OUTFALL MONITORING SCIENCE ADVISORY PANEL (OMSAP)
FLOUNDER LESION FOCUS GROUP MEETING
Thursday, March 31, 2005, 10:00 AM - 1:00 PM
Smith Conference Room, WHOI

MINUTES

ATTENDANCE

Focus Group Members: Andy Solow, WHOI (chair); Sal Frasca, U. Connecticut; Mark Hahn, WHOI; Grace Klein-MacPhee, URI-GSO; Anne McElroy, SUNY Stonybrook; John Stegeman, WHOI; and Scott Weber, New England Aquarium.

Observers: Sandy Baldwin, USGS; Ellen Baptiste Carpenter, Battelle; Bruce Berman, Save the Harbor/Save the Bay (SH/SB); Michael Bothner, USGS; Jeanine Boyle, Battelle; Todd Callaghan, MCZM; David Dow, NMFS; Patty Foley, SH/SB; Maurice Hall, MWRA; Bob Hillman, Battelle; Carlton Hunt, Battelle; Ken Keay, MWRA; Paul Kennedy, MWRA; Matt Liebman, EPA; Megan Lim, SH/SB; Michael Moore, WHOI; Judith Pederson, MITSG; Andrea Rex, MWRA; and Cathy Vakalopoulos, MADEP.

SUMMARY OF FOCUS GROUP RECOMMENDATIONS

The focus group recommends that the following be considered:

High Priority

- Etiology Study
  - To be designed by M. Moore in consultation with S. Frasca and S. Weber and reviewed by the flounder lesion focus group.
- Additional field studies at multiple sites and seasons
  - Focus on January, February, and March at station FF09
- Other suggested diagnostic techniques
  - Co-culture
  - Electronmicroscopy
  - Shotgun sequencing
  - Consensus primers
- Study the progression of lesions in laboratory flounder caught at FF09 in January
  - Morphology – gross and histological through time (early, open, healing, and healed lesions)
  - Immune function – blood and lymph
  - Experimental sediment exposure
  - Experimental transmissibility

Moderate Priority

- Tagging
  - Use spaghetti, not acoustic tags (model after Chesapeake Mycobacteriosis studies), to study healing and migration.
- Field caging study
- Novel contaminants to consider (pharmaceuticals and personal care products)

MINUTES

Welcome and Introductions

A. Solow welcomed everyone and outlined the discussion questions for the meeting. P. Foley introduced herself. She is the president of Save the Harbor/Save the Bay in Boston and chair of the Public Interest Advisory Committee (PIAC). The role of PIAC is to inform the public about the health of the harbor and the bay and to inform the Outfall Monitoring Science Advisory Panel (OMSAP) about the public concerns. She thanked everyone for taking the time and offering their expertise today to discuss this important subject.

The focus group members then introduced themselves. S. Weber is the head veterinarian at the New England Aquarium in Boston. His background is aquatic pathobiology and veterinary medicine. A. McElroy received her degree at WHOI. She worked on flounder aromatic hydrocarbon interactions at U Mass Boston. She is currently at Stonybrook researching flounder and sewage contaminant related interactions. J. Stegeman has worked with M. Moore in examining hepatic neoplasms in winter flounder has also been involved in studies researching chemical effects on fish. M. Hahn was originally trained as a toxicologist. He works in the WHOI biology department and works on comparative biochemistry and molecular biology studying mechanisms of toxicity. G. Klein-MacPhee recently retired from URI but has worked for 30 years on winter flounder. She worked for EPA raising flounder for toxicology studies, has studied flounder exposure to dredge material in Narragansett Bay, and surveyed flounder numbers for RIDEM. They have not seen many lesions off of Rhode Island, but there have not been many flounder either. S. Frasca is an associate professor of pathology at U. Connecticut. He is a veterinary pathologist for the Connecticut Veterinary Medical Diagnostic Laboratory at U. Connecticut. He has an interest in fish pathology. He recently received some of the flounder with lesions to evaluate histologically.

Findings to date

M. Moore summarized what is known to date. He acknowledged the others that have worked on this project: Roxanna Smolowitz (Marine Biological Laboratory), Kevin Uhlinger (MBL), Lisa Lefkovitz (Battelle), John Ziskowski (National Marine Fisheries Service Milford CT), George Sennefelder (NMFS Milford CT), Jeremy King (MA Division of Marine Fisheries), Maurice Hall (Massachusetts Water Resources Authority), Jack Schwartz (MADMF), and David Pierce (MADMF). The work has been funded by the MWRA through a prime contract with Battelle.

MWRA began examining flounder at Deer Island Flats in the mid-1980's. They began routinely surveying five stations every April (Deer Island Flats, new outfall site, Broad Sound, Nantasket Beach, and eastern Cape Cod Bay) in 1992. In 2004, they added additional surveys and sites as part of the flounder lesion investigation. He listed their fishing effort per year at each station and noted that the new outfall went on-line in September 2000. The station at the outfall is actually about a mile away because there is no fishable bottom around the outfall. They try to sample 50 fish, 30 cm or longer, at each station. He then showed photographs of lesions. They mainly began to see the lesions in 2003, although there were some reports of lesions in 2002. All lesions, with one exception, are on the blind side of the flounder and it has been confirmed histologically that this is a classic case of ulcerative dermatitis. He showed photographs of lesions from the June and September 2004 surveys that appear to be in various stages of healing. NMFS data suggest that this is a seasonal occurrence because they had seen ulcers in the spring and not the fall. When M. Moore first saw the lesions a couple of years ago, he had never seen anything like this in winter flounder. He asked other researchers and no else had either. John Ziskowski, Bob Murkolano, and others from NMFS had an extensive dataset from the

1980's in Boston Harbor that summarized visible external pathologies such as fin rot and bent fin and ulcers were statistically absent from the population.

M. Moore said that when he discovered the lesions, he brought samples to Roxanna Smolowitz at MBL for routine histology and microbiology and she came up with no diagnosis. In 2004, MWRA expanded their surveys and worked with NMFS and MADMF so that lesion identification was consistent.

M. Moore then presented MWRA, NMFS, and MADMF data for the spring 2004 survey. There is a fairly broad elevated prevalence in western Mass Bay. NMFS has stations throughout the Gulf of Maine and ulcers were very rare in the rest of GOM (one found off of Portland, Maine and one found off of Yarmouth, Nova Scotia). He then presented a table of prevalence data. He pointed out that there was an absence of fish inshore in the winter and fall and that this was not the case 15 years ago.

A. Rex asked whether the percent healed on the prevalence table is the percent of fish that had healed ulcers, or the percent of ulcers that were healed. M. Moore replied that it's the percent of fish showing a scar from an ulcer. B. Berman said that means a fish with two healing ulcers would count as one. A. Rex asked how a flounder with two healing ulcers and one unhealed ulcer would be counted. M. Moore replied that the fish would be listed on the table twice. S. Frasca asked if there are ulcers of different chronicity on the same animal. M. Moore replied occasionally, yes.

M. Moore said that he was able to advertise the problem to the scientific community and get feedback during his presentation at the NOAA flatfish meeting in Milford in December 2004. The only person that reported lesions was Donald Danila from Connecticut who recently found a small number of lesioned flounder in the Niantic River potentially comparable to what has been seen in Mass Bay. G. Klein-MacPhee added that D. Danila is sampling fish near the nuclear power plant on the Niantic River.

M. Moore then reviewed Roxanna Smolowitz's and Kevin Uhlinger's work in the lab. They grew 21 cold-loving bacterial species from between 1-10 fish per species of bacteria. Many fish had multiple organisms, 7 were isolated from normal skin samples, 19 were isolated from 28 ulcerated samples. He showed a list of the bacterial species found. There is a broad spectrum of diverse microflora on the skin and ulcers. No fungal or viral elements were identified in either routine or specially stained tissue samples. They did see trematodes in the gills and skin and lymphocystis which are common conditions. A. Solow asked if this kind of analysis was also done on fish without ulcers. M. Moore replied yes. There was a broader, more diverse population of bacteria in the ulcers.

A. Solow noted that there is a rarefaction problem here because the sample size is 10 non-ulcerated fish and 28 ulcerated fish. M. Moore explained that in terms of their sample size for controls and ulcerated fish, they had to make a decision as to how much focus they gave to the controls versus trying to isolate a pathogen. A. Solow understood but wanted to point out that the sampling effort is important. They sampled roughly three times as many ulcerated fish and found roughly two times as many species. M. Moore said that the sampling was designed to maximize the chance of encountering the pathogen.

M. Moore then summarized the features of the syndrome. It appears to be seasonal and began in 2002, possibly 2001, but certainly not prior to that. It seems to mostly be limited to western Mass Bay with the highest prevalence near the Mass Bay Disposal Site (MBDS). George Gardner (EPA) did a study on flounder pathology at the MBDS and he will try to find that report. In the last 10 years, there has

been a major source reduction of contaminants as well as improved treatment of the MWRA effluent. The toxic burden has been reduced enormously and that is reflected in the reduction of contaminated-associated liver lesions in the flounder. The treatment plant is currently full primary and 90% secondary treatment. A. Rex added MWRA is using much less chlorine because of the dilution at the discharge and the increased contact time in the disinfection basins. They are also dechlorinating the effluent. A. McElroy asked when MWRA upgraded to secondary. A. Rex replied that one battery of secondary went on-line in 1997 and they ramped up treatment with two additional batteries over the next few years.

M. Moore then discussed flounder migration. The last flounder migration study in this area was conducted by Howe and Coates in 1975. It is his sense from the literature that flounder are driven by an avoidance of 15 degrees Centigrade or higher. Flounder can find cooler water by going into trenches without having to migrate long distances. It was thought that winter flounder don't migrate far in Mass Bay but he is not sure that this is true anymore. This may have changed due to a change in nutrition. Flounder used to feed at Deer Island Flats which was full of polychaete mats. This is no longer the case with the cleaner effluent and outfall relocation. M. Moore speculated that this could have changed the whole migratory pattern of winter flounder.

D. Dow said that NMFS has outlined essential fish habitats for different species including winter flounder. These documents examine changes over time since the 1970's. M. Moore said that NMFS only fishes during the spring and fall, and not during the winter which is an important time, but he will look at their winter flounder report. He pointed out that on 2/5/91 and 3/12/91, the winter flounder in Broad Sound were abundant, but there were none on 3/10/05. M. Liebman asked if there has been a dramatic increase in flounder numbers at the new outfall in the last few years. M. Moore replied yes, but the increase in numbers began before the outfall went on-line.

M. Moore said that he sent S. Frasca three flounder with early ulcers that they found on March 10th. The earlier the lesion is found, the more likely the agent causing them can be found. S. Frasca has a broader diagnostic artillery arsenal than R. Smolowitz so the chances of finding the causative agent are greater this year.

M. Moore pointed out that there are many seasonal and inter-annual factors which co-vary and could hypothetically be associated with the ulcers. A partial listing includes:

- Changes in the fish during spawning.
- Changes in seasonal precipitation.
- Sediment resuspension and transport.
- Inter-annual climate variations – e.g. North Atlantic Oscillation (NAO).
- Undetected changes in effluent chemistry.
- Seasonal pathogen bloom.
- Seasonal predation.

He noted that J. Ziskowski theorized that hagfish could be attacking flounder as they hide in the sediments but hagfish are scavengers so he does not agree with that theory. He showed a lesion that resembled a bite mark and said that hagfish are mainly found in Great South Channel. S. Weber said that hagfish will attack live things as well. G. Klein-MacPhee said that hagfish are opportunistic. M. Bothner asked how hagfish attack. M. Moore replied that hagfish tend to dissolve the tissue of dead fish. They crawl inside the fish and fill the entire cavity like a "bag of worms". S. Weber added that tend to attack fish in cages where their prey can't get away.

M. Moore showed a diagram of winter/spring bottom temperatures by region. He then listed ulcerogenic infectious agents in fish: *Aeromonas salmonicida, Pseudomonas, Vibrio*, and *Proteus* and outlined other studies on cod, flounder, and menhaden diseases. He then recommended that they: (1) continue to quantify prevalence and severity of the lesion during April 2005 flounder monitoring studies; (2) continue to cooperate with state and federal fisheries agencies and scientists to evaluate the condition; (3) further evaluate the long-term USGS mooring, sediment trap, and surficial sediment data for seasonal or interannual changes in sediment quality, either natural or anthropogenic, that might be related to the syndrome; and (4) discuss next steps if any regarding diagnosis (pending results from the U. Connecticut lab).

J. Stegeman said that he was struck by the limited instances of ulcers lesions in the literature and wondered if there are more studies that have identified ulcers in fish. M. Moore acknowledged that he has not done an exhaustive review of the literature. M. Hahn added that there has been a lot of work on flatfish in Puget Sound. He asked if anyone knew whether they have seen anything similar out there. M. Moore said that he spoke to someone there two years ago and at the time, the lesions were rare.

S. Weber asked if they have bottom temperature data from the September collection when the animals were healing because temperature is a temporal factor that plays an important role in the activation of the fish's immune system. M. Moore said they do not measure bottom temperatures during the survey work. M. Hall added that MWRA does have temperature data for that particular time and the area.

J. Stegeman asked if there are there other instances of progression of the condition from obvious pathology to healing and scaring in the literature. M. Moore said that he has not seen such studies. S. Weber noted that the ulcers look fairly defined, circumscribed, and they heal. M. Moore replied yes, and he would describe them as unlikely to have been of clinical significance (i.e. the lesions do not seem to affect the health of the fish).

Focus group discussion

Questions for discussion:

1. What kind of study could be conducted to determine whether or not the lesions are attributable to the outfall?
2. Is there a study that would help us definitively understand what is causing this phenomenon?
3. Discuss theories as to the cause of the lesions: e.g. hagfish predation and fish net trauma.
4. Is there agreement that this is a seasonal phenomenon and that the lesions appear to heal over the summer and fall?
5. Is there a risk to human health?
6. Are there alternative laboratory techniques that might help better identify the pathogens in the flounder lesions?

A. Solow began the discussion with question #5, "Is there a risk to human health?" G. Klein-MacPhee asked if people are likely to eat a flounder with a lesion. M. Moore replied yes, because most of the flounder that people eat will not have the skin on it. G. Klein-MacPhee thinks that it's difficult to say whether there is a risk to human health if the cause of the lesions is not known. The rest of the focus group agreed.

A. Solow then moved to question #4, "Is there agreement that this is a seasonal phenomenon and that the lesions appear to heal over the summer and fall?" G. Klein-MacPhee and J. Stegeman thought that based on M. Moore's and John Ziskowski's work that this seemed to be the case.

M. Hahn's only reservation to the conclusion that this is a seasonal phenomenon is that the seasonal incidence data are superimposed on seasonal migration data thus they may not be sampling the same group of fish. It is not clear to him how this is taken into account in interpreting these data. M. Moore noted that the long term data set on hepatic lesions is secure to the extent that they return to each station at the same time each year, however migration is an issue. J. Stegeman thinks that if this is associated with migration, then one population would have many ulcers and another population would have healed ulcers. He finds this less credible than having a seasonal change in the severity of the condition.

S. Frasca thinks it is important to examine fish with healing lesions histologically because the quality of the healing will also speak to the health of the fish. Animals that have a healthy immune system and are on par nutritionally will demonstrate better healing histologically than animals that are in some ways debilitated.

J. Stegeman asked if they are able to get the fish alive on the deck. M. Moore replied yes, but a lot of stress is put on the fish when it is caught and kept. It also takes about three months to reclimatize a flounder in a tank. G. Klein-MacPhee agreed and added that if they are caught with lesions, they tend to get worse rather than better.

A. McElroy asked about tagging. S. Weber noted that they are tagging the striped bass in the mycobacteriosis studies in Chesapeake Bay. He suggested offering a reward to fishermen that turn in tagged animals. M. Moore asked if they should use spaghetti tags. A. McElroy said yes, and she thinks that the additional cost of tagging the flounder would be trivial. A. Solow pointed out that the sample size would have to be much larger. M. Moore said that based on the resource available now, they work hard to catch 50 fish at a station. Catching 250 fish at one station could take an entire day of trawling. The resource isn't that that great now. Back in the 1980's when Deer Island was discharging, the polychaetes were a yard deep there and the boat would be full of fish after only 10 minutes of trawling. A. McElroy did not think that they would need a lot of fish for a tagging study. A. Solow said that they could figure that number out.

C. Hunt asked if the purpose of a tagging study would be to look at migration or healing. The focus group members replied that the purpose would be to see if the lesions are healing. M. Bothner added that it's also important to find out if the lesions heal but leave scars. The first step of a tagging program would be to prove whether there was a seasonal cycle to the lesions. M. Moore thinks that the lesions heal completely. G. Klein-MacPhee suggested that they could save some of the flounder with healed scars in a tank to see if they completely disappear but if flounder with open ulcers were collected, they would probably die. M. Moore thought that he could keep them alive. J. Stegeman thinks that this kind of study is worth doing with ulcerated/non-ulcerated fish but that seasonality should also be looked at. S. Weber said that if they couldn't bring them back alive, they could instead collect blood and lymph fairly readily. B. Berman thinks that a tagging study would be useful not only to see if the lesions heal completely, but also to learn about flounder migration which he thinks is a critical issue.

A. Solow noted that the group's answer to question #4 "Is there agreement that this is a seasonal phenomenon and that the lesions appear to heal over the summer and fall?" is a qualified yes. If there

is a need to get a better answer to that question, then the group has provided some ideas about how to learn whether the lesions heal completely or leave scars. He the moved on to question #6: "Are there alternative laboratory techniques that might help better identify the pathogens in the flounder lesions?"

J. Stegeman asked about pathogens that may be unculturable. S. Frasca said that there are a number of well-known diseases caused by bacteria that are quite difficult to culture such as mycoplasmas and they require specific culture techniques. He thinks that the histopathological work-up that they have done here is certainly thorough. However, there is only so much that can be done conventionally and there are agents that may be unculturable. There are a number of factors that make diagnosis difficult and so alternative diagnostics need to be considered. Perhaps molecular techniques, additional morphologic techniques like electronmicroscopy, or alternative culture techniques (e.g. co-cultures with different agents). These methods are difficult and time consuming and thus would have to be a part of another directed study. Another option is to modify current techniques to optimize for the host in question.

J. Stegeman pointed out that this appears to be a self-limiting condition, i.e. it doesn't spread to cover the whole underside of the fish. So the question is, when is the earliest that they can sample to find the lesions when they are just forming and are the size of a pinprick. M. Moore said that they looked in January and didn't find any lesions. S. Weber said that many fish diseases manifest themselves at different times of the year at certain water temperatures and many viruses have a very narrow temperature range at which they will manifest themselves. He agreed with S. Frasca in that it is difficult to identify many viruses – although a presumptive diagnosis could be made. He agrees that M. Moore's group did an excellent job on their survey work and a thorough job at the pathology. If there were more funding, sampling in different areas and utilizing other laboratory techniques would be useful. He does not know if the genesis of the lesions begins as a pinprick because many of the ulcerative lesions he has seen have not developed like that.

J. Stegeman asked if they thought that based on the appearance of the lesions that this is more likely a viral rather than a bacterial infection. S. Weber replied that a lot more work needs to be done but the lesions do not suggest some of the common bacterial infections that he has seen before. S. Frasca said that usually *Aeromonas* and *Vibrio* are culturable. The fact that there has been difficulty isolating the agent with the conventional methodologies used argues for something that's harder to culture such as viruses. A fundamental question when trying understand the cause (etiology), is to determine whether or not the ulcer forms from the inside out, or from the outside in. The problem is that the ulcers here have rounded edges and it is difficult to tell whether they formed from the inside or the out. The earliest lesions will give some insight as to how they formed. He thinks that one important study would be one that attempts to determine the pathogenesis of a lesion. He thinks the group should also discuss other factors that could alter the immune function of fish such as exogenous compounds that may affect the endocrine and immune systems. One thing they may want to consider is incubating animals with the lesions along with animals without the lesions to see whether or not transmission occurs.

J. Stegeman agreed that the question of chemicals altering the endocrine and immune systems is interesting. However, looking back to when there was an enormous chemical burden discharged at Deer Island Flats, ulcerative lesions were not detected. He thinks this argues against a chemical involvement in the etiology. A. McElroy agreed and said that no one has seen this at very contaminated sites. She works in an estuary off of New York that gets most of its fresh water from sewage and the fish don't have these lesions. Although the numbers of lesions are certainly higher near the new outfall site, she sees an overall urban signal because the numbers are elevated in the

whole area. She thinks that something has changed in the 2001-2003 time frame that may be affecting their immune system and/or making conditions more favorable for supporting some type of virus.

S. Weber said that there are several confounding factors here. One is that the immune function of fish, which are cold blooded, varies greatly during the winter versus the summer. Just a few degrees change in temperature can activate certain immunological factors but it can take some weeks to for the fish to adapt to the increase in temperature and make all of the proteins that fight off different infections. This means that since it's winter, their immune systems are probably suppressed during this time. The second confounder is nutrition. When working in Alaska, he learned that when sport fishing for halibut, the best place to find them is at the outfalls. Flatfish seek outfalls to feed, and now that the effluent is cleaner, there is no longer this great food source, and poor nutrition can also affect immune function. The third confounder is urban runoff which contains toxins that may influence immune function. This is a multifactorial problem and researching it will take more than one study.

J. Stegeman asked if S. Weber thought that sufficient differences immune function can be detected in the March versus June fish. S. Weber replied that the lab he worked at in Aberdene used flatfish as their models and they canulated (inserted a catheter into a lymph vessel to draw lymph) them. They noticed temporal differences in their immune function. Flatfish are very good models for canulation because lymph can easily be drawn from them. If an infectious agent was involved here, animals could be infected to see how their lymph changes over time. J. Stegeman thinks that it would certainly be worth doing some studies on immune function.

A. Rex asked if, as in this case, it was common to have histopathological work show no evidence of a viral infection. S. Frasca replied yes, this is not uncommon. One coarse way of organizing viruses is DNA-containing viruses and RNA-containing viruses. DNA-containing viruses can leave behind histologic evidence and there are stains that are more readily available to detect them. RNA-containing viruses can also produce histologic evidence, but they are sometimes more difficult to identify with special stains. There are also other types of viruses such as retroviruses that don't leave a histologic legacy of their presence, however, they can be identified molecularly. S. Weber added that it's probably not uncommon with even the best tissue samples to find any agent. This happens at the Aquarium – despite best efforts, sometimes they can't even determine whether the problem is a virus, bacteria, worm, or even trauma. S. Frasca noted that the same situation occurs in humans. For example, there are no etiologic agents for diseases such as Chrohn's Disease yet this disease has been studied for decades.

A. Solow brought up the point that the new outfall was not only a relocation, but also a change in the treatment of the sewage. A. Rex added that these two things did not occur at the same time. MWRA stopped dumping sludge in 1991, the quality of the influent going into Deer Island improved dramatically in the late 1980's. There isn't any change of the quality of the effluent that one can point to at any one point of time. G. Klein-MacPhee asked when the decrease in chlorination occurred. A. Rex replied that the level of chlorination decreased dramatically when the outfall went on-line in September 2000.

M. Hahn asked if there have been microbiological surveys before and after that change in the chlorine to measure the effect on Mass Bay. A. Rex replied yes. They almost never detect sewage indicator bacteria in the area around the outfall. Viral levels were so low both before and after the outfall relocation that they can be considered the same. M. Hahn asked if those surveys were done in the water or the sediments. A. Rex replied that all of the microbial surveys measured sewage indicator bacteria (except for *Clostridium*) in the water column in the direct outfall area. Sediment surveys for

Clostridium perfringens are also conducted and sediment traps are deployed by USGS. S. Weber noted that the New England Aquarium always measures sewage bacteria at their intake in Boston Harbor and it has been a very clean source, at least in the five years that he has been there.

S. Frasca asked if lesions have been seen in other types of fish in the area. M. Moore replied that they routinely catch other bottom fish and they do not see lesions on them. The fishermen say that they only see lesions on winter flounder. B. Berman asked if they took 50 fish a month before the time that the lesions are usually seen from a location with the highest incidence historically and put them in a tank, would they expect the lesions to form on some of them? M. Moore replied that on the basis of the statistics, yes. However, the act of catching them and putting them into the tank may stress them out to the point that they may either die, change their physiology in some way, and/or become immunosuppressed. M. Hahn agrees with a point that S. Frasca brought up – if there was more known about the pathogenicity of the lesions, there would be more by the way of clues to the cause. Trying to define pathogenicity is probably a priority item – to define the pathogenicity, find the earliest evidence of the lesions, and describe them carefully both histologically and microbiologically.

M. Moore said that this work will require a proposal. He thinks S. Frasca and S. Weber could provide laboratory perspectives and he could prepare the field side of the proposal. However, the experimental design will be difficult to develop. The focus group then listed alternative laboratory techniques discussed earlier: alternate culturing techniques including co-cultures, electronmicroscopy inspection of the tissues, molecular shotgun sequencing of the lesions, and consensus primers for certain virus groups.

J. Stegeman then summarized a collection of studies in Marine Ecology Progress conducted in Oslo Fjord in 1986. Animals were sampled in Oslo's Fjord and European flounder were held on chemically characterized sediments collected from various regions within Oslo Fjord. Circumscribed lesions with a depth formed on the animals that were held in captivity on the sediments from the most highly contaminated regions. The focus group discussed these studies and J. Stegeman noted that the 1986 report only provided observations and no conclusions.

A. Rex asked the focus group if the fact that the lesions are forming on the blind side of the flounder has anything to do with the sediments. S. Frasca replied that it's difficult to say. It doesn't necessarily match with what people conventionally think of as a systemic agent. *Salmonicida* infections in fish don't have a side preference. But these fish are bottom dwellers and one side does have a completely different exposure and that could play a significant role. He couldn't answer A. Rex's question but thinks that is an important element to future evaluation of the pathogenesis. M. Moore thinks it's reasonable to assume that they are not going to find one single agent that is causing this problem. Factors that are probably involved are an infectious agent, some type of physical traumatic event, and temperature causing immunosuppression. S. Frasca noted that shell disease in lobster is always seen on the dorsal side.

A. Rex asked if the bottom side of the flounder is tougher. G. Klein-MacPhee replied that the scales are ctenoid (pricklier) on the top and cycloid (smoother) and smaller on the bottom on females. M. Moore added that the males are rough on the bottom. A. Solow asked if there was there any difference in prevalence in males versus females. M. Moore replied that they have not compared males versus females because there are many more females being caught. G. Klein-MacPhee said that they have the same problem off of Rhode Island – it has become much tougher to find flounder, and when they do, there now tends to be many more females than males. M. Hall said that at least 80% of the flounder they catch in Mass Bay are females.

M. Hahn thought that thought this may not be a factor in this case, but that he should at least mention a group of chemicals – pharmaceuticals and personal health care products. These are an emerging group of contaminants from birth control hormones to antibiotics and as far as he knows, there are no data from this area. A. McElroy said that she knows someone that is taking measurements of these types of chemicals and conducted a very small fish study off of New York. The fish were kept on the contaminated sediments for a short period of time. After exposure, they were found to have an elevated protein indicative of exposure to estrogen mimics. Interestingly, three out of four fish also picked up a nematode that was not seen in any of the fish or the sediments.

M. Hahn asked if there are other species besides hagfish that may prey on flounder. He also asked if there have been any new benthic invertebrates detected. K. Keay replied that MWRA has an extensive benthic sampling program and overall, they have not seen any changes in the benthic communities in the four years since the outfall went on-line. However, spatial scales are important and if a predator lives at a density of one per square meter, it probably wouldn't be captured by the sampling program. M. Hahn asked how much time flounder spend off of the bottom. M. Moore replied only in short bursts. G. Klein-MacPhee added that they don't do a lot of swimming in the water column.

J. Stegeman asked about the hydrography in the area of the outfall and whether the effluent is distributed over the same areas where the lesions are found. B. Berman noted that migration should also be taken into account. A. Rex said that the pattern of flounder lesions is not consistent with the presence of the outfall in terms of the gradient of dilution. C. Hunt said that the effluent plume is generally located further inshore and to the south. J. Stegeman concluded that if the lesions are outfall related, it has to be due to a combination of the movement of the water and the movement of the fish. A. Rex also noted that it also depends on the time of the year. During the spring when the lesions are forming, there is no water column stratification and so the effluent is more dilute. During the summer, the effluent is more concentrated on the bottom.

A. McElroy asked if they will continue the surveys and study the seasonality of the lesion formation. A. Rex replied that they have not decided what they will do in addition to their annual spring survey. A. McElroy thinks that they should at least sample at some of the stations in January/February. A. Solow suggested that the focus group ask for M. Moore to outline a proposed plan of research to address the questions discussed today.

T. Callaghan asked if a study similar to the one in Norway could be conducted here. Lesion-free flounder could be placed on sediments collected from where the lesions are prevalent to see if any ulcers form. M. Moore said that he has had problems in the past conducting this kind of study. M. Hahn suggested some type of caging study although he thinks that this would also be very difficult logistically. S. Weber asked about sediment chemistry studies. A. Rex replied that USGS is conducting a large sediment quality study and MWRA has been sampling sediment since 1992. MWRA also monitors the benthos, water quality, and effluent. M. Hahn asked if pharmacological and personal products can be added to the list of chemicals monitored. A. Rex replied that it's possible to test for these chemicals but there are a lot of them, and we need to know what to look for. J. Stegeman asked if MWRA conducts sediment toxicity bioassays. K. Keay replied no. They measure PAHs, PCBs, metals, *Clostridium*, grain size, and TOC. J. Stegeman asked if the results have been relatively static. K. Keay said that there is a rough onshore to offshore gradient from historical sources and little or no change in anything except for a localized increase in the vicinity of the outfall of *Clostridium* since the discharge went on-line.

M. Liebman suggested that the focus group prioritize the list of suggestions (see list on page 1). G. Klein-MacPhee thinks that studying the progression of lesions was a high priority. J. Stegeman suggested that during their next survey, they fix material appropriately for doing sequencing. C. Hunt said that the next survey is in late April. M. Moore said that he could freeze the entire range of ulcer stages that he finds in April. S. Frasca said that they could potentially store tissues for three to six months for future work-up. He suggested that progression studies be conducted to see how the lesion changes under different temperatures and conditions. He noted that his lab does not have the capacity to deal with large projects using complicated diagnostics without having individuals directly associated with the work at U. Connecticut.

A. Solow said that the focus group will present the study proposal to OMSAP who could then recommend to EPA/MADEP that the study be conducted. He understands how complicated these types of problems are and thinks that though this may never be solved, more progress should be made. The focus group agreed. P. Foley said that after having conversations with some of the other PIAC members, PIAC will support OMSAP to recommend fast track funding, because this is an important issue.

Adjourned

Summary prepared by C. Vakalopoulos.

OUTFALL MONITORING SCIENCE ADVISORY PANEL (OMSAP) MEETING
Thursday, August 11, 2005, 10:00 AM to 2:00 PM, Battelle, Duxbury, MA

MINUTES

AGENDA TOPICS

- 2005 *Alexandrium* bloom
- Update on mooring enhancements
- Winter flounder lesion update
- Model Evaluation Group
- MWRA permit renewal

ATTENDANCE

Members Present: Norb Jaworski, retired; Bob Kenney, U. Rhode Island; Scott Nixon, U. Rhode Island; Judy Pederson, MIT/Sea Grant (co-chair); Jim Shine, Harvard School of Public Health; Andy Solow (co-chair); and Juanita Urban-Rich, U. Mass Boston.

Observers: Don Anderson, WHOI; Michele Barden, EPA; Bruce Berman, Save the Harbor/Save the Bay; Todd Borci, EPA; Peter Borrelli, Center for Coastal Studies; Jeanine Boyle, Battelle; John Brawley, Battelle; Martin Dowgert, USFDA; Paul Dragos, Battelle; Dave Duest, MWRA; Patty Foley, Save the Harbor/Save the Bay; Chris Gagnon, Battelle; Sal Genovese, Safer Waters in Massachusetts; Maury Hall, MWRA; Carlton Hunt, Battelle; Mingshun Jiang, U. Mass Boston; Ken Keay, MWRA; Yong Lao, MWRA; Wendy Leo, MWRA; Scott Libby, Battelle; Mike Mickelson, MWRA; Michael Moore, WHOI; Ann Pembroke, Normandeau; Andrea Rex, MWRA; Jack Schwartz, MADMF; Steve Tucker, Cape Cod Commission/Mass. Bays Program; Cathy Vakalopoulos, MADEP; Gordon Wallace, U. Mass Boston; and Meng Zhou, U. Mass Boston.

MINUTES

OMSAP approved the September 27, 2004 meeting summary with no amendments.

2005 *ALEXANDRIUM* BLOOM

D. Anderson presented preliminary observations and data from the 2005 New England *Alexandrium fundyense* bloom. The MWRA Contingency Plan includes a nearfield *A. fundyense* caution threshold of 100 cells/liter. The threshold was exceeded triggering MWRA notification to regulators and the public and the implementation of their *Alexandrium* Rapid Response Survey Plan. The high numbers of *A. fundyense* in the nearfield were part of an extensive bloom that extended from western Maine to Martha's Vineyard and Buzzards Bay near the Cape Cod Canal. Growth of *A. fundyense* was favored in 2005 due to several factors: (1) high numbers of *A. fundyense* cysts in western Gulf of Maine sediments, (2) heavy winter snowfall and spring rainfall, and (3) two Nor'easter storms in May that helped move the cells into Massachusetts Bay. Agencies and institutions have been working cooperatively to track the extent of the bloom and monitor the toxicity of shellfish beds. In Massachusetts, these include the Woods Hole Oceanographic Institution, Massachusetts Division of Marine Fisheries, MWRA, and the Center for Coastal Studies. Future work includes surveying *A. fundyense* cysts in sediments, modeling bloom development, and additional data and laboratory analyses.

ACTION: OMSAP will review MWRA's Rapid Response Survey Plan and J. Pederson will find out if MIT SeaGrant can host a 2005 Red Tide Symposium. [OMSAP has reviewed the Rapid Response Survey Plan and the Symposium was held on April 18, 2006. A summary report will be posted at: http://web.mit.edu/seagrant/].

UPDATE ON MOORING ENHANCEMENTS

M. Mickelson presented an update on progress on the augmentation of moorings in Massachusetts Bay with additional instrumentation. The Gulf of Maine Ocean Observing System (GoMOOS) is installing a surface chlorophyll fluorometer on their mooring off of Cape Ann. USGS's long term Buoy LT-A will be taken out of service in February 2006 because the study period for their project will be completed. MWRA will work with MIT SeaGrant to determine what an appropriate replacement would be. There are several problems that need to be addressed: collecting real time data at the Buoy LT-A location has been difficult, ship traffic, corrosion, and fouling. NOAA has requested that their regions identify priorities for platform augmentation. MWRA will work with GoMOOS, Todd Callaghan (MA Coastal Zone Management), and others to coordinate a response to NOAA on monitoring needs in Massachusetts Bay. J. Pederson suggested that OMSAP reconvene the mooring technologies focus group.

WINTER FLOUNDER LESION UPDATE

C. Hunt and M. Moore presented the latest information on flounder lesions in Massachusetts Bay. Lesions on winter flounder have been noted in the spring in Massachusetts Bay since 2002. To date, even with additional sampling and laboratory tests, there has been no diagnosis of the cause of the lesions. The lesions almost always appear on the blind (i.e. bottom) side of the flounder and they seem to heal over the course of the summer and fall. The highest numbers of ulcers have been measured in northwestern Massachusetts Bay. MWRA will continue to monitor flounder as part of their Ambient Outfall Monitoring Program. OMSAP convened a flounder lesion focus group that met in March 2005 [for a summary of the meeting, go to: http://www.epa.gov/region01/omsap/pdfs/OMSAP0503-flfgm.pdf].

ACTION: OMSAP members agreed that since the flounder biologists have been unable to diagnose the cause of the lesions, and there is no evidence to date that the outfall is causing the lesions, the best thing is for MWRA to do is to continue the same annual flounder monitoring.

MODEL EVALUATION GROUP

W. Leo presented background the MEG meeting that is scheduled for September 12, 2005. The Bays Eutrophication Model Evaluation Group has guided the development and evolution of the coupled hydrodynamic (USGS) and water quality (HydroQual) models since 1992. The MEG is currently a subcommittee of OMSAP. These two models are now maintained by U. Mass Boston and MWRA. As part of MWRA's NPDES permit, MWRA is required to update, maintain, and run the models at least once a year. The current MEG members are: Dr. Eric Adams, chair (MIT), Dr. Steve Chapra (Tufts), Dr. Jack Kelly (EPA, Duluth MN), Dr. Pierre Lermusiaux (Harvard), Dr. John Paul (EPA Narragansett RI), Dr. Rich Signell (USGS), and Dr. Huijie Xue (U. Maine).

ACTION: OMSAP suggested that the MEG include additional expertise in biological modeling. They also suggested that MEG examine closely what the models are not modeling well. It would also be useful to examine why the models model some parameters very well.

MWRA PERMIT RENEWAL

M. Barden gave an update on MWRA's NPDES permit. The permit expired yesterday (August 10, 2005). MWRA has sent in a timely application and their current discharge permit will remain in effect until the new one is in place. Now we have the benefit of having outfall data since it went on-line in September 2000 whereas when the last permit was written, there were no outfall data. EPA is hoping to have a draft

completed in six months to a year, depending on how long EPA's effluent data review takes to complete. During the permitting process, people can contact EPA with concerns and comments. The draft permit will go to public notice and there will be public hearings.

ACTION: OMSAP members request a list of EPA's proposed changes to the permit when one becomes available. OMSAP also requests that MWRA provide a list of what they would like to see changed in the permit.

ADJOURNED

MEETING HANDOUTS:

- Agenda
- September 2004 draft OMSAP meeting summary
- September 2004 draft Public Interest Advisory Committee meeting summary
- March 2005 flounder lesion focus group meeting summary
- EPA and MWRA information briefings

Summary prepared by C. Vakalopoulos. Post-meeting comments are included in [brackets]. All such comments have been inserted for clarification only. They do not, nor are they intended to, suggest that such insertions were part of the live meeting components and have been expressly set-off so as to avoid such inference.

Summary of Model Evaluation Group (MEG) Meeting
Monday, September 12, 2005
10:00 AM to 3:00 PM
MIT Sea Grant, Cambridge, MA

Attendance

MEG Members:
Eric Adams (chair), MIT
Jack Kelly, EPA Duluth MN Research Lab
Pierre Lermusiaux, Harvard
Rich Signell, USGS
Steve Chapra, Tufts
Jim Kremer, U. Connecticut
John Paul, EPA Research Triangle, NC
Huijie Xue, U. Maine.

Other Attendees:
Martin Dowgert, FDA
Ferdi Hellweger, Northeastern U.
Mingshun Jiang, U. Mass Boston
Wendy Leo, MWRA
Mike Mickelson, MWRA
Andrea Rex, MWRA
Cathy Vakalopoulos, MADEP
Meng Zhou, U. Mass Boston
Jim Fitzpatrick, HydroQual
Carlton Hunt, Battelle
Yong Lao, MWRA
Matt Liebman, EPA
Judy Pederson, MIT, OMSAP
Larry Schafer, retired
Gordon Wallace, U. Mass Boston

Purpose and Process

The Model Evaluation Group (MEG) convened on 9/12/2005. The purpose of this meeting was to review the modeling reports[1] for years 2000-2001 written by the U Mass Boston modeling team and to provide advice on the future direction of the modeling.

The meeting was tape-recorded and transcribed by Cathy Vakalopoulos. An unsorted list of recommendations made by MEG and audience members was culled from the transcripts and is included as an attachment. The following draft summary was compiled by Cathy, Mike Mickelson and Eric Adams.

Background

MEG is a focus group of the Outfall Monitoring Science Advisory Panel (OMSAP) that advises EPA and MADEP on MWRA's outfall monitoring. Since 1992, MEG has provided independent technical advice on the development, maintenance, and running of MWRA's Bays Eutrophication Model (BEM). BEM consists of a hydrodynamic (HD) model[2] coupled to a water quality (WQ) model[3], both adapted to cover Massachusetts Bay and several miles beyond into the Gulf of Maine (GoM).

MEG last convened in 2002[4] and MWRA has since been working on addressing MEG's recommendations. Over the years, many of MEG's recommendations have been implemented. For example:

- Add more monitoring stations near the boundary with GoM
- Add an algal growth carbon/chlorophyll model to simulate the subsurface chlorophyll maximum.
- Add a third algal group to simulate high chlorophyll in fall blooms.
- Increase the horizontal resolution of the WQ model to match that of the HD model.

In addition, MEG recommended that an academic home be found for BEM so it could continue to evolve and be available to other agencies or groups. HydroQual and USGS had used BEM to model years 1990-1999 (with a gap in 1995-1997), and then U Mass Boston modeled 2000-2001.

[1] These are reports # 2004-08 and 2004-09 at http://www.mwra.state.ma.us/harbor/enquad/trlist.html
[2] ECOM-si: Estuarine, Coastal and Ocean Model (semi-implicit). http://woodshole.er.usgs.gov/operations/modeling/ecomsi.html
[3] RCA: Row Column AESOP (Advanced Ecological Systems Operating Program). http://www.hydroqual.com/wr_rca.html
[4] The 2002 MEG report is located at: http://www.epa.gov/region01/omsap/meg0302.html

[Note added after the meeting: MEG comments on the 2000-2001 reports are reflected in improvements in the 2002-2004 reports[5] which were completed after the MEG meeting.]

BEM was developed to model the effects of MWRA's 9.5-mile-long outfall located offshore in Massachusetts Bay. Important modeling results from BEM include

- Harbor DO is improved by secondary treatment; Harbor chlorophyll is improved by outfall relocation.
- Relocation of the outfall has only minor effects on Bay DO and chlorophyll.
- The MWRA outfall contributes only 3% of the total nitrogen load to the Bay.
- Low DO episodes in the Bay are mostly due to inflow from GoM.

Those model results were especially useful as predictions needed before the outfall went on-line, in September 2000. Later, modeling became a requirement in MWRA's National Pollutant Discharge Elimination System (NPDES) permit, with MWRA required to run BEM on an annual basis (hindcasting). MWRA's permit however is currently in the renewal process and MEG's recommendations will help define how to best use the model and assist EPA in determining what (if any) modeling requirements will exist in the new permit. EPA remains interested in whether the model can help discern negative environmental effects of the outfall on Mass Bay.

MEG Review and Recommendations

1 General Conclusions

MEG expressed overall satisfaction with the modeling efforts.

1.1 The model.

A considerable investment has been made in the existing model. While newer models are available, they generally don't differ from the current model in fundamental ways. Improvements can be made to the existing models through increased resolution, improved data, and/or additional processes. While there is little sentiment to change models, there would be merit in increased collaboration with other modelers and in comparing the results of the existing model with results from other models with overlapping domains.

1.2 The modelers.

It is good to have found a home for the model. The baton seems to have been passed successfully, and the modelers are engaged in some interesting studies related to mesoscale processes, sample design, boundary forcing and forecasting that will extend model utility.

1.3 The model results.

The apparent level of agreement between model results and observations seems reasonable and is typical of other models. But the MEG has many questions/comments as indicated below.

2 Questions guiding future model effort

Future modeling effort, and an assessment of how successful this effort is, should be viewed in terms of a number of factors.

2.1 Who is using the model (MWRA, other agencies, other scientists)?

2.2 How will the model be used (hindcasting, nowcasting, forecasting)?

2.3 What is the simulation time frame (average year, interannual variability, monthly variability)?

2.4 How will output be aggregated (predicted concentrations as a function of space and time or aggregated fluxes attributed to boundaries, internal processes, point sources)?

2.5 Will predictions be absolute, e.g., c(x,y,z,t), or relative, e.g., incremental Δc due to outfall?

[5] These are reports # 2006-12 and 2006-13 at http://www.mwra.state.ma.us/harbor/enquad/trlist.html

3 Suggestions to improve model validity and presentation

3.1 Model documentation

The model reports lack information about how the modeling was done objectively, including how boundary data were assimilated. Inclusion of this information would improve confidence in model validity.

Model calibrations, changes, etc. should be cataloged in one place. Differences in model formulations, parameters, and data sources used from year to year should be noted.

Be careful with terminology: don't interchange "calibration" and "validation".

Be consistent and use either calendar or Julian days.

3.2 Model-data comparison

The same filters and depths should be used when comparing model results and observations.

The same forcing functions (irradiance and extinction) should be used when comparing calculated values such as primary production.

Be careful how different phytoplankton groups are defined. It is difficult to defend how the summer, winter and fall groups differ using empirical data, especially field data.

4 Assessment measures

4.1 Summer salinity

The model misses certain aspects of the summer salinity structure. The biological implications could be assessed by plotting predicted bottom as well as surface currents to see how salinity affects transport.

4.2 Interannual comparisons

It is not clear if the model can distinguish different years. To test this capability, important attributes, e.g., yearly minimum dissolved oxygen, should be computed for each year and the years should be tabulated by rank order, for comparison with similar rankings based on observations.

4.3 Effects of outfall

Predictions from post-outfall and pre-outfall years should be compared. If a negative event is observed in a post-outfall year, the outfall should be "subtracted" to see if the event still occurs. Likewise, in a pre-outfall year, one could see how much outfall loading would need to be "added" to trigger concern.

4.4 Focus on boundary conditions

Understanding how open boundary conditions affect interannual variability is critical. Boundary salinities should be displayed to demonstrate whether boundary assimilation is working.

4.5 Algal respiration

Care should be taken to distinguish between net primary production and net community production.

4.6 Additional variables

Model results could be compared with satellite imagery to see if the fronts align.

Observed and predicted extinction coefficients should be compared.

4.7 Other approaches besides simply "observed" vs. "predicted"

Sensitivity to model forcing should be examined. For example as nutrient loading is varied from low to high, do annual productivities, maximum chlorophylls, and sediment deposition totals fit expected patterns?

Look for correlations in related variable (e.g., chlorophyll should be inversely related to nitrogen).

Compare predictions of a climatological year with that of actual years.

Develop a dissolved oxygen budget like the nitrogen budget conducted previously.

5 Can the modeling be improved?

5.1 Use additional data to force the model?

One could obtain atmospheric-ocean flux predictions from weather models, to either drive model or compare with model results. (The model currently computes fluxes from computed surface temperatures, which provides some degree of self-correction.)

The UNH monitoring program (monthly, beginning in 2003) may have useful data to share.

5.2 Nest the model?

Test whether using the Gulf of Maine model for outer boundary conditions is better than the current approach based on limited sampling.

5.3 Improve vertical hydrographic structure?

The thermocline is not sharp enough in many of the modeled vertical sections. Remedial options include a revised turbulence closure model (not strongly endorsed), increased vertical resolution (by a factor of 1.5 to 2 beyond the current 12 layers), and use of 2D atmospheric forcing (as long as it resolves the sea breeze). The need for additional improvement here should be gauged by their effect on the biology.

5.4 Increase the horizontal resolution?

U Mass Boston is evaluating this.

5.5 Increase the complexity of the zooplankton model?

The current model with a simple instantaneous temperature-dependent death rate for phytoplankton is unable to model several observed features, such as the lag between phytoplankton stock and grazing rate. However, zooplankton are very difficult to simulate and augmenting the zooplankton component is not recommended.

6 Future directions

6.1 Model every year?

Yes. An important use of the model is hindcasting: using the model to help determine the cause of observed events. Everyone learns when a model fails, especially for a new reason. We should look at all previous model failures to learn how to make the model better. While not all years are equally “interesting”, if you don’t model every year, you lose expertise.

6.2 Relationship to monitoring

It’s very important to have a monitoring program coupled to the modeling. Having each run continuously allows all the monitoring data to be tied together. The model is available to help answer questions raised by the data, to guide strategic sampling, and to fill in gaps between monitoring.

6.3 Stellwagen Basin

Monitoring data suggest particle transport that focuses in Stellwagen Basin. How much of that carbon is from the outfall or from productivity stimulated by the outfall could be addressed by turning off the MWRA sources of carbon and nitrogen.

6.4 Tighter nutrient management

A sensitivity analysis could be conducted to determine what combinations of effluent/physical conditions promote low DO. This would help if a Mass Bay-wide TMDL is ever developed in the for nutrients or organics.

6.5 Additional output

The state shellfish sanitation program is interested in seeing additional modeling work that involves plume tracking and following a discrete patch of effluent.

MWRA could identify a set of treatment-plant-failure scenarios, feed those into the model, and use the results to determine the scope of ambient monitoring to detect effects on the Bay.

Attachment
Draft attributed unsorted Recommendations

MEG Recommendations

1. Compare interannual variability and minimum dissolved oxygen. (Signell) A table of each year compared side by side would be useful. U Mass Boston has so far modeled 2000-1 and 2003-4 and are looking at the variation between years. The outfall went on-line September 2000.

2. Be careful with terminology: don't interchange "calibration" and "validation". (Paul)

3. Obtain real time forcings or previous analyses from others (e.g. Collier at URI) to calculate proper fluxes. (Lermusiaux) **Counterpoint:** fluxes should be based on model predictions so that if there is some automatic correction in the model. (Adams)

4. Model-data comparisons are best done when data are plentiful. If there is a large gap in field data, don't interpolate in case there is a feature there. (Kremer)

5. Check to see that temperature and salinity are filtered the same way. (Signell)

6. Make sure that there is averaging of the observed temperature field, i.e. confirm smoothing (overhead #24). (Signell)

7. Buoy A is near station N18. Buoy A data are from 5m and N18 data are from the surface. There is a temperature difference between these two locations of 10 degrees. (Xue) Both should be compared at 5 m. (Signell) Could compare with satellite imagery to see if there is a front. (Paul)

8. In general, it would be useful to compare satellite imagery to model results. (Paul)

9. UNH monitoring program may have useful data to share. (Xue)

10. Salinity tends to not be modeled accurately, especially during summer stratification. How good does the model have to be if salinity, for example, does not affect surface currents? (Kelly) The U Mass Boston group noted that data assimilation significantly improved the salinity.

11. It would be helpful to show plots of modeled bottom currents as well as modeled surface currents. The model has a problem with currents at depth because they are affected by salinity. (Kelly)

12. How important is the apparent inability to predict the summer salinities? Are there biological and ecological implications for this in terms of vertical exchanges, i.e. water column stability? (Kremer)

13. The thermocline is too spread out in most of the modeled vertical sections compared to reality. Have there been any trials done on increasing the vertical resolution (currently use 12 layers) in the HD model to see if the stability of the water column improves? Vertical resolution could be increased by 1.5 to 2. Or as a quick improvement, 2D atmospheric forcing could be used (as long as it resolved seabreeze). (Signell & Lermusiaux) **Counterpoint:** Don't necessarily have to go to a more complicated physical structure. If it is decided to improve the vertical physics, keep it in the context of how well the biology is going to work. It is an open question as to whether improving physics will improve the biology. (Kremer)

14. There has been some work done by Bouchard and others on turbulence closures that compare different schemes. Lermusiaux can provide references.

15. To test the model's ability to predict temperature, salinity, and currents, run the model for 2001 (post-outfall) and compare the results to pre-outfall years. (Kelly)

16. Test whether using the Gulf of Maine model for boundary conditions is better than the current approach of using the limited sampling and buoy data available from the boundary. Set up three experiments – model one year, model key events over a year, and use a weighted average of the two. The third approach is potentially the most effective since care must be taken when two models with slightly different scales are forced. It's usually better to use the open boundary model if it captures the mean correctly, instead of manipulating the data. However, problems arise if the mean of the GOM model is slightly inaccurate. (Lermusiaux)

17. If the overall focus is to have an accurate model, then all of these year-by-year questions need to be addressed. If MWRA only needs a model that is representative of the types of features seen in that environment, then in an abstract way, ask the model questions such as the relative influence of the outfall pollutants. If this is adequate, then this becomes less of a challenge. (Kremer)

18. Significant improvement in the WQ model vertical resolution was accomplished when the subsurface chlorophyll maximum could be reproduced. (Paul)

19. To examine how accurate the model needs to be, pick one year and run the WQ model with "what-ifs" on extreme conditions and see how model improvements affect the modeled water quality results (Kelly). If a negative event occurs, figure out how to model it correctly, then "subtract" the outfall and see you the event still occurs. (Adams)

20. One of the important uses of the model is hindcasting: if an event occurs in Mass Bay, use the model to help determine the cause. This may mean that the model does not have to keep running. (Signell) **Counterpoints:** If you don't plan to run the model every year, then you lose expertise. (Paul) If you want the model to guide strategic sampling, then there is value to continually run the model. (Xue)

21. Understanding how open boundary conditions affect interannual variability is critical. (Signell)

22. Now is a good time for MWRA to identify the future direction of the modeling. For example, rapid response studies or trying to integrate the monitoring and the modeling programs better (by trying to improve the design or getting the critical data). Are there any other potential collaborations with other universities or organizations? Would it be advantageous to combine other models? (Lermusiaux)

23. In the context of Mass Bay, the models available are all based on the same physics. Some have slightly better advection schemes or handle vertical coordinates slightly differently, but they all have similar vertical mixing schemes and are capable of the same kinds of resolutions. At the current model resolution of 1 km, one model is not better than another. (Signell) Keep the current model – a lot of work and resources have already been spent on it, and it would take two to three years to get a new model up and running. (Lermusiaux)

24. The modeling field is moving away from competing models and towards using a combination of models. (Lermusiaux) In meteorology, modelers run a suite of models each with their strengths and weaknesses. When they converge, it increases faith in their forecast. There might be some benefit to this approach with oceanic modeling. (Kremer)

25. To help in the evaluation of the modeling results, add a table of observed and predicted extinction coefficients. (Kremer) Need to make sure the same forcing functions are used to make calculations. (Kelly)

26. Be careful when measuring algal respiration. There is a difference between net primary production and net community production. How algal and entire plankton community respiration are calculated makes a significant difference in the observed vs. predicted results. There is a great deal of difficulty in establishing what algal respiration should be. The physiological algal respiration model is based on only a few cultures that cannot possibly represent algal respiration in the field. Our data suggest that algal respiration might reach as high as 50-70% of production at certain times. Algal respiration, grazing, and death should be in the range of 60-80%. It's difficult to evaluate the ecology without some information on the relative balance of those rates and what fraction of those productions go into the various losses seasonally. (Kremer)

27. In reports and presentations, be consistent and either use calendar or Julian days. (Paul)

28. The general approach has been to compare "observed" vs. "predicted" but there are other approaches that can test whether the model is capturing some of the essential features. For example, cross-correlations between related variables, e.g. chlorophyll is inversely related to nitrogen. Cross-system comparisons - load model with very low to very high loading rates, should get annual productivities, maximum chlorophylls, and sediment deposition totals to fit accepted patterns. (Kremer) Could also run a simple baseline model (e.g. a climatology model) to see what additional benefit is gained from the complexity of the model. (Signell)

29. A simpler model doesn't require specifying information that is unknown or can't be measured. (Kremer)

30. Be careful how the different phytoplankton groups are defined. It's difficult to defend how the summer, winter, and fall groups differ using empirical data, especially field data. (Kremer)

31. Zooplankton are very difficult to simulate and so augmenting the zooplankton component is not recommended. Presently, there may not even be enough data available. (Chapra) Progressing with the zooplankton leads up the food chain and predictive value is reduced. Though this would be good research, it wouldn't be helpful in terms of answering questions related to the outfall. (Kremer)

32. The idea that the grazing rate should lag the phytoplankton stock allows for a very simple proxy for a zooplankton and pseudo-predator/prey dynamics. However, this pattern was not shown in the modeling results. Instead, the result was constant high phytoplankton. This may be due to an incorrect magnitude of the grazing. If you increase the magnitude, make sure it is changed all the way through because it may have and effect on sediment deposition and bottom oxygen concentrations. (Kremer)

33. U Mass Boston has shown that there are some interesting and useful uses of the model that investigate some processes, scales, and dynamics that, in combination with the monitoring and buoy data, give some insights on Mass Bay. (Kelly)

34. The question of "is the model good enough?" is difficult to answer because there hasn't been a synthesis shown of how the model and observational data relate. It would also be useful to catalog all calibrations, changes, etc. in one place. We want to make sure that the model produces the right answers based on the right reasons. (Kelly)

35. Conduct a dissolved oxygen budget much like the nitrogen budget that was done a few years ago. (Fitzpatrick) If this is conducted, it should be done looking back at previous years. There are times when the dissolved oxygen minimum was not captured by the model and this would help learn what can be fixed in the model. (Kelly)

36. All models have flaws. We should never take much solace when they appear to work. (Kremer) **Counterpoint:** Because models are based on mass balance, they do bound the problem and do an accounting. So though there are uncertainties, we are not creating something out of nothing. (Chapra)

37. The BEM is typical of other models. (Kremer & Chapra)

38. A sensitivity analysis could be conducted to ask what combinations of effluent and/or physical conditions would promote low dissolved oxygen. This would also help if in the future a Mass Bay-wide total maximum daily limit (TMDL) is developed for nutrients or organics. (Kelly)

39. It's very important to have a monitoring program with a modeling program (Signell). Modeling and data go together. Having monitoring and modeling that runs on a continuous basis from year to year provides a means to tie all the monitoring data together and model is available to help answer questions. (Chapra) Modeling is important to show what is happening when there is no monitoring. (Xue)

40. Everyone learns when the model fails. The only reason to run it every year is if there is an interesting event and the model fails for a new reason. Should make sure have looked at all of the times that the model has failed from the past so that we can learn how to make the model better. (Signell)

41. Overall progress was very good. The future directions identified in the presentations (mesoscale effects, sampling design, open-boundary procedures and schemes, etc) were appropriate. (Lermusiaux)

42. Modeling Investigations (Lermusiaux)
    - Vertical resolution
    - Utilization of SST, SSH and SSC, both for model calibration and data assimilation
    - Turbulence closures
    - Atmospheric forcing (x,y,t)
    - Sensitivity to selected biological processes (e.g. what happens if a given process, or a term in the equations, is removed)
    - Parameter estimation

43. Potential Additional Directions (Lermusiaux)
    - Collaborations among, and integration of, modeling and monitoring/sampling efforts
    - Rapid responses (on demand investigations, events, at-sea accidents, pollutions, homeland security, etc)
    - Seasonal and smaller scales
    - Sampling array design and adaptive sampling
    - Participations in planned academic and business-related research efforts in Mass Bay
    - Diversifications: e.g. nowcasts/forecasts for recreational/tourism activities and their monitoring

Audience Recommendations

44. One thing that was learned from the 1993 *Asterionellopsi*s bloom is that there is a lot of variability in the carbon to chlorophyll ratio due to the particulate organic carbon (POC). POC and the respiration that is associated with it should be considered. (Fitzpatrick)

45. We are seeing particle transport that seems to focus in Stellwagen Basin. How much of that carbon is from the outfall and how much is from productivity stimulated by the outfall? This is a chronic, subtle, long term effect. (Pederson & Hunt) Should be able to address the effect of the outfall by turning off the MWRA carbon and nitrogen. This would help answer how much the long term carbon accumulation (and potentially metals) there is in Stellwagen Basin. (Fitzpatrick)

46. U Mass Boston showed phytoplankton biomass, but they should also show how the nutrients were affected. (Fitzpatrick)

47. Dowgert: With respect to the shellfish sanitation program any additional modeling work that involves plume tracing and following a discrete patch of effluent would be of interest. Questions include the following:
    1. What would be the time of travel, dilution and fate of a patch of effluent that represents a 24 hour failure of disinfection and secondary treatment at the upper limit hydraulic flowrate for adequate primary treatment and under several wind, tidal and seasonal regimes?
    2. What if the above disinfection and treatment failure and elevated flowrate persisted for 72 hours?
    3. What would be the time of travel of the leading edge and the dilution of the plume if the plant provides disinfection at the permit upper limit of 14,000 FC and at the upper limit flowrate that provides for full secondary treatment on an ongoing basis? I would suggest that any FC die-off factors used be very conservative to represent the low light conditions encountered at depth and during the winter season.

48. Schafer recommends:
    1. MWRA establish a set of failure episodes to feed into the model
    2. concerned scientists develop a final version of the model, with the goal of
        a. having a working tool for predicting the consequence of any variation in treatment.
        b. define just what on-going monitoring should be set up for the Bay
        c. run the MWRA criteria and write up the results.