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Appendix A:

**Importance of Primary and Secondary Production in
Controlling Fish Tissue Mercury Concentrations**

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DISCLAIMER

This document is a draft report by staff of the California Regional Water Quality Control Board, Central Valley Region. No policy or regulation is either expressed or intended.

Appendix A: Importance of Primary and Secondary Production in Controlling Fish Tissue Mercury Concentrations will be appended to the **Statewide Mercury Control Program for Reservoirs** technical staff report, currently under development. *Appendix A* describes the scientific basis of the mercury conceptual model and linkage analysis, which will be fully described in the technical staff report. *Appendix A* will be submitted to formal scientific peer review together with the technical staff report.

Appendix A: Importance of Primary and Secondary Production in Controlling Fish Tissue Mercury Concentrations

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Introduction

The purpose of Appendix A is to discuss the importance of primary and secondary production in the bioaccumulation of methylmercury in fish in freshwater reservoirs. The paper is divided into six parts. First, statistical evidence is presented that the ratio of methylmercury to chlorophyll in a water body is positively correlated with fish tissue mercury concentrations. Second, the mechanisms whereby chlorophyll and primary production control the accumulation and transfer of methylmercury in aquatic food webs are described. Third, the phenomenon of cultural oligotrophication is reviewed with descriptions of how oligotrophication decreases nutrient concentrations resulting in reductions in primary and secondary production, decreases in fish yield, and likely increases in biotic mercury levels. A fourth section describes the results of nutrient fertilization programs in lakes and rivers to reverse cultural oligotrophication and restore economically important commercial and recreational fisheries. This section is included because “lessons learned” from decades of fertilization work elsewhere may be of interest to the State of California should it decide to consider fertilization as a temporary implementation option for reducing fish mercury levels while longer term mercury control measures are implemented. A fifth section estimates the amount of additional chlorophyll needed from a fertilization program to reduce fish mercury concentrations to safe levels in California reservoirs. The final section discusses the California Nutrient Criteria Program and how it is unlikely to negatively affect the implementation of a mercury fertilization program.

Three Recommendations of Appendix A:

- Programs are needed to conduct experimental nutrient fertilization studies in selected reservoirs to determine the feasibility of using nutrients to reduce fish tissue mercury

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concentrations. The program should also document the unintended negative consequences of nutrient additions.

- Programs are needed to restore, to the maximum extent possible, historic salmon runs in watersheds with elevated fish mercury levels.
- The State of California should consider protecting aquatic resources from both cultural oligotrophication and cultural eutrophication when developing a freshwater nutrient criteria policy. A scientifically based program should protect all the beneficial uses of the State's water bodies including human and wildlife consumption of fish.

Section 1: Methylmercury to Chlorophyll Ratio

A statistical evaluation was conducted to determine important factors in California reservoirs that were correlated with fish mercury concentrations. The statistical analysis is described fully in the staff report. Briefly, mercury concentrations in 350-mm length bass were employed as the dependent variable. Factors that had previously been reported in the peer reviewed literature to explain a significant amount of fish tissue mercury concentration were employed as independent variables. Not all variables could be transformed to fit the assumptions of parametric statistics so, both parametric and nonparametric analyses were conducted.

The ratio of unfiltered aqueous methylmercury to chlorophyll explained the largest amount of variation in mercury concentration in bass of any single variable evaluated for California reservoirs (Table 1A, Pearson's $r=0.67$, $P<10^{-6}$)². Larger ratios were positively correlated with higher fish tissue concentrations. It is important to note that the ratio explains statistically more variation than either aqueous methylmercury or chlorophyll alone. This observation emphasizes that it is the interaction of the two variables in each reservoir that is most important in determining fish mercury concentrations. To our knowledge no other study has used this ratio to predict biotic mercury levels although Watras and others (1998) and Sunda and Huntsman (1998) recommended a similar method for evaluating the biomagnification of other bioaccumulative substances in aquatic food webs

A multiple regression analysis was conducted to determine which combination of physical and chemical variables in Table 1A best correlated with fish mercury levels in California reservoirs. Several equations explained similar amounts of variation. All equations included a term for chlorophyll, average maximum annual reservoir water level fluctuation (AnnFluc), and some form of mercury. Equation 1A (adjusted $R^2=0.81$, $P<10^{-3}$, $N=26$) was selected to describe mercury bioaccumulation as it included the ratio of methylmercury to chlorophyll a. Including the ratio was believed important as it explained the largest amount of variation of any single physical or chemical factor evaluated in Table 1A. Equation 1A is:

$$\text{LnFish Hg} = 0.34(\text{MeHg/Chl } \underline{a}) + 0.39\text{AnnFluc} + 0.56(\text{aqueous THg}) - 0.91$$

² Although not reported here, a similar relationship was observed when the average mercury concentration of all top trophic level fish was analyzed.

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The ratio of methylmercury to chlorophyll a incorporates several important and largely independent processes and is used here as a quantitative measure of these separate factors. The aqueous methylmercury concentration of a reservoir is a proxy for the net result of all processes that produce and degrade methylmercury and for the amount of toxic metal available for biomagnification in the aquatic food chain. Increasing methylmercury concentrations in water are positively correlated with increasing fish tissue concentrations in California and elsewhere (Table 1A for California reservoirs, Brumbaugh et al. 2001; Sveinsdottier and Mason 2005; Rolfhus et al. 2011; Simonin et al. 2008; Wood et al. 2010). More details about the relationship between aqueous methylmercury and fish mercury levels are contained in the staff report.

The chlorophyll concentration of a reservoir is the net result of all the processes producing and consuming algae and is a measure of the amount of labile organic matter available as an energy source for secondary production. A positive correlation is often observed in lakes and reservoirs between chlorophyll a and fish yield (Hanson and Leggett 1982; Downing et al. 1990; Lee et al. 1991) suggesting that pelagic primary production is a factor influencing fish production. The importance of the positive relationship between the ratio of methylmercury to chlorophyll and fish tissue mercury concentrations emphasizes that a successful mercury control program must either decrease methylmercury production, increase primary production, or act on a combination of both processes.

The biomagnification of methylmercury in aquatic ecosystems is a dietary phenomenon (Harris and Bodaly 1998; Watras and Bloom 1992). Phytoplankton is the primary entry point for methylmercury into the food chain. Incorporation of methylmercury from water into phytoplankton is the largest single increase in the biomagnifications pathway. Methylmercury concentrations in phytoplankton increase 10^5 to 10^6 fold over concentrations in water (Mason et al. 1996; Pickhardt and Fisher 2007; Gorski et al. 2006 & 2008) while methylmercury increases at each subsequent trophic level are typically only a factor of two to five fold (Wood et al. 2010; Stewart et al. 2008; Cabana et al. 1994; Peterson et al. 2007). Therefore, phytoplankton is important both as a mercury concentration mechanism and as a source of organic energy for aquatic ecosystems. As will be shown later, changes in primary production rates, phytoplankton species composition, algal abundance and nutritional value all affect the relative transfer rate of methylmercury in aquatic systems and help determine final biotic methylmercury levels.

Key Points:

- The ratio of methylmercury to chlorophyll a explains the largest amount of the variation in mercury concentrations in bass of any single physical or chemical variable evaluated for California reservoirs.
- The ratio emphasizes that a successful mercury control program must either decrease methylmercury concentrations, increase primary production, or act on a combination of the two processes.

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Section 2: Primary Production and Mercury Bioaccumulation

Multiple studies have demonstrated that water bodies with higher chlorophyll levels have lower biotic mercury concentrations. These observations come from both descriptive field studies and from nutrient amendments to whole lakes and laboratory mesocosms. The purpose of the present review is to summarize the results of these studies and to identify the responsible biological mechanisms.

Descriptive Field Studies

Multiple papers have observed negative correlations in field studies between chlorophyll a and fish tissue mercury concentrations (Lange et al. 1993; Allen et al. 2005; Simonin et al. 2008; Melwani et al. 2010; Negrey et al. 2010). Lange and others (1993) evaluated the relationship between a suite of physical and chemical factors and mercury concentration in 3-year old largemouth bass from 53 Florida lakes. A negative correlation was observed with chlorophyll a ($r = -0.50$, $P < 0.001$). A best fit multiple regression model demonstrated that chlorophyll a and alkalinity were the two most important variables and together explained 45-percent of the variation in fish tissue levels. Allen and others (2005) examined methylmercury concentrations in macro invertebrates and brook stickleback in 12 Canadian boreal lakes. Mercury concentrations in aquatic biota were negatively associated with pH, water hardness, and \log_{10} chlorophyll a levels. The correlation between \log_{10} chlorophyll a and brook stickleback, the only fish species evaluated, was also significant ($r = -0.78$, $P < 0.01$). Significant negative correlations also existed for amphipods and several, but not all, macro invertebrates. No relationship was found for *Notonecta*. A potential explanation for the lack of a correlation with *Notonecta* is that the insects are semi aquatic and capable of migrating between lakes and feeding on terrestrial organic material. Simonin and others (2008) collected water samples and fish from 131 New York lakes. Target fish were large and smallmouth bass, walleye, and yellow perch. Chlorophyll a was negatively correlated with mercury levels in size-adjusted large and smallmouth bass ($r = -0.34$, $P < 0.05$) but not with yellow perch or walleye. Finally, Melwani and others (2010), Negrey and others (2010) and this study found a negative correlation (Table 1A) between mercury in 350-mm largemouth bass and chlorophyll a in California reservoirs.

A number of other descriptive field studies did not measure chlorophyll a but instead inferred productivity from nutrient concentrations (Kamman et al. 2004; Soneston 2003; Chen et al. 2005). This is a reasonable inference as strong positive relationships have been observed between nutrient concentrations, particularly phosphorus, and chlorophyll levels in freshwater lakes and reservoirs (Schindler 1977; Carpenter 2008; Schindler et al. 2008). Kamman and others (2004) examined a suite of water quality parameters in 92 New England lakes and compared the results to mercury concentrations in age-adjusted yellow perch. The authors divided their lakes into trophic states based on total phosphorus (TP) levels (oligotrophic < 8 $\mu\text{g/L}$, mesotrophic 8-16 $\mu\text{g/L}$ and eutrophic > 16 $\mu\text{g/L}$). Age-adjusted yellow perch mercury levels were significantly lower in eutrophic lakes ($P < 0.005$). Soneston (2003) measured fish mercury concentrations in 78 Swedish lakes and correlated tissue concentrations with 48

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physical and chemical parameters. The trophic status of each lake was established by measuring total phosphorus and total nitrogen (TN). A partial least square analysis was used to establish the relationship between mercury tissue concentrations and environmental variables. Lake nutrient status was found to have a statistically significant negative effect on perch mercury levels. Higher nutrient concentrations were associated with lower tissue concentrations. Finally, Chen and others (2005) used US EPA Environmental Monitoring and Assessment Program data from four different US EPA surveys of 150 lakes in the Northeastern United States. The primary purpose of the study was to identify lake attributes most likely associated with high fish tissue levels. Lakes were selected based on a stratified probability design to ensure that lake types were sampled proportional to their relative abundance. The study suffered from the fact that not all variables were measured in each study. Chlorophyll *a* was only analyzed in one survey. In this survey, pigment concentration was negatively associated with methylmercury in zooplankton. Nutrient concentrations (TN and TP) were measured in the other three surveys. Negative correlations were observed between nutrients and fish tissue levels in each of these surveys. Chen and others (2005) concluded that fish collected from lakes that were poorly buffered and had low pH, low productivity, and forested watersheds were at a higher risk of having fish with elevated mercury levels.

Controlled Laboratory and Field Nutrient Amendment Studies

Several laboratory and field fertilization experiments have been conducted to examine the effect of increasing phytoplankton density on methylmercury accumulation in aquatic food webs (Pickhardt et al. 2002 & 2005; Essington and Houser 2003; Kidd et al. 1999). In two separate laboratory studies Pickhardt and others (2002 & 2005) produced a phytoplankton concentration gradient in a series of otherwise identical mesocosms by the addition of increasing nutrient concentrations. The authors also added the same amount of a stable methylmercury isotope and similar numbers of filter feeding daphnids (*Daphnia mendotae*) to each tank. In a second study the authors repeated the earlier experimental design but, in addition to the daphnids, added two copepod species, *Leptodiaptomus minutus* and *Mesocyclops edax*. In both studies methylmercury concentrations were measured in water, phytoplankton and zooplankton for up to three weeks. Phytoplankton density increased 4-fold across the nutrient gradient while methylmercury concentrations declined in phytoplankton 3 to 4-fold and in zooplankton 2 to 3-fold. A decrease in methylmercury was observed in all three zooplankton species at the higher phytoplankton concentration. The authors also found a significant difference in mercury concentration in the three zooplankton species at the same phytoplankton level. The daphnids accumulated 2 to 3 times more methylmercury per gram dry weight than did either of the two copepod species. The authors note that daphnids are a preferred prey organism for many lake fish species and speculate that daphnids may represent a keystone species for the transfer of methylmercury between phytoplankton and fish.

Essington and Houser (2003) and Kidd and others (1999) studied methylmercury accumulation in lakes that had been artificially fertilized to increase phytoplankton concentrations. Essington and Houser (2003) reported on the effect of whole-lake nutrient enrichment on mercury concentrations in age-1 yellow perch. Two Wisconsin lakes were fertilized for four years with both TN and TP at concentrations several times higher than the background level. Mercury concentrations in fish from the two fertilized lakes were compared with concentrations in eight

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similar nearby unfertilized reference lakes. Atmospheric deposition was the primary source of mercury in the region and deposition rates were assumed to be similar at all sites. Mercury concentrations in yellow perch from all lakes were highly correlated with lake pH and nutrient enrichment ($R^2 = 0.87$). Age-1 perch in the two enriched lakes had half the mercury concentration of fish in the reference lakes with an equivalent pH (0.11 and 0.24 mg/kg, respectively). Finally, Kidd and others (1999) measured mercury concentration in zooplankton and cyprinid fish in an oligotrophic and a eutrophic lake at the experimental lakes area in Northwestern Ontario, Canada. Both lakes are remote from point sources of pollution and were chosen for a comparative study because of their similar physical characteristics. The primary source of mercury in the region is atmospheric deposition. The nutrient poor lake was naturally oligotrophic while the eutrophic one had been fertilized for about 25 years. Average chlorophyll levels were 8 times higher in the eutrophic than in the oligotrophic lake (25.7 and 3.2 $\mu\text{g/L}$, respectively). Methylmercury concentrations in zooplankton in the eutrophic lake were consistently about 4 times lower than in the oligotrophic lake (35 and 146 $\mu\text{g/g}$ dry weight, respectively). Mercury concentrations in fathead minnow and redbelly dace were also significantly lower on 3 of 4 sampling occasions. The average difference in fish tissue concentrations between the eutrophic and oligotrophic lake was about 3-fold (50 and 150 ng/g wet weight, respectively).

No studies have been encountered in the literature where either increases in chlorophyll or increases in nutrient concentrations were correlated with an increase in fish tissue mercury concentrations.

Summary

A combination of descriptive field studies and nutrient amendments in the laboratory and in the field support the hypothesis that increasing phytoplankton density decreases biotic mercury concentrations. The next section describes the biological processes responsible for the reduction in methylmercury concentrations.

Trophic Transfer

Two primary and several minor biological processes have been proposed to explain why the trophic status of a lake might reduce fish tissue concentrations. The two primary mechanisms are called algal bloom dilution and somatic growth dilution. Both processes can occur simultaneously and, while different, are impossible to separate quantitatively without explicit experimental laboratory procedures.

Algal Bloom Dilution

Algal bloom dilution occurs when a constant mass of methylmercury is distributed among a larger biomass of algae. The result is a lower concentration of methylmercury per unit of algal food. Evidence for the importance of algal bloom dilution comes from the results of the previously described mesocosm experiments of Pickhardt and others (2002 & 2005). Briefly, in these experiments the same mass of methylmercury was amended into identical tanks with an increasing biomass of algae. Zooplankton were subsequently added and allowed to feed on the contaminated algal cells. Mass balance calculations, confirmed by analytical

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measurements, demonstrated that the concentration of methylmercury per algal cell was lower in tanks with higher algal biomass because the contaminant was partitioned among a greater number of phytoplankton cells. As a result, zooplankton fed on an algal diet with lower methylmercury concentrations in tanks with more fertilizer. Tissue analysis demonstrated that zooplankton reflected their diet; less contaminated algal cells resulted in less contaminated zooplankton. A 3 to 4-fold increase in phytoplankton biomass resulted in a 2 to 3-fold decrease in zooplankton methylmercury in spite of the fact that the same initial mass of methylmercury was added to each tank. The authors noted that zooplankton is the preferred prey for many planktivorous fish. Because fish acquire most of their methylmercury through their diet, lower methylmercury concentrations in zooplankton were predicted to result in lower fish tissue levels.

Somatic Growth Dilution

Somatic growth dilution occurs when organism adds new biomass faster than methylmercury is incorporated into it. An organism grows when more energy is ingested than needed for basal metabolism. If the amount and quality of food is only sufficient for basal metabolic needs, then no tissue growth occurs. However, methylmercury concentrations in the organisms increase as food is ingested to support basal metabolism. The result is a gradual increase in methylmercury concentrations in the organism over time. Alternatively, as food resources increase above basal metabolic needs, then both tissue growth and methylmercury uptake occur. The result is a reduction in mercury tissue concentrations in the growing organism in comparison with animals existing only on a subsistence diet. The dilution in mercury with increasing tissue growth is called somatic growth dilution. Somatic growth dilution, unlike algal bloom dilution, can happen at all levels of the aquatic food chain. Decreasing tissue mercury concentrations with an increase in tissue growth is taken as empirical evidence for the occurrence of somatic growth dilution³. Evidence supporting somatic growth dilution comes from both descriptive field studies and controlled laboratory and field work.

Descriptive Field Studies

Results from three correlative field studies support the occurrence of somatic growth dilution (Simoneau et al. 2005; Sonesten, 2003; Larsson et al. 1992). Simoneau and others (2005) examined the relationship between mercury in walleye and their growth rate in twelve lakes in eastern Canada. Fish growth rates were inversely correlated with mercury concentration ($R^2=0.86$; $P<0.001$). Faster growing walleye had a lower mercury concentration than did slower growing individuals of the same size. The authors report that growth rate dominated all other environmental factors in accounting for differences in mercury concentrations in walleye populations. Sonesten (2003) measured mercury concentrations in perch in 78 Swedish lakes. Mercury concentrations were found to be negatively associated with TP and TN and fish tissue growth. Higher nutrient levels resulted in faster fish growth and lower fish tissue concentrations. The results were significant in a partial least square regression analysis. The author did not measure primary production rates or chlorophyll concentrations but assumed that the trophic status of the lake explained both the increased fish growth rate and the lower

³ Additional evidence for somatic growth dilution is when the mercury content of a growing organism (methylmercury/individual) remains constant but tissue concentrations (methylmercury/tissue) decline.

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mercury concentration. Finally, Larsson and others (1992) measured Σ PCB concentrations, another bioaccumulative substance, in northern pike in 61 Scandinavian lakes. The primary source of PCBs was atmospheric deposition which was assumed to be similar in all the lakes. The authors also measured TN and TP and chlorophyll concentrations as surrogates for lake productivity. The chlorophyll level of the lakes was positively correlated with TN and TP levels ($P < 0.01$). The growth rate of pike was 11 to 15 percent higher in the more eutrophic, nutrient rich lakes. The faster growth rate was associated with a 69 to 75 percent decrease in Σ PCB concentrations. The authors ascribed the lower Σ PCB concentrations to somatic growth dilution.

Controlled Laboratory and Field Work

Somatic growth dilution was tested experimentally in the laboratory with *D. pulex* (Karimi et al. 2007). The green alga *Ankistrodesmus falcatus* was cultured at high (110:1) and low (15:1) nitrogen to phosphorus ratios⁴ (N:P). Algae grown in low N:P cultures (more phosphorus (P)) are more nutritious and support faster daphnid growth and higher reproduction rates. Algae cultured under both nutrient regimes were amended with the same amount of labeled methylmercury. Daphnids were placed in replicate flasks for five days and fed identical algal concentrations while measuring growth and methylmercury assimilation efficiency. Assimilation efficiencies were unaffected by the nutrient level of the algae. Similar amounts of methylmercury were present in each individual organism (ng-methylmercury per individual) on the high and low quality ration. However, daphnid growth was 3.5 times faster on the high quality diet. As a result, methylmercury concentrations in daphnids on the high-quality food resource were only one third that of individuals fed low-quality algae (ng-methylmercury per gram-wet weight). The authors conclude that somatic growth dilution can occur when growth rates increase with relatively little change in mercury uptake. The authors also note that a disproportionate increase in tissue growth with no change in mercury uptake can also occur if animal activity or respiration rates are low or food quantity or quality is high.

Somatic growth dilution has also been tested in the field (Gothberg 1983; Verta 1990; Lepak et al. 2012; Ward et al. 2010; Essington and Houser 2003). Gothberg (1983) and Verta (1990) intensively fished two small lakes and removed about half of the biomass of top predator fish. In both cases the remaining predacious fish grew at about twice their original rate for several years because of a decrease in competition for food. Mercury concentrations in the rapidly growing fish decreased while concentrations in forage organisms did not change. Verta (1990) observed no change in either fish growth or mercury concentrations in a control lake that had not been manipulated. Gothberg (1983) did not have a control lake. Both authors ascribe the decrease in methylmercury to somatic growth dilution as mass balance calculations indicate that the same mass of mercury was present in individual fish before and after the intensive fishing program. However, the biomass of individual organisms doubled in the intensively fished lakes and this resulted in a decrease in mercury concentrations on a tissue basis (ng-methylmercury per gram wet weight).

A stocking experiment was conducted in a small Colorado lake to test somatic growth dilution (Lepak et al. 2012). Rainbow trout with low mercury concentrations were added to the lake as

⁴ Molar basis

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forage organisms for northern pike. Several nearby ponds served as reference sites. Pike weighed more and had lower mercury concentrations 50-days after the stocking event (both at $P < 0.01$). The average weight gain was about 15 percent while the average decrease in mercury concentration was about 20 percent. No change was observed in the reference ponds. A bioenergetics model suggested that the change in mercury concentrations was most parsimoniously explained by a combination of lower mercury concentrations in prey organisms and somatic growth dilution. The results were transitory. Pike growth rates and mercury concentrations returned to their original value a year after stocking ceased.

The relationship between fish growth and mercury concentration was also assessed in a watershed stocking study (Ward et al. 2010). Atlantic salmon fry hatched under identical conditions were released at 18 stream sites in the Connecticut River basin in New Hampshire and Massachusetts. There was no natural salmon reproduction in these streams; the only salmon present were those stocked in 2005 and 2006 for this experiment. Fish were recovered 116 to 143 days after stocking and their foreguts emptied and the mercury content of the ingested invertebrate prey measured to assess mercury concentrations in their food. Fish weights and tissue mercury concentrations were also measured. The mercury concentration in the recaptured salmon was positively correlated with mercury concentrations in prey ($P < 0.009$ for both years) and inversely correlated with fish growth ($P < 0.0001$ and $P < 0.04$ for 2005 and 2006, respectively). A hierarchical partitioning of mercury concentrations demonstrated that prey concentrations explained 59-percent of the variation in mercury concentrations at each site while differential growth rates accounted for 38 percent of the variation. Inter-annual variation explained the remaining 3 percent. The authors note that, consistent with predictions of somatic growth dilution, larger, faster growing fish had lower mercury concentrations. The authors conclude that factors which suppress tissue growth rates increase the susceptibility of organisms to mercury contamination.

Finally, the whole-lake nutrient amendment study of Essington and Houser (2003) was previously described. Briefly, two Wisconsin lakes were fertilized for several years with both TN and TP and mercury concentrations in age-1 yellow perch compared with concentrations in eight nearby unfertilized control lakes. Yellow perch mercury concentrations were highly correlated with lake pH and nutrient enrichment ($R^2 = 0.87$). Age-1 yellow perch were 4 to 5 times larger and had 50 percent lower mercury concentrations in the two enriched lakes than in reference water bodies of similar pH. A mass balance model indicated that somatic growth dilution could only explain 30 to 40 percent of the reduction in yellow perch mercury concentrations. Fertilization produced additional unidentified effects on fish mercury concentrations. The authors did not account for potential reductions caused by algal bloom dilution.

Other Processes

A number of other algal related processes may also influence--positively or negatively--changes in methylmercury bioaccumulation in aquatic systems. These processes have not received as much attention as algal bloom or somatic growth dilution but might be important in

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some environmental circumstances. The processes include methylmercury production from algal derived organic matter, changes in algal cell size, assimilation efficiency, and DOC.

Methylmercury Production

The production of methylmercury in sediment is a positive function of carbon content (Lambertsson and Nilsson 2006; Benoit et al. 2003; Wiener et al. 2003). Increased abundance of phytoplankton may also result in the increased availability of organic carbon settling to the sediment. This organic carbon may become available for microbial decomposition which can produce anoxic, reducing conditions that stimulate methylation by sulfate-reducing bacteria. A positive correlation was observed in California reservoirs between aqueous methylmercury and chlorophyll *a* concentrations (**Table X in staff report, $r = 0.49$, $P < 0.001$**). This suggests that increased algal production is stimulating microbial activity and methylmercury production in California. Nonetheless, as reviewed previously, the net effect of increasing primary production and chlorophyll *a* in California and elsewhere has been a decrease in fish tissue mercury concentrations. This suggests that the positive effects of algal bloom and somatic growth dilution outweigh the negative effect of an increase in aqueous methylmercury concentration on the trophic transfer rate of methylmercury. This conclusion is consistent with the conceptual model of Driscoll and others (2012) for nutrient methylmercury dynamics in coastal marine ecosystems.

Algal Cell Size

Several researchers have noted that smaller-sized algal cells accumulate more methylmercury on a weight basis than do larger ones (Kim et al. 2011; Pickardt and Fisher 2007; Mason et al. 1996). Mason and others (1996) used this information and developed a methylmercury food web model and tested it with data from lakes in Russia, the Adirondacks and Wisconsin. The model assumed that methylmercury uptake in algae was inversely proportional to cell radius. The model was able to predict fish tissue concentrations in the different lake systems ($r = 0.65$ to 0.81 , $p < 0.05$). Kim et al. (2011) cultured six marine phytoplankton species and measured methylmercury bioconcentration rates. Like Mason and others (1996), uptake was inversely proportional to cell size among the different salt water algal species. Finally, Pickardt and Fisher (2007) cultured four freshwater algal species and also measured methylmercury uptake rates. The test species were a diatom, green alga, cryptophyte and blue-green alga. Again, methylmercury uptake was independent of species but inversely proportional to cell size. Smaller cells took up more methylmercury than did species with larger cell sizes.

Eutrophication increases algal biomass but also typically causes shifts in algal species composition and cell size. Oligotrophic freshwater systems are characterized by pico and nano sized plankton while mesotrophic systems are dominated by larger sized algal species including diatoms (Stockner et al. 2000). Therefore, the phenomenon of algal bloom dilution may be amplified by the tendency of more eutrophic water bodies to contain larger bodied phytoplankton with a lower methylmercury bioaccumulation potential.

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Methylmercury Assimilation Efficiency

Laboratory studies demonstrate that methylmercury assimilation efficiency by herbivores is inversely proportional to the size of their food ration. Tsui and Wang (2004) cultured *D. magna* in the laboratory and fed them two green algal species with isotopically labeled methylmercury. The study confirmed that methylmercury uptake in daphnids was predominately from their diet. Methylmercury assimilation efficiencies were higher at low phytoplankton concentrations and decreased as carbon levels increased. The *D. magna* assimilation efficiency at an algal concentration less than 0.15 mg-C/L averaged about 90-percent but fell to 60-percent at concentrations greater than 2.7 mg-C/L. Tsui and Wang (2004) did not measure chlorophyll *a*. However, a concentration of 0.15 mg-C/L is equivalent to about 7- μ g/L chlorophyll⁵. These observations are important because they suggest that methylmercury accumulation in filter feeding herbivores is inversely proportional to ration and algal biomass. Higher methylmercury assimilation efficiencies occur in more oligotrophic lakes characterized by lower chlorophyll and algal biomass concentrations. Furthermore, larger methylmercury assimilation efficiency at low chlorophyll would compound the negative effect of low somatic growth dilution in oligotrophic systems.

Dissolved Organic Carbon

Eutrophic systems often have higher dissolved organic carbon (DOC) concentrations than oligotrophic waters. Much of the DOC in eutrophic systems is derived from the lysing and decomposition of algal material. Laboratory studies demonstrate that DOC can out-compete algae for dissolved methylmercury, reducing intracellular phytoplankton concentrations and potential uptake by herbivores (Gorski et al. 2008; Luengen et al. 2011a & 2011b) but see Pickardt and Fisher (2007) for alternate results. Luengen and others cultured the diatom *Cyclotella* in the laboratory and measured methylmercury uptake as a function of both increasing DOC concentrations and different DOC types from San Francisco Bay. The DOC tested also included organic material produced from ruptured algal cells. The highest volume concentration factors⁶ (VCF) were measured for *Cyclotella* in water without DOC (30×10^4). VCFs declined as DOC concentrations increased to 1.5-mg C/L and dropped as low as 2×10^4 at concentrations of 20-mg C/L. Gorski and others (2008) obtained similar results. They cultured *Selenastrum* in bioassays containing field collected freshwater with varying DOC concentrations. Bioconcentration factors were measured by stable methylmercury isotopes. Bioconcentration factors were significantly higher in low DOC water than in mixtures of lake and high DOC river water. Similar to the results obtained by Luengen and others algal uptake declined rapidly when DOC exceeded about 5-mg C/L. These results suggest an additional reason why resident fish in more eutrophic systems have less mercury. A portion of the methylmercury may remain complexed to DOC and thus be biologically unavailable to herbivores.

⁵ Chlorophyll to carbon ratio of 5%

⁶ Amount of intra cellular methylmercury divided by the amount of methylmercury in an equivalent volume of water.

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Key Points:

- Multiple descriptive field and controlled laboratory and field studies have observed a decrease in biotic mercury concentrations with an increase in primary production and chlorophyll a.
- The two primary biological mechanisms responsible for the reduction in fish tissue mercury concentrations with increasing primary production are algal bloom dilution and somatic growth dilution.

Summary

Predatory fish tissue mercury concentrations decline as chlorophyll a levels rise. A combination of controlled laboratory and field experiments demonstrate that the primary biological mechanisms responsible for the decline in fish mercury levels are algal bloom dilution and somatic growth dilution. Both processes can be modified in both a positive and negative fashion by a suite of other physical and biological processes. However, the net effect of increasing primary production and chlorophyll a in field and laboratory studies is a decrease in fish mercury concentrations.

Section 3: Cultural Oligotrophication

Cultural oligotrophication is defined as an anthropogenically induced decrease in nutrient concentrations and aquatic primary production (Stockner et al. 2000; Stockner and Ashley 2003). One consequence of cultural oligotrophication is a gradual decline in fish tissue growth in impounded water bodies and in downstream creeks and rivers. There are multiple causes of cultural oligotrophication. These have been reviewed by Stockner and others (2000), Stockner and Ashley (2003), Anders and Ashley (2007), and Milbrink and others (2011). Cultural oligotrophication has not been explicitly documented in California but many of the processes responsible for the phenomenon elsewhere also occur in California reservoirs. One such process is the large annual fluctuation in water elevation levels in California reservoirs. The large fluctuations result in eroded banks and denuded, armored sides limiting benthic primary production. Another process that supports cultural oligotrophication in California reservoirs is that reservoirs trap and settle incoming suspended sediment and their associated attached nutrients, particularly TP (Finger et al. 2007; Vorosmarty et al. 2003; Matzinger et al. 2007). This loss of suspended nutrients reduces pelagic primary production (Schindler 1977; Carpenter 2008; Schindler et al. 2008). Benthic and pelagic primary production are responsible for the majority of organic matter fueling fish tissue growth in lakes and reservoirs (Vadeboncoeur et al. 2003; Vander Zanden and Vaeboncoeur 2002; Karlsson and Bystrom 2005; Karsson et al. 2009). Finally, reservoir dams block the return migration of anadromous salmon and their large marine derived nutrient loads. Salmon carcasses are both a food resource for benthic invertebrates and larval fish and, after mineralization, become a nutrient source for benthic and pelagic primary production (Nakajima and Ito 2003; Bilby et al. 1996; 1998 & 2001; Mundie et al. 1991; Quamme and Slaney 2003; Kiernan et al. 2010; Kline et al. 1990 & 1993; Richey et al. 1975). Elsewhere, the net effect of reducing *in situ* benthic and

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pelagic primary production and eliminating salmon migrations has been a gradual multi-decadal decline in ecosystem productivity. A similar process must also have occurred in California. We hypothesize that an additional indirect effect of cultural oligotrophication is a concomitant increase in fish mercury concentrations because of the loss of primary and secondary production. This is the reverse of the phenomena of algal bloom and somatic growth dilution discussed earlier. It is also consistent with the nutrient-mercury conceptual model of Driscoll and others (2012) for marine ecosystems.

Benthic Primary Production

Reservoirs are susceptible to cultural oligotrophication because of a gradual reduction in benthic primary production. Benthic littoral production is a major pathway for the production of organic matter in oligotrophic lakes and, in some instances, can equal or exceed pelagic primary production (Vadeboncoeur et al. 2001 and 2003; Vander Zanden and Vadeboncoeur, 2002; Karlsson and Bystrom 2005; Karlsson et al. 2009; Ask et al. 2009). Positive correlations have been measured in oligotrophic lakes between benthic primary production and fish biomass (Karlsson et al. 2009; Karlsson and Bystrom 2005; Vander Zanden and Vadeboncoeur 2002). Benthic algae acquire nutrients, including TP, from both the sediment and the water column (Hansson 1992; Vadeboncoeur et al. 2000). The ability of benthic algae to take up nutrients from sediment reduces their dependence on ambient water column concentrations. Acquisition of nutrients from sediment is restricted though, to algae growing on soft bottoms. Benthic algae attached to wood, cobble, or other hard surfaces must acquire nutrients from the water column making their primary production rate dependent on ambient nutrient concentrations, like pelagic algae (Vadeboncoeur et al. 2001).

Benthic algal production is normally light limited (Hansson, 1992; Karlsson et al. 2009). This restricts periphyton production to shallow areas, mostly reservoir margins. In Mediterranean climates water demand for downstream irrigation and hydropower production often results in large annual fluctuations in reservoir water elevation levels. As water levels fall, wave actions strip reservoir margins of vegetation, eroding the fine grain material around their margins leaving the terraces composed of cobble and gravel. Hard substrates in oligotrophic watersheds do not support luxuriant benthic algal production because they lack the fine grained nutrient rich organic sediment which is the primary source of nutrients for periphyton. So, an indirect effect of water regulation is the creation of terraces with reduced benthic primary production around the margin of reservoirs. The result is that secondary production in reservoirs gradually becomes increasingly dependent on dissolved nutrients and pelagic primary production.

Descriptive Field Studies

Evidence for the importance of reduced benthic algal production on inducing reduced fish growth comes from Sweden. Milbrink and others (2011) compared the size of arctic charr in nine regulated Swedish reservoirs and eight nearly unregulated lakes. The water bodies were selected for study because fishing pressure was light and reservoir construction was the only major anthropogenic disturbance in each watershed. All the reservoirs had been dammed 40 to 65 years earlier. The growth rate of charr began to decline in the reservoirs after about

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seven years and continued to decrease with increasing reservoir age. The average weight of a 4-year old reservoir charr was 49 percent less after 10-38 years and 72 percent less after 40-65 years of impoundment than similar aged fish in nearby reference lakes. The progressive decline in fish growth in reservoirs, but not in nearby reference lakes, was ascribed to a number of factors including the loss of benthic algal production because of water level fluctuations that had eroded reservoir banks and reduced periphyton production.

No information is available on the effect of fluctuations in water elevation levels on benthic primary production or fish growth in California reservoirs. However, there is a positive relationship between mercury concentrations in 350-mm largemouth bass and the mean annual maximum change in water depth in California reservoirs (nonparametric correlation coefficients between 0.23-0.35, $P < 0.006$, Table 1A). A multiple regression analysis was conducted to determine the factors most strongly correlated with fish mercury concentrations. The average annual fluctuation in reservoir water depth was determined to be an important factor (Equation 1A). Greater annual drawdown in reservoir water levels was associated with higher fish tissue mercury concentrations.

The phenomenon of somatic growth dilution is the most parsimonious biological explanation for why annual fluctuation in reservoir water elevation would be positively correlated with fish tissue mercury concentrations. Larger fluctuations in water levels result in larger expanses of eroded banks and greater reductions in benthic littoral periphyton production. As previously described, the loss of benthic production has been documented to contribute to reductions in fish tissue growth and these reductions likely accounts for the increase in fish mercury levels.

Pelagic Primary Production

This section describes how reservoirs are also susceptible to cultural oligotrophication because they gradually undergo a process of reduced pelagic primary production. Reservoir pelagic primary production decreases because of a gradual decline in the amount of P available for algal production. P is the limiting nutrient for pelagic primary production in many lentic water bodies (Schindler 1977; Schindler et al. 2008; Lee et al. 1991). P strongly binds with suspended matter making the element susceptible to loss by sedimentation. The estimated irreversible annual water column loss of P to sediment ranges between 34 and 90 percent of the incoming load (Rydin et al. 2008; Moosmann et al. 2006; Nurnberg 1984). Sedimentation and P loss is a natural phenomenon in lakes and other static water bodies. This loss occurs naturally in lakes but is usually compensated for by an increase in benthic algal production because of the greater amount of light reaching the bottom. However, as discussed earlier, an increase in benthic algal production is not possible in reservoirs located in California's Mediterranean climate.

Descriptive Field Studies

Loss of P to lake and reservoir sediment is a natural process and has been investigated extensively. Annual P mass balances were constructed for five Swiss lakes (Moosmann et al. 2006). The purpose of the Swiss study was to determine how multi-decadal sedimentation loss rates changed as a function of decreasing P loads and artificial aeration. Three lakes were

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aerated for 10 to 20 years while two control water bodies were allowed to develop a summer anaerobic hypolimnion. Ambient multi-decadal P concentrations in the Swiss lakes varied between 38 and 457 $\mu\text{g P/L}$ at spring overturn. Annual irreversible loss of P to bottom sediment ranged between 40 and 80 percent of the incoming load. The study also concluded that the proportion of P lost to sediment was inversely proportional to the incoming load. Water bodies with smaller loads had higher loss rates. This was hypothesized to occur because the sedimentation of P was a function of the number of iron binding sites on suspended sediment. There was always more dissolved P molecules than available sediment iron binding sites. Therefore, lakes with larger P load had a lower proportional P sedimentation loss rate than did watersheds with smaller loads.

The inverse relationship between loads and sedimentation explains why oligotrophic water bodies are more vulnerable to the loss of P and to becoming ultra-oligotrophic with exceptionally low primary production rates. Moosmann and others (2006) also concluded that the sediment induced loss of P was independent of aeration and the oxygen content of the hypolimnion. Internal release of P from anaerobic sediment increased sediment-water exchange and hypolimnetic P concentrations but did not alter the annual P sedimentation loss rate. These conclusions about P loss to sediment are consistent with the findings of Rydin and others (2008). Rydin and others added P to the water column of a reservoir for a final concentration of 6 $\mu\text{g P/L}$ and determined that about half the amended P was lost annually as organic matter to bottom sediment. In contrast, Nurnberg (1984) found through mass balance studies of 54 oxic and 33 anoxic lakes that P loss was statistically lower in oxic water bodies. On average, oxic and anoxic lakes lost 34 and 58 percent of their annual incoming P loads to bottom sediment, respectively.

All studies have concluded that reservoirs trap and irreversibly settle P, driving water bodies oligotrophic unless new sources of the nutrient are found. However, the peer reviewed literature does not agree on whether oxic water bodies lose P faster than do anoxic ones.

Simulation Modeling

Lowering P concentrations reduce pelagic primary production, and this has been shown to result in a decrease in fish growth (Hanson and Leggett, 1982; Downing et al 1990; Lee et al. 1991). Numerical models supported by field observations have been used to determine the effect of decreasing ambient P concentrations on both lake primary production rates and fish population levels (Finger et al. 2007; Matzinger et al. 2007). Lake Brienz, Switzerland was eutrophic in the 1960s. However, anthropogenic actions resulted in the lake's oligotrophication and a collapse in the fish population by the early 2000s (Finger et al. 2007). Lake Brienz's oligotrophication was due to a decrease in the loading of P, which was attributed to a ban on P-containing detergents, reduced discharges from local sewage treatment plants, and the construction of seven upstream hydroelectric reservoirs. Mass balance estimates suggested that P loading was reduced by 80 percent from urban sources and 25 percent from reservoir construction in the upstream watershed. Simulation modeling was used to evaluate whether the decrease in P was responsible for the collapse of the fishery (Finger et al. 2007). Modeling confirmed that the collapse was consistent with decreased P loading rates. Interestingly, both monitoring and modeling revealed that the collapse in the fishery was not due to a reduction of

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algal biomass as a result of the reduction in P, but a decrease in algal nutritional value by increasing the N:P ratio of the algae. Simulation modeling predicted that zooplankton and fish biomass would increase with additional P.

Monitoring and numerical modeling was also used to determine the cause of the collapse of the kokanee fish population on the Columbia River in the Canadian Arrow Lakes (Matzinger et al. 2007). The Hugh Keeleyside Dam was constructed at the outlet of the Arrow Lakes to increase downstream flood protection and to produce hydroelectric power. The Arrow Lakes are naturally oligotrophic and P-limited. Several hypotheses were evaluated to determine the cause of the kokanee collapse. The hypotheses included reduced primary production due to reductions in P from construction of two upstream reservoirs and hydrologic changes to the downstream reservoir. Monitoring determined that the upstream Kinbasket and Revelstoke Reservoirs trapped about 75 percent of the incoming P load (Pieters et al. 2003). Model simulations estimated what Arrow Lake primary production rates would have been without construction of the upstream dams. Phosphorus retention in upstream reservoirs was determined to be responsible for 40-50 percent of the decrease in Arrow Lake primary production. The remaining loss in primary production was attributed to hydraulic changes in the Arrow Lakes from the construction and operation of the Hugh Keeleyside Dam.

Controlled Field Work

Several whole-lake nutrient addition experiments have been completed to test nutrient limitation effects on secondary productivity. In upper Arrow Lake, a five year nutrient addition experiment was begun in 1999 to test the hypothesis that the collapse of the kokanee salmon population was, at least partially, the result of nutrient limitation (Pieters et al. 2003). Preliminary results after two years are consistent with the nutrient limitation hypothesis. After fertilization, annual average TP concentrations in the upper Arrow Lake were about 4 µg/L. Primary production increased 1.5-fold with increased densities of micro flagellates and very large late summer blooms of diatoms. The biomass of zooplankton doubled with much of the increase being due to increased numbers of *Daphnia* sp. Finally, the number, size and fecundity of kokanee salmon began to increase and were the largest observed in the past ten years.

Further evidence that the effect of nutrient limitation and that cultural oligotrophication can be reversed comes from a 4-year whole-reservoir nutrient enrichment study in several of the reservoirs used by Milbrink and others (2011), discussed earlier. Persson and others (2008) and Rydin and others (2008) used an upstream unfertilized reservoir as a control while nutrient additions were made in summer to a downstream water body. Fertilization doubled ambient TP concentrations from 3 to about 6 µg P/L. The fertilization did not alter benthic littoral production but phytoplankton concentrations doubled with only minor changes in algal species composition. After two years of fertilization, cladoceran and rotifer densities were 2-3 and 3-4 times higher, respectively, than in the unfertilized upstream control water body. By the end of the study the average weight of a 4-year old charr had doubled in the fertilized reservoir while no change had occurred in the weight of charr in the unfertilized control water body.

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Summary

Lakes and reservoirs naturally trap P, and this decreases pelagic primary production over time. The decrease in pelagic primary production gradually increases photic depth, and this stimulates additional benthic algal production in lakes because benthic algae are usually light limited (Hansson 1992; Karlsson et al. 2009). However, the additional light penetration does not result in enhanced benthic production in reservoirs because erosion has removed the fine grained, nutrient rich sediment around their margins where most of the benthic algal production would have occurred. The result is a greater overall decrease in primary production in reservoirs than in lakes because both forms of primary production have decreased. The decrease in reservoir primary production initiates a trophic cascade with reductions in secondary production and fish growth. However, there is evidence that cultural oligotrophication can be reversed; whole-lake experiments have increased both primary and secondary productivity through nutrient additions.

Key Points:

- Benthic and pelagic primary production is an important factor controlling fish yields in lakes and reservoirs.
- Benthic algal production is normally light limited. Fluctuations in water levels reduce benthic primary production in oligotrophic reservoirs because wave actions erode the soft sediment around reservoir margins where there is sufficient light for photosynthesis and this erosion deprives benthic algae of the nutrients needed for growth.
- P is the limiting nutrient for pelagic primary production in most freshwater lakes and reservoirs.
- Lakes and reservoirs naturally lose P to bottom sediment.
- The natural loss of P and reduction in pelagic primary production in lakes is usually compensated for by an increase in photic depth and an increase in benthic primary production. The shift from pelagic to benthic production does not occur in reservoirs because the loss of nutrient rich sediment around their eroded margins restricts benthic algal growth.
- Fish mercury concentrations in California reservoirs are positively correlated with annual changes in water elevations and inversely correlated to chlorophyll a concentrations. These statistical relationships are consistent with fish tissue growth being controlled by benthic and pelagic primary production. The loss of both forms of primary production results in an increase in fish mercury levels because of the inverse of algal bloom and somatic growth dilution.
- Whole-lake experiments have demonstrated that it may be possible to reverse cultural oligotrophication.

Salmon Migration

The previous section described the phenomenon of cultural oligotrophication and loss of primary production because of the physical characteristics of reservoirs--sediment related P deposition and water level fluctuations. Cultural oligotrophication also occurs in many west

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coast ecosystems because a critical, natural pathway for importing nutrients no longer exists. The next two sections describe the loss of nutrients formerly provided by salmon migrations and some activities that might be undertaken to reverse the nutrient loss and resulting cultural oligotrophication and increased fish mercury concentrations.

Dam and reservoir construction contributes to cultural oligotrophication by blocking salmonid migrations. Historically, five to six million salmon returned annually to California waterways (Gresh et al. 2000) but overfishing and reservoir construction reduced the runs by blocking access to spawning areas above the dams (Smith and Kato 1979). Present runs are between 1 and 6 percent of their original size (Gresh et al. 2000; Schoonmaker et al. 2003) and are mostly restricted to major rivers below rim dams⁷ in the Central Valley. Originally, adult salmon returned to both coastal and inland streams including many oligotrophic water bodies high in the watershed below waterfalls. Salmon die after spawning and their carcasses are food for both scavenging invertebrates and fish and the uneaten flesh mineralized back to dissolved N and P to supplement ambient nutrient concentrations (Nakajima and Ito 2003; Bilby et al. 1996, 1998 & 2001; Mundie et al. 1991; Quamme and Slaney 2003; Kiernan et al. 2010; Kline et al. 1990 & 1993; Richey et al. 1975). Almost all of a salmon's body weight is of marine origin (Larkin and Slaney 1997). As such, salmon migrations transport nutrients from marine to freshwater ecosystems and have been called the "Anadromous Nutrient Pump" by Stockner and Ashley (2003).

Marine derived nutrients are ecologically important in oligotrophic watersheds as the addition of salmon carcasses has been found to increase ambient nutrient levels and stimulate primary and secondary production (Mundie et al. 1991; Quamme and Slaney 2003; Kiernan et al. 2010; Richey et al. 1975). Stable isotope measurements demonstrate that up to 100-percent of the N in aquatic biota in oligotrophic waters can be from the marine environment (Bilby et al. 1996, 1998 & 2001; Kline et al. 1990 & 1993). Salmon are low in mercury⁸ and were a predictable high quality protein source for American Indians⁹ and riparian wildlife (as reviewed in Stockner and Ashley 2003; Naiman et al. 2002). We hypothesize that the catastrophic loss of salmon runs and their marine derived nutrients were not only a tragedy for native people and local wildlife but also contributed to the cultural oligotrophication of many California water bodies.

A number of studies have quantified the loss of nutrients in inland ecosystems because of the collapse of salmon runs. The loss of marine derived salmon nutrients has been estimated by Gresh and others (2000) and Schoonmaker and others (2003). Gresh and others (2000) calculated pre-European salmon escapement¹⁰ in California from cannery records while current escapement was obtained from fish management agencies after excluding hatchery

⁷ A rim dam is defined here as the last major high elevation dam before a river empties onto the valley floor. Most rim dams do not have fish ladders and terminate the upstream migration of adult salmon.

⁸ The average mercury concentration of returning adult salmon is 0.08 mg·kg⁻¹. This is less than half of the proposed State Board tissue objective of 0.2 mg·kg⁻¹ and among the lowest recorded concentrations of any California freshwater game fish (Gassel et al. 2012).

⁹ Native consumption and trade may have been as high as 57-million kg of salmon per year (as reported in Gresh et al. 2000).

¹⁰ Escapement is defined as the number of returning adult salmon successfully making it to their natal streams to spawn.

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returns. The difference between historic and current escapement was estimated at 5.5 to 6.4-million fish or 24.8 to 28.6-million kg of biomass. The N and P content of salmon are 3.03 and 0.35-percent wet weight, respectively (Larkin and Slaney 1997). A nutrient deficit for California was estimated by multiplying the loss of biomass by its nutrient content. Gresh and others (2000) calculated that construction of dams and overfishing resulted in N and P losses in California of between 750,000-867,000 kg-N and 87,000-100,000 kg-P annually. This corresponds to a 95-percent loss of marine derived nutrients. Schoonmaker and others (2003) made a similar computation and concluded that present salmon escapement in California is about 1-percent of historic value. The difference between the two estimates is because Gresh and others (2000) used actual escapement to estimate present values while Schoonmaker and others (2003) relied on predictions from a fish population model. Regardless, both studies concluded that there has been a very large decline in returning salmon and that this has resulted in a decrease in marine derived nutrients to natal streams. Of equal significance is the fact that nutrients from salmon carcasses were historically distributed in small amounts across thousands of miles of streams while now they are mostly restricted to a few major Central Valley Rivers below rim dams.

Isotopic analysis has been used to determine the amount and biological importance of marine derived nutrients in freshwater ecosystems (Bilby et al. 1996; Kline et al. 1990 & 1993). Marine derived organic matter in anadromous salmon contain higher concentrations of the heavier stable isotopes of C, N, and S than organic matter of terrestrial origin (Kline et al. 1990). Variation in the natural abundance of ^{15}N has been used as a tracer to quantify the proportion of marine derived N in freshwater ecosystems. The use of stable isotope N ratios to represent the uptake of marine derived nutrients does not imply that N is controlling freshwater aquatic production (Bilby et al. 2001) because P or other elements likely limit production in oligotrophic systems. However, P does not have multiple stable isotopes so similar calculations cannot be made to determine the importance of marine derived P. It is assumed that P of a marine origin is biologically recycled in an analogous fashion as marine derived N.

First, evidence is presented that salmon derived nutrients are important for primary and secondary production in oligotrophic streams and rivers. Next, similar evidence is presented for nutrient poor lakes. Together, the results indicate the near universal importance of nutrients from salmon carcasses in freshwater ecosystems and how these oligotrophic systems consistently respond in a positive "bottom up" fashion to nutrient additions.

Streams and Rivers

Bilby and others (1996, 1998 & 2001) and Kline and others (1990) used stable isotopes to determine the amount and biological significance of marine derived nutrients in small streams without fish hatcheries in southeastern Alaska and southwestern Washington State. Both studies found that ^{15}N concentrations were lowest in all trophic levels immediately before a salmon run and increased rapidly when carcasses were present. In Washington, the annual average percentage of marine derived N in epilithic organic matter (benthic algae, fungus, and bacteria) was 18 percent while the proportion in invertebrates ranged between 11 and 25 percent (Bilby et al. 1996). Streams in Washington have a variety of fish species. The proportion of marine derived N in cutthroat trout, age-0 coho salmon, and age-1 steelhead was

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between 19 and 46 percent (Bilby et al. 1998). To quantify the ecological significance of salmon migrations, spent salmon carcasses were added to one stream reach and the growth of juvenile coho salmon compared to an adjacent waterway with few carcasses. The growth rate in the carcass amended stream was double that of the reference site and similar to that of hatchery reared salmon with unlimited food (Bilby et al. 2001). Fish residing at the carcass amended site had nearly 20 times as much material in their gut as those at the reference location. More than half the material consisted of salmon eggs and flesh. In the Alaskan stream a greater proportion of the N was of a marine origin. At least half of the N in benthic algae was of a marine origin, and this increased to 100-percent after the arrival of adult salmon (Kline et al. 1990). Resident caddisfly and stonefly nymphs were composed of between 50 and 100 percent marine derived N. The only fish sampled in the Alaskan study were rainbow trout. They contained between 75 and 100 percent marine derived N. Bilby and others (1996, 1998 & 2001) and Kline and others (1990) concluded that ^{15}N uptake in the aquatic food chain is from both the autotrophic utilization of dissolved mineralized nutrients by benthic algae and from the scavenging of salmon carcasses by macro invertebrates and juvenile fish.

The conclusion that the uptake of marine derived N in streams and rivers is both from mineralization and from scavenging is consistent with the results of nutrient amendment studies by Kiernan and others (2010), Quamme and Slaney (2003), Mundie and others (1991) and Richey and others (1975). Kiernan and others (2010) placed artificial stream channels in the Navarro River, California, to determine the effect of the addition of salmon carcasses and dissolved carcass nutrients on periphyton, invertebrate, and fish growth. Periphyton chlorophyll concentrations and total invertebrate biomass increased statistically in carcass amended treatments. The growth of age 0+ steelhead trout was statistically greater in both the nutrient only and the nutrient plus carcass treatments than in the unamended control. The authors concluded that salmon carcasses can enhance the short-term growth of juvenile salmonids via a bottom up pathway. However, a more substantial enhancement of invertebrate and fish production resulted from the scavenging of carcasses. Quamme and Slaney (2003) placed replicated troughs adjacent to the Keogh River, British Columbia, and dosed them with an increasing concentration of dissolved P at an N:P ratio of 1:1 to determine the effect on periphyton and stream insect abundance. Periphyton biomass increased linearly with nutrient additions up to 2.5- μg P/L. Likewise, the abundance of adult and nymphal baetids and benthic nemourids, perlodids, and hydroptilids increased rapidly at nutrient concentrations up to 2.5- μg P/L. The researchers excluded grazing insects from a single unfertilized trough and found increased periphyton abundance suggesting that invertebrate grazing was limiting algal biomass. The authors concluded that aquatic insect population densities are food limited to nutrient concentrations of about 10.0- μg P/L. Mundie and others (1991) placed twelve replicated troughs adjacent to Carnation Creek, British Columbia, and dosed six with 10- μg P/L (8X the control) while the other six were left as controls. Controls and treatments were continuously colonized with small aquatic biota by diverting some of Carnation Creek through the troughs. After two weeks the algal biomass in the dosed treatment was 3.5 times greater than in the controls and after seven weeks the number of benthic invertebrates was 1.8 times greater than in the controls. The authors concluded that the addition of dissolved P increased insect periphyton food and resulted in about a doubling of both insect carrying capacity and survival to emergence. Finally, Richey and others (1975) examined the effect of native

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kokanee salmon carcass decomposition in Taylor Creek, a tributary to Lake Tahoe, California. Primary production, periphyton biomass and nutrient concentrations were higher in the creek reach with carcasses than in an upstream reference area. An overall conclusion from all these natural and controlled field experiments is that nutrients from historical salmon migrations are ecologically important to primary and secondary production in oligotrophic stream and rivers, and that dam and reservoir construction contributes to the cultural oligotrophication in California and other locations where salmon migration once existed. In addition, biota in oligotrophic streams and rivers (dissolved P < several $\mu\text{g-P/L}$) respond positively when amended with salmon carcasses to simulate historic salmon migrations.

Lakes

Evidence for the importance of marine derived N also comes from whole lake studies. Kline and others (1993) measured ^{15}N in biota from Lliamna Lake in the Kvichak River watershed in southwestern Alaska when escapement was high and low. The Kvichak basin is the largest producer of sockeye salmon for the Bristol Bay fishery¹¹. Benthic periphyton samples were collected from sites around the lake with high and low densities of spent salmon carcasses. The average annual percentage of marine derived nitrogen in benthic algae in low and high density carcass sites was 46 and 87 percent, respectively. Net plankton (mostly algae and zooplankton) samples were also collected. The percentage of marine derived nitrogen varied between 49 and 73 percent. Both fry and yearling sockeye showed shifts in the concentration of ^{15}N in spring in concert with adult salmon escapement during the previous fall. The highest percentage of marine derived N was observed in the lake after the arrival of an escapement of more than 10-million salmon. Conversely, the lowest concentration occurred after years with a low escapement of less than 7-million adults. In high escapement years fry averaged 71-percent marine derived N while in poor years the value dropped to 27-percent. The authors conclude that the primary route of incorporation of marine derived N in Lliamna Lake was from benthic algal production after the mineralization of salmon carcasses. Hyatt and Stockner (1985) fertilized several British Columbia freshwater lakes to simulate nutrient mineralization from salmon carcasses. The fertilization resulted in enhanced primary and secondary production, larger standing stocks of zooplankton, increased growth of juvenile sockeye salmon, and larger out-migrating smolt. Finally, Finney and others (2000) used lake sediment records of ^{15}N to reconstruct salmon abundance near Bristol Bay, Alaska, over the past 300 years. The sediment data suggested a bottom-up positive feedback loop. Higher adult salmon escapement resulted in increased nutrient loads to the lake and higher primary and secondary production rates. The enhanced primary and secondary production resulted in greater numbers of larger out-migrating smolt. In other words, “bottom up” nutrient control appeared important in the sediment record in establishing the minimum number of returning adult salmon in the Bristol Bay fishery.

¹¹ The average twenty year (1971-1990) escapement for Lliamna Lake is 5.7 million fish.

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Key Points:

- Dam and reservoir creation contributes to cultural oligotrophication and the loss of primary and secondary production in California by blocking adult salmon migrations and the return of marine derived nutrients.
- Present salmon runs in California are 1 to 6 percent of their historic size.
- Stable nitrogen isotope studies in lakes and rivers with salmon demonstrate that the mineralization and scavenging of salmon carcasses can contribute up to 100 percent of the N and P in primary and secondary production in oligotrophic water bodies.
- Dissolved nutrients and salmon carcass additions to nutrient poor lakes and rivers demonstrate that both systems consistently respond in a positive “bottom up” fashion by increasing primary and secondary production.

Summary

A multiple regression equation was developed to predict mercury concentrations in bass in California reservoirs (Equation 1A). The best fit multiple regression equation predicts that fish tissue mercury concentration is directly proportional to water elevation changes and inversely proportional to chlorophyll levels. As previously noted, the changes in water elevation should be negatively correlated with benthic algal production while reductions in chlorophyll and pelagic primary production are directly correlated with reduced P levels. While losses of pelagic primary production in lakes are compensated for by increases in benthic production, both forms of primary production simultaneously decrease over time in reservoirs. The inclusion of both forms of primary production in the multiple regression equation is consistent with the importance of primary production in regulating fish tissue mercury levels through the processes of algal bloom and somatic growth dilution. So, elevated concentrations of mercury in fish are an unintended, indirect consequence of reservoir construction and operation.

Section 4: Artificial Fertilization Programs

The previously mentioned oligotrophication led the Canadian Department of Fisheries and Oceans to experimentally fertilize a variety of lakes and rivers in an attempt to recover important commercial and recreational fisheries. Some of the lessons learned from these lake and river fertilization efforts are summarized in the next section. The section also includes a summary of “lessons learned” from the Canadian fertilization program. This information may be important for California because mercury research has demonstrated that increased primary and secondary production results in reduced fish tissue mercury concentrations. Artificial fertilization of fresh water systems may be a temporary method of reversing the oligotrophication of California reservoirs while simultaneously reducing the threat posed by mercury to human and wildlife consumers until a successful long term mercury control program is developed and implemented.

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Watershed Nutrient Additions

Cultural oligotrophication has been successfully combated through targeted stream and reservoir fertilization programs (Hyatt et al. 2004; Stockner and MacIlsac 1996; and Stockner and Ashley 2003). The Canadian Department of Fisheries and Oceans began a Lake Enrichment Program (LEP) in 1975. The program grew over three decades to include 25 lakes (Stockner and Ashley 2003). The goal of the program was to aerially fertilize sockeye salmon nursery lakes to increase primary and secondary production, improve juvenile salmon growth and survival, and increase the commercial catch of adult salmon (Stockner and MacIlsac 1996). The Alaskan Fisheries Research and Enhancement Division (FRED) conducted a similar program to restore the productivity of Alaskan sockeye salmon nursery lakes. Lake fertilization was also part of the recovery effort for the endangered sockeye population in Redfish Lake, Idaho (Griswold et al. 2003). The Canadian Department of Fisheries and Oceans has responded to frequently asked questions about LEP at <http://www.pac.dfo-mpo.gc.ca/sep-pmvs/lep-pel/lep-pel-eng.htm>. The Canadian federal agencies have now largely abandoned their fertilization programs, not because of a lack of success in increasing recreational and commercial catch, but because the goal of their agencies has shifted to stock conservation (Hyatt et al. 2004). The Federal Agencies believe that enhancing the catch of common, commercial species by fertilization may increase the danger that threatened fish populations might also be taken in greater numbers and, possibly, driven to extinction.

Hyatt and others (2004) reviewed 24 sockeye salmon nursery lake studies involving whole-lake fertilization. These include nutrient enhancement lakes from Canada, Alaska and Idaho. The authors concluded that, "...21 of 21 studies showed that fertilization was associated with increased chlorophyll *a* concentrations, 16 of 16 showed increased zooplankton biomass, 16 of 16 demonstrated increased average smolt weights and 11 of 13 showed increased smolt biomass...Studies involving increased smolt to adult survival (i.e. marine survival) were even rarer but all (3 of 3) showed that lake fertilization and increased smolt size were associated with increased marine survival..."

In 1992 the British Columbian Ministry of Environment, Land and Parks (MELP) initiated a fertilization program on inland reservoirs that had been impounded for hydroelectric production. These water bodies had experienced a loss of primary and secondary production and a collapse of their recreational kokanee salmon populations because of the sedimentation of nutrients, particularly P. Kootenay Lake was selected for the initial study but the program was expanded to the upper and lower Arrow reservoirs on the Columbia River and to several smaller coastal reservoirs (Askley et al. 1997; Stockner and Ashley 2003). All reservoirs have shown a strong positive response at each trophic level including the kokanee salmon population (Stockner and Ashley 2003; Pieter et al. 2003; Ashley et al. 1997). Whole-river fertilization was also evaluated in British Columbia, Canada (Wilson et al. 2003; Slaney et al. 2003; Johnston et al. 1990; Perrin et al. 1987). Initial fertilization work began on the Keogh River in British Columbia. Perrin and others (1987) and Johnston and others (1990) evaluated various nutrient formulations and loading rates. Both studies found that nutrient additions increased periphyton standing crop by up to an order of magnitude. Johnston and others (1990) monitored fish production and recorded a 1.5 to 2.0-fold increase in coho and steelhead fry weight. Slaney and others (2003) followed up by evaluating the response of the

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biotic community to inorganic nutrient additions. Periphyton biomass increased 5 to 10-fold and the average weights of steelhead trout and coho fry in treated reaches were 1.4 to 2.0 fold higher. Average steelhead smolt abundance in the three years of fertilization increased 60-percent over the pre-fertilization control period. The response of coho salmon smolts was more modest and their abundance increased by only about 20 percent. The fertilization studies were extended to include the Salmon and Adams Rivers and Big Silver Creek (Wilson et al. 2003; Slaney et al. 2003). Again, the results were positive and confirmed that modest fertilization produced a positive trophic response.

Summary

A conclusion of the whole-lake and whole-river nutrient addition studies is that all oligotrophic systems studied responded by producing more primary and secondary production and this led to increased fish yields. These results are consistent with the conclusions of Finney and others (2000) from an evaluation of 300 years of the sediment record of lakes around Bristol Bay. Oligotrophic Pacific slope aquatic systems appear to be controlled by “bottom up” nutrient processes.

Fertilization “Lessons Learned”

Lessons learned from three decades of whole ecosystem fertilization studies have been reviewed in Stockner and MacIassac (1996), Stockner and Ashley (2003), Hyatt and others (2004) and Ashley and Stockner (2003). These include concentration and formulation of fertilizers, seasonal timing and frequency of application, location of application sites, N:P ratios, and application techniques. Not all these are reviewed here. However, Stockner and MacIassac (1996) point out that the best candidate lakes for fertilization are oligotrophic water bodies with chlorophyll a less than 3- μg Chl/L and TP less than 3- μg P/L. After several years of trial and error, systems were found to respond best when N and P were added weekly for about 5 months in the summer (Stockner and MacIassac, 1996). The P-load was lake specific and adjusted to produce an increase in ambient epilimnetic P concentrations of between 0.2 and 0.7- μg P/L. The goal of the fertilization program was to double chlorophyll concentrations. In no case should a program attempt to change the basic oligotrophic nature of a water body. The N:P ratio in the fertilizer was kept high (25:1) to promote chrysophytes and cryptophytes production within an edible size range for herbivorous zooplankton. There did not appear to be any long term build up in nutrients because fertilization was light. The lakes returned to background conditions several years after the addition of nutrients ceased (Stockner and MacIassac 1996)

Several formulations and methods of applying fertilizer were tried in lakes and rivers. In the lake's program, high concentration solutions of stock liquid fertilizer using mixtures of ammonium polyphosphate and urea-ammonium nitrate were determined to be best, and these were applied from fixed wing aircraft after several years of experimentation (Stockner and MacIassac 1996). Several river application methods have also been evaluated. These include a flow proportional fertilizer injection system using real time stage discharge data (Wilson et al. 2003). The method was valuable for maintaining a precise concentration of nutrients in the river. However, the system had the draw back that the large battery packs had to be recharged every few weeks. Another technique that has been developed and tested is the use of slow

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release inorganic nutrient briquettes (Sterling and Ashley 2003; Sterling et al. 2000). The briquettes have the advantage of being able to be distributed once a year through out a river system. Finally, Pearsons and others (2007) has developed a salmon carcass analog (SCA) pellet using ground, dried, and pasteurized Chinook salmon carcasses from hatcheries. The pellets are distributed throughout a stream system where they sink to the bottom and are available for consumption by macro invertebrates and juvenile fish. In addition, the pellets mineralize in the rivers and support periphyton production. Kohler and others (2012) analyzed the ecological effect of the addition of SCA in 15 streams across the Columbia River basin. Periphyton standing crop, macroinvertebrate density and salmonid fish growth rates and stomach fullness increased after each addition. ^{15}N signatures confirmed the trophic transfer of SCA to lower trophic levels but the signal was weak in fish despite their increased growth and stomach fullness.

Several problems were encountered and only partially solved in the fertilization program (summarized in Hyatt et al 2004; Stockner and MacIsaac 1996; Ashley and Stockner 2003). Epilimnetic blue-green algal blooms and hypolimnetic blue-green algal plates periodically developed. These disappeared when the water body was fertilized at a higher N:P ratio (more N applied). However, the diatom *Rhizosolenia* is an undesirable species that can proliferate in high N environments (Hyatt et al. 2004). *Rhizosolenia* has a large cylindrical shape with spines on either end and is poorly grazed by zooplankton. A potential *Rhizosolenia* control method is to reduce N:P ratios (more P) which can push the competitive advantage back to blue-green algae. The interim solution developed by the Canadians was to conduct regular monitoring to determine nutrient status and phytoplankton species composition and make adjustments in fertilizer formulation if an undesirable algal species developed.

Key Points:

- The Canadian government and the States of Alaska and Idaho have fertilized lakes and rivers for up to 25-years to recover important recreational and commercial fish stock.
- All oligotrophic lakes and rivers responded positively to fertilization by producing more primary and secondary production and increased fish yields.
- The best candidate lakes for fertilization were oligotrophic water bodies with a chlorophyll a concentration less than 3- μg Chl/L.
- The goal of the nutrient fertilization program was to double chlorophyll a concentrations. In no case should a program attempt to change the basic oligotrophic nature of a water body.
- The effect of fertilization is reversible. All water bodies returned to their original nutrient status several years after fertilization ceased.

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Section 5: Possible California Reservoir Fertilization Program

An analysis was conducted to determine whether a reservoir nutrient fertilization program would help in reducing fish tissue mercury concentrations to safe levels in California. Table 2A lists the geometric mean epilimnetic chlorophyll a concentration of lakes and reservoirs in California with 0.2-ppm or higher mercury concentrations in largemouth bass. Some of these water bodies are on the 2010 Clean Water Act 303(d) list. The rest are expected to be added in the next listing cycle. Lake specific chlorophyll concentrations reported in Table 2A should be considered preliminary as some values are not based on many measurements. As such, the predictions from this analysis should also be considered preliminary until a more comprehensive chlorophyll data set is collected. No chlorophyll data are available for some listed reservoirs. These are not included in Table 2A. Stockner and MacIassac (1996) recommended that oligotrophic lakes with summer epilimnetic chlorophyll concentrations less than 3- μg Chl/L were candidates for possible fertilization as their experience indicated that water bodies with these characteristics were sufficiently nutrient poor to consistently respond in a positive “bottom up” fashion to the addition of small amounts of N and P. Stockner and MacIassac (1996) cautioned that a fertilization program should not attempt to alter the basic oligotrophic character of a water body. This includes changes in algal species composition, hypolimnetic dissolved oxygen concentrations or water column secchi depth. To ensure this, Stockner and MacIassac (1996) recommended that fertilization not increase ambient summer chlorophyll levels more than two-fold. Finally, Stockner and MacIassac (1996) cautioned that a fertilization program should only be considered a temporary solution until a permanent fix could be devised.

Eighty-three percent (20/24) of the listed reservoirs in Table 2A without an adopted mercury control plans have geometric mean chlorophyll concentrations less than 3- μg Chl/L. This makes them potential candidates for possible fertilization. It also suggests that their oligotrophic nature may contribute to their mercury problem as would be predicted from algal bloom and somatic growth dilution mechanisms. Candidate lakes are widely distributed geographically across the State. They include water bodies from the Coast Range, Trinity Alps, low and high elevations in the Sierra Nevada Mountains, and Southern California.

A multiple linear regression equation¹² was developed to predict the chlorophyll concentration associated with different fish tissue mercury levels ($R^2=0.65$, $N=41$, $P<10^{-4}$). The best fit equation (Equation 2A) was:

$$\ln(\text{Chl } a) = 33.4 - 0.46\ln(\text{Fish Hg}) + 0.42\ln(\text{geomean MeHg water}) - 8.76$$

ln(Longitude)

Equation 2A was used to predict the required increase in chlorophyll needed to reduce 350-mm bass fish tissue mercury levels to 0.2-ppm wet weight, the proposed State Board fish tissue objective. The required new chlorophyll values are included in Table 2A for each water

¹² A new regression equation was developed instead of rearranging the terms in equation 1A as the new equation is based on a larger sample size (41 samples instead of 26). The larger sample size is believed to result in more robust predictions.

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body. The ratio of required to ambient chlorophyll *a* concentrations has also been calculated for each reservoir and is included in Table 2A. Stockner and MacIsaac (1996) caution that this ratio should not exceed a factor of two to ensure that the basic oligotrophic character of a water body not change. Equation 2A predicts that 90-percent (18/20) of the candidate reservoirs would need less than a doubling in chlorophyll to meet the proposed fish tissue objective while two would require more primary production to be in full compliance. One of these is in a watershed with an inoperative mercury mine¹³. The presence of mercury mines emphasizes that additional controls, including removal/sequestration of inorganic mercury waste piles, will also be required to reduce mercury concentration to acceptable levels.

Summary

The analysis suggests that 83-percent of listed reservoirs with elevated predatory fish tissue concentrations are candidates for fertilization because they now have ambient chlorophyll concentrations less than 3- μ g Chl/L. Ninety percent of these reservoirs could be brought into full compliance with the proposed fish tissue objective of 0.2-ppm by implementing a light fertilization program. The remaining two water bodies would benefit from nutrient additions but other control measures will also be required. Based on these results, the State of California needs a program that includes experimental nutrient fertilization studies in representative reservoirs to determine the feasibility of using nutrient additions to reduce fish tissue mercury concentrations.

Key Points:

- Eighty-three percent of the reservoirs in California with elevated fish tissue mercury concentrations are good candidates for a nutrient fertilization program because their ambient chlorophyll levels are less than 3- μ g Chl/L.
- Three quarters of these candidate reservoirs could be brought into full compliance with the proposed fish tissue objective of 0.2-ppm mercury with a light fertilization program that increased ambient chlorophyll *a* by a factor of 2 or less.
- One reservoir requiring more than a doubling in chlorophyll also has an inoperative mercury mine in its watershed. This water body would benefit from an inorganic mercury control program.
- A nutrient fertilization program should only be considered as a temporary solution until a permanent mercury fix is developed.

Section 6: National Nutrient Criteria Program

The U.S. EPA has established the National Nutrient Criteria Program because about half of the Nation's waters are impaired by excess nutrients and cultural eutrophication (USEPA 2000a). The water bodies of concern include lakes, reservoirs, streams and rivers. The program requires that the U.S. EPA or the States develop nutrient criteria for each region of the nation.

¹³ Nacimiento Reservoir.

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The criteria would be the basis for developing site-specific narrative numerical endpoints to protect the designated beneficial uses of each water body. The State of California has committed to consider adopting a nutrient policy for inland fresh waters by January 2014. Tetra Tech (2006) has developed a technical support document to guide the State of California in this effort. Unfortunately, neither the U.S. EPA (2000a) nor Tetra Tech (2006) has considered cultural oligotrophication. Cultural oligotrophication appears to be a widespread phenomenon and is responsible for an unnatural reduction in fish production and an increase in fish tissue mercury concentrations. Cultural oligotrophication, like cultural eutrophication, should be evaluated as part of the National Nutrient Criteria Program to determine whether it is a violation of beneficial uses and of the anti-degradation policy in the Federal Clean Water Act (Lackey 2003; Anders and Ashley 2007). Beneficial uses potentially at risk in California from cultural oligotrophication include Commercial and Sport Fishing (Comm)¹⁴ and Water Contact Recreation (Rec-1)¹⁵. These beneficial uses are intended, at least in part, to ensure that resident fish populations are safe for consumption by humans and wildlife.

Tetra Tech (2006) has recommended chlorophyll criteria to protect California reservoirs from the negative effects of cultural eutrophication. Water bodies with a “cold” designation are considered unimpaired and impaired if average summer epilimnetic chlorophyll concentrations are less than 5 and greater than 10 µg Chl/L, respectively. Likewise, water bodies with a “warm” designation are unimpaired and impaired if concentrations are below 10 and above 25 µg Chl/L, respectively. The recommended chlorophyll criteria should not conflict with an experimental fertilization program to reduce fish tissue mercury concentrations to safe levels. Nonetheless, reservoirs with drinking water intakes should not be included in the fertilization program to eliminate the possibility that nutrient additions might contribute to a blue-green algal bloom in a potable water supply.

Tetra Tech (2006) also includes a BATHTUB model to estimate algal growth responses to changes in reservoir nutrients. The model was developed by the U.S. Army Corp of Engineers for use in U.S. lakes and reservoirs. The model requires hydraulic residence time, light availability and other key input variables for each reservoir. The BATHTUB model may be a valuable tool for reservoir operators to use in calculating the appropriate amount of fertilizer required to double pigment levels in an experimental fish tissue mercury reduction program.

¹⁴ Comm is defined as the use of water for commercial or recreational collection of fish...intended for human consumption...

¹⁵ Rec-1 is defined as the use of water for recreational activities including fishing.

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Key points:

- The U.S. EPA has established the National Nutrient Criteria Program because about half of the Nation's waters are impaired by excess nutrients and cultural eutrophication.
- The National Nutrient Criteria Program does not consider the negative effects of cultural oligotrophication, including increasing fish tissue mercury concentrations to unsafe levels for human and wildlife consumption. The National Nutrient Criteria Program should evaluate the negative effects of both cultural eutrophication and cultural oligotrophication in the development of chlorophyll criteria.
- The recommended chlorophyll criterion for California reservoirs is sufficiently high to not be in conflict with an experimental nutrient addition program for reducing fish tissue mercury reductions.

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Table 1A. Parametric and nonparametric correlation coefficients for bass mercury concentrations and a suite of physical and chemical variables measured in California reservoirs. A description of each variable and the statistical **analysis is included in chapter X of the staff report**. Colored boxes indicate $p < 0.05$ in a 2-tailed test of significance. Green colored boxes indicate correlations where no data transformation was found to meet all the assumptions of parametric statistics.

Variable	Variable Code	350-mm Bass Hg tissue Concentration					
		Pearson's r		Spearman's Rho		Kendall's Tau-a ^a	
		Correlation Coefficient	P-Value ^b	Correlation Coefficient	P-Value ^b	Correlation Coefficient	P-Value ^b
Ratio Aq MeHg Geomean to Chl-a Geomean (ng/ug)	MeHg/Chla	0.67	0.000001	0.70	0.0000002	0.50	0.000002
Sed THg Geomean (mg/kg)	STHg	0.50	0.00003	0.47	0.0001	0.33	0.0001
Upland Soil THg Geomean (mg/kg)	SoilTHg	0.40	0.0016	0.44	0.0004	0.28	0.0016
Longitude (decimal degrees)	Long	0.39	0.0001	0.40	0.0001	0.27	0.0001
Chl-a Geomean (ug/L)	Chla	-0.34	0.0181	-0.27	0.0568	-0.16	0.1028
Average Maximum annual Water Level Fluctuation (ft)	AnnFluc	0.33	0.0078	0.35	0.0040	0.23	0.0061
Percent Vegetation in Watershed	%Veg	0.32	0.0035	0.29	0.0091	0.19	0.0114
Aq MeHg Geomean (ng/L)	AMeHg	0.31	0.0254	0.38	0.0049	0.27	0.0049
Aq THg Geomean (ng/L)	ATHg	0.30	0.0431	0.25	0.0908	0.18	0.0811
Percent Open Water in watershed	%OpenWater	-0.27	0.0150	-0.30	0.0065	-0.20	0.0088
Reservoir Dam Height (ft)	DamHeight	0.25	0.0136	0.34	0.0009	0.23	0.0011
Reservoir Elevation (ft)	Elevation	-0.22	0.0276	-0.27	0.0091	-0.17	0.0162
Percent Forest in Watershed	%Forest+1	0.22	0.0484	0.12	0.2999	0.08	0.2722
Atm THg dep to Watershed from CA sources (g/km/yr)	CADepShed	0.19	0.0873	0.17	0.1409	0.10	0.1971
Watershed Productive Mine Density (productive mines/sq mile)	ProdMineDensity+1	0.17	0.1191	0.05	0.6390	0.04	0.6022
Number of Mines in Watershed	PAMP+1	0.15	0.1696	0.17	0.1295	0.11	0.1291
Year Dam Built	YrBuilt	0.15	0.1423	0.19	0.0683	0.13	0.0632
Watershed Mine Density (mines/sq mile)	MineDensity+1	0.14	0.2290	0.01	0.9261	0.01	0.8891
Number of Dams Upstream of Reservoir	u/sDams+1	0.13	0.2546	0.06	0.5883	0.05	0.5567
Maximum Reservoir Capacity (ac-ft)	MaxCap	0.10	0.3469	0.17	0.0891	0.12	0.0726
Ratio of reservoir to watershed surface area	SA:WA	0.09	0.3912	0.19	0.0666	0.13	0.0682
Atm THg Dep to Reservoir from Ca sources (g/km/yr)	CADepRes	0.08	0.4166	0.12	0.2551	0.07	0.2901
Latitude (decimal degrees)	Lat	0.08	0.4484	0.04	0.7097	0.02	0.7611
Atm THg dep to Watershed from Ca sources (g/yr)	CADepShedL	0.07	0.5597	0.07	0.5178	0.04	0.6186
Atm THg Dep to Reservoir from Ca sources (g/yr)	CADepResL	0.06	0.5514	0.14	0.1719	0.11	0.1091
Atm THg Dep to Watershed (g/yr)	DepShedL	-0.05	0.6378	0.00	0.9875	0.01	0.9350
Watershed Surface Area (acres)	WA	-0.05	0.6308	0.13	0.1935	0.09	0.1909
Atm THg Wet Dep to Watershed (g/yr)	WDepShedL	-0.05	0.6893	0.00	0.9691	0.00	0.9512
Atm THg Dep to Watershed (g/km/yr)	DepShed	0.03	0.8085	0.02	0.8857	0.01	0.8955
Atm THg wet Dep to Reservoir (g/km/yr)	WDepRes	-0.03	0.7951	0.03	0.7602	0.02	0.7635
Number of Productive Mines in Watershed (MRDS)	ProdMines+1	0.03	0.8228	0.00	0.9887	0.00	0.9670
Percent Wetlands in watershed	%Wet+1	-0.02	0.8322	0.00	0.9846	-0.02	0.7471
Atm THg Dep to Reservoir (g/km/yr)	DepRes	-0.02	0.8503	0.05	0.6541	0.03	0.6492
Atm THg wet Dep to Watershed (g/km/yr)	WDepShed	0.01	0.9243	-0.04	0.7069	-0.03	0.7379
Percent Agriculture in watershed	%Agr+1	-0.01	0.9589	-0.08	0.4700	-0.06	0.4407
Reservoir surface area	SA	0.01	0.9581	0.05	0.6565	0.02	0.7371
Atm THg Dep to Reservoir (g/yr)	DepResL	0.00	0.9790	0.07	0.5243	0.04	0.5369
Number of Mines in Watershed (MRDS)	MRDSMines+1	0.00	0.9888	-0.03	0.8148	-0.01	0.9346
Atm THg wet Dep to Reservoir (g/yr)	WDepResL	0.00	0.9908	0.09	0.3948	0.06	0.3701

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Table 2A. Predicted increase in chlorophyll required to reduce bass mercury levels in 303(d) listed reservoirs to a concentration less than the proposed 0.2-ppm mercury objective.

Reservoir	303 (d) listed waterbodies ^{a/}	Observed bass Hg (mg/kg)	Ambient Chl a ^{b/}	Predicted Chl-a @ bass = 0.2 mg/kg ^{c/}	Chl-a increase ^{d/}
Hodges, Lake	1	0.29	18.1	19.0	1
Silverwood Lake	2	0.49	10.1	5.5	1
San Antonio, Lake	1	0.3	6.2	4.0	1
Lexington Reservoir	2	0.6	5.3	6.0	1
Irvine, Lake	2	0.48	5.3	10.5	2
Guadalupe Reservoir	3	6.1	4.6	12.4	3
Castaic Lake	1	0.32	3.9	3.8	1
San Luis Reservoir	1	0.57	3.3	2.6	1
Almaden Reservoir	3	4.3	2.8	13.0	5
Nacimiento, Lake	1	0.99	2.2	6.1	3
Calero Reservoir	2	1.05	1.6	7.7	5
Oroville, Lake	1	0.44	1.5	1.1	1
McSwain Reservoir	2	0.54	1.4	2.2	2
Folsom Lake	1	0.47	1.3	1.8	1
Pillsbury, Lake	1	1.32	1.3	2.8	2
Indian Valley Reservoir	1	0.97	1.3	2.7	2
Oneil Forebay	1	0.24	1.3	3.0	2
Mendocino, Lake	1	0.55	1.2	1.8	1
Natoma, Lake	1	0.54	1.1	1.7	2
Thermalito Afterbay	1	0.22	1.1	1.1	1
McClure, Lake	1	0.77	1.1	2.2	2
New Melones Reservoir	1	1.13	0.9	1.4	2
Sonoma, Lake	1	0.68	0.9	1.9	2
Camp Far West Reservoir	1	0.65	0.8	2.3	3
Don Pedro Reservoir	1	0.44	0.8	1.4	2
French Meadows Reservoir	2	0.21	0.6	0.8	1
Trinity Lake	1	0.45	0.6	0.8	1
Rollins Reservoir	1	0.75	0.6	1.4	2
Hell Hole Reservoir	1	0.92	0.5	1.3	3
Oxbow Reservoir	1	0.29	0.5	1.1	2
Englebright, Lake	1	0.52	0.5	1.6	4
Slab Creek Reservoir	1	0.47	0.4	1.3	4

a/ 1=Listed on 2010 303(d) list. 2=Expected to be listed in next 303(d) cycle 3=Adopted Hg control program

b/ Geometric mean of chlorophyll concentration

c/ Predicted chlorophyll concentration needed to produce bass with 0.2 mg/kg-Hg

d/ Required increase in chlorophyll to achieve proposed fish tissue objective (column 5 divided by column 4)

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