RECENT BROWN TIDE ACTIVITY

Between August and December 2002 (the most current brown tide counts from Suffolk County), brown tide counts for sampled stations in the Peconic Estuary remained below 2,000 cells per milliliter. In August in Great South Bay, brown tide cell counts reached a high of 8,700 cells per milliliter at one sampling station then remained below 2,200 cells per milliliter through December with the majority of counts measuring below 1,000 cells per milliliter.

Thus far for 2003, there are no indications of brown tide bloom activity in the Peconic Estuary or South Shore Estuary.

Editor's Note

The following project updates summarize the final results of the three BTRI projects that ran from 1999-2001 (see BTRI Reports #5-7 for more details and previous updates: http://www.seagrant.sunysb.edu/BTRI/btripublications.htm).

The BTRI program, administered by New York Sea Grant (NYSG), is not the only organization funding brown tide research on Long Island. The Ecology and Oceanography of Harmful Algal Blooms (ECOHAB) program, New York Sea Grant and Suffolk County are supporting multiple brown tide projects that complement the BTRI and other past brown tide related efforts. A few of these projects are summarized in this issue.

BTRI Report #8 builds on the preceding 7 BTRI Reports and follows a similar format as the previous issues for easy project tracking.

Boldface terms are defined under Key Terms, adding to those defined in the earlier reports.

WHAT'S NEXT

Synthesizing the results of the past five years of laboratory and fieldwork continues to be a priority for BTRI. To this end, NYSG, in collaboration with the BTRI Steering Committee, put together a team of researchers to synthesize results of various brown tide projects since 1997. The synthesis team includes Drs. Darcy Lonsdale, (pictured) Stony Brook University; Gregory Boyer, SUNY College of Environmental Science and Forestry; and Christopher Gobler, Long Island University, Southampton College. This team is well on its way to producing an article for a peer reviewed journal. Once this synthesis document is accepted for publication, a version written for a non-scientific audience will appear as BTRI Report #9 and coincide with the last BTRI Public Symposium (date to be determined).
BTRI Projects 1999-2001

Kana, MacIntyre, Cornwell & Lomas: Benthic-Pelagic Coupling and Long Island Brown Tide

It is hypothesized that brown tide events are related to the availability of dissolved organic nutrients in the water. A potential source of dissolved organic compounds is the sediment. Sediments were suspected to play a critical role in regulating the quantity and type of nutrients found in the water column that are available to the plankton community including brown tide, (Aureococcus anophagefferens.) This study documented seasonal changes in phytoplankton nutrients in the water column, the uptake and release of nutrients by the sediments, the amount and activity of primary producers associated with the sediment layer and water column, and the occurrence of brown tide.

Two east end bays, Quantuck Bay and Flanders Bay were sampled during the summers of 2000 and 2001. However, a brown tide event (300,000 cells per milliliter) was observed only in June of 2000 in Quantuck Bay. Results indicated a shift in the relative control of primary production from the algae growing on the bay bottom (benthos), called microphytobenthos, to the phytoplankton in the water column (pelagic) around the time of the brown tide event. The shift in primary production from the benthos to a pelagic dominated system correlated with a change in the processing of nutrients by the sediment. In spring of 2000, nitrogen was released from the sediment predominantly as dissolved inorganic nitrogen (DIN), whereas in the summer, when A. anophagefferens bloomed, nitrogen from the sediment was predominantly released as dissolved organic nitrogen (DON). This demonstrated a change in the way the benthos processed sediment nutrients. These results support the idea that organic nutrients play an important role in the dynamics of A. anophagefferens in Long Island bays and that there can be interplay between processes that occur in the sediment and processes that occur in the water column (benthic-pelagic coupling). Mesocosm experiments conducted in 2001 (a non-brown tide year) also showed the importance of organic nutrients. Aureococcus anophagefferens grew only in the organic nutrient additions treatments. This supports previous research showing that A. anophagefferens blooms when DON levels are high.

BACKGROUND

The Brown Tide Research Initiative (BTRI) is funded by the National Oceanic and Atmospheric Administration's Coastal Ocean Program and administered by New York Sea Grant. The first three-year (1996-1999) $1.5 million BTRI program was developed to increase knowledge concerning brown tide by identifying the factors and understanding the processes that stimulate and sustain brown tide blooms. Continued funding for BTRI came from NOAA's COP as a $1.5 million three-year effort (1999-2001) effort. The COP National Sea Grant Office, National Science Foundation, Environmental Protection Agency, office of Naval Research, and National Aeronautics and Space Administration are jointly sponsoring research on Harmful Algal Blooms (HAB) ecology and oceanography in the interagency research program, Ecology and Oceanography of Harmful Algal Blooms (ECOHAB).

There were eight projects in the first three-year effort, and three projects in the next three-year effort. All BTRI projects were selected from national calls for proposals. The research projects chosen for BTRI funding were selected following peer review and evaluation by a technical review panel. To involve concerned parties and aid in decision-making, New York Sea Grant also formed the BTRI Steering Committee of invited state, local and government agency representatives, and citizen's groups to evaluate the programmatic value of the proposals. This Report Series will aid in the dissemination of general brown tide information. The results and conclusions of the projects will help determine the directions of potential management and future research.
Lonsdale, Caron & Cerrato:
Causes and Prevention of Long Island Brown Tide

This team has proposed that a central issue in the initiation of brown tides involves a shift in the group of organisms that controls the food web (species composition and abundance). The dominant group is comprised of either the organisms on the bottom, such as hard clams (Mercenaria mercenaria), or the organisms in the water column (pelagic microbial grazers), such as protozoa.

Through their suspension-feeding activity, bivalves (e.g., hard clams) can exert significant grazing pressure on phytoplankton, particularly in shallow waters. Prior to the 1985 brown tide outbreak, there was a drastic decline in the hard clam population. This resulted in approximately a ten-fold decrease in their grazing impact on phytoplankton. Some microbial grazers, such as protozoa, may prefer to consume phytoplankton other than Aureococcus anophagefferens, whereas suspension-feeding bivalves, however, may be relatively non-selective and feed on whatever phytoplankton is currently in the water column. This included A. anophagefferens at low abundance. The reduction in the hard clam population and their concomitant planktonic grazing has led this team to hypothesize that A. anophagefferens benefits from this shift from benthic suspension-feeding to microbial grazing pressure in the water column.

This team tested the hypothesis that the presence of significant grazing pressure by a hard clam population prevents phytoplankton from reaching bloom conditions, and prevents a shift in the phytoplankton community to a brown tide. Using 300-liter experimental mesocosm tanks containing a natural mixture of plankton species, they have found repeatedly that the density of hard clams can play a pivotal role in determining whether or not blooms of A. anophagefferens become established. The magnitude of hard clam filtration pressure needed to prevent brown tide from developing in the mesocosm tanks was similar to hard clam population estimates for Great South Bay two decades ago. The researchers suggest, however, that the decrease in bivalves may only be an important pre-condition for brown tide. Light, temperature and nutrient conditions would also have to be right for a brown tide to occur.

This team concludes that the feeding activities of hard clams in shallow bays could exert considerable control on total phytoplankton biomass in the overlying water column, and specifically on the ability of A. anophagefferens to dominate the phytoplankton community and form brown tides. Moreover, the restoration of these bivalve populations may constitute a feasible mechanism for improving water quality and preventing brown tides.

Figure 2:
The presence of brown tide on Great South Bay during the summer of 2001 had little impact on recreational activities. [Photo by Branca]
Other Brown Tide Work/Projects

**ECOHAB**

(ECology and OceAnography of Harmful Algal Blooms)

ECOHAB provides a scientific framework designed to increase our understanding of the fundamental processes underlying the impacts and population dynamics of Harmful Algal Blooms (HABs). The overall objective of ECOHAB is to investigate fundamental physical, biological, and chemical oceanographic questions critical to scientifically based management of fisheries resources, public health, and ecosystem health in regions threatened by toxic and harmful algae.

**Giner & Wikfors:**

HAB Sterols and Their Effects on Shellfish and Crustaceans

You probably don’t need any extra cholesterol in your diet, but clams and scallops might. Cholesterol is a requirement for all animals. Unlike people, mollusks and crustaceans are unable to make their own cholesterol de novo, but have to get it from the food they eat. Most of the algae they consume do not contain cholesterol, but shellfish are equipped to make cholesterol out of the typical phytosterols found in most algae. Many harmful algae, however, do not contain typical phytosterols. These algae contain unusual sterols that are very different in their chemical structure from cholesterol. Although unusual sterols have been identified for many years, their function was not known. This team recently proposed that unusual sterols benefit the algae because they cannot be converted into cholesterol. If the animals that eat the algae cannot get enough cholesterol, they will not grow and eat more algae. By interfering with the cholesterol requirements of invertebrate animals, the
unusual sterols protect the algae from their predators; the algae thrive and form harmful algal blooms (HABs). Examples of unusual sterols discovered in Giner’s lab come from the brown tide algae (*Aureococcus anophagefferens*), as well as from toxic red tide organisms (*Karenia breve*).

This new hypothesis is being examined in Giner’s laboratory at SUNY College of Environmental Science and Forestry (see Figure 3). Chemical synthesis has provided a way of getting sufficient amounts of harmful algal sterols so that their effects can be tested. Studies of the responses of shellfish to these sterols are being carried out in collaboration with Wikfors at the NOAA Fisheries Lab in Milford, Connecticut. This research will lead to a better understanding of the nutritional requirements of economically and ecologically important invertebrates. It is hoped that methods to mitigate the effects of harmful algae will be developed, such as providing supplementary cholesterol to enable the growth of shellfish and crustaceans during bloom events. The information gained in these studies will also be important for the utilization of HAB sterols as food chain and environmental biomarkers, and as biomarkers for HAB paleochronology.

(Text written by Giner and modified by Dooley.)

**Figure 3:**
Dr. José Giner (center) and SUNY-CESF chemistry graduate students Hui Zhao and Ming He.
[Photo courtesy of Giner]
Mulholland & Minor:
Nutritional Factors Promoting the Growth and Dominance of Aureococcus anophagefferens in Coastal Waterways

Dissolved organic nitrogen (DON) has been implicated as a causative agent in the formation of brown tides. DON may not serve strictly as a nitrogen source, however, as A. anophagefferens can also use organic carbon to supplement carbon produced during photosynthesis. This project is aimed at understanding how the role of dissolved organic matter can influence the growth and establishment of brown tide blooms. This research team (see Figure 4) is comparing physically similar coastal sites in Maryland and Virginia to determine why blooms form in some areas and not others. In Chincoteague Bay, there are a number of sites in MD and VA that experience intense seasonal brown tide blooms. In both 2002 and 2003, blooms in the Bay were in excess of 1 million cells per milliliter. The researchers found that high dissolved organic carbon concentrations are characteristic of areas experiencing blooms. It is still not clear whether blooms result from the high dissolved organic carbon concentrations or contribute to them. In addition, in 2002, they found that photosynthetic carbon uptake could not supply the carbon demand of cells as estimated from nitrogen uptake. During the day, organic carbon uptake did not contribute enough additional carbon to fill this gap. This year, the team conducted a series of diel studies to determine whether dissolved organic carbon taken up during the dark cycle could supply the missing carbon. The team is currently analyzing those results.

In contrast to a previous study in Quantuck Bay on Long Island in 2000, but consistent with studies in Shinnecock Bay, Long Island in 1995, researchers found that urea was the dominant nitrogen source fueling the 2002 bloom in Chincoteague Bay. However, most of the urea carbon was not assimilated suggesting that urea is not a good carbon source for A. anophagefferens. In Quantuck Bay, ammonium and amino acids contributed most of the nitrogen to support the growth of A. anophagefferens. Amino acid carbon and nitrogen both contributed substantially to the carbon and nitrogen nutrient of the bloom in Quantuck Bay but contributed little to the nitrogen nutrition in Chincoteague Bay during 2002. In Quantuck Bay, high rates of peptide hydrolysis may have facilitated the production of amino acids or small peptides that could be used for growth. In Chincoteague Bay, peptide hydrolysis rates were much lower, either because there were inappropriate substrates for hydrolysis or because hydrolysis was inhibited by the presence of some other compound (e.g., urea or some other source of organic carbon). It appears that A. anophagefferens can exploit different nitrogen sources to support its growth but that organic carbon compounds are required to compensate for insufficient photosynthetic carbon acquisition during the day.

(Text written by Mulholland and modified by Dooley.)

Figure 4:
Mulholland’s research team at work at the Virginia Institute of Marine Science’s Eastern Shore Laboratory in Wachapreague, VA. From left to right are: Michelle Watson, Pete Bernhardt, Dr. Margaret Mulholland, Esther Cornfeld, George and Andrea Rocha. [Photo courtesy of Mulholland]
The potential for viral activity to serve as a control on population densities of the bloom forming alga, *Aureococcus anophagefferens*, will be evaluated to contribute to the knowledge of the complexity and dynamic interplay of the virus-host interaction, and to an understanding of why these algae continue to bloom in certain areas at certain times and then quickly disappear. The hypothesis being tested is that viral infection will account for greater mortality in blooms of natural populations of *A. anophagefferens* than other environmental or biological factors (e.g., grazing, water quality parameters, etc.). The objectives of this study are to: 1) determine the frequency that viral-like particles infect and lyse natural populations of *A. anophagefferens* in coastal bays of New York and New Jersey. 2) Isolate viruses specific to *A. anophagefferens* and establish baseline information on the genetic diversity of viruses that infect *A. anophagefferens*. 3) Determine the influence of viral activity on the proliferation of *A. anophagefferens* and in situ bloom termination.

The combined results of this study and previous studies clearly show continuing evidence of a persistent viral infection of natural populations of *A. anophagefferens* (see Figure 5) occurring over a regional geographic range over several bloom years. The sampling frequency in 2002 provided a better definition of the different stages of the bloom in Little Egg Harbor, NJ. The transmission electron microscopy results clearly characterized the percentage of viral-like particles infected *A. anophagefferens* in natural populations throughout the bloom period. Although these results also corroborated previous studies, this team’s research provided additional evidence of an increased percentage of infection by viral-like particles infection at the end of the bloom.

The laboratory results confirmed the lytic activity of the viral isolates from New Jersey and New York waters that were specific to *A. anophagefferens in situ*. Transmission electron microscopy results confirmed the presence of intracellular viral-like particles in healthy cultures infected with the lytic virus. These results corroborated previous studies of re-infection experiments of viral isolates from New York waters. The transmission electron microscopy results showed that at least two types of viruses were present in unpurified viral isolates that caused the lysis of some healthy cultures of *A. anophagefferens*. Results will be published in *Estuaries*. (Text written by Gastrich and modified by Dooley).

**Figure 5:**
Hard clams, *Mercenaria mercenaria*, have been an important resource in Great South Bay, NY for decades despite a precipitous decline in abundance in recent years. One suspected cause of this decline is recurring brown tides, *Aureococcus anophagefferens*. Brown tide has been shown to be harmful to several shellfish species by causing gill structures to stop pumping water, resulting in a slowing or cessation of feeding activity. In this study, marked hard clams were planted in Great South Bay for two years to determine the effects of brown tide on their growth. Growth and chemical isotope analyses of shells recovered from the mark-recovery experiment and archived shells obtained from the Town of Islip confirmed the presence of daily growth increments during the growing season and the formation of growth bands in winter and summer that can be used to determine age (see Figure 6). During the first year of the mark-recovery experiment, a brown tide at a concentration above that known to cause growth cessation in laboratory studies (35,000 cells per milliliter) was present during 88% of the growing season. In the second year, brown tide was present above the effects concentration only during 6.5% of the growing season. Clams grew comparable amounts between the two years of the mark-recovery experiment and also relative to studies from years prior to brown tides. Archived shells from the Town of Islip’s annual shellfish surveys showed no differences in shell growth between brown tide and pre-brown tide years. While no relationship was observed between brown tide concentration and shell growth, a strong relationship was observed with temperature.

Unlike the case for other shellfish such as bay scallops and mussels, extended exposure to brown tide did not appear to have a large negative effect on the growth of hard clams in Great South Bay.

The lack of a severe brown tide effect on adult hard clam growth suggests that some restoration efforts can be attempted in Great South Bay without concern for brown tide effects on adult growth.

(Text written by Cerrato and modified by Dooley.)
Giner & Zhao:  
Investigation of the Past Occurrence of Brown Tides by Sediment Analysis for Specific Sterol Biomarkers

The rare sterol 24-propylidenecholesterol is found in the brown tide alga Aureococcus anophagefferens, making it a potential chemical marker for this organism. The existence of this biomarker makes it possible to look for A. anophagefferens in sediments that were deposited over the course of centuries at the bottom of Long Island bays. Its detection could answer how long the brown tide have been present in these waters. However, it was uncertain whether this sterol is really a valid biomarker for A. anophagefferens. To find out, all of the strains of A. anophagefferens that are stored at Bigelow Laboratory were analyzed by this team (see Figure 7), and all of the other examples from the same class of algae were also analyzed.

Results showed that all of the A. anophagefferens strains contained 24-propylidenecholesterol, and all of them contained the two chemical isomers in about a 3:1 ratio. Most of the other algae contained one or the other of the two isomers. The 3:1 ratio of 24-propylidenecholesterols is therefore a valid biomarker for A. anophagefferens. In the analysis of seawater samples taken during brown tides, the researchers found the same ratio of the biomarkers to be present. The same ratio was also found in the top layer of mud that settles to the bottom of the bay. Sediments going down 70 cm that had been deposited over the past 700 years were taken and cut into 20-year sections. Analysis of the sterols showed that the A. anophagefferens biomarkers were clearly present over the past 120 years. The most likely conclusion is that the brown tide alga has been present in Long Island waters for at least 120 years.

(Text written by Giner and modified by Dooley).

Figure 7:  
Dr. José Giner and SUNY-CESF chemistry graduate students Hui Zhao. [Photo by Dooley]

Suffolk County

Taylor, Sañudo-Wilhelmy & Gobler:  
Influence of Groundwater Constituents on Initiation of the Brown Tide in the Peconics Bay System

In this Suffolk County Office of Ecologysponsored project, researchers are examining the effects of submarine groundwater discharges into shallow bays on relative densities of A. anophagefferens and co-occurring algal species. For example, growth of A. anophagefferens cultures is inhibited by higher exposures to Flanders Bay submarine groundwater discharges and not influenced by West Neck Bay submarine groundwater discharges (Figure 8).

Continued on page 10

Figure 8:  
Example of bioassay results. Cultures of the brown tide organism, Aureococcus anophagefferens, and a diatom, Thalassiosira pseudonana, were exposed to increasing concentrations of groundwater from Flanders (FB) and West Neck Bays (WNB) while maintaining constant salinity at 27‰. Growth rates were calculated from maximum slope of growth curves to yield divisions per day.
In contrast, growth of the diatom, Thalassiosira pseudonana, is stimulated by Flanders Bay submarine groundwater discharges and inhibited by higher exposures to West Neck Bay submarine groundwater discharges. Causes for these varying responses are likely to be complex and multi-factorial. The researchers are examining whether biocides carried by groundwaters select for or against proliferation of A. anophagefferens or its competing phytoplankton species. Results clearly suggest that groundwater from different sources can vary selective pressures on phytoplankton species and influence their relative abundances (see Figure 8).

In the laboratory, this team is exposing cultures of A. anophagefferens with four co-occurring phytoplankton species (diatom, dinoflagellate, cyanobacterium and chlorophyte) to varying concentrations of pesticides, submarine groundwater discharge samples, metals or macronutrients to assess effects on growth performance. In the field, the team is examining relationships between brown tides, trace metals, rates of groundwater discharge and nutrient chemistry. (Text written by Taylor and modified by Dooley).

**BIBLIOGRAPHY**


*Published papers and graduate student theses from BTRI and other NYSG funded brown tide projects since 1996.*
KEY TERMS

For a complete running list of all Key Terms in the BTRI Report Series, visit: http://www.seagrant.sunysb.edu/BTRI/britems.htm

alga(e) Primitive, often aquatic, plants that carry on photosynthesis but lack the flowers, roots, stems, and leaves of higher plants.
amino acids The building blocks for the synthesis of proteins. Can be a source of nitrogen and/or carbon.
ammonium An ion, NH4+, derived from ammonia by combination with a hydrogen ion that can be an inorganic source of nitrogen.
assimilate Conversion of nutritive materials into a living organism.
benthic-pelagic coupling The interaction between the benthos or bottom, with the water column, or pelagic ecosystem. It refers to how the dynamics of one ecosystem influences the dynamics of the other.
benthos The floor of a sea or ocean; also includes the bottom-dwelling organisms that live there.
biodiversity Chemicals used to kill living organisms (e.g., pesticides, algacides).
biomarker A chemical compound produced by a specific organism that can be used as an indicator for the presence of that organism.
chlorophyte A green alga.
crustacean Any mainly aquatic arthropod usually having a segmented body and chitinous exoskeleton such as lobster, shrimps, crabs, woodlice, water fleas and barnacles.
cyanobacteria Blue-green bacteria, sometimes called blue-green algae.
dark cycle (or Calvin Cycle) The portion of photosynthesis that does not require light.
diatom Single-celled algae, mostly photosynthetic, that form silica cell walls, can grow singly in chains or in simple colonies.
diel A 24-hour period.
DIN Dissolved inorganic nitrogen (e.g., nitrate, nitrite and sometimes ammonium).
dinoflagellate A single-celled organism found in fresh and marine waters with characteristics of both plants (e.g., photosynthesis) and animals (e.g., uses outside organic sources of nutrition). Many harmful algae blooms are caused by dinoflagellates.
dissolved organic matter Dissolved organic compounds ranging from macromolecules to low molecular weight compounds such as simple organic acids and short-chained hydrocarbons.
DON Dissolved organic nitrogen, (e.g., urea).
ECOHAB Ecology and Oceanography of Harmful Algal Blooms, an interagency research effort headed by NOAA’s Coastal Ocean Program.
eukaryotic A cell with a distinct membrane-bound nucleus.
graze To feed by browsing on, cropping, or eating.
growth rate Increase in the number of individuals in a population per unit time.
HAB Harmful algal bloom.
hydrolysis Chemical reaction of a compound with water, usually resulting in the formation of one or more new compounds.
hypothesis An idea or statement that must be tested before it can be stated as fact.
in situ In the original location (e.g., water column or within the organism).
invertebrates Animals lacking a backbone and internal skeleton.

lytic or lysis The process of disintegration or destruction of a cell.
mesocosm Experimental apparatus or enclosure designed to approximate natural conditions, and in which environmental factors can be manipulated.
micrometer (mm) or micron One millionth of a meter (1 inch = 25,400 mm). 1 millimeter = 1,000 microns.
mollusk A group of invertebrate animals including the snail, clam and octopus, that are characterized by the presence of an internal or external calcium shell.
nutrient Substances such as nitrogen and phosphorus, used by organisms to grow.
paleochronology A chronology of ancient events based on information obtained from geology and fossils.
pelagic Open water that is above the bottom and below the surface.
peptide A compound of two or more amino acids joined by peptide bonds. Proteins are formed by the linkage of many peptides.
peptide hydrolysis The splitting of a peptide compound molecule (protein or polypeptide) by the addition of water.
photosynthesis The physicochemical process by which plants, algae and some bacteria can utilize the energy of sunlight to power the biosynthesis of organic molecules, using carbon dioxide as the carbon source.
phytoplankton Microscopic, photosynthetic plants that are suspended in the water column.
phytosterol Plant sterols found in all living plants and are natural constituents of the human diet.
picoalgae Very small, single-celled planktonic algae in a size range of 0.2 – 2.0 microns.
picoalgae niche hypothesis The hypothesis suggests a succession from larger to smaller algal cells in Long Island bays between April and May. Typically, Synechococcus dominates the smaller picoalgae size class; however, if Synechococcus is selectively removed or its density is reduced, the picoalgae niche opens for some other similar sized algae, such as Aureococcus anophagefferens.
plankton Organisms, both plant and animal, that are suspended in the water column and transported by tides and current.
primary producers An organism that uses light to synthesize new organic material from carbon dioxide and water, also called autotrophic autotroph.
primary productivity The total amount of new organic matter produced by photosynthesis.
protist A group of simple organisms not distinguished as animals or plants, though having some characteristics common to both.
treatment Controlled technique or action applied in a specified process or experiment.
urea An organic waste product that can be used by phytoplankton a source of nitrogen.

NOTE:
This glossary was compiled with input and definitions from an assortment of sources:

And the Waters Turned to Blood (1997), by Rodney Barker.
Various BTRI investigators & Steering committee members.
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If you have any questions about brown tide, would like a copy of Report #1, 2, 3, 4, 5, 6 or 7, or would like to be added to our mailing list, please contact Patrick Dooley at New York Sea Grant (patrick.dooley@stonybrook.edu or 631-632-9123).

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