GREAT LAKES FISHERY COMMISSION

2006 Project Progress Report[1]

Early Mortality Syndrome Research and Information Coordination Meetings

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GREAT LAKES FISHERY COMMISSION
RESEARCH STATUS REPORT

EARLY MORTALITY SYNDROME WORKSHOP

September 22, 2005
Ann Arbor, MI

GLFC-Sponsored Research Coordination Meeting on Early Mortality Syndrome

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Abstract

Early mortality syndrome (EMS) is the term used to describe embryonic mortality affecting the offspring of salmonids in the Great Lakes Basin. A workshop was held in Ann Arbor, MI September 2005 brought together 37 federal, state, provincial and tribal scientists and interested resource personnel to share information, present data and discuss the latest observations on EMS and other early life stage moralities. Sixteen speakers presented current EMS research in the Great Lakes Basin. In 2004, EMS continued to be an issue for Lake Michigan and Lake Ontario salmonine populations similar to previous years. However, in Lake Huron, record low alewife abundance was associated with improved egg thiamine concentrations in salmonines and for the first time in 20 years evidence of natural reproduction in lake trout in USGS trawl surveys. The situation in Lake Huron bears close monitoring in subsequent years. Establishment of a more tightly integrated monitoring program, not only for Lake Huron but also Lakes Michigan and Ontario is recommended. Evidence was presented to suggest that thiamine deficiency in egg and early fry development has long lasting affects. Behavioral patterns and immunological responses between thiamine replete and thiamine deficient sac-fry differed months after the deficiency was corrected. Fish behavior toward water borne iron and zinc sulfate differed according to previous thiamine status and specific lymphocytes (T-cells) involved in immune response were lower in thiamine deficient lake trout, suggesting possible impacts for thiamine deficient fish to resist a disease challenge. The use of cDNA microarray technology identified multiple genes regulating important biochemical pathways that were affected by thiamine deficiency. This work opens the door to possibilities such as genetic selection for fish less susceptible to thiamine deficiency. Overall there is a need to establish thresholds for the various latent and secondary effects of thiamine deficiency. Additional study is needed to determine the relevance of ecosystem thiaminase because reported thiaminase activity in net plankton may have implications for larval fish feeding on plankton.

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Introduction

Early mortality syndrome (EMS) is the term used to describe an embryonic mortality affecting the offspring of salmonids (coho salmon, chinook salmon, Atlantic salmon, steelhead trout, brown trout and lake trout) in Lakes Michigan and Ontario and to a lesser extent Huron and Erie. Clinical symptoms of EMS include loss of equilibrium, swimming in a spiral pattern, lethargy, hyper-excitability, hemorrhage and death occurring between hatch and first feeding (Marquenski and Brown 1997; McDonald et al., 1998). Salmonine EMS is caused by a maternal thiamine deficiency that likely results from a diet with a high proportion of thiaminase containing alewife (Honeyfield et al. 2005). In addition to negative affects on fry, there is evidence of impacts due to thiamine deficiency on adults (Brown et al 2005a; Fitzsimons et al 2005a; Ketola et al. 2005). The consequences of the thiamine deficiency on salmonine survival, behavior and physiology are still not fully appreciated. As part of our effort to communicate current information about thiamine deficiency and recent progress in EMS research to fishery managers and other researchers, a workshop was held in Ann Arbor, MI, September 22, 2005. The workshop brought together 37 federal, state, provincial and tribal scientists and interested natural resource personnel to share information, present data and discuss the latest observations on EMS and other thiamine deficiencies in aquatic animals. There were 16 presentations describing the status of current EMS research. We document the current extent to EMS and report a brief synopsis of workshop findings. Based on presentations, an open discussion session and recent literature, directions of future studies are recommended.

Incidence of EMS and thiamine status in Great Lakes Salmonines

Lake Michigan. Lake trout spawning biomass an is important component for successful natural reproduction. Bronte et al (page 14) described site-specific stocking efforts for re-colonizing historically important spawning reefs in Lake Michigan. Densities of adult fish were significantly higher on stocked onshore and offshore sites than on non-stocked reefs, and suggest that site-specific stocking is effective at establishing spawning aggregations as opposed to relying on self-colonization. Overall adult spawning densities were low and consisted of young adults at most sites, which indicate that lake trout parental reproductive stock size is limiting and may partly explain the lack of significant natural reproduction in Lake Michigan. The extent of EMS mortality in lake trout from Lake Michigan has been monitored since 1975 (**Figure 1**) and remained elevated at 48% in 2004. This corresponds with the continued low egg thiamine content of lake trout from Lake Michigan (Honeyfield and Sweet, page 15). Based on egg thiamine level, Honeyfield and Sweet estimate that 37% of lake trout families from Lake Michigan are susceptible to overt mortality due to EMS and that 60% of families may experience secondary effects. Thiamine deficient fry mortality continues to be a concern for other Lake Michigan salmonines. Brown et al (page 14) summarized the mortality rates attributed to EMS in Lake Michigan Pacific salmon offspring. The prevalence of EMS in coho salmon was elevated (range 35-90%) in 1993, 1996, 1999-2003 (**Figure 2**). While perhaps slightly lower in 2004, the incidence of EMS still falls in the 25-35% range. For 2004, EMS was also somewhat lower (31%) relative to 2003 estimates (58%) in chinook salmon from the Little Manistee River (**Figure 3**).

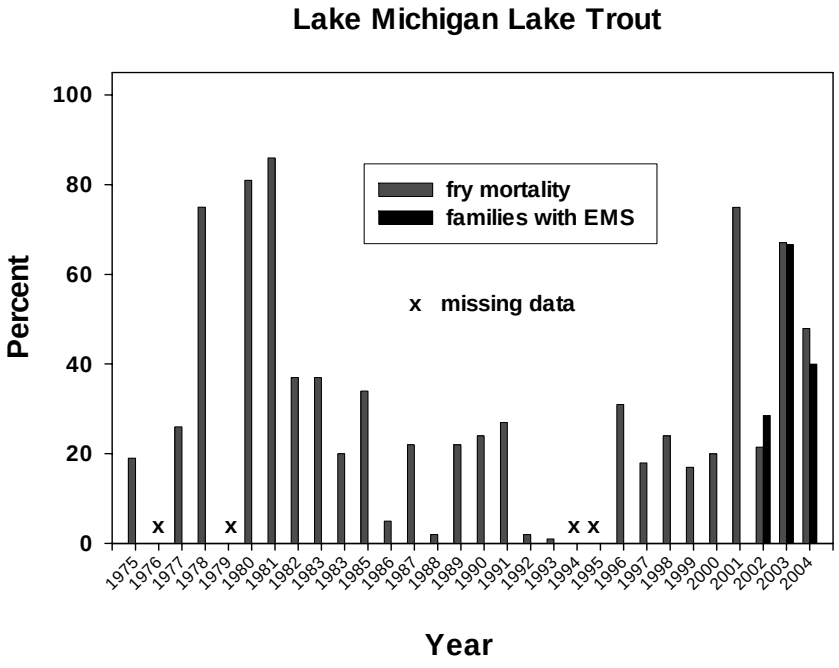

Figure 1. EMS in lake trout in Lake Michigan from 1975-2004 (data provided by Carol Edsall, USGS, Great Lakes Science Center, Ann Arbor, MI)

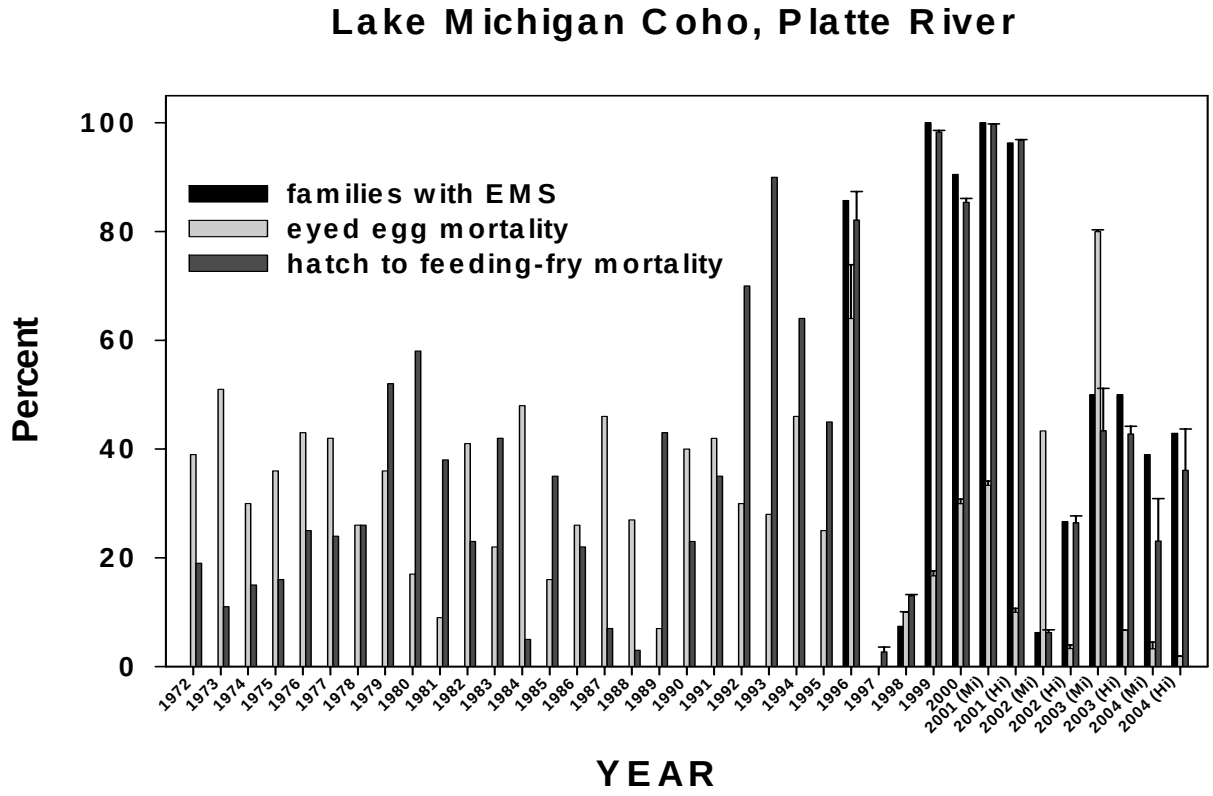

Figure 2. EMS in coho salmon in the Platte River 1972-2004 (data provided by Martha Wolgamood, Michigan DNR)

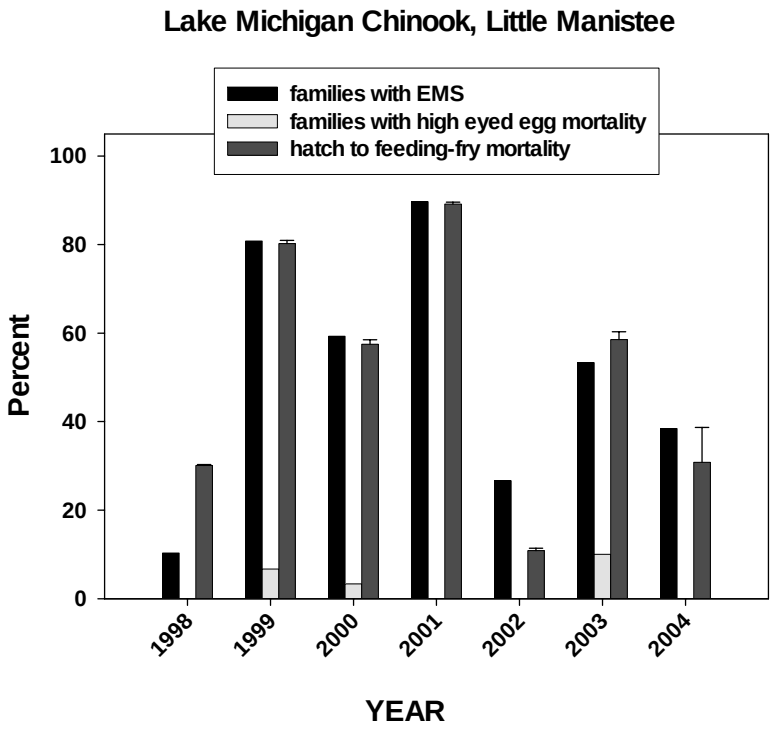

Figure 3. EMS in Lake Michigan chinook salmon 1998-2004 at Little Manistee River (data provided by Martha Wolgamood, Michigan DNR).

Lake Ontario. Monitoring of EMS in Lake Ontario Lake trout was not conducted in 2004. However, EMS has been consistently elevated in lake trout since about 1990 (**Figure 4**). Monitoring for EMS in chinook salmon from the Credit River and in coho from the Salmon River showed that overt mortality was fairly low (<10%), however there were observations of the behavioral symptoms associated with EMS in 40% and 30% of chinook and coho families, respectively (Brown and Daniels, page 16).

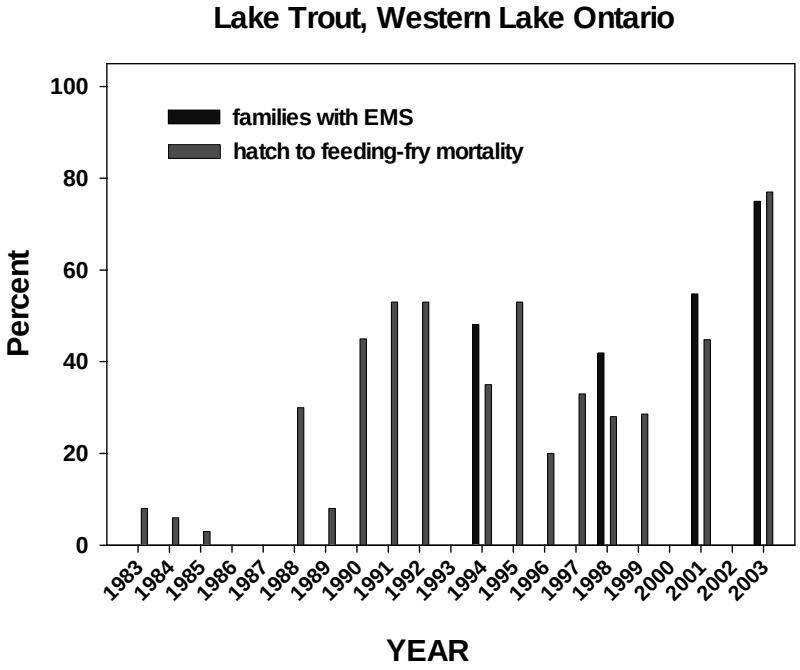

Figure 4. EMS in lake trout from Lake Ontario, 1983-2003. (data provided by John Fitzsimons, Department of Fisheries and Oceans, Canada)

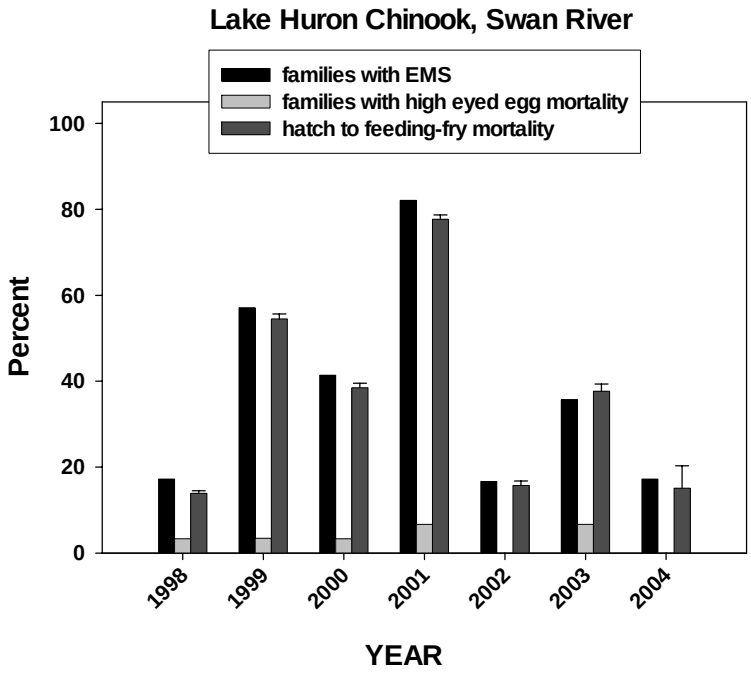

Figure 5. EMS in Lake Huron chinook salmon 1998-2004 at Swan River (data provided by Martha Wolgamood, Michigan DNR).

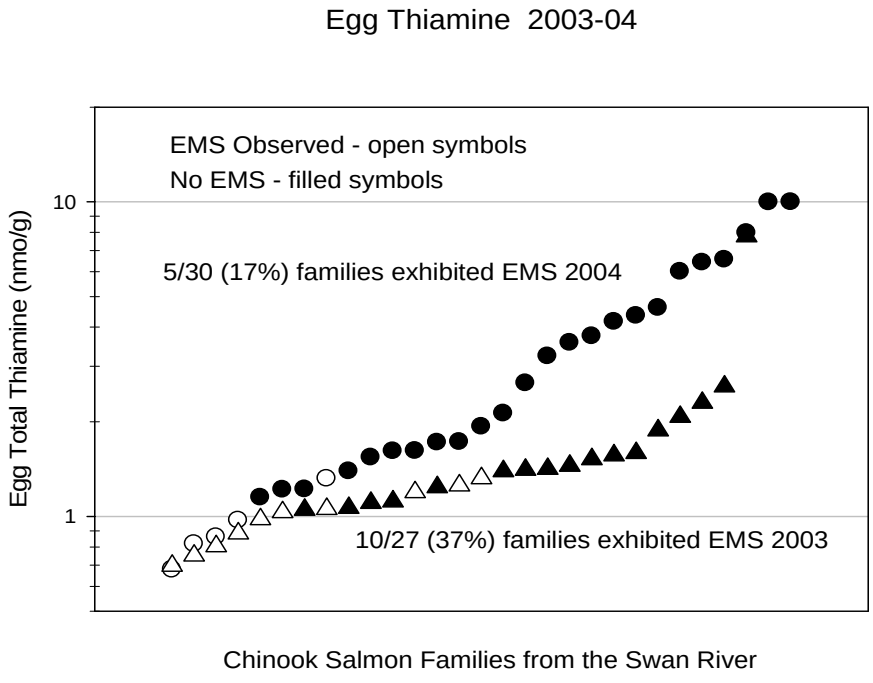

Figure 6. Thiamine concentrations in eggs of Lake Huron chinook salmon 2003 (△) and 2004 (○) at Swan River (data provided by Dale Honeyfield, USGS Wellsboro).

Lake Huron. Brown et al (page 14) reported that EMS in Swan River chinook salmon from Lake Huron decreased from 37% in 2003 to 17% in 2004 (**Figure 5**). For a majority of fish this was also associated with a marked improvement in egg thiamine concentrations. About 30% of chinook eggs would be considered as thiamine replete (> 4 nmol/g) in 2004 compared to 4 % in 2003 (**Figure 6**). Rook and Werner (page 15) reported on EMS status of the offspring of Atlantic salmon spawned from the St. Mary's River in 2003 and 2004. This stock is believed to forage mainly in Lake Huron. EMS was relatively low in the last two years compared to rates observed in the offspring of fish collected in previous years. Lake trout egg thiamine has also increased in several but not all areas of Lake Huron (**Table 1**, Honeyfield, unpublished).

Table 1. Lake Huron lake trout mean egg total thiamine (nmol/g) concentration and the percentage of the egg samples with < 1.5 nmol/g and percentage of the egg samples with < 4.0 nmol/g.

Spawning Location	Year	N	Egg Total Thiamine	% < 1.5	% <4.0
Parry Sound	2001	29	3.91[a]	17	66
	2004	10	7.50[b]	0	20
Owen Sound	01.'02,'03, '04	42	6.28	7	36
South Bay	2003	21	7.79[a]	0	14
	2004	12	10.46[b]	0	0
Yankee Reef	'01, '02	19	2.51[a]	37	84
	2004	14	5.36[b]	21	50
Six Fathom Bank	'01, '02, '04	35	3.17	40	74
Drummond Island	'02, '03, '04	45	3.90	20	71

[a b] Means within location with different letters are significantly different P<0.05).

We conclude that EMS continues to be an issue in Lake Michigan, and Lake Ontario but it appears to have diminished in Lake Huron chinook and Atlantic salmon and this trend corresponds to improvements in egg thiamine concentrations in all salmonine species where egg thiamine concentrations have been measured.

Relationships between Salmonine Recruitment and Prey Abundance

Based experimental work and ecoepidemiological assessments, we hypothesized that thiamine deficiency induced by alewife was important variable in natural recruitment of Great Lakes salmonids (Brown et al. 2005c). Therefore measuring population change of forage species, especially alewife and recruitment of top predators provides evidence to support or refute this position. Schaeffer and Riley (page 19) reported on alewife population trends from USGS trawl surveys in Lake Huron. They reported recent dramatic declines of alewife abundance throughout Lake Huron, such that in 2003 and 2004 alewife abundance was at the lowest levels in the last 20 years **(Figure 7)**. More importantly, evidence of natural reproduction of lake trout in Michigan waters of Lake Huron was noted. Natural reproduction in lake trout has been extremely limited in recent decades. Rarely have unclipped wild juvenile lake trout fry have been captured by USGS bottom trawl surveys. In 2004-2005, the USGS bottom trawl surveys captured unclipped juvenile lake trout throughout the Michigan waters of Lake Huron. The last time significant numbers of unclipped juvenile lake trout was observed occurred in the 1984 year class which also followed a period of extremely low alewife abundance. The years 1983-1984 and 2003-2004 represent the lowest levels of alewife abundance in the entire 30 year monitoring period. The correspondence of significant natural reproduction in lake trout following these periods of all time low alewife abundance is remarkable. Schaeffer and Riley (page 19) suggested two potential mechanisms to explain reduced lake trout reproduction in the presence of alewife: 1) alewife predation on lake trout fry

or 2) Thiamine Deficiency Complex. They concluded that current data are insufficient to confirm mechanisms contributing to the association between alewife abundance and low lake trout reproduction and recommended an integrative monitoring program to help establish cause and effect relationships. These changes occurring in Lake Huron fishery are of considerable scientific and managerial significance and needs to be monitored closely.

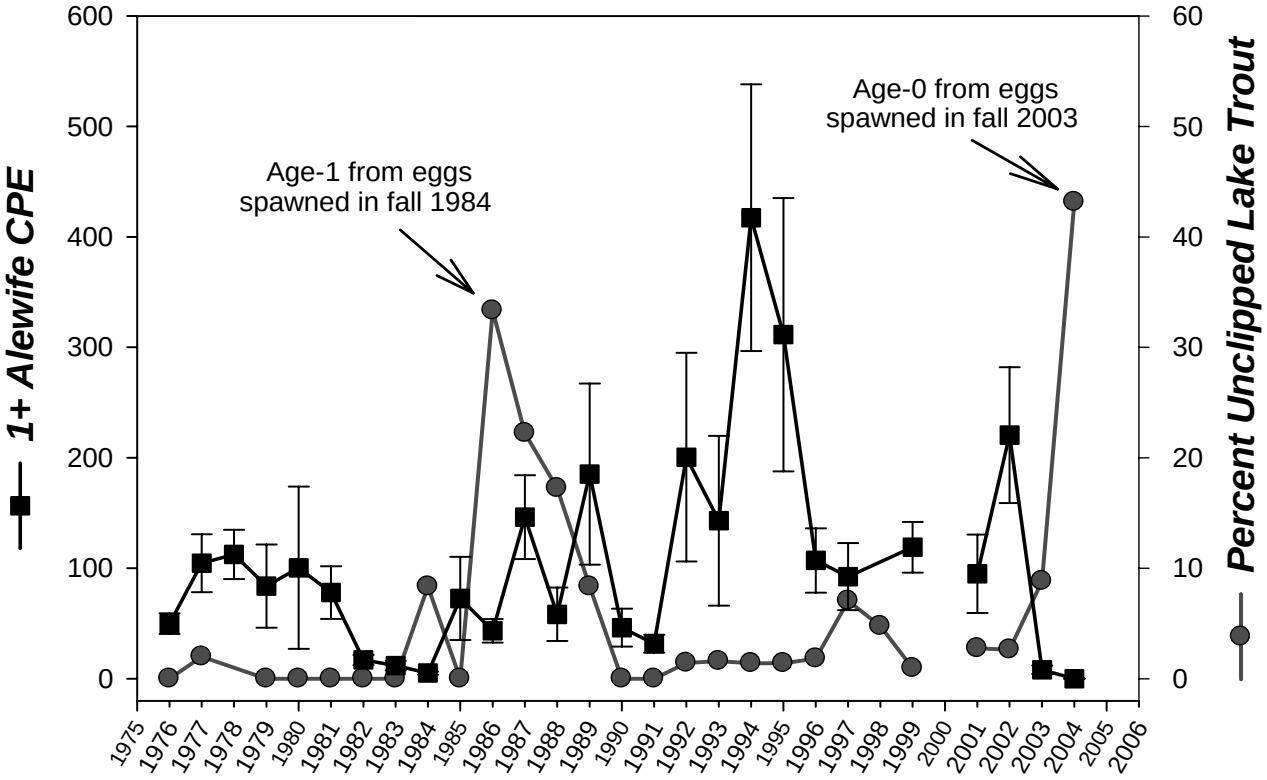

Figure 7. Alewife abundance estimates and unclipped lake trout captured in USGS assessment trawls from Michigan State waters of Lake Huron (data provided by Jeffrey Schaeffer and Stephan Riley, USGS, Great Lakes Science Center).

Consequences of Low Egg Thiamine

Thiamine bath treatment of fry has been shown to be effective in preventing mortality (McDonald et al 1998; Brown et al 2005b). Such rehabilitated fry appear normal and grow well, but possible long-term subtle effects on their behavior or physiology are unknown. Ketola et al (page 17) examined the effect of transitory deficiency in steelhead fry from Lake Ontario and landlocked salmon from Cayuga Lake that developed signs of deficiency and were reversed by immersion for 24 hr in water containing thiamine. Control fry were immersed as eggs during water hardening and when first hatched. All fry were reared and fed a standard feed containing adequate thiamine. After they became fingerlings and yearlings, their avoidance responses to culture water containing added zinc sulfate or iron sulfate were tested. Results showed that both control and thiamine rehabilitated steelhead and Atlantic salmon altered their behavior

to the test chemicals. However, the responses differed between control and thiamine rehabilitated fish. It appears that thiamine deficiency in fertilized ova of steelhead and landlocked salmon fry may cause long-term neurological impacts as evidenced by their significantly different responses to sub-lethal water borne zinc sulfate and iron sulfate. Gene expression may also provide evidence linking low egg thiamine to potential long term outcomes. Rise et al (page 18) used genomics tools and techniques to study the molecular pathogenesis of EMS. So far they found altered expression of several genes in thiamine deficient lake trout eggs, embryos, and larvae, and hypothesized that the functional annotations of these genes will point to molecular pathways in EMS. In the egg microarray experiment, differences in genes had functional annotations such as iron ion homeostasis, cell cycle regulation, transcription factor activity, and metallopeptidase activity; in the sac fry microarray experiment, differences in gene functional annotations included intra-Golgi traffic, ubiquitin-dependent protein catabolism, protein biosynthesis, and proteolysis. Quantitative RT-PCR confirmed significant alterations in 12 molecular biomarkers of thiamine deficiency in unfertilized eggs, including ribonucleotide reductase M2, cathepsin B, beta-2 microglobulin, ferritin H and upstream transcription factor 1. Planned research will include further characterization of the subtracted egg libraries, as well as construction and characterization of embryonic and larval lake trout cDNA libraries enriched for genes responsive to maternal thiamine deficiency. The goal of this research is to provide a molecular picture of EMS which will increase our understanding of the current fishery problem and provide a model that can be used to understand other non-infectious and infectious diseases affecting lake trout and their management. Honeyfield et al (page 19) examined the previously unexplored effects of thiamine deficiency on immune function and disease resistance. Thiamine is a critical co-factor in the metabolic pathway for energy and the production of the essential sugar ribose found in DNA and RNA. In thiamine replete and depleted lake trout, lymphocyte activity was differentially impacted. T-cell populations exhibited less proliferation following mitogen stimulation, while B-cells did not appear to be impacted. T-cells play a critical role in immunity to intracellular pathogens such as viruses (e.g. IPNV) and some bacteria (BKD). Macrophage bactericidal activity and cytotoxic cell activity did not appear to be impacted. Thiamine depleted lake trout may be more susceptible to diseases caused by intracellular pathogens as a result of T-cell dysfunction. Direct disease challenge studies are needed to confirm the implications of thiamine deficiency.

Thiaminase in the Great Lake Ecosystem

Because salmonine EMS is likely the result of a dietary predominance of prey species like alewives that are high in thiaminase (Honeyfield et al. 2005), understanding the factors influencing thiaminase activity in alewives is important for managing the consequences of salmonine thiamine deficiency in the Great Lakes. Previous work has shown that the thiaminase activity of alewives exhibits high between and within lake variability but the causes of the variations are unknown (Fitzsimons et al., 2005b; Tillitt et al. 2005). Consequently, Lepak and Kraft (page 20) evaluated stress as a factor affecting thiaminase measured in alewife. Because thiaminase expression may be linked to visceral bacteria in alewife, they hypothesized that stress related suppression of immune response may allow for bacterial proliferation and a subsequent increase in thiaminolytic activity. However, the stressors examined (e.g. low temperature, conductivity, nutrient/sediment loads) did not influence alewife thiaminase. The best predictor of thiaminase expression in alewife and gizzard shad was fish condition as measured by water content. Reduced water content was strongly correlated to elevated thiaminase expression (Kraft et al, page 20). It has previously been found that among-stock variation in thiaminase activity of Finger Lakes alewives was correlated with their lipid content (Fitzsimons et al., 2005b). To investigate the relationship further Fitzsimons (page 21) collected alewives from Conesus Lake (New York) in the fall and fed them isonitrogenous diets having differing lipid content over a four month period. Although fish grew in length and weight and their lipid content increased over this period, there was no relationship between thiaminase activity and either dietary or tissue lipid level suggesting that the earlier correlation may be associated with some as yet unidentified factor(s). The development of a non-radioactive thiaminase

assay continues to be a challenge. Kraft et al (page 20) provided an update on development of a non-radiometric thiaminase assay by Tadgh Begley (Cornell University). Substrate design based on three dimensional rendering of the thiaminase active site is being evaluated. However, results have not been promising and there is still an important requirement for quantitative non-radiometric thiaminase assay.

Mitigation of Thiamine Deficiency in Broodstock

Meuninck (page 14) reported on investigative efforts by the Bodine State Fish Hatchery to improve offspring survival from summer-run steelhead (Skamania) from the St. Joseph River. One half of the captured steelhead adults were IP injected with thiamine. At hatch, half of all the sac fry will be given a thiamine flow-through treatment. This first phase of this investigation will determine whether the thiamine injection will improve survival of adults during the holding period prior to spawning. The second will investigate the efficacy of maternal thiamine treatment versus water-borne treatments. The results may lead to changes in the thiamine treatment protocols with a substantial savings in the money spent on thiamine. IP injection has not been evaluated to eliminate long term effects resulting from thiamine deficiency in egg development but in theory should be successful. Along similar lines, Ketola et al (page 18) investigated the feasibility of enhancing first spawning and recruitment of steelhead and landlocked salmon stocked in waters with alewives or other forage fishes containing thiaminase by injecting fingerlings with an internal supply of long-lasting thiamine or one of its analogs. Five analogs were evaluated by injecting fry or fingerling steelhead trout held in the laboratory. While metabolically available thiamine was noted for all forms evaluated, in fry dibenzoyl-thiamine retention was greater than any other analog. Testing is continuing and these compounds will be evaluated at 1 to 2 years to determine their potential for application in hatchery fish stocked in lakes where recruitment may be impaired by thiaminase-containing forage fishes.

Recommendations

The following is the list of recommended research issues developed during the Group Discussion period at the meeting.

1. Coordinated EMS Research Effort

- Develop an integrated sampling protocol that can be sustained with limited resources to optimize time and effort expended to gather data addressing the many factors thought to be involved in reproductive failure of lake trout within Lakes Michigan, Huron and Ontario. The protocol should also take into account existing historical data sets and collected site specific data to link egg thiamine, lake trout recruitment and biomass of prey species. Sampling should include age-0 recruits.

2. Implications of Thiaminase

- Determine source(s) of thiaminase in clupeids and investigate the presence or absence of de novo thiaminase synthesis in alewife. As an ancillary question, determine how alewife handle or compartmentalize thiaminase while protecting their own physiological thiamine requirements.

- Determine factors that modulate thiaminase activity in alewife. Dietary lipid did not appear to affect thiaminase activity in alewife but are there other dietary items that affect alewife

thiaminase content? Is there a relationship between alewife food items and their thiaminase activity?

- Develop a PCR assay or functional ELISA to measure thiaminase. Without an assay that does not depend on radioactive thiamine data collections will not be comprehensive and analysis limited to licensed facilities.

- Determine the temporal and spatial distribution of thiaminase in the foodweb. Preliminary data from net plankton has shown the presence of thiaminase in the lower foodweb, but when and where it is found is not documented. Secondly, the timing and location may affect larval fish. What are the consequences of thiaminase in lower trophic food items for larval fish? For example, does lower trophic thiaminase affect larval fish survival and fitness of lake trout or other important species?

3. Implications of Thiamine Deficiency on Physiology and Behavior

- Determine the long term affects on behavior, immune function and physiological functions of rehabilitated fish after thiamine deficiency in egg and early fry development. Latten affects such as behavior to chemical stimuli or inability to regulate immune system appear to have a profound affect on overall animal fitness. What are the latent affects and how important are they?

- Differences in gene expression between thiamine deficient and replete lake trout have been found but does it translate into functional differences that affect a fish survival?

- Develop a complete genomic library for lake trout to enhance discovery of potential genes affected by thiamine deficiency or other gene functions changes specific to lake trout.

- Use genomic techniques to develop a set of EMS resistance biomarkers. For example, examine the adaptive advantage for the apparent lower thiamine concentrations observed in chinook salmon.

- Conduct disease challenge studies to determine the affect of reduced lymphocyte T-cell proliferation or other immune functions observed in thiamine deficiency lake trout.

4. Consideration for Data Management

- Develop infrastructure and support for a consolidated accessible database to support fishery management data. This should be a lake committee (or higher organizational level) commitment. Data may include but is not limited to: egg thiamine, incidence of EMS, forage fish composition/abundance, lower trophic foodweb inventory and associated biochemical composition, top predator population estimates, invasive species, and lake physical, chemical and limnological variables. Finally a listing of sample sources and contacts for back-testing,

Acknowledgments

We sincerely thank the Great Lakes Fishery Commission and staff for their long-standing support of EMS research in the Great Lakes Basin. We especially want to extend our appreciation for the efforts of our speakers and other meeting attendees who took time from their busy schedules to share their work and

ideas for EMS research. We thank Lisa Brown (National Water Research Institute, Burlington) for help with the meeting arrangements.

References

Brown, S.B., D. C. Honeyfield, J. G. Hnath, M. Wolgamood, S. V. Marcquenski, J. D. Fitzsimons and D. E. Tillitt. 2005a. Thiamine Status in Adult Salmonines in the Great Lakes. Journal of Aquatic Animal Health 17: 59-64.

Brown, S.B., L. R. Brown, M. Brown, K Moore, M. Villella, J. D. Fitzsimons, B. Williston, D. C. Honeyfield, J. P. Hinterkopf, D. E. Tillitt, J. L. Zajicek and M. Wolgamood. 2005b. Effectiveness of Egg Immersion in Aqueous Solutions of Thiamine and Thiamine Analogs for Reducing Early Mortality Syndrome. Journal of Aquatic Animal Health 17: 106-112.

Brown, S.B., J. D. Fitzsimons, D. C. Honeyfield and D. E. Tillitt. 2005c. Implications of Thiamine Deficiency in Great Lakes Salmonines. Journal of Aquatic Animal Health 17:113-124.

Fitzsimons, J. D., B. Williston, P. Amcoff, L. Balk, C. Pecor, H. G. Ketola, J. P. Hinterkopf and D. C. Honeyfield. 2005. The Effect of Thiamine Injection on Upstream Migration, Survival, and Thiamine Status of Putative Thiamine-Deficient Coho Salmon. Journal of Aquatic Animal Health 17: 48-58.

Fitzsimons, J. D., B. Williston, J. L. Zajicek, D. E. Tillitt, S. B. Brown, L. R. Brown, D. C. Honeyfield, D. M. Warner, L. G. Rudstam and W. Pearsall. 2005. Thiamine Content and Thiaminase Activity of Ten Freshwater Stocks and One Marine Stock of Alewives. Journal of Aquatic Animal Health 17: 26-35.

Honeyfield, D. C., J. P. Hinterkopf, J. D. Fitzsimons, D. E. Tillitt, J. L. Zajicek and S. B. Brown. 2005. Development of Thiamine Deficiencies and Early Mortality Syndrome in Lake Trout by Feeding Experimental and Feral Fish Diets Containing Thiaminase. Journal of Aquatic Animal Health 17: 4-12.

Ketola, H. G., T. L. Chiotti, R. S. Rathman, J. D. Fitzsimons, D. C. Honeyfield, P. J. VanDusen and G. E. Lewis. 2005. Thiamine status of Cayuga Lake rainbow trout and its influence on spawning migration. North American Journal of Fisheries Management. 25:1281–1287.

Marquenski, S.V. and S.B. Brown. 1997. Early mortality syndrome in the Great Lakes, p. 135-152 *In* Rolland, R.M., M.Gilbertson, R.E. Peterson (Eds.) Chemically Induced Alterations in Functional Development and Reproduction in Fishes. SETAC Press Pensacola FL.

McDonald, G., J. Fitzsimons, and D.C. Honeyfield (Eds). 1998. Early life stage mortality syndrome in fishes of the Great Lakes and Baltic Sea. Symposium 21, p 1-177. American Fisheries Society, Bethesda, MD 20814.

Tillitt, D. E., J. L. Zajicek, S. B. Brown, L. R. Brown, J. D. Fitzsimons, D. C. Honeyfield, M. E. Holey and G. M. Wright. 2005. Thiamine and Thiaminase Status in Forage Fish of Salmonines from Lake Michigan. Journal of Aquatic Animal Health 17: 13-25.

Abstracts of Presentations

Introduction: EMS in the Great Lakes

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John D. Fitzsimons, Bayfield Institute, Department of Fisheries & Oceans, Burlington, ON, Canada
Carol Edsall, Great Lakes Science Centre, U.S. Geological Survey, Ann Arbor, MI. USA.

The incidence of EMS in Great Lakes salmonids has been highly variable in the past. Therefore, it is important to continue to monitor EMS in Lake Michigan. Starting in the early 1970's, EMS was reported and has increased with time. Year to year variation in EMS that has been observed in conjunction with changes seen at the various trophic levels is one clue that may be useful for us to ultimately understand the root cause of this environmental problem. Eggs from thirty randomly selected females will be collected and reared by Martha Wolgamood, Michigan DNR, Wolf Lake State Hatchery. Percent EMS and egg thiamine were continuously recorded for Platte River Coho and Little Manistee chinook stocks. Very high levels of EMS coupled with very low thiamine levels occurred in Lake Michigan coho and chinook salmon for 1999 to 2001. The incidence of EMS dropped dramatically in 2002 with only an average of 16.4 and 26.7% of coho and chinook families, respectively, exhibiting mortality. In 2003, the incidence of EMS has increased to involve approximately 50% of reared families. EMS remains highly prevalent among Great Lakes Salmonines.

Thiamine Injection of Skamania Steelhead Broodstock Investigation

Dave Meuninck, Bodine State Fish Hatchery, Indiana DNR, Mishawaka, IN, USA

The best time to prevent the occurrence of early mortality syndrome in salmonids is to treat the spawners with an injection of thiamine hydrochloride (Honeyfield, personal communication). The Bodine State Fish Hatchery collects adult summer-run steelhead (Skamania) from the St. Joseph River for use as broodstock for Indiana's Skamania program. Half of all harvested steelhead will be IP injected with thiamine. At hatch, half of all the sac fry will be given a thiamine flow-through treatment. The first phase of this investigation will determine whether the thiamine injection will improve survival of adults during the holding period prior to spawning. For the second phase, a 2 X 2 treatment matrix will be used to establish four treatment groups. At one extreme, there will be a treated group having parents and sac fry that were thiamine treated. At the other extreme, neither the parents nor sac fry will have been given thiamine. It is hoped this investigation will support Dr. Honeyfield's findings and lead to a change in the thiamine treatment protocol at Bodine resulting with improved survival of captive adult and early life stage Skamania and a substantial savings in the money spent on thiamine.

Evaluation of Stocking Strategies to Re-colonize Lake Trout Spawning Reefs in Lake Michigan

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Jory L. Jonas, Randall M. Claramunt, Michigan DNR, Charlevoix, MI, USA
Brian Breidert, Indiana DNR, Michigan City, IN, USA
Mark P. Ebener, Chippewa/Ottawa Resource Authority, Sault Ste. Marie, MI, USA
Guy W. Fleischer, U.S. Geological Survey, Ann Arbor, MI, USA
Richard Hess, Illinois Department of Conservation, Des Plaines, IL, USA
Archie W. Martell, Jr., Little River Band of Ottawa Indians, Manistee, MI, USA

Erik J. Olsen, Grand Traverse Band of Ottawa and Chippewa Indians, Suttons Bay, MI, USA
Patrick C. McKee, Michael L. Toneys, Wisconsin DNR, Sturgeon Bay, WI, USA

Our objective was to determine how effective site-specific stocking is for re-colonizing historically important spawning reefs. We compared the relative abundance of spawners during fall 1999-2001 on 19 stocked spawning sites to 25 sites that received no fish. Densities of adult fish were significantly higher on stocked onshore and offshore sites than on unstocked reefs, and suggest that site-specific stocking is effective at establishing spawning aggregations as opposed to relying on self-colonization. Overall spawner densities were low and young at most sites, which indicate that lake trout parental stock size is low and may partly explain the lack of significant natural reproduction in Lake Michigan. Less than 3% of all spawners could have been wild fish and indicates little natural reproduction in past years. Recaptures of coded wire tagged fish of five separate strains indicated that most fish returned to their stocking location or to a site nearby and that dispersal from stocking locations was around 100 km for most strains. The relative survival of the Seneca Lake strain was higher than that of the Jenny Lakes-Lewis Lake strain or the Superior –Marquette for older year classes examined but was less evident for younger year classes just recruiting into the parental stock These results will be used to draft a new management plan that will increase the probability of sustained natural reproduction by lake trout in Lake Michigan.

Lake Michigan Lake Trout Egg Thiamine Content

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Egg thiamine concentrations are an indicator of the risk of lake trout developing early mortality syndrome (EMS) within a family group. Egg thiamine values less than 1.5 nmol/g are associated with high incidence of fry mortality. Fry hatching from eggs with greater than 1.5 nmol/g but less than 4 nmol/g are at significant risk of the secondary effects of thiamine deficiency. These risks include impaired growth, vision, predator avoidance, prey capture and immune function. From the 2004 spawning season, 120 lake trout egg samples from 12 sites were collected. Analysis of egg thiamine was by HPLC method (Brown et al, 1998). For all sites the mean was 4.87 nmol/g with median value of 2.53 nmol/g. Thiamine ranged from 0.4 to 29.1 nmol/g. Thirty-seven percent of the families were at risk of overt fry losses from EMS and 60% were at risk from EMS and the secondary effects of thiamine deficiency. The site with the lowest egg thiamine was Grand Traverse Bay. Thiamine in all fish from this site were below 1.5 nmol/g and the number of samples available was low. The site with the highest mean egg thiamine was East Reef (6.38 nmol/g) in Wisconsin waters. The median egg thiamine value (5.68 nmol/g). Among the four reef sites, egg thiamine was the highest at East Reef and lowest at Beaver Island. Mean and median egg thiamine concentrations within the four off-shore reefs (Beaver Island, East Reef, Julian's Reef and Waukegan Reef) were 5.4 and 3.4 nmol/g, respectively. By comparison mean and median egg thiamine of 2004 Lake Huron (Parry Sound) samples were 7.5 and 6.3 nmol/g, respectively. Parry Sound is considered a success for lake trout restoration of a self-sustaining stock. Although egg thiamine values appear to be edging upward slightly when compared to eggs collected between 1996 and 2003, most sites appear to have eggs that are at significant risk of either direct fry mortality or impaired functions due to thiamine deficiency

Relationship of St. Mary's River Atlantic Salmon (Salmo salar) Population Factors with Forage-Base Population Factors

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Early mortality syndrome (EMS) is the term used to describe the trend of increased mortality currently affecting the developing offspring of salmonids in the Great Lakes region. This trend of increased mortality is linked to the presence of a thiamine degrading enzyme known as thiaminase in Alewives (*Alosa pseudoharengus*), the main food source for salmonids throughout the Great Lakes. The Atlantic salmon (*Salmo salar*) population which spawns in the St. Mary's River region near Sault Ste. Marie, Michigan is known to be heavily dependent upon alewives as a food source. In 2003 and 2004, a study was designed to monitor egg-thiamine levels in spawning Atlantic salmon and monitor EMS related hatchery mortality. In addition to data collected from spawning Atlantic salmon in 2003 and 2004, analysis of LSSU's historic salmon data was performed. Results indicated mean size, growth rate, clutch size, and the overall condition of individuals from the 2003 and 2004 populations were lower than corresponding values observed in previously sampled populations. EMS related mortality rates were relatively low in the offspring of fish collected in both 2003 and 2004 compared to rates observed in the offspring of fish collected in previous years. These trends were found to correspond to an overall decrease in alewife numbers throughout Lake Huron in 2003 and 2004. The relationship to these trends and historical data going back to 1990 will also be discussed.

Thiamine deficiency complex in Lake Ontario Pacific Salmon

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Marion Daniels, Lake Ontario Management Unit Ontario Ministry of Natural Resources, Peterborough, ON, Canada.

To determine incidence of EMS and establish thiamine deficiency thresholds for occurrence of EMS in Lake Ontario salmonines, we conducted 3 experiments. Each experiment tracked 20-30 individual families of chinook and coho salmon from the Credit River and Salmon Rivers. Egg and fry samples were taken to determine thiamine concentrations and mortalities from fertilization to 3 weeks post-feed were tallied along with the clinical signs of EMS. In Credit River chinook, clinical signs of EMS were found in about 40 % of families, there were associates with thiamine deficiency but only small differences in d15N for EMS positive families. Credit River coho salmon exhibited thiamine levels mostly above threshold for EMS-related effects and the overall amount of EMS was too low for meaningful interpretation of possible associated factors. In the Salmon River coho, egg thiamine concentrations were mostly near and below threshold for EMS related effects. The location differences in fatty acids, carotenoids and thiamine all imply different forage-related factors in Salmon River coho relative to Credit River coho. These findings suggest that greater incidences of EMS and lower thiamine levels in Salmon River coho stocks may be related to forage differences.

Among-Stock Variation in the Thiamine-EMS Dose Response – Possible Causes

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Earlier work based on the thiamine-EMS dose-response suggested that Lake Ontario lake trout embryos might be more sensitive to the effects of a thiamine deficiency than embryos from other stocks also suffering from a thiamine deficiency. However it is difficult to draw definitive conclusions from such data

because of year to year and fish to fish variation and the lack of perfect coincidence among stocks in egg thiamine concentration. To investigate this further we used a mix of thiamine replete (Lake Manitou) and thiamine deficient (Lake Ontario, Seneca, Cayuga, and Charleston Lakes) stocks to look at 1) the response to the model thiamine antagonist oxythiamine as a measure of sensitivity to a thiamine deficiency and 2) loss rate of thiamine by the developing embryo as a measure of vulnerability to developing a thiamine deficiency. Compared to sac-fry from Seneca Lake, those from Lake Ontario proved less sensitive to the effects of oxythiamine and exhibited a similar dose-response relationship to sac-fry from Lake Erie. Loss of total thiamine (e.g. sum of TPP, TMP and free thiamine) between fertilization and swim-up showed significant among stock variation with greatest losses occurring in the thiamine replete stock. Among the thiamine deficient stocks, loss rates of total thiamine were much lower and appeared to be inversely correlated with swim-up thiamine concentration although the Seneca Lake stock was anomalous in having a much lower rate of loss for its swim-up thiamine concentration than the other stocks. With regards to TPP, the thiamine replete stock was unique in gaining TPP over the period fertilization to swim-up whereas all of the thiamine deficient stocks lost TPP over this same period with the exception of Seneca Lake where no change occurred. Based on our investigations to date Lake Ontario embryos are neither overly sensitive to the effects of a thiamine deficiency nor exhibit anomalous embryonic thiamine clearance rates. In contras,t the Seneca Lake stock while apparently more sensitive to the effects of a thiamine deficiency may compensate for this by reduced clearance and as a result conserving levels.

Influence of Transitory Deficiency of Thiamine in Fry on Avoidance Behavior of Fingerling Steelhead and Landlocked Salmon

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Jeff Robins, New York State Department of Environmental Conservation, Cortland, NY, USA

Often hatchery fry of several species of feral salmonid brood stocks develop thiamine deficiency that is reversed by immersing fry in water containing added thiamine. Such rehabilitated fry appear normal and grow well and may be stocked, but possible long-term subtle effects on their behavior or physiology are unknown. Therefore we examined the effect of transitory deficiency in fry of steelhead from Lake Ontario and landlocked salmon from Cayuga Lake that developed signs of deficiency and were reversed by immersion for 24 hr in water containing 1,000 PPM thiamine. Control fry were immersed as eggs during water hardening and when first hatched. These fry were reared and fed a standard feed containing adequate thiamine. After they became fingerlings and yearlings, we tested their avoidance responses to culture water containing added zinc sulfate or iron sulfate. Fingerlings and yearlings were tested in special paired avoidance units each constructed with three adjacent chambers arranged in a line with passageways between adjacent chambers (2.1 liters water capacity/chamber). The outer chambers were supplied (200 ml/min) with either laboratory culture water (control) or the same water with additions of zinc sulfate or iron sulfate. Control water had a hardness of 240 ppm as CaCO3; the levels of zinc and iron were not detectable (<0.003 ppm) by inductively coupled plasma spectrophotometry. Behavior was recorded simultaneously for 60 minutes for two sets of fish (control and rehabilitated) by use of a video camera to avoid human disturbance. From the video recordings, observations and records of location of each of five fish (control *vs* test chambers) were recorded every minute between 30-60 minutes during a one hour test. Tests were repeated 6 or more times, reversing the positions of the fish and locations of introduction of test water. Results showed that both control and rehabilitated steelhead significantly avoided 0.03 to 0.2 ppm added zinc (as zinc sulfate), while rehabilitated steelhead avoided it significantly more strongly than controls. In contrast, rehabilitated steelhead significantly preferred 0.5 ppm added iron (as iron sulfate) more than control fish, but both groups strongly preferred iron at higher levels (2 ppm). Landlocked salmon also avoided zinc but only when added at much a higher concentration than that for

steelhead, i.e., 0.1 vs 0.03 ppm. Further, rehabilitated salmon were significantly less responsive to added zinc than controls. In contrast to steelhead, control salmon significantly avoided 0.5 to 0.75 ppm iron, whereas rehabilitated salmon did not significantly respond to iron added at any level tested. In conclusion, transitory deficiency of thiamine in fry of steelhead and landlocked salmon causes long-term neurological impacts as evidenced by their significantly different responses to sub-lethal additions of zinc sulfate and iron sulfate in water.

Activity and Retention of Thiamine and its Analogs in Steelhead and Landlocked Salmon

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Jeff Robins, New York State Department of Environmental Conservation, Cortland, NY

We investigated the feasibility of enhancing first spawning and recruitment of steelhead and landlocked salmon stocked in waters with alewives or other forage fishes containing thiaminase. Thiaminase is an enzyme that destroys thiamine (vitamin B1) in predators that consume them, thereby reducing deposition of thiamine in their eggs to levels insufficient for normal survival of their fry. As a possible way to enhance recruitment, we assessed the feasibility of injecting fingerlings with an internal supply of long-lasting thiamine or one of its analogs. Specifically, we examined metabolic activity, tolerance, and retention of thiamine and five analogs injected into fry or fingerling steelhead trout held in the laboratory. While activity of all forms was demonstrated in fry, tolerance and storage up to six months was significantly greater for dibenzoyl-thiamine (DBT) than any other. These retention results will be evaluated at 1 to 2 years to confirm their potential for application in hatchery fish stocked in lakes where recruitment is normally impaired by thiaminase-containing forage fishes.

Identifying Gene Expression Signatures of Maternal Thiamine Deficiency in Lake Trout Eggs and Larvae

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Dale C. Honeyfield U.S. Geological Survey, Northern Appalachian Research Laboratory, Wellsboro, PA, USA

We use genomics tools and techniques to study the molecular pathogenesis of Early Mortality Syndrome (EMS). EMS, a disease associated with low egg thiamine levels, causes early life stage mortality and low recruitment of Great Lakes salmonids including lake trout (*Salvelinus namaycush*). We aim to identify the key genes dysregulated in thiamine deficient lake trout eggs, embryos, and larvae, and hypothesize that the functional annotations of these genes will point to molecular pathways altered in EMS. We have created reciprocal subtracted cDNA libraries from thiamine deficient (total thiamine 259-443 pmol/g) and thiamine replete (9439-9793 pmol/g) lake trout eggs. Approximately 200 clones from these libraries have been sequenced and identified, including genes involved in DNA synthesis, antigen processing, cytoskeleton organization, proteolysis, and regulation of transcription. In addition we have used a ~3500 gene salmonid cDNA microarray to identify lake trout egg and sac fry genes responsive to maternal thiamine depletion. In the egg microarray experiment, dysregulated genes had functional annotations such as iron ion homeostasis, cell cycle regulation, transcription factor activity, and metallopeptidase activity; in the sac fry microarray experiment, dysregulated gene functional annotations included intra-Golgi traffic, ubiquitin-dependent protein catabolism, protein biosynthesis, and proteolysis. Quantitative RT-PCR (QPCR) using histone H2AZ as a normalizer gene confirmed 12 molecular biomarkers of thiamine deficiency in unfertilized eggs, including ribonucleotide reductase M2 (1512±131 fold suppression), cathepsin B (30.4±0.7 fold

suppression), beta-2 microglobulin (7.7±0.3 fold suppression), ferritin H (5.9±0.2 fold suppression), and upstream transcription factor 1 (1.4±0.1 fold induction). Future research includes further characterization of the subtracted egg libraries, as well as construction and characterization of embryonic and larval lake trout cDNA libraries enriched for genes responsive to maternal thiamine deficiency. In addition, 16,000 gene salmonid cDNA microarrays will be used to further characterize the global gene expression responses of lake trout eggs, embryos, and larvae to maternal thiamine depletion. Molecular biomarkers of thiamine deficiency will be tested in QPCR-based assays on templates prepared from feral lake trout eggs of various thiamine levels. The goal of this research is to provide a molecular picture of EMS which will increase our understanding of the current fishery problem and provide a model that can be used to understand other non-infectious and infectious diseases affecting lake trout management.

Evaluation of Immune Function in Thiamine Deficient Lake Trout

Dale C. Honeyfield, U.S. Geological Survey, Northern Appalachian Research Laboratory, Wellsboro, PA, USA
Chris Ottinger, Christine Densmore, and Phil McAllister, U.S. Geological Survey, Leetown Science Center, Kearneysville, WV

One unexplored aspect of early mortality syndrome (EMS) is the effect of thiamine deficiency on immune function and disease resistance. Thiamine is a critical co-factor in the metabolic pathway for energy and the production of the essential sugar ribose found in DNA and RNA. In this report the effect of thiamine deficiency on immune function is presented. Lake trout (200-250 g) were reared on a diet that limited body stores of thiamine but was adequate for growth and survival. The fish were then allocated to two treatment groups; adequate or marginal thiamine. Fish in the marginal group were first fed a thiamine deficient diet based on casein- gelatin with bacterial thiaminase (CBT) until signs of deficiency were observed. During the tissue sampling phase, fish were maintained on either a semi purified diet with adequate thiamine (2 mg/kg feed) or marginal thiamine (0.4 mg/kg feed). In vitro immunoassays were conducted. Anterior kidney leukocytes from thiamine replete and thiamine depleted lake trout was purified with on Percoll density gradients. Cytotoxic cell activity, macrophage bactericidal activity, and lymphocyte mitogenesis compared using in vitro microplate assays. Leukocyte functions in thiamine replete and depleted fish compare by t-test with $P \leq 0.025$ taken to represent a significant difference. Macrophage bactericidal activity and cytotoxic cell activity did not appear to be impacted by the level of thiamine deficiency used in this study. However, lymphocyte activity was differentially impacted with T-cell populations exhibiting reduced proliferation following mitogen stimulation. B-cells do not appear to be impacted. T-cells play a critical role in immunity to intracellular pathogens such as viruses (e.g. IPNV) and some bacteria (BKD). Thiamine depleted lake trout may be more susceptible to diseases caused by intracellular pathogens as a result of T-cell dysfunction.

Prey Fish Trends in the Upper Great Lakes and Possible Links to Thiamine Deficiency

Jeffrey S. Schaeffer and Stephen C. Riley, Great Lakes Science Centre, U.S. Geological Survey, Ann Arbor, MI. USA.

Evidence of natural reproduction of lake trout in Lake Huron has been extremely limited in recent decades, and unclipped wild juvenile lake trout fry have rarely been captured by USGS bottom trawl surveys. In 1984 and in 2004-2005, however, USGS bottom trawl surveys captured unclipped juvenile lake trout throughout the Michigan waters of Lake Huron. Based on alewife population trends from bottom trawl surveys, these years represent the lowest levels of alewife abundance in the 30 year time series, suggesting that lake trout reproduction may occur only in years of low alewife abundance. Two

potential mechanisms to explain reduced lake trout reproduction in the presence of alewife are alewife predation on lake trout fry or Thiamine Deficiency Syndrome, whereby lake trout egg thiamine levels are suppressed by alewife consumption. There are currently insufficient data to explore either mechanism. We recommend that a lake trout egg thiamine monitoring program be developed in Lake Huron.

Evaluating the Effects of Environment and Stressors on Thiaminase Expression in Alewives and Gizzard Shad

Jesse Lepak and Cliff Kraft, Department of Natural Resources, Cornell University, Ithaca, NY, USA

Variability in alewife thiaminase activity has been associated with cyanobacteria blooms, fish condition, season, location, diet composition and population density, but no single or combination of these factors appears to adequately explain the wide range of variability. This study evaluated stress as a source of thiaminase expression in alewife. Cortisol is an immunosupressor that reduces the number of circulating lymphocytes available to suppress bacteria in fish, as a response to stress. Since thiaminase expression has been linked to visceral bacteria in alewife, it was hypothesized that a lowered immune response would allow for bacterial proliferation and a subsequent increase in thiaminolytic activity. Alewife were subjected to cold temperatures (0-4 °C), low conductivity (< 100 µs) and starvation trials to induce stress. Additionally, gizzard shad in ponds were subjected to different nutrient and sediment loads. Alewife stress was unrelated to thiaminase expression in all cases. The best predictor of thiaminase expression in alewife and gizzard shad was condition as measured by water content. Reduced water content was strongly correlated to elevated thiaminase expression.

Factors Influencing Thiaminase Levels in Gizzard Shad, Plus an Update on Thiaminase Assay Development

Clifford Kraft, Jesse Lepak, and Tadgh Begley, Departments of Natural Resources and Chemistry & Chemical Biology, Cornell University, Ithaca, NY, USA
Mike Vanni, Department of Zoology, Miami University, Oxford, OH, USA

Two aspects of a New York Sea Grant-funded project examining thiaminase in fishes will be reported. First, we will provide results from an experimental pond manipulation in which gizzard shad were reared in replicated ponds exposed to nutrient and sediment additions in a 2 X 2 factorial design. These experiments were conducted in summer 2004 at Miami University of Ohio's Ecology Research Center. Thiaminase levels in gizzard shad were greater in all treatments (nutrient addition, sediment addition, and sediment + nutrient addition; mean thiaminase level = 30,500 pmol thiamin/g/min) than plastic-lined control ponds without sediments (mean thiaminase level = 22,000 pmol thiamin/g/min). Ponds in which fish had high thiaminase levels also were higher in chlorophyll *a* than in control ponds, and gizzard shad within these ponds exhibited better condition than fish in control ponds. In the second portion of this presentation we will report results from an effort currently being undertaken within the lab of Tadgh Begley (Cornell University) to develop a non-radiometric thiaminase assay. Recent efforts within the Begley lab have identified similarities within the active sites of thiaminase I and thiaminase II that are responsible for the breakdown of thiamine (Begley's lab has also confirmed that thiaminase II is common within many taxa and is generally involved in thiamin synthesis). The substrate design effort for a new thiaminase assay has been guided by identification of the 3-D structure of thiaminase active sites, and a new strategy has been identified to develop an assay that will allow thiaminase activity to be assessed by

using a UV-visible spectrometer or colorimeter. The molecule required for this assay will be synthesized and tested as a substrate for thiaminase I and II by December 2005.

Effect of Dietary Lipid Level on Thiaminase Activity in Alewives

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Scott B. Brown and Lisa Brown, National Water Research Institute, Environment Canada, Burlington, ON, Canada
Rick Barrows, U.S. Department of Agriculture, Hagerman Fish Culture Experiment Station, Hagerman, Montana
Dale C. Honeyfield, Northern Appalachian Research Laboratory, U.S. Geological Survey, Wellsboro, PA, USA
Donald E. Tillitt, Columbia Environmental Research Center, U.S. Geological Survey, Columbia, MO, USA

Among-stock variation in thiaminase activity of Finger Lakes alewives was correlated with their lipid content. To investigate the relationship further we collected alewives from Conesus Lake (New York) in the fall and fed them isonitrogenous diets having low (8%), medium (14%), or high (20%) lipid content over a four month period. Although fish grew in length and weight and increased their lipid content over this period there was no significant relationship between thiaminase activity and either dietary or tissue lipid level for any of the sampling periods suggesting that the earlier correlation may represent autocorrelation with some as yet unidentified factor or factors.

Roster of Attendees to EMS Task Meeting, Ann Arbor, MI, September 22, 2005

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Agenda EMS Workshop September 22, 2005, Weber's Inn, Ann Arbor, MI
Sponsored by the Great Lakes Fishery Commission

Thursday September 22

08:30 Introduction: EMS in the Great Lakes ……Scott Brown, NWRI, Burlington.
08:50 Thiamine injection of Skamania Steelhead broodstock investigation. ……
……. Dave Meuninck, Indiana DNR, Bodine State Fish Hatchery.
09:10 Relative abundance, survival and site affinity of lake trout spawners in Lake Michigan.
…….Chuck Bronte, USFWS, Green Bay Fishery Resources Office
09:30 Lake Michigan lake trout egg thiamine……..……
…….Dale C. Honeyfield, USGS, Wellsboro
09:50 Relationship of St. Mary's River Atlantic Salmon (*Salmo salar*) population factors with forage-base population factors. ….Benjamin J. Rook and R. Marshall Werner, Lake Superior State University

10:10 Break

10:30 Thiamine deficiency complex in Lake Ontario Pacific Salmon……
……Scott Brown, NWRI, Burlington and Marion Daniels, OMNR, Peterborough.
10: 50 Lake to lake variation in embryonic clearance of thiamine and possible factors involved.
…… John Fitzsimons, DFO, Burlington
11:10 Influence of transitory deficiency as fry on avoidance behavior of fingerling steelhead and landlocked salmon. ……..George Ketola. USGS, Cortland
11:30 Activity and storage of thiamine and its analogs by steelhead and landlocked salmon.
……..George Ketola, USGS, Cortland

11:50 Lunch

13:00 Identifying gene expression signatures of maternal thiamine deficiency in lake trout eggs and larvae. ……Matthew Rise and Marlies Rise, Great Lakes WATER Institute
13:20 Immune function in thiamine deficient lake trout……
…… Dale Honeyfield, USGS, Wellsboro
13:40 Prey fish trends in the upper Great Lakes and possible links to thiamine deficiency.
…… Jeff Schaeffer. USGS, Ann Arbor
14:00 Evaluating the effects of environment and stressors on thiaminase expression in alewives.
…… Jesse Lepak, Cornell University, Ithaca
14:20 Factors influencing thiaminase levels in gizzard shad, plus an update on thiaminase assay development. ……..Cliff Kraft, Cornell University, Ithaca
14:40 The effect of dietary lipid on thiaminase activity in alewives…..……
……..…….. John Fitzsimons, DFO, Burlington
15:00 Great Lakes Science Center program direction……
…….Jerri Nichols and Stephen Riley, USGS, Ann Arbor

15:15 Break

15:30 Group Discussion