Red Hill Marina.

The rotifer *Brachionus rotundiformis* is extremely abundant component of the zooplankton in summer. The *Brachionus* in the Sea have in previous reports been called *B. plicatilis* - a closely related form - but one now considered distinct from *B. rotundiformis*. This rotifer grazes on algae and bacteria and in turn is consumed by fish, copepods, and possibly barnacles.

The Phylum Annelida (segmented worms) is represented primarily by the pileworm *Neanthes succinea*, long known to be a major prey item for both fish and birds in the Salton Sea. Four additional species of annelids, all only a few mm in length, were found by our surveys. The polychaete *Streblospio benedicti* (Family Spionidae) is present at the

Salton Sea in greatest abundance in clay sediments in shallow (ca. 2 m) water. It has been previously recorded from coastal marine environments of North and South America and Europe. Three oligochaetes are abundant within the barnacle shell and sandy substrates along the shoreline. Two species - *Thalassodrilides gurwitschi* and *T. belli* - are members of the Family Tubificidae, a family known for species tolerant of polluted waters. The first of these species has a wide distribution but has not been previously reported from North America. The second is previously known only from the Caribbean and Gulf of Mexico. A third oligochaete is as yet unidentified but belongs to the Family Enchytraeidae.

In the Phylum Arthropoda, two crustacean species new to the Sea were found. The ostracod *Cyprideis beaconensis* (Order Podocopida: Family Cytherideidae) is found in algal mats (*Chaetomorpha linum*) associated with rocky substrates, in shoreline barnacle shell substrates, and in muddy sediments at extremely shallow depths along the shoreline, especially at the southern end. The amphipod *Corophium louisianum* (Family Corophiidae) constructs a tube of mud and detritus attached to rocks, algae, or the interiors of empty barnacle shells, or imbedded within soft sediments. It is smaller (< 4 mm) and much less abundant than the larger amphipod present, *Gammarus mucronatus*.

BENTHOS DYNAMICS

(Progress Report 13)

The principal infaunal invertebrates (those living within the sediments) found were the polychaetes *Neanthes succinea* and *Streblospio benedicti*, and the oligochaetes *Thalassodrilides gurwitschi*, *T. belli*, and an enchytraeid. Benthic crustacean species found were the amphipods *Gammarus mucronatus* and *Corophium louisianum* and the barnacle *Balanus amphitrite*.

The pileworm *Neanthes succinea* was the dominant macroinvertebrate on the Sea bottom at depths of 2-12 m. During 1999, its abundance varied spatially and temporally. Abundance declined greatly in offshore sediments at depths >2m during summer and fall, due to decreasing oxygen levels at the sediment surface. In contrast, *Neanthes* persisted year round on shoreline rocks, where densities of all invertebrate species and biomass of *Neanthes* increased from January to November.

Area-weighted estimates of standing stock of *N. succinea* in September and November 1999 were two orders of magnitude less than biomass estimated to be present in September and November 1956. We speculate that oxygen conditions in the Sea may have deteriorated over the last half century and, along with a 30 percent increase in salinity, be responsible for this difference.

The rocky shoreline had the highest numbers of organisms per unit area. In that habitat maximum densities of *Neanthes* and the amphipods *Gammarus mucronatus* and *Corophium louisianum* exceeded previously reported values for those species from other parts of their range. This demonstrates the high productivity of the Salton Sea, and the

importance of the rocky shoreline habitat as a refuge for *Neanthes* and other food organisms for fish and birds during seasonal anoxia in deeper waters.

PHYTOPLANKTON DYNAMICS

(Progress Reports 2, 7, 8, 25)

Three algal groups dominated the phytoplankton: dinoflagellates, diatoms and a raphidophyte. Cryptomonads, a coccolithophore, non-motile chlorophytes and a euglenoid were also important. Total phytoplankton abundance was highest in late winter, when it was dominated by dinoflagellates, especially *Gyrodinium uncatenum*, *Heterocapsa niei* and a species of *Gymnodinium*. A drop in phytoplankton density each April was likely caused by increased feeding by the planktivorous fish, tilapia, *Oreochromis mossambicus*, as the lake warmed. As the fish were forced into nearshore environments in summer by anoxia, the mid-lake phytoplankton rebounded and was dominated primarily by *Chattonella marina*, *Thalassionema* sp., scrippsielloid dinoflagellates, and *Gonyaulax grindleyi*. Overturn events occurring in late summer or early fall were associated with low oxygen and high sulfide concentrations in the water, and phytoplankton abundance declined abruptly by an order of magnitude or more. After the crashes, phytoplankton density showed a gradual increase throughout the fall and winter, mostly involving diatoms and dinoflagellates. These late summer crashes in plankton did not occur in the 1950s. Species composition has also changed since the 1950s. We report a number of dinoflagellate and diatom species and one raphidophyte species that have not previously been documented, but some of which now dominate the phytoplankton. Dinoflagellate and diatom species now dominant are larger than those dominant in the 1950s.

Two species of phytoplankton were of special interest. *Chattonella marina* was found in high abundance from April to November. Maximum mean density was over 600 cells ml⁻¹ mid-lake. Algae in this genus are known to be toxic and produce brevetoxins and superoxide radicals, both of which may be lethal to fish. Fish kills occur frequently in the Salton Sea and may be due in part to this organism. Gills of fish collected at the height of the *Chattonella* bloom showed damage to the primary and secondary lamellae and increased numbers of mucus cells. To our knowledge this is the first report of a species of *Chattonella* in a salt lake.

Dense populations of the coccolithophore *Pleurochrysis pseudoroscoffensis* were found in surface films at several locations around the Salton Sea on various occasions between February and August. To our knowledge, this is the first record of this widespread marine species in any lake. The cells of this species are covered with tiny, highly reflective, calcium carbonate plates (or coccoliths). Surface concentrations of this alga no doubt account for some of the swirls seen in satellite images of the Salton Sea. The nature of these has been a mystery since the first satellite images were taken.

ZOOPLANKTON DYNAMICS

(Progress Reports 3, 20)

Total zooplankton abundance was highest in summer following late winter/early spring phytoplankton blooms. At this time metazooplankton consisted mostly of the copepod, *Apocyclops dengizicus*, and the rotifer, *Brachionus rotundiformis*.

In August or September severe crashes in the metazooplankton populations occurred each year in mid-lake due to strong wind events that increased mixing and caused low oxygen and high sulfide concentrations throughout the water column. Larvae of the polychaete worm, *Neanthes succinea* and the barnacle, *Balanus amphitrite* were present mostly in late winter and spring. Their scarcity in summer is due in part to persistent anoxic bottom conditions that decrease adult populations and in part to predation by tilapia, an omnivorous fish that has become abundant in the lake since the 1960s.

Synchaeta spp., rotifers not previously reported from the Sea, were abundant in winter and spring and predation on these may have permitted the copepod to persist at low levels through the winter.

There have been two major changes in metazooplankton dynamics since 1954-1956 in addition to the appearance of two synchaetid rotifers in the fauna. First, there are now much lower densities of barnacle and polychaete larvae in the fall, probably due to the invasion of the zooplanktivorous fish, tilapia. Second the precipitous crashes now seen in zooplankton densities, especially the copepod, in late summer-early fall did not occur in the 1950s possibly because fall overturn events did not result in such high sulfide levels then.

PHYSICAL AND CHEMICAL LIMNOLOGY

Thermal and mixing regimes (Progress Reports 1, 18)

The Salton Sea is a wind driven system, with predominant winds paralleling the long axis of the lake, being strongest in spring and weakest in summer and fall. The Sea mixed daily or nearly daily between September and January. During this cooling period, moderate to high levels of dissolved oxygen (3-11 mg l⁻¹) were found throughout the water column.

Mean water column temperature ranged from a minimum of 13-14 °C in early January to a maximum of 30-34 °C in July-September. During most of this warming period the Sea was thermally stratified but subject to periodic wind driven mixing events. Winds were stronger in spring 1998 than in 1997 or 1999, causing more rapid heating of the lake that year and also delaying onset of anoxic conditions in bottom waters.

In the southern basin, freshwater inflows tended to move out over the surface of the Sea mixing with saline lake water as a function of wind conditions. Salinity gradients often

contributed more to water column stability than did thermal gradients in the southeasternmost portion of the lake.

Oxygen and sulfide regimes (Progress Reports 1, 18)

During summer months, mid-lake surface waters were sometimes supersaturated with oxygen, and bottom waters were hypoxic or anoxic with sulfide concentrations sometimes $> 5 \text{ mg } \Gamma^1$. Oxic conditions ($> 1 \text{ mg } \text{O}_2 \Gamma^1$) often extended a few meters deeper nearshore than they did well offshore as a consequence of greater mixing nearshore.

Mixing events in late summer deoxygenated the entire water column for a period of days. Consumption of oxygen by sulfide oxidation likely was the principal mechanism for these deoxygenation events. Sulfide concentrations in surface waters were $0.5\text{-}1 \text{ mg } \Gamma^1$ approximately three days after one mixing event in mid-August 1999. Sulfide is highly toxic to most aerobic organisms, and these mixing events were associated with population crashes of phytoplankters and zooplankters and with large fish kills.

Conditions with respect to these variables have changed since the 1950s. The limnological studies carried out in 1954-1956 never found sulfide at as high concentrations in surface waters as we did. They also found that surface waters experienced both hypoxia *and* oxygen supersaturation less frequently than we did. In other words, oxygen levels in the 1950s showed less dramatic fluctuations than they do now.

Nitrogen, phosphorus and silicon (Progress Reports 1, 19)

These elements are of special importance to the ecosystem as they tend to be present in low concentrations relative to the demand for them by algae. They thus often determine the abundance of algae and other photosynthetic organisms and the availability of energy to the rest of the food web.

The dissolved inorganic, dissolved organic, and particulate forms of nitrogen and phosphorus were measured. The mean molar ratio of nitrogen to phosphorus was ~ 70 , suggesting phosphorus was the nutrient limiting phytoplankton growth throughout the study. Total nitrogen concentrations (mean = $5.9 \text{ mg } \text{N } \Gamma^1$) were always higher than phosphorus throughout the study. Nitrogen was found primarily as dissolved organic nitrogen (mean = $4.8 \text{ mg } \text{N } \Gamma^1$), concentrations of which varied little throughout the year. Ammonia (mean = $0.9 \text{ mg } \text{N } \Gamma^1$), nitrate (mean = $0.08 \text{ mg } \text{N } \Gamma^1$), and nitrite (mean = $0.007 \text{ mg } \text{N } \Gamma^1$) exhibited greater seasonal variation. Ammonia concentrations were up to $2.2 \text{ mg } \text{N } \Gamma^1$ in bottom waters during periods of stratification, whereas nitrate and nitrite were undetected. During fall and winter nitrate and nitrite were highest with ammonia concentrations at seasonal lows. Particulate nitrogen (mean = $0.25 \text{ mg } \text{N } \Gamma^1$) varied with phytoplankton abundance.

As in the case of nitrogen, total phosphorus (0.16 mg P l^{-1}) was primarily made up of the dissolved organic fraction. Orthophosphate concentrations (mean = 0.02 mg P l^{-1}) and dissolved organic phosphorus (mean = 0.1 mg P l^{-1}) exhibited little seasonal variation although orthophosphate vertical gradients did exist during periods of stratification. Particulate phosphorus (mean = $0.036 \text{ mg P l}^{-1}$) exhibited seasonal and vertical variation that followed changes in phytoplankton abundance. Nutrient loadings from the three main inflows to the Salton Sea were $1.6 \text{ g/m}^2/\text{yr}$ of phosphorus and $12 \text{ g/m}^2/\text{yr}$ of nitrogen.

Silicon, in the form of silica or glass is the main component of the cell walls of diatoms, one of the most abundant types of algae in the Sea. Concentrations of dissolved forms of silicon are high throughout the year with a mean concentration of 6.1 mg Si l^{-1} . Lowest silica concentrations occurred in surface waters during periods of stratification and high diatom abundance, but it seems likely that silicon levels never fall to levels low enough to be limiting to diatom populations.

Light regime (Progress Reports 1, 26)

The penetration of light into the Sea is of significance in that it determines to what depths photosynthesis can be carried out by both planktonic and benthic algae, and also in that it is determined in part by the abundance of plankton populations. The light regime of the Salton Sea has seasonal variability that is partly a function of changes in the abundance of phytoplankton, as measured by chlorophyll *a* concentrations.

Water transparency, as measured by depth to which a 20-cm black and white Secchi disk remained visible, increased during the warming period and then decreased during the cooling period. Differences among stations were found, but each station followed the same general pattern. The shallowest Secchi disk depths were recorded for a station several km downcurrent from the New River inflow, perhaps reflecting both the suspended silt brought in by the rivers or indirectly by the supply of nutrients, which may increase phytoplankton abundance. The pattern of light penetration as measured with the radiometer followed that of water transparency as determined with the Secchi disk.

Chlorophyll *a* concentrations increase during the warming period and then a decline during the cooling period. Large differences in chlorophyll *a* concentrations over depths were found on dates when the lake was stratified, while minimal differences were found when the lake was well mixed. Although the light regime of the Salton Sea is influenced by the abundance of phytoplankton, there is evidence that other dissolved and particulate materials are also very important in regulating the availability of light.

SURVEY FOR ALGAL TOXINS

(Progress Reports 2, 18, 16)

Algal toxins have been known to cause disease and mortality in fish, birds, mammals and even humans. Several groups contain species that are known to produce toxins including Prymnesiophyceae, Dinophyceae, Raphidophyceae and Bacillariophyceae. In response

to wildlife mortality including unexplained eared grebe (*Podiceps nigricollis*) dieoff events in 1992 and 1994 and other mortality events such as large fish kills, a survey was conducted for the presence of algal toxins in the Salton Sea. Goals of this survey were to determine if and when algal toxins are present in the Salton Sea and to describe the phytoplankton composition during those times.

A total of 25 samples were collected for toxicity analysis. Dinoflagellates and diatoms dominated most samples, but some were dominated by a prymnesiophyte (*Pleurochrysis pseudoroscoffensis*) or a raphidophyte (*Chattonella marina*). Several types of blooms were observed and sampled. The dinoflagellate *Gyrodinium uncatenum* formed an extensive, dense (up to 310,000 cells ml⁻¹) and long-lasting bloom during the winter. A coccolithophore, *Pleurochrysis pseudoroscoffensis*, was found in high densities (up to 260,000 cells ml⁻¹) in surface films during spring and summer. These surface films also contained high densities of one or two other species (an unidentified scrippsielloid, *Heterocapsa niei*, *Chattonella marina*). Localized blooms were also observed in the Salton Sea. Several species of *Gymnodinium* reached high densities (110,000 cells ml⁻¹) inside Varner Harbor, and an unidentified species of *Gymnodinium* formed a dense (270,000 cells ml⁻¹) band along part of the southern shoreline during the summer. Three known toxic species were among the phytoplankton sampled during this study. *Gonyaulax grindleyi* and *Chattonella marina* were found in several samples taken during summer months, and *Prorocentrum minimum* was found in low densities in several samples.

Most samples, including those containing known toxic species, showed a low level of activity in the brine shrimp lethality assay. All samples tested in the mouse bioassay showed no activity. ¹H NMR spectra from these samples did not indicate the presence of toxins. One sample taken from the bloom of *Gymnodinium* spp. was highly active in the brine shrimp lethality assay. Although the ¹H NMR spectrum from this sample was extremely complex, the active material was lost during fractionation and we were not able to isolate a toxin.

While dense algal blooms are common at the Salton Sea, no evidence gathered in this study suggests that algal toxins are responsible for past eared grebe mortality events. Blooms of phytoplankton, however, may contribute to wildlife mortality at the Salton Sea in other ways. Dissolved organic material released by phytoplankton can lower the surface tension of water. Because surface tension is a critical factor to the waterproofing properties of avian plumage, a change in surface tension may cause aquatic birds to become waterlogged. Eventually, the birds may die of hypothermia. This might explain the eared grebe dieoffs, but further research needs to be conducted on the effect of algal blooms on surface tension in the Salton Sea and on how changes in surface tension affect eared grebes.

CONCLUSIONS

This four-year project has tremendously expanded our understanding of the Salton Sea. Hundreds of species, mostly protozoans and algae, not previously known from the Sea have been found there. Some of these are completely new to science. We remain ignorant of the role of most of these species, old or new, in the Salton Sea ecosystem. Some are important by virtue of their abundance and their role in providing food resources to fish and birds in the upper part of the food web. Others are capable of producing substances that may be toxic to fish or birds, though our preliminary work did not document a single event of such intoxication. And still others are important as parasites or predators that have large effects on individual populations or the whole food web.

The lake is a wind-driven ecosystem, more so than would be the case with lakes much deeper or much shallower than the Sea. Year-to-year variations in the frequency and strength of wind events cause much corresponding variation in the biology and chemistry of the lake. No year is typical. Had we not initiated our studies two years prior to receipt of EPA funding our conclusions would likely have been a bit simplistic.

Wind is especially important in the summer and early fall when it determines when and to what degree anoxic, low quality bottom waters are mixed into the surface waters. When such mixing events occur there typically are massive mortalities of fish, plankton, and benthos. Anoxia at the sediment surface following the first prolonged periods of stratification in the spring likely is the cause for the absence of the pileworm, one of the most important members of the lake's food web, over most of the lake bottom during the warmer half of the year. And the first overturns, or mixing of bottom and surface waters, following the first periods of stratification in spring may be what drive most of the fish to the shallow nearshore waters for the spring and summer months.

Comparison of our results with those gathered in the 1950s by UCLA and California Department of Fish and Game researchers suggest that the dynamics of the lake's plankton and benthos have changed over the last half century. New species have appeared, the dynamics of other species have changed, and episodes of lethally poor water quality seem to have increased. Over this period of time, salinity has increased 25 percent, large amounts of additional nutrients have come into the lake, and a new fish species, tilapia, has invaded and become the most abundant fish in the Sea, exerting strong effects via its grazing and predation on the plankton and benthos. The separate influences of these factors are difficult to disentangle. It is well understood that the system will change greatly once it becomes too saline for fish. But we can also expect that as salinity rises gradually, other changes will take place in the lake long before the fish disappear. These short-term changes could be dramatic but they are much less predictable than those that would result from large salinity increases, e.g. one of 30-40 g/L.

Our understanding of Salton Sea ecology, of its likely future, and of possible impacts of engineering projects aimed at restoration would be facilitated by new mechanisms for attracting and supporting research efforts that better build on existing information than do current ones. Critical insights into what is going on at the Sea are not being taken advantage of. An open-topic RFP and award process would attract research proposals

that themselves would be great eye-openers for the EPA, Salton Sea Authority, and Salton Sea Science Office. Had this mechanism been in place earlier, we would be a couple of years ahead of where we are now, in the opinion of many.

The sort of process needed would be much like that of the National Science Foundation or the National Institutes of Health. At intervals (e.g. 4-6 months) SSA and SSSO could put out a call for proposals, for example, for analysis of any aspect of the Salton Sea system.

The RFP would indicate that proposals would be evaluated *on the basis of the proposed work being likely to provide information valuable to the lake's restoration and management*. A significant fraction, say 30-60 percent, of funds available for the applied research program might be allocated to such work. Depending on funding and on the number and quality of proposals received in response to an open topic RFP, anywhere from no to several proposals might be funded at each round in the process. Remaining funds would be for the usual narrowly focused, committee-defined, topic-specific work awarded to single contractors via the usual RFP process.

Whatever the obstacles may be to implementing open topic RFPs, they should be overcome. There is no question they will get SSA "more bang for the buck."

ACKNOWLEDGMENTS

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SCIENTIFIC PUBLICATIONS FROM THE BIOLOGICAL LIMNOLOGY RECONNAISSANCE

Papers already published

Kuperman, B. I., and V. E. Matey. 1999. Massive infestation by *Amyloodinium ocellatum* (Dinoflagellida) of fish in a highly saline lake, Salton Sea, California, USA. *Diseases of Aquatic Organisms*. 39:65-73.

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Reifel, K. M. M. P. McCoy, M. A. Tiffany, T. E. Rocke, C. C. Trees, S. B. Barlow, D. J. Faulkner and S. H. Hurlbert. 2001. *Pleurochrysis pseudoroscoffensis* (Prymnesiophyceae) blooms on the surface of the Salton Sea, California. (*Hydrobiologia* (in press)).

Tiffany, M. A., S. B. Barlow, V. E. Matey and S. H. Hurlbert, 2001. *Chattonella marina* (Raphidophyceae), a potentially toxic alga in the Salton Sea, California. *Hydrobiologia*, in press).

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Small, E. B., G. F. Gebler, and J. C. Cole. 2001. A survey of the Ciliophora (Protista) of the Salton Sea, CA, USA. *Hydrobiologia* (in press).

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Tiffany M. A., B. Swan, J. Watts, and S. H. Hurlbert, 2001. Metazooplankton dynamics of the Salton Sea 1997-1999. *Hydrobiologia* (in press).

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Wood, A. M., S. M. Miller, S. Augustine, and R. W. Castenholz. Fine structure and spectral analysis of an unusual free-living marine cyanobacterium which contains chlorophyll-d as its major photosynthetic pigment.

QUALITY ASSURANCE REPORT

Quality assurance protocols were carried out to check the accuracy of identifications and counts of organisms in samples of the benthos, phytoplankton, and zooplankton. This entailed having subsets of samples recounted by qualified persons other than the person who made the original accounts.

Detailed tabulations showing original counts and recounts for all samples so treated have been presented in Progress Reports as follows: benthos (Progress Report 13, Appendix C), phytoplankton (Progress Report 25, Appendix C), zooplankton (Progress Report 20, Appendices A and E). Below we summarize the results of these procedures.

Benthos

In the Quality Assurance Project Plan for this project, we specified that a count would be considered acceptable if the recount value (R) fell within an acceptable range (A) of values about the value of the original count (C). These acceptable ranges were determined as follows:

- 1) For each species where $C > 50$, then $A = 0.5C < R < 2C$
- 2) For each species where $50 > C > 20$, then $A = 0.32C < R < 3.2C$
- 3) For each species where $20 > C > 5$, then $A = 0.2C < R < 5C$
- 4) For no species where $C = 0$ is $R > 5$; for no species where $R = 0$ is $C > 5$

In all benthic habitats sampled, 5-6 taxa could be represented. Of the 457 samples obtained, 55 were selected for QA/QC. There were approximately 275 individual recounts of individual taxa. Following the initial counts made after each sampling date, at least 10% of the samples were randomly picked and recounted by an individual in the benthic research group who did not count the sample originally, and who did not know the original count results until after the recount was completed.

A total of 325 benthic grab samples from offshore sediments were obtained between January and November 1999, and 31 samples were processed for QA/QC.

A total of 18 macroplankton tow samples were obtained between January and November 1999. One of the triplicate samples (6 samples or 33%) obtained on each sampling date was randomly selected and recounted by a second enumerator on the benthic team.

Five quadrat samples were obtained on two different rocky substrates on each sampling date, for a total of 60 samples over the sampling period. One replicate of each set of samples (12 samples or 20%) collected from each substrate type was randomly picked and recounted by a second enumerator.

Triplicate samples of barnacle shell substrate were collected at each of three littoral stations on each sampling dates, yielding 54 samples over the sampling period. One sample from each sampling date (6 samples) was randomly selected for recount by a second enumerator.

In all cases, recounts were within the specified acceptable range. For the 220 recounts conducted on benthic and shoreline samples (exclusive of algal and barnacle quadrats), there were 15 instances (6.8%) where recounts differed from original counts, and in all cases only by <2 individuals. Similarly, for the 60 recounts of algal and barnacle quadrat samples, all were within the specified acceptable range. On two occasions, recounts of *Balanus* were 10-20% lower than the original count. These discrepancies occurred because most of the barnacle shells had to be cracked open during the initial count to verify whether the organisms were alive at time of sampling. This procedure destroyed smaller barnacles growing on top of larger ones, and those former were absent from the recount. Nonetheless, all of the variations in counts met the QA/QC requirements.

Phytoplankton

A total of nine samples, which equated to 261 individual recounts of individual taxa (given that 29 taxa constituted the assemblage enumerated), were selected for conducting recounts to assess the reliability of the numerical abundance data. These samples were randomly chosen to be enumerated by a second individual. This second enumerator had previous experience with both the phytoplankton species found in the Salton Sea and the specific counting procedures employed for such samples.

Acceptable ranges for recount values were defined exactly as they were for the benthos data (above). For the 261 recounts conducted there were 25 instances where the recount lay outside the specified acceptable range. Some recounts were only slightly outside the range and in most cases, small-sized, difficult to see and identify taxa were involved. These instances represent 9.6 percent of the total recounts conducted. In every case, the effect on figures showing population changes, of using the re-count value instead of the original value was so small as to have no visible effect on the trends shown.

Zooplankton

A total of 60 zooplankton samples were re-enumerated to check the accuracy of the abundance estimates for each species or groups of species. There were ten species that were enumerated individually in each sample. Therefore a total of 600 recounts were conducted.

Acceptable ranges for recount values were defined exactly as they were for the benthos data (above). There were eight instances where the recount lay outside the acceptable range specified for a count. These instances represented 1.3 percent of the total recounts conducted. In every case, the effect on figures showing density variations, of using the recount value instead of the original value was so small as to have no effect on the trends shown.

Based on all the above findings, we conclude that for all three groups of organisms, counts were in general of acceptable quality and that the stochastic factors producing discrepancies between original counts and recounts are without significance for the findings and conclusions of our reports.

**MBE/WBE REPORTS
(1998 - 2001)**

OMB CONTROL NO. 2830-
APPROVED: 9/12/96
APPROVAL EXPIRES: 9/30/98

**U. S. ENVIRONMENTAL PROTECTION AGENCY
MBE/WBE UTILIZATION UNDER FEDERAL GRANTS, COOPERATIVE
AGREEMENTS, AND INTERAGENCY AGREEMENTS**

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PART 1 (NEGATIVE REPORTS MAY BE REQUIRED)

1A. FEDERAL FISCAL YEAR 19 <u>98-99</u>		1B. REPORTING QUARTER (Check appropriate box) <input checked="" type="checkbox"/> 1st (Oct-Dec) <input type="checkbox"/> 2nd (Jan-Mar) <input checked="" type="checkbox"/> 3rd (Apr-Jun) <input type="checkbox"/> 4th (Jul-Sep) <input checked="" type="checkbox"/> Annual	
2. FEDERAL FINANCIAL ASSISTANCE AGENCY (Department/Agency, Bureau/Administering Office, Address) SALTON SEA AUTHORITY 78-035 CALLE ESTADO LA QUINTA, CA 92253		3. REPORTING RECIPIENT (Name and Address) SAN DIEGO STATE UNIVERSITY FOUNDATION 5250 CAMPANILE DRIVE SAN DIEGO, CA 92182-1947	
2A. REPORTING CONTACT TOM KIRK	PHONE: 716-564-4888	3A. REPORTING CONTACT ALICIA G. VALLEJO	PHONE: (619) 594-5769
4A. FINANCIAL ASSISTANCE AGREEMENT ID NUMBER Document dated 11/20/98		4B. FEDERAL FINANCIAL ASSISTANCE PROGRAM N/A	
5A. TOTAL GRANT AMOUNT \$746,471.00		5B. TOTAL CONTRACT/PROCUREMENT AMOUNT THIS QUARTER \$ N/A	
		5C. RECIPIENT'S MBE/WBE GOALS MBE 14.9 % WBE 9.6	
5D. ACTUAL MBE/WBE PROCUREMENT ACCOMPLISHED THIS REPORTING PERIOD MBE \$ 38.12 WBE \$ 0.00		5E. NEGATIVE REPORT (Check) <input type="checkbox"/> SEE INSTRUCTIONS	
6. COMMENTS DUE TO THE NATURE AND TYPE OF EQUIPMENT ORDERED FOR THIS PROJECT, WE WERE UNABLE TO SECURE PROSPECTIVE MBE/WBE SUPPLIERS. A SEARCH WAS DONE IN THE SMALL BUSINESS SOURCE DIRECTORY, PUBLISHED BY GOVERNMENT DATA PUBLICATIONS; THE WOMAN'S BUSINESS DIRECTORY AND RESOURCE GUIDE, PUBLISHED BY YMCA OF SAN DIEGO COUNTY; AND THE MINORITY BUSINESS COUNCIL WAS CONTACTED FOR PROSPECTIVE SUPPLIERS. THEY HAD NONE REGISTERED IN THE AREAS OF NEED.			
7. NAME OF AUTHORIZED REPRESENTATIVE MICHELE G. GOETZ		TITLE DIRECTOR, RESEARCH AND COMMUNITY SERVICES ADMINISTRATION	
8. SIGNATURE OF AUTHORIZED REPRESENTATIVE		DATE	

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U. S. ENVIRONMENTAL PROTECTION AGENCY
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AGREEMENTS, AND INTERAGENCY AGREEMENTS

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 50870A/50871A/50872A/50873A SJT

PART 1 (NEGATIVE REPORTS MAY BE REQUIRED)

1A. FEDERAL FISCAL YEAR 00-01		1B. REPORTING QUARTER (Check appropriate box) <input type="checkbox"/> 1st (Oct-Dec) <input type="checkbox"/> 2nd (Jan-Mar) <input type="checkbox"/> 3rd (Apr-Jun) <input type="checkbox"/> 4th (Jul-Sep) <input checked="" type="checkbox"/> Annual	
2. FEDERAL FINANCIAL ASSISTANCE AGENCY (Department/Agency, Bureau/Administering Office, Address) SALTON SEA AUTHORITY 78-401 HIGHWAY 111, STE T LA QUINTA, CA 92253		3. REPORTING RECIPIENT (Name and Address) SAN DIEGO STATE UNIVERSITY FOUNDATION 5250 CAMPANILE DRIVE SAN DIEGO, CA 92182-1947	
2A. REPORTING CONTACT LINDA QUESNELL	PHONE: 760-564-4888	3A. REPORTING CONTACT ALICIA G. VALLEJO	PHONE: (619) 594-5769
4A. FINANCIAL ASSISTANCE AGREEMENT ID NUMBER Document dated 11/20/98		4B. FEDERAL FINANCIAL ASSISTANCE PROGRAM N/A	
5A. TOTAL GRANT AMOUNT \$746,471.00		5B. TOTAL CONTRACT/PROCUREMENT AMOUNT THIS QUARTER \$ N/A	
		5C. RECIPIENT'S MBE/WBE GOALS MBE 14.9 % WBE 9.6 %	
5D. ACTUAL MBE/WBE PROCUREMENT ACCOMPLISHED THIS REPORTING PERIOD MBE \$ 0.00 WBE \$ 0.00		5E. NEGATIVE REPORT (Check) <input type="checkbox"/> SEE INSTRUCTIONS	
6. COMMENTS DUE TO THE NATURE AND TYPE OF EQUIPMENT ORDERED FOR THIS PROJECT, WE WERE UNABLE TO SECURE PROSPECTIVE MBE/WBE SUPPLIERS. A SEARCH WAS DONE IN THE SMALL BUSINESS SOURCE DIRECTORY, PUBLISHED BY GOVERNMENT DATA PUBLICATIONS; THE WOMAN'S BUSINESS DIRECTORY AND RESOURCE GUIDE, PUBLISHED BY YMCA OF SAN DIEGO COUNTY; AND THE MINORITY BUSINESS COUNCIL WAS CONTACTED FOR PROSPECTIVE SUPPLIERS. THEY HAD NONE REGISTERED IN THE AREAS OF NEED. SUBCONTRACTORS SUBMITTED MBE/WBE REPORT AT THE END OF THE AWARD DATE; HOWEVER, NO MBE/WBE TO REPORT.			
7. NAME OF AUTHORIZED REPRESENTATIVE MICHELE G. GOETZ		TITLE DIRECTOR, RESEARCH AND COMMUNITY SERVICES ADMINISTRATION	
8. SIGNATURE OF AUTHORIZED REPRESENTATIVE		DATE	

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