

**CACHE CREEK NATURE PRESERVE  
MERCURY MONITORING PROGRAM**

YOLO COUNTY, CALIFORNIA

**FIFTH SEMI-ANNUAL DATA REPORT**  
**(FALL 2002– WINTER 2002/2003)**

June 23, 2003

*prepared for*  
**Yolo County, California**

***Study and Report by***

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## EXECUTIVE SUMMARY

This fifth semi-annual report is the next to last in this three year mercury study at the Cache Creek Nature Preserve and environs. In this work, which began in the fall of 2000, mercury bioaccumulation has been characterized within the Nature Preserve wetlands in ten quarterly samplings to date. A more extensive additional series of collections was made semi-annually at strategically located adjacent sites (five events to date). The data collected thus far indicate that Gordon Slough is generally a significantly lower mercury environment than adjacent Cache Creek. Mercury bioaccumulation in the Nature Preserve has been substantially elevated over that in its source water environment (Gordon Slough), bringing it to a level similar to that found in the most elevated Cache Creek location, upstream of the project area. The Nature Preserve has thus functioned to some extent as a source of methylmercury. This is consistent with most wetland environments. The effect has been confined mainly to the wetland itself. An additional localized elevation in invertebrate mercury bioaccumulation has been noted on some dates in Cache Creek immediately downstream of the Nature Preserve outlet, though not in the most recent sampling. In any case, a short distance downstream of the Preserve, in Cache Creek below Gordon Slough, mercury bioaccumulation has consistently been similar to or lower than corresponding levels in Cache Creek upstream of the project.

A fascinating seasonal trend is increasingly apparent in the long-term data. The small fish exhibited a pronounced drop in their mercury levels in all three spring samplings to date, relative to adjacent fall collections. The fall concentrations were generally between 50% and over 400% greater than corresponding spring levels. We believe that these trends in the fish data are actually offset by a period of one to several months from the underlying cycles of mercury methylation and bioavailability. Other project data are consistent with these cycles being maximal in the mid-summer and lowest in the winter. The invertebrate data did not consistently capture these apparent seasonal trends. This may be due to a more rapid equilibration between mercury exposure conditions and bioaccumulation in these organisms.

As initiated at the beginning of the second year of the project, organic (methyl) mercury analyses have been added to the standard total mercury assays for biological samples. As seen in reports generated since that time, these additional data again closely followed the total mercury values, at high percentages of the total mercury concentrations. Data interpretation would be very similar using either mercury parameter as a monitoring tool. However, we felt that it was important, for this demonstration wetland effects study, to obtain a solid set of representative methyl vs total mercury data.

Also because of the likely future importance of this demonstration project, we continued to conduct substantially increased numbers of some of the analyses relative to the numbers required (502 mercury analyses this period vs the contracted 75), in order to obtain tighter confidence intervals, facilitating the statistical differentiation of mercury exposure conditions between individual sites and dates.

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## BACKGROUND AND INTRODUCTION

The Cache Creek Nature Preserve is a constructed wetland reserve in Yolo County, built in 1999-2000 at the site of a former gravel mining operation. It is located between Gordon Slough and Cache Creek, just west of County Road 94B and the Gordon Slough-Cache Creek confluence, on the north side of Cache Creek. The Preserve was built to provide new habitat for wildlife and recreational and educational opportunities for the public. A contoured pond system with several islands was created and landscaped with native vegetation. A visitor's center and a system of walkways and overlooks were built. The Preserve has attracted numerous wildlife species and is becoming a popular educational and recreational destination.

As constructed, the wetland is intended to occasionally have its water exchanged between the months of approximately April and September (irrigation season). During these months, water is periodically diverted from Gordon Slough to the wetland, exiting the wetland to Cache Creek approximately 0.5 km upstream of the Gordon Slough - Cache Creek confluence. Gordon Slough is a seasonally operated irrigation drainage canal, which contains agricultural return flows of water that originates from upstream Cache Creek. Wetland water exchanges typically occur for 1/2 – 2 days and are conducted one or two times per month during irrigation season. As is typical for the fall-winter period, no exchange of water occurred during this reporting period. It was further notable that no exchanges were conducted during the previous summer season.

Cache Creek is known to transport seasonally elevated loads of mercury (Hg) from historic mining districts in the upper watershed (Slotton et al. 1997, Foe and Croyle 1998). Consequently, there is a general interest in mercury and mercury bioaccumulation throughout the watershed. Because wetland habitats have been demonstrated to enhance the conversion of inorganic mercury to bioaccumulating methylmercury in certain environments (Rudd 1995), it was felt that the issues of mercury and mercury bioaccumulation should be investigated at the Cache Creek Nature Preserve, both within the Preserve wetlands and in adjacent Cache Creek upstream and downstream of the Nature Preserve outlet. Mercury dynamics in this wetland are additionally of interest as a model that may help in the planning of potential future wetland preserves at other sites in the region.

A three-year mercury monitoring study was initiated in the fall of 2000. The primary purpose of this monitoring program is to indicate the potential role, if any, that Nature Preserve water discharges may have on mercury levels in adjacent Cache Creek. Secondly, the study seeks to quantify the relative changes, if any, in mercury bioaccumulation within the constructed wetland system itself, relative to conditions in both its source waters and adjacent Cache Creek. The monitoring includes a series of quarterly collections, with data reports issued semi-annually. This is the fifth of these reports. In it, we discuss findings from collections made in Fall 2002 and Winter 2002/2003, and compare to results from previous sampling for the project (Fall 2000 through Summer 2002).

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## METHODS

As designed, the monitoring program focuses on mercury levels in selected types of localized aquatic biota as the primary indicators of relative mercury conditions at several key locations. These locations are:

- Cache Creek upstream of the Preserve and the Preserve water outlet.
- Gordon Slough, near the Preserve intake and the confluence with Cache Creek.
- The Nature Preserve wetlands.
- Cache Creek downstream of Gordon Slough and the Preserve.

The Cache Creek sites for invertebrate collections are located approximately 1 km upstream of the Preserve and 1 km downstream (~0.7 km downstream of the Cache Creek - Gordon Slough confluence). Small fish are taken along the Creek from these points and continuing away from the Preserve for a further 0.3-0.7 km. An additional site was added for invertebrate sampling only at:

- Cache Creek between the Preserve outlet and the Gordon-Cache confluence.

Fish cannot be effectively sampled from this intermediate site because (a) the reach between the Preserve outlet and the Cache-Gordon confluence is too small to contain sufficient, reliable samples and (b) due to fish movement, it is not possible to ensure that samples taken in this location are not derived from adjacent sites upstream and downstream. However, with some effort, it has been possible to collect adequate samples of invertebrate bioindicators that we believe are representative of mercury exposure conditions at the intermediate stretch of Cache Creek just downstream of the Preserve outlet.

By comparing mercury concentrations in similar local biota taken at the different sites, it can be determined whether statistically significant differences exist among the sites. The potential role of Nature Preserve water discharges on mercury levels in Cache Creek can also be assessed.

All of the sites are scheduled for sampling on a semi-annual basis, once in the fall and once in the spring. Because aquatic organisms accumulate their mercury burdens over time, biotic sampling can provide information on average conditions throughout the previous months. The fall samplings are intended to integrate relative mercury bioaccumulation throughout the warm season period of irrigation flows and maximal biological activity. The spring collections monitor conditions during the previous cool season, when the Preserve is not designed to exchange water. Supplementing the primary multi-site, semi-annual collections are additional quarterly samplings from the Nature Preserve wetlands site alone, conducted in summer and winter.

The target biota for use as mercury bioindicators in this project are small or juvenile fishes and aquatic insects that tend to remain within fairly localized ranges. These

organisms have been found to be ideal monitoring tools to indicate relative mercury bioaccumulation between locations, in numerous research studies by the authors throughout California and by other researchers elsewhere. Because these organisms are generally well under a year old when collected, they can also reflect potential seasonal changes in mercury exposure more directly than large fish. In contrast, large fish mercury burdens represent the combined mercury accumulations of several years time. Large fish are also far less likely to have remained in the immediate sampling region throughout their lives.

The localized availability of potential mercury bioindicator species was determined at each of the sites in the initial Fall 2000 work. Sampling was directed at taxa that were prevalent and would not be impacted by collections. Efforts have been made to collect similar sample types from the different sites and between collection dates. Whenever possible, composite samples of multiple individuals were collected. For the samples that were most inter-comparable between sites and/or dates, replicate composites were taken when possible. When three or more identical composites could be taken, each containing multiple small individual fish or invertebrates, statistical confidence intervals of the mean mercury levels were determined.

Benthic aquatic insects were collected with research kick screens and various nets. Small fishes were taken using a backpack electro-shock unit, beach seines, and other nets. Samples were identified, measured, cleaned, and sorted using well-established laboratory protocols. Composite and individual samples were prepared and analyzed for both total and methyl mercury at the UC Davis Environmental Mercury Laboratory. Research-level Quality Assurance / Quality Control (QA/QC) was employed throughout.

Following the first year of the project, in which we analyzed strictly for total mercury according to the monitoring protocol, the concern was raised that tracking of total mercury alone in project bioindicator samples might not provide an accurate measure of methylmercury trends. We have therefore conducted double analyses of all samples since that time, generating both organic (methyl) and total mercury data. Using our extensively tested procedures, we analyze both total and methyl fractions for each sample within the same analytical run, thereby further reducing potential laboratory-based variability. Additionally, because of high levels of natural variability seen occasionally in the creek invertebrate samples, all of the invertebrate samples were further analyzed in duplicate, using laboratory split samples. This again doubled the analytical load but improved the statistical confidence intervals for each sample.

For this last semi-annual period, we tested individual variability relative to the former approach of 3-6 composites, each of multiple individuals. Therefore, in this report alone, we present mean data resulting from numerous (6-10) analyses of individual small fish, together with associated statistics. Smaller sizes of mosquitofish and all of the invertebrates continued to be analyzed in multi-individual composites due to analytical constraints.

## RESULTS AND DISCUSSION

This fifth semi-annual data report discusses samples taken in the fall of 2002 (November 12-15) and the winter of 2002/2003 (February 14). A total of 178 fish samples, including both individuals and composites of multiple similar individual small fish, were prepared and analyzed from the four fish sampling locations in Fall 2002. Thirty-eight composite samples of aquatic insects were assembled from the fall collections as well. Winter 2002/2003 collections at the Nature Preserve site alone included 35 composite samples of fish. Samples analyzed for both total and methyl mercury during this reporting period are summarized in Table 1 (below). Additional data tables and associated figures are presented together following the text. Figures 6(a-e) and 7(a-c) compare selected parameters seasonally across the entire monitoring project to-date.

**Table 1. Summary of Biological Samples Analyzed for Mercury in This Six Month Period (Fall 2002 and Winter 2003 Collections).**

<u>Site</u>	<u>Small Fish</u>	<u>Aquatic Insect</u>	<u>Totals</u>
<u>FALL 2002</u>			
Cache Ck upstream of Preserve	32	12	<b>44</b>
Gordon Slough	29		<b>29</b>
Nature Preserve Wetlands	83		<b>83</b>
Cache Ck btw. Preserve & Gordon Sl.		13	<b>13</b>
Cache Ck downstream of Gordon Sl.	<u>34</u>	<u>13</u>	<b><u>47</u></b>
TOT. FALL 2002 SAMPLES (216 total):	<b>178</b>	<b>38</b>	<b>216</b>
<u>Winter 2003</u>			
Nature Preserve Wetlands	<u>35</u>		<b><u>35</u></b>
TOT. WINTER 2003 SAMPLES (35 total):	<b>35</b>		<b>35</b>
TOTAL SAMPLES FOR THg ( <u>251 total</u> ):	<b>213</b>	<b>38</b>	<b>251</b>
TOTAL SAMPLES FOR MeHg ( <u>251 total</u> ):	<b>213</b>	<b>38</b>	<b>251</b>

These 251 total mercury and 251 methylmercury samples were prepared and analyzed using research levels of QA/QC. Associated QA/QC data are presented in Table 2(a-b). No problems were encountered in the analyses and the associated data for these two quarters of sampling, as those from the preceding quarters, can be treated as very reliable.

## **SITE TO SITE (SPATIAL) VARIATION IN MERCURY**

### **Mercury in Small/Juvenile Fishes**

The new multi-site fish mercury data from Fall 2002 are presented in Tables 3-7 and are shown graphically in Figures 1-4. In fish sampling since Year 1, we are focusing our efforts on non-native species that can be obtained in replicate composites from the maximum number of sites. While the composite and individual samples were all powdered and analyzed on a dry weight basis for consistency, the values as presented have been converted to wet (fresh) weight concentrations so as to be comparable to various guidelines. Moisture percentage was determined for each individual sample to convert the consistent data from the dry, powdered samples to a fresh/wet weight basis. Replicate composite samples were generally similar in mercury concentration. Replicate individual single fish were found to be far more variable. The data are displayed so as to allow inter-comparison of the sites and dates, for each sample type. The added parameter of methylmercury will be additionally discussed in a following section, though it can be summarized as having virtually mirrored the total mercury data, at a consistent high percentage of corresponding total mercury. Identical mercury trends were described by both data sets and either would lead to the same conclusions. Below, we discuss the data trends relative to total mercury, corresponding to the previous four reports.

Red shiners (Table 3, Figure 1) were initially the most readily available of the fish species present at all of the fish sampling locations. And, being an invasive, non-native species, we could sample intensively without impacting the local aquatic environment. This species thus provided the strongest measure of inter-site variability in fish mercury bioaccumulation. Red shiners have remained abundant at the Gordon Slough and Cache Creek sites but, over the course of the project, shiners in the Nature Preserve itself have declined to low densities. They were initially largely displaced by fathead minnows as the new wetlands evolved. Parallel sampling has been conducted with both species to the extent possible during the period of overlap. It appears now that both of these indicator species may be disappearing at the Preserve site, which continues to evolve. Red shiners remain an excellent indicator species at the Gordon Slough and Cache Creek sites.

Partly due to the difficulty of obtaining additional samples at the Nature Preserve site, we decided to test the approach of analyzing multiple individual fish rather than multiple composites of numerous individuals. For this reporting period, we tested this approach at all of the fish sites and with all small fish samples other than the tiny mosquitofish. We were additionally interested in potential individual variation in mercury bioaccumulation



among similar individuals. Up to 10 individual analyses of similar single fish were performed for each site and size class, resulting in an approximate doubling in the total number of analyses in this period. Unfortunately, as the figures portray, individual variation was not insignificant. The resulting statistical confidence intervals for these sets of fish were considerably less precise than in previous samplings, where multiple composites, each containing numerous similar individuals, were analyzed. This is an important finding. In ongoing sampling, we will preferentially utilize multi-individual composites (taken in up to six replicates as available), with analysis of replicate individual fish only when constrained by very low sample numbers. It is important to note, however, that we have found greater variation in fall samplings to date, as compared to spring samplings, even among multi-individual composites. Thus, not all of the decrease in precision in this report was due to the use of individuals rather than multi-individual composites.

Fall 2002 data are shown in Table 3 and are plotted in Figure 1. Mean mercury concentrations from multiple individual analyses ranged between 0.08 and 0.19 ppm in all samples except one. The larger size class from the Cache Creek above Preserve site was considerably elevated at 0.36 ppm. As compared to corresponding samples from the previous spring sampling, these concentrations were notably greater, by approximately two to three fold. This spring low and fall high trend has been consistent across the span of the study. The larger size class (51-65 mm, Class 3) exhibited higher concentrations than the mid-size class (36-50 mm, Class 2) by up to approximately double. The difference was statistically significant at the two Cache Creek sites. Between sites, the Fall 2002 data set also exhibited some statistical differences. Among the smaller size class fish, which can be expected to have their mercury accumulations most dominated by conditions of the recent season, trends were similar to those seen in past years. Gordon Slough fish were notably lower than all others, the Cache Creek below Gordon Slough site was intermediate, and the Nature Preserve and upper Cache Creek sites exhibited approximately double the concentrations found in Gordon Slough. At the 95% level of significance, Nature Preserve Class 2 shiners were statistically elevated over both the Gordon Slough and Cache Creek below Gordon Slough sites. Among the larger size class, however, fish from the Nature Preserve, Gordon Slough, and Cache Creek below Gordon Slough were indistinguishable statistically. The sample from Cache Creek above the Preserve was statistically elevated above all of these sites.

Methylmercury spatial trends corresponded to those seen for total mercury and were also statistically significant in most cases. It is interesting that the methyl:total mercury percentage in this sampling, following the warm season, was greater, at 93-99% (mean = 96.5%), than the post-cold season spring sampling, when the corresponding percentages were 83-93% (mean = 87.6%).

Fathead minnow data from the Fall 2002 collections are displayed in Table 4 and are shown graphically in Figure 2. This species was available only from the Nature Preserve wetlands, and it was nearly absent there as well. With much work, it was possible to obtain six individuals from each of the two size classes. These were analyzed individually. Concentrations ranged between 0.09 and 0.13 ppm. The larger size class



was somewhat *lower* in mercury concentration than the mid size class, though the difference was not statistically significant. As compared to red shiners of the same sizes from the Nature Preserve site, the Fall 2002 fathead minnows were approximately 24-50 % lower in overall mercury levels (0.13 and 0.10 vs 0.17 and 0.19 ppm). The same trends were apparent in the methylmercury data. Fathead minnows have been sampled primarily as an alternate to the red shiners at the Nature Preserve site, where the shiners have been declining. However, it appears that both species may soon be largely absent from that site in the near future. Because of the normal successional evolution of newly created wetland systems, it will be important to utilize an adaptive monitoring system in any similar monitoring, expecting changes in available bioindicator species.

Juvenile green sunfish (Table 5, Figure 3) were taken at all four of the fish sampling sites in the Fall 2002 sampling. Individual fish were analyzed in numbers sufficient to provide statistical confidence intervals for seven of the eight site/size class samples. Green sunfish mercury concentrations from Fall 2002 ranged between 0.04 and 0.22 ppm. As in the previous two fall seasons, concentrations and inter-site trends diverged fairly sharply from the previous spring sampling. While levels at Gordon Slough and Cache Creek below Gordon Slough remained low or increased moderately, concentrations at Cache Creek above the Preserve and, particularly, the Nature Preserve itself exhibited dramatic increases. These elevations were statistically significant, both spatially and seasonally. Use of multiple individual analyses vs replicate, multi-individual composites led to less precise statistical confidences despite the increased analytical load. In any case, we can assert that, between Spring 2002 and Fall 2002, mercury concentrations in the Nature Preserve wetlands green sunfish increased by 138-139 % in both size classes.

Comparable seasonal changes at the other fish sites were: 62-161% at the upper Cache Creek site, 10-59% at Cache Creek below Gordon Slough, and -34-46% in Gordon Slough. Spatially, in the Fall 2002 sampling, the upper Cache Creek site was statistically elevated, at the 95% confidence level, for both size classes relative to corresponding green sunfish from the Gordon Slough and Cache Creek below Gordon Slough sites. We note that, again, fish from Cache Creek below Gordon Slough and the Preserve were statistically *lower* in mercury bioaccumulation than corresponding fish taken from Cache Creek above the Preserve. Among Size Class 1 fish (25-50 mm), those from the Nature Preserve site were most highly elevated; this elevation was statistically significant over corresponding samples from the Gordon Slough and Cache Creek below Gordon Slough sites. For Class 2 sunfish (51-80 mm), the upper Cache Creek site showed the greatest elevation; Nature Preserve sunfish of this size group could be shown to be statistically elevated over Gordon Slough fish but not those from Cache Creek below Gordon Slough. Methylmercury trends followed those noted for total mercury, continuing as in previous samplings at a high percentage of total mercury (91-100%).

Mosquitofish (Table 6, Figure 4) While this species has been very difficult to obtain during spring collections, conditions throughout the warm season allowed the establishment of good populations at each of the sites. We were able to obtain excellent, replicate composite samples, each of multiple individuals, at each of the sites for the smaller two size classes (20-30 mm and 31-40 mm), with less precise replicate (3-4) individual samples from each site for Class 3 fish (41-50 mm). We are thus able to

provide statistical confidence intervals for the means of each site/size sample. Concentrations in the Fall 2002 samples ranged between 0.06 and 0.27 ppm across all four sites. Spatial trends were consistent with the other Fall 2002 fish. In the small and mid sized mosquitofish, for which best statistics were available, lowest levels came from Gordon Slough, slightly higher levels from Cache Creek below Gordon Slough, and notably elevated concentrations from the Nature Preserve and Cache Creek upstream of the Preserve. These elevations were statistically significant for Class 1 fish from both high sites and for Class 2 fish from upper Cache Creek. The upper Cache Creek site had the highest concentrations for each of the three size classes. Methylmercury trends followed those seen for total mercury, also at a high percentage of total mercury (74-100%, mean = 88%). Interestingly, the youngest, smallest fish (Size Class 1, 20-30 mm) consistently demonstrated the highest methylmercury percentages (89-100%, mean = 95%).

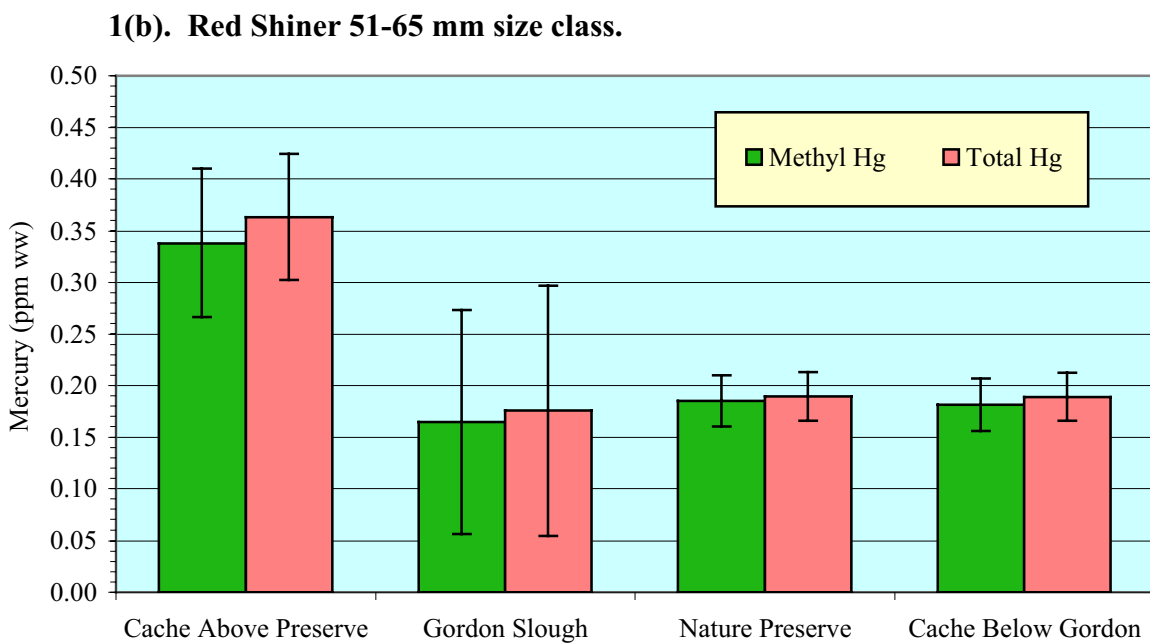
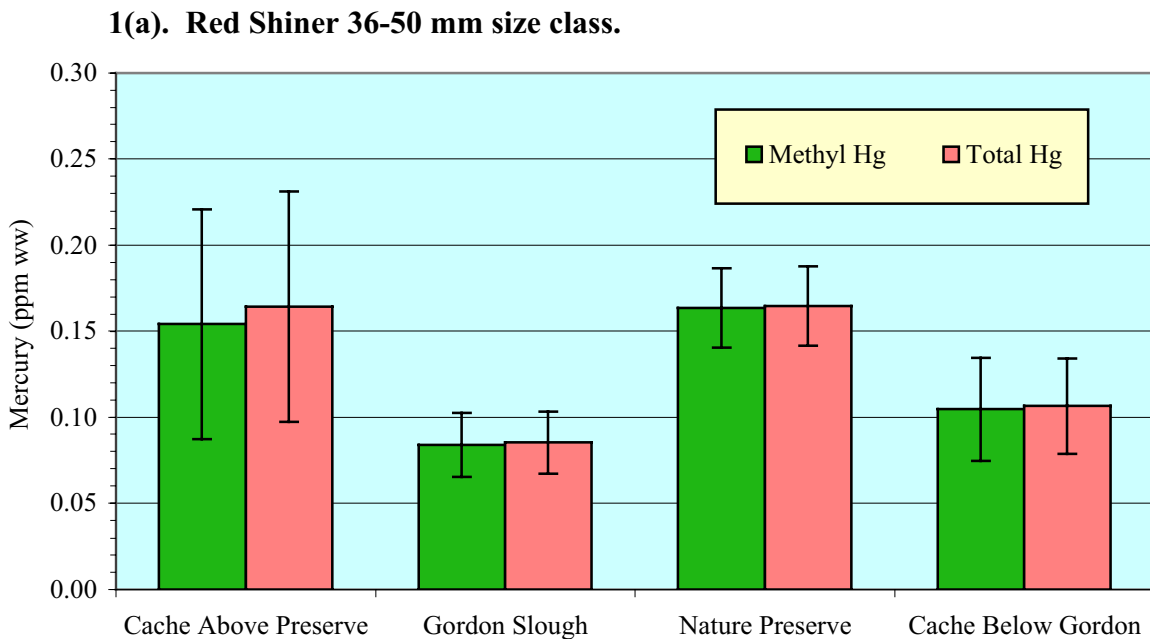
Additional Fish Species Taken From the Nature Preserve Site (Table 7). As the Nature Preserve wetlands continue to evolve, the fish assemblage there continues to change. Some of the originally prevalent species have become scarce, while several other species have colonized and gradually or rapidly increased. Inland silversides, bluegill sunfish, and black crappie have become populous enough to sample to some extent in the Nature Preserve, though they remain uneven or absent at the creek and slough sites. Like the other species utilized in this project for long-term bioindicator monitoring, these are also non-native, introduced fish species. They were sampled in the Nature Preserve during this Fall 2002 collection to obtain some comparative mercury information within the wetland environment. Between three and eight individuals were analyzed for each of two size classes for the silversides and bluegill. One size class of black crappie was sampled with six individual analyses. Data are presented in Table 7.

Inland Silversides samples were obtainable from the 51-65 mm and 66-80 mm size classes, comparable to silversides sampled at this site in the spring. Concentrations were dramatically elevated in Fall 2002. The 51-65 mm size class averaged 0.19 ppm (vs 0.05 ppm in Spring 2002, a 280% increase), and the 66-80 mm fish contained 0.26 ppm (vs 0.07 ppm in Spring 2002, a 270% increase). These dramatic increases were comparable to those seen in most of the other fish samples taken from the Nature Preserve in the Fall 2002 sampling. Also as in the other samples, methylmercury was found at a high percentage of total mercury (90-91%). Silversides exhibited the highest mercury content of any of the small fish sampled from the Nature Preserve in Fall 2002.

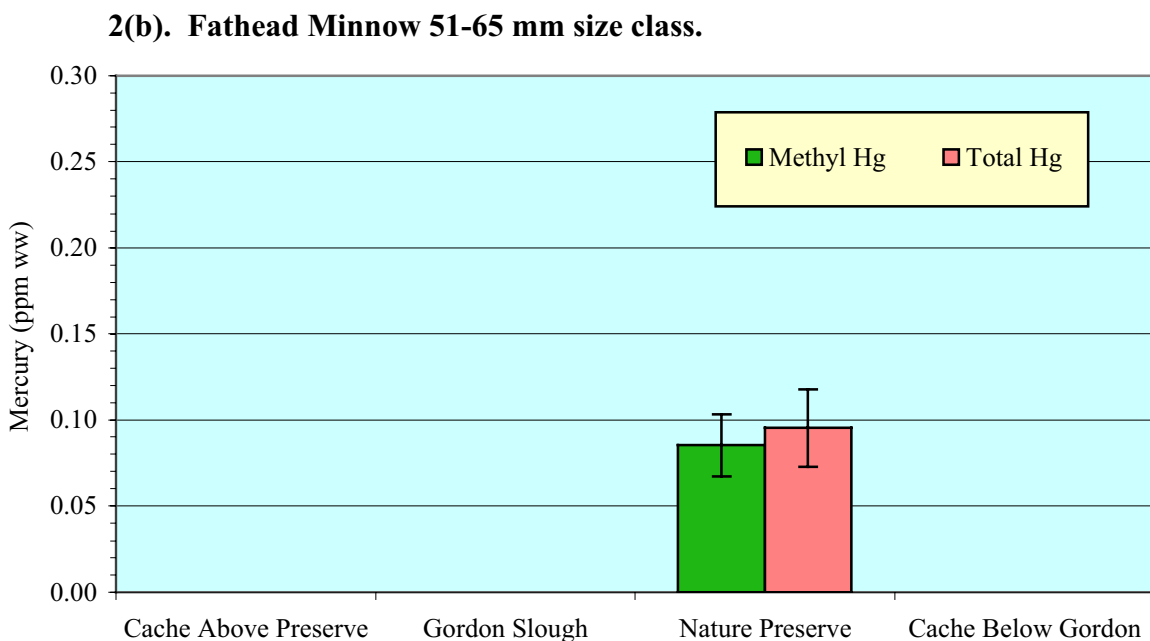
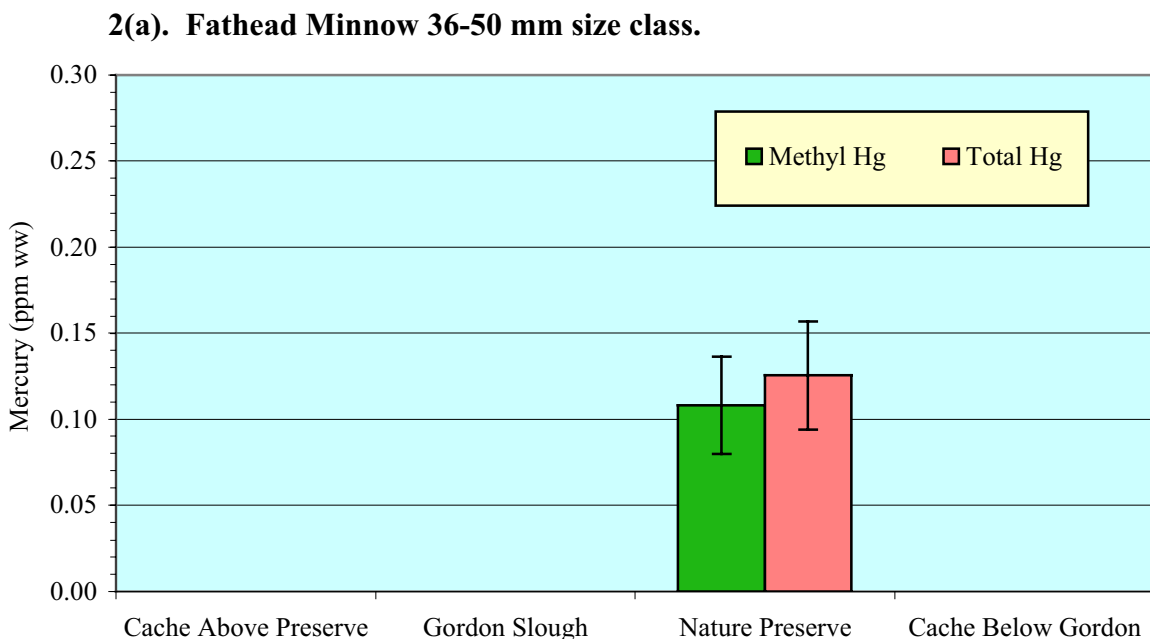
Bluegill Sunfish were taken in the two size classes utilized for green sunfish (25-50 mm and 51-80 mm). This species may be replacing green sunfish in the wetlands. Five to six individual analyses were conducted from each class. Concentrations averaged a very similar 0.12-0.14 ppm in both sizes. This was somewhat lower than the corresponding green sunfish concentrations of 0.16-0.17 ppm, reflecting the smaller mouth size and generally lower trophic level diet of the bluegill. Methyl percentages were high, at 92-100%

Black Crappie juveniles were available in one size range; 6 individuals between 70 and 100 mm were analyzed. Mercury averaged 0.15 ppm; methyl percentage was 91%. As crappie grow larger, they become piscivorous (eating fish) and can be expected to reach relatively high mercury concentrations (similar to bass if they were present).

**Fig. 1. FISH Composite Hg (wet wt ppm) VS LOCATION**  
**Red Shiner (*Notropis lutrensis*)**  
*(mean values plotted for replicate composites)*  
*(95% confidence intervals shown for samples with  $\geq 3$  replicates)*

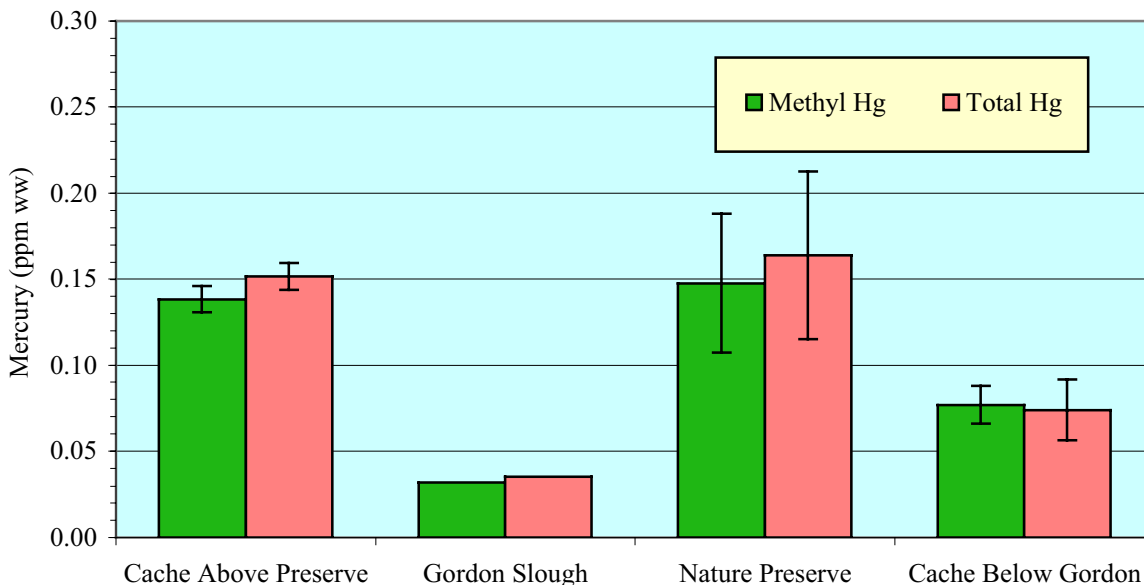


**Fig. 2. FISH Composite Hg (wet wt ppm) VS LOCATION**  
**Fathead Minnow (*Pimephales promelas*)**  
*(mean values plotted for replicate composites)*  
*(95% confidence intervals shown for samples with  $\geq 3$  replicates)*

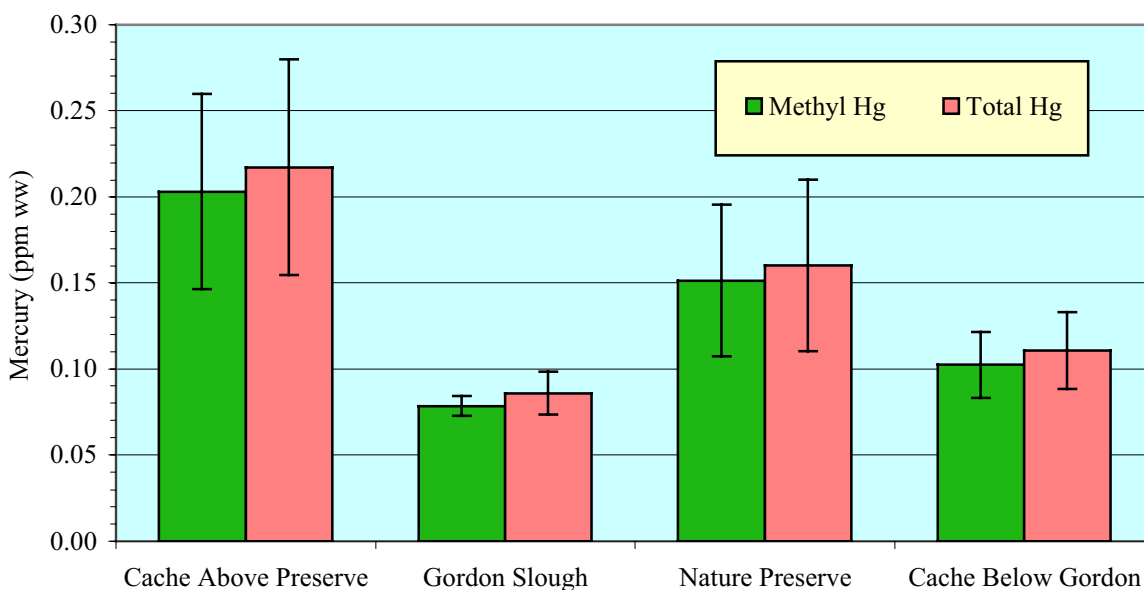


**Fig. 3. FISH Composite Hg (wet wt ppm) VS LOCATION**  
**Green Sunfish (*Lepomis cyanellus*)**  
*(mean values plotted for replicate composites)*  
*(95% confidence intervals shown for samples with  $\geq 3$  replicates)*

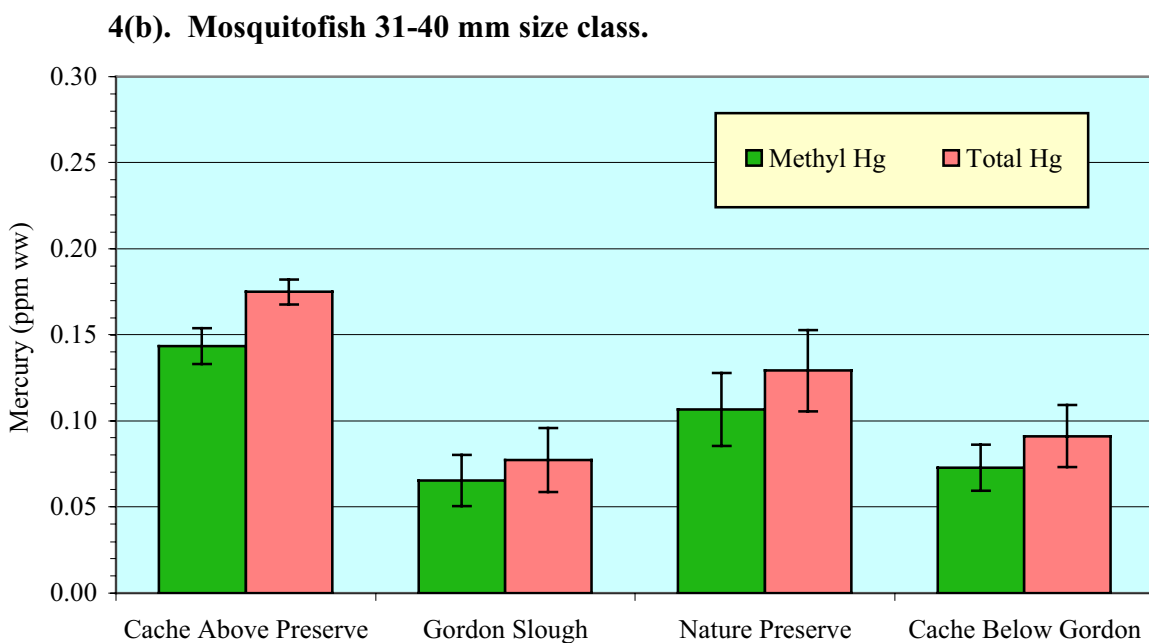
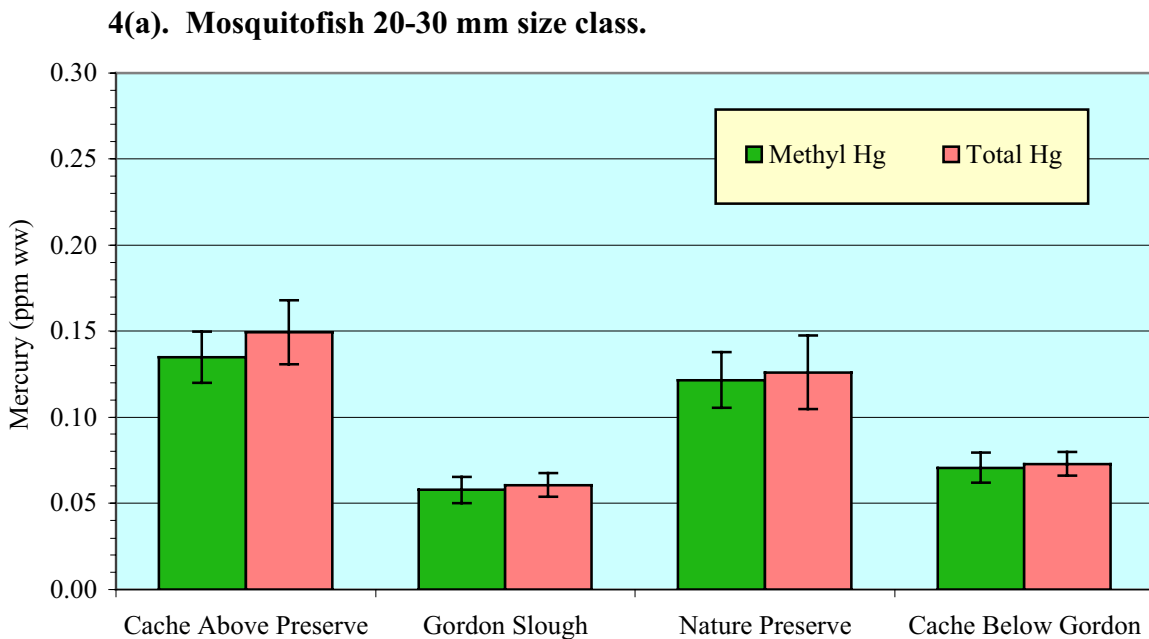
**3(a). Green Sunfish 25-50 mm size class.**

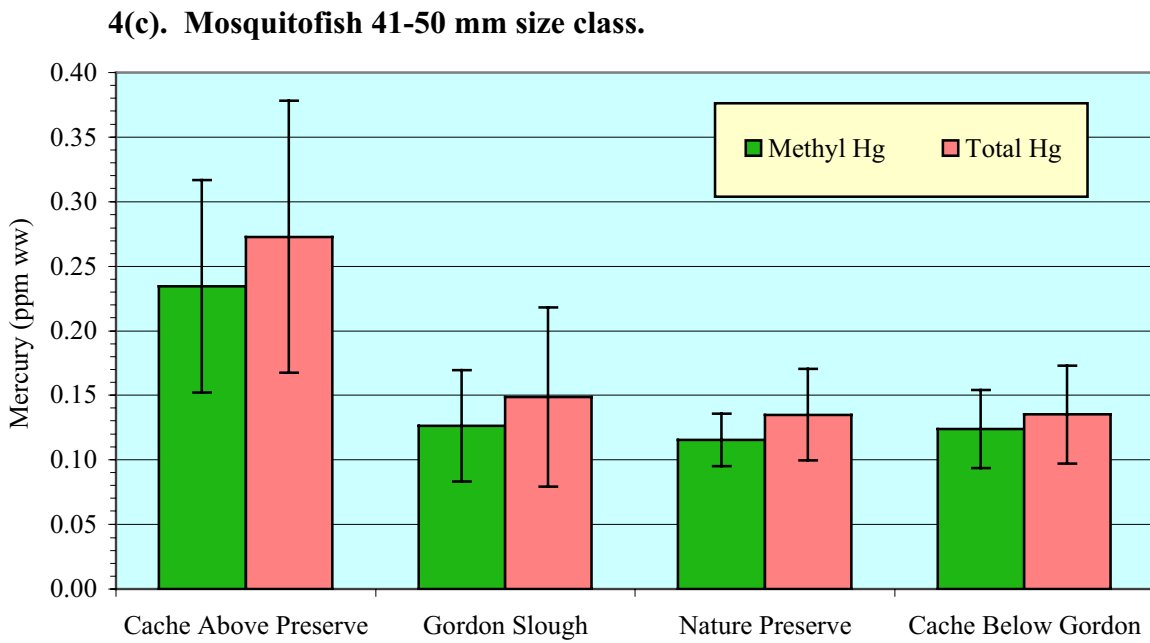


**3(b). Green Sunfish 51-80 mm size class.**



**Fig. 4. FISH Composite Hg (wet wt ppm) VS LOCATION**  
**Mosquitofish (*Gambusia affinis*)**  
*(mean values plotted for replicate composites)*  
*(95% confidence intervals shown for samples with  $\geq 3$  replicates)*







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## Mercury in Aquatic Insects

Aquatic insect mercury data from the Fall 2002 collections are presented in Tables 8-10 and Figures 5(a-d). These data, as initiated in Year 2 of the project, are displayed in units of *fresh/wet weight* parts per million total or methyl mercury, corresponding to the fish data. We have developed an accurate methodology to sequentially obtain the fresh and then dry weights of each invertebrate sample. This allows us to directly convert the invertebrate mercury dry weight, powdered concentrations, as analyzed, to a wet weight basis, for comparability with the fish. Because of the generally lower range of mercury concentrations in the invertebrate samples relative to the fish, the invertebrate data are graphically plotted with 0.00-0.10 ppm axes rather than the 0.00-0.30 range generally used for the fish figures, and the data are discussed at the 0.001 ppm level. One group (Naucorid predaceous water bugs) was somewhat elevated over the other invertebrate samples and was plotted on a 0.00-0.15 ppm axis.

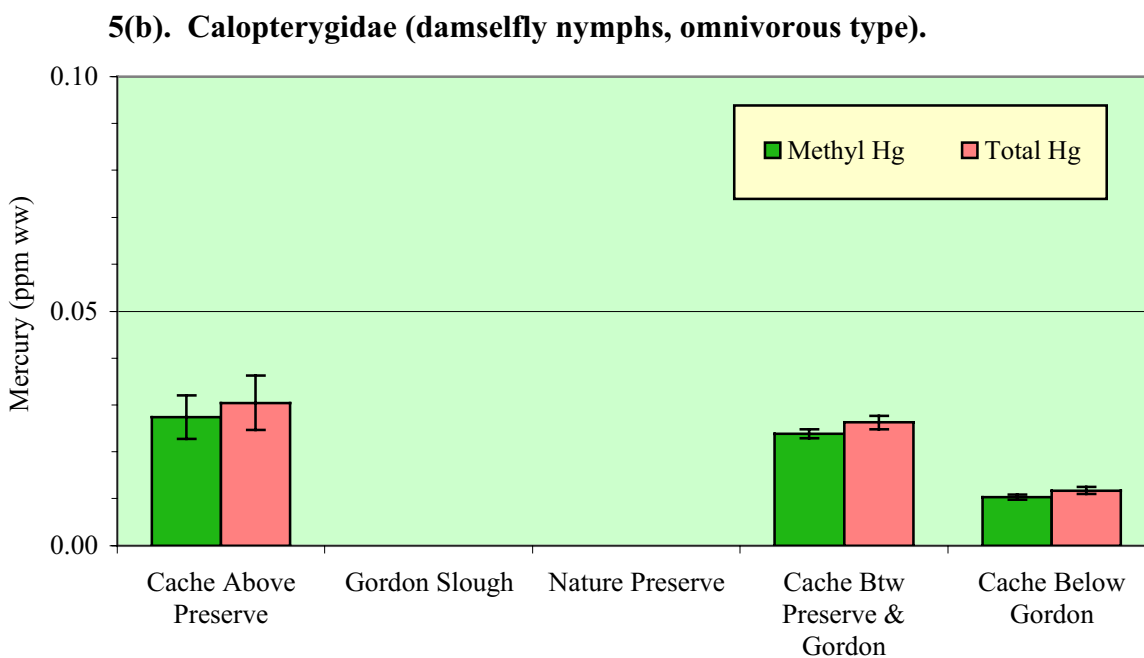
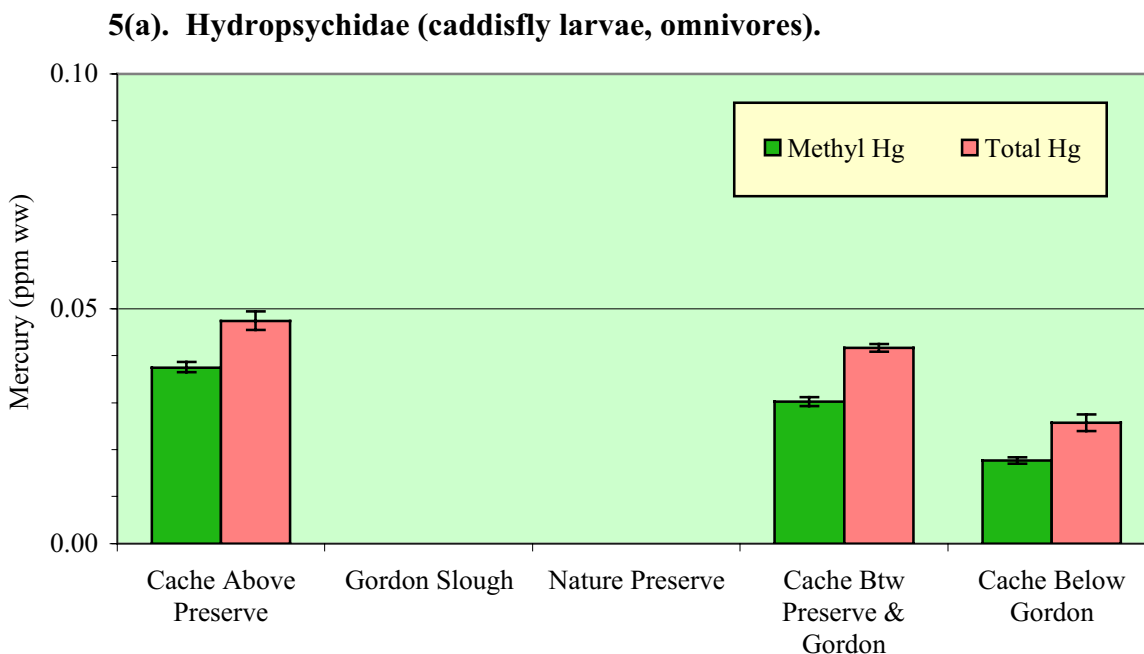
As in previous samplings, aquatic insects were not available from the Nature Preserve or Gordon Slough in sufficient numbers for meaningful comparative analysis. Fall 2002 aquatic insect data therefore come from the strategic series of Cache Creek sites including: Cache Creek above the Nature Preserve, Cache Creek between the Nature Preserve outlet and Gordon Slough, and Cache Creek below Gordon Slough.

Hydropsychid caddisfly larvae (Table 8, Figure 5a) were again taken in four replicate composite samples of numerous multiple individuals (40-70 each) from each of the three Cache Creek sites. We further analyzed each of the composites in duplicate, to lessen potential anomalous variation seen in earlier samplings. These intensive analyses again resulted in very good accuracy and precision in the statistics generated. Fall 2002 caddisfly mercury ranged between 0.025 and 0.049 ppm among all 9 of the composites. All were significantly elevated relative to corresponding samples taken in the previous spring, by the following percentages: 62% at the upper Cache Creek site, 20% between the Preserve outlet and Gordon Slough, and 30% at the Cache Creek site located downstream of both the Preserve and the Gordon Slough confluence. Spatially, in the Fall 2002 sampling, concentrations were found to decline somewhat, moving downstream from the site above the Preserve (0.047 ppm) to the site located between the Preserve outlet and Gordon Slough (0.042 ppm). This 11% decline was statistically significant. A more dramatic decline was noted at the most downstream Cache Creek site, located downstream of the Gordon Slough confluence (0.026 ppm). These identical caddisfly samples were 38% lower in mercury than those from the intermediate site and 45% lower than samples from the upstream site above the Preserve. Each of these mean site concentrations were statistically different from each of the others. The caddisfly methylmercury data showed matching trends at 67-83% of corresponding total mercury levels (mean = 74%). It is interesting to note that this percentage was consistently elevated relative to identical samples from Spring 2002 (58-64%, mean = 61%). Additionally, the Fall 2002 series exhibited a spatial decrease in methyl:total mercury percentage, moving from upstream to downstream (79%, 73%, 69%).

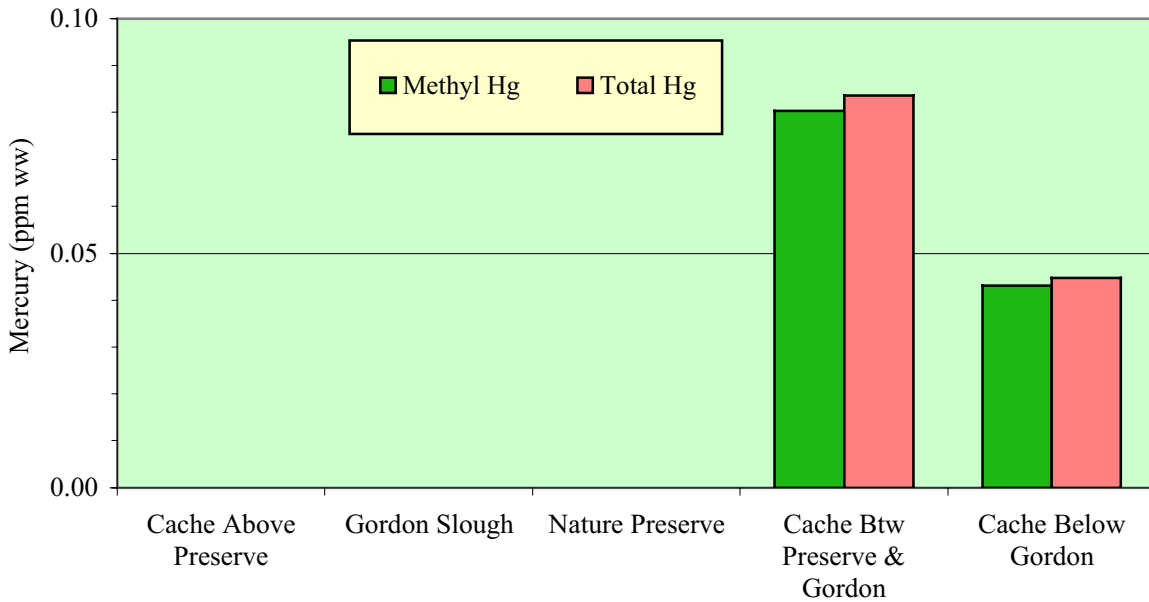
Calopterygid damselfly nymphs (Table 9, Figure 5b) were also taken from each of the three Cache Creek invertebrate sampling locations, with four replicate composites of multiple individuals (16-28 each) collected at each of the sites. These composites were also analyzed in duplicate to further lessen potential variability. The approach generated quite good precision and accuracy. Fall 2002 Calopterygid mercury ranged between 0.012 and 0.030 ppm. Similar to the caddisfly data, concentrations declined at this time along a downstream transect. Greatest concentrations occurred at the upstream site above the Preserve (0.030 ppm), declining somewhat to 0.26 ppm at the intermediate site located below the Preserve outlet and above Gordon Slough. This 13% decline was not statistically significant, due to variability in the upstream composites. However, a dramatic and statistically significant further decline was seen at the downstream site below Gordon Slough (0.012 ppm). Damselflies from the downstream site were 54% lower in mercury than identical samples from the intermediate site and 60% lower than identical samples from the upstream site. Methylmercury trends generally tracked those seen for total mercury, at 82-94% of total mercury (mean = 90%). Similar to the caddisfly data, the methyl:total mercury percentages from the Fall samplings were elevated relative to corresponding percentages from the previous spring (mean = 83%).

Data from additional aquatic insect samples (Table 10, Figures 5c-d), to the extent they were available, were consistent with those from the primary two invertebrate indicator species noted above. Libellulid dragonflies were not available at this time and Coenagrionid damselflies were available only as single, multi-individual composites at two of the three sites, precluding the generation of statistics. The Coenagrionid data were similar to those seen from the Hydropsychids and Calopterygids, declining between the intermediate site (0.084 ppm) and the downstream site below Gordon Slough (0.045 ppm), a 46% drop. On this sampling, it was possible to collect replicate, multi-individual composites of Naucorid predaceous water bugs at each of the three Cache Creek sites, comparable to the intensive samples of the primary bioindicator species. As seen in those samples, Naucorid mercury was significantly lower at the downstream site below Gordon Slough (0.046 ppm vs 0.111-0.128 ppm). In percentage terms, this downstream concentration was 59% lower than the upstream site and 64% lower than the intermediate location. The intermediate site (0.128 ppm) was somewhat higher than the upstream site (0.111 ppm) for this species and date, though the difference was not statistically significant. The overall Coenagrionid and Naucorid mercury concentrations were higher than those in the Hydropsychid and Calopterygid indicator species, with the Naucorids being most elevated. This is consistent with their more predaceous feeding habits relative to the primary species.

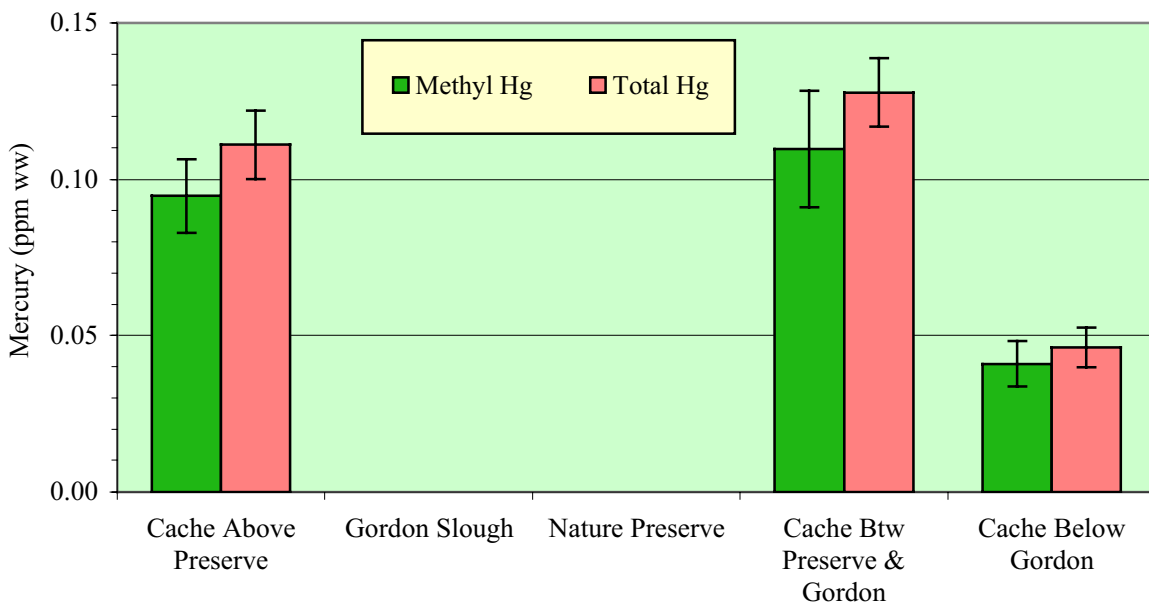
**Fig. 5. INVERTEBRATE Composite Hg (WET wt ppm)  
VS LOCATION**  
*(mean values plotted for replicate composites)*  
*(95% confidence intervals shown for samples with  $\geq 3$  replicates)*



**5(c). Coenagrionidae (predatory damselfly nymphs).**



**5(d). Naucoridae (predaceous creeping water bugs).**



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## SEASONAL VARIATION IN MERCURY LEVELS

In addition to the multi-site primary sampling of November 2002, a winter sampling of biota was made at the Nature Preserve wetlands site in February 2003. Data from that sampling are presented in Table 11. At this point in the project, we now have five semi-annual data sets from the Cache Creek and Gordon Slough sites (Fall 2000, Spring 2001, Fall 2001, Spring 2002, and Fall 2002) and ten quarterly collections from the Nature Preserve (Fall 2000, Winter, Spring, Summer, and Fall 2001, Winter, Spring, Summer, and Fall 2002, and Winter 2003). Graphic comparisons of selected, sequential, semi-annual fish data from Cache Creek and Gordon Slough are shown in Figures 6a, 6b, and 6c. Graphic comparisons of selected Nature Preserve fish data from all ten seasonal samplings to date are displayed in Figures 6d and 6e. The seasonal trends for invertebrates from the series of Cache Creek sampling locations are plotted in Figures 7(a-c).

The fish data from all of the sites now clearly demonstrate a consistent, strong seasonal pattern. This cycle includes a maximum in the fall, and a distinctive minimum level of fish mercury concentrations in the spring samplings. Where sufficient numbers of replicate composites have been available for statistical comparison, this fall high vs. spring low phenomenon was significant at the 95% confidence level. Though our experimental technique of using replicate individual samples (rather than multi-individual composites) resulted in wider statistical confidence intervals during this latest series of spring collections, the fall/spring differences were also generally statistically significant at the 95% level of confidence. The fathead minnow data continued to exhibit the smallest relative seasonal changes. But among the other species, the seasonal fluctuation in mercury content continued to be relatively enormous, with fall levels typically ranging from 50% to over 400% greater than corresponding concentrations measured in the spring. All three fall samplings to date (2000, 2001, and 2002) exhibited this distinctive elevation over the previous spring data. The most recent contrast between Fall 2002 and Spring 2003 concentrations are discussed above within the spatial treatment of individual species. The higher resolution quarterly data from the Nature Preserve wetlands provide additional information, also describing an annual pattern of minimal fish mercury concentrations in spring, increasing toward maximal concentrations in fall, with transitions between the spring low and fall high levels generally present in the winter and summer.

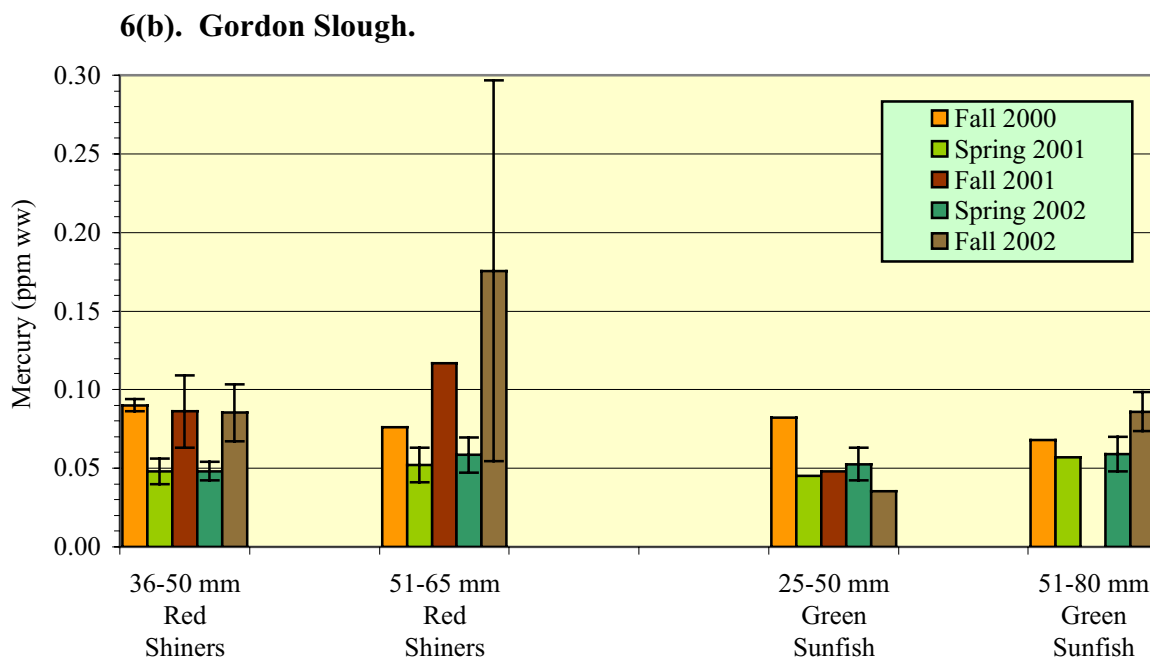
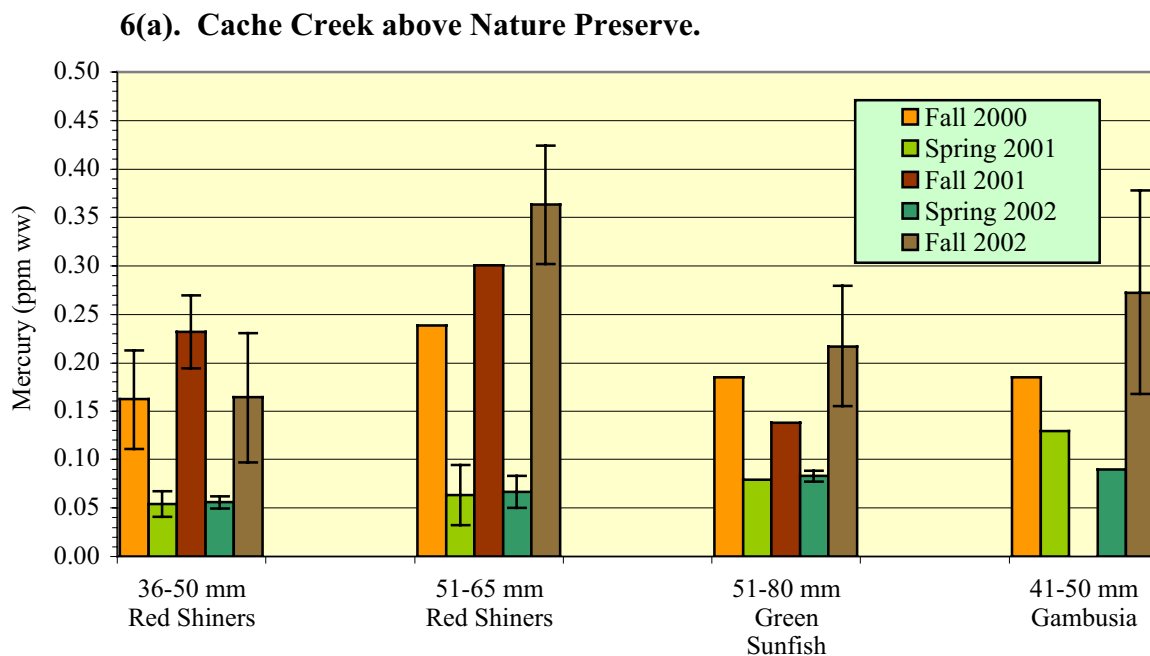
As fish mercury accumulates over time, it is likely that these peaks and lows exhibited in their concentrations are not precisely matched with the underlying seasonal cycles in methylmercury production and bioavailability. It is likely that those processes somewhat precede their appearance in the fish tissue record; i.e. lowest in the winter and highest in the summer. This would be consistent with the data set we have for methylmercury in water.

In contrast to the dramatic seasonal trends noted nearly uniformly for fish, the invertebrate seasonal data (Figures 7a-c) exhibited no consistent seasonal trend. For

some sites and/or organisms, spring data were lower than corresponding fall values. For others, spring levels were higher. In the case of Spring 2001 Hydropsychidae (caddisfly larvae) at the upstream Cache Creek site, this apparent spring increase was statistically significant. A significant inter-annual decline in concentrations was noted in the previous report in all but one of the comparisons where statistics were available, between Spring 2001 and Spring 2002. With the inclusion of the most recent Fall 2002 data, some of the invertebrate data sets are beginning to more closely resemble the fish trends, with lower levels in the spring and higher levels in the fall.

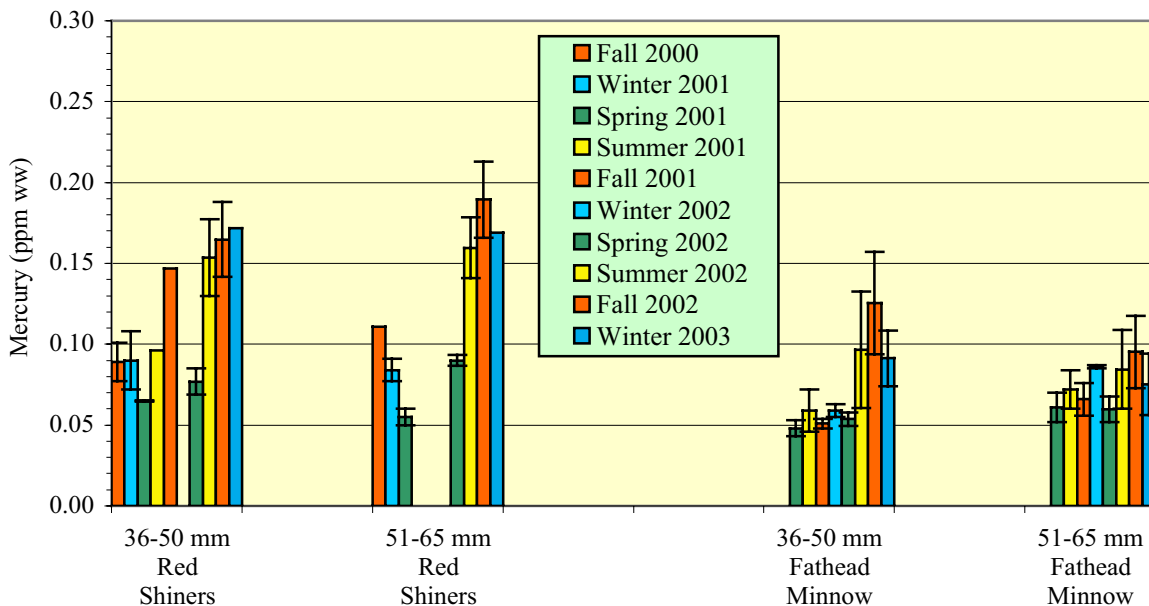
The invertebrate mercury concentrations were generally more consistent than the fish across seasons. This runs counter what we would intuitively expect. The relative lack of a strong seasonal pattern in most of the Cache Creek invertebrate data may be due to a different invertebrate response to changes in localized methylmercury exposure, relative to the small fishes. Some of our other recent research in the watershed and elsewhere suggests that this difference is related to a more rapid invertebrate response to changing mercury exposure conditions. Under this scenario, methylmercury production and exposure may indeed be maximal in the summer and at lowest levels in the winter. The fish may be taking a period of one to several months for their accumulated mercury burdens to reflect recent conditions, while the invertebrates may change more rapidly. This could be due to physiological differences or simply greater relative growth rates and turnover in the seasonal invertebrates as compared to the fish. Exposure and rapid equilibration in mercury accumulations may be fairly similar for the invertebrates in the beginning of the warm season in the spring and at the end of the warm season in the fall.

**Fig. 6 (a-e). FISH Composite Hg VS SEASON**  
(mean values plotted for replicate composites)  
(95% confidence intervals shown for samples with  $\geq 3$  replicates)

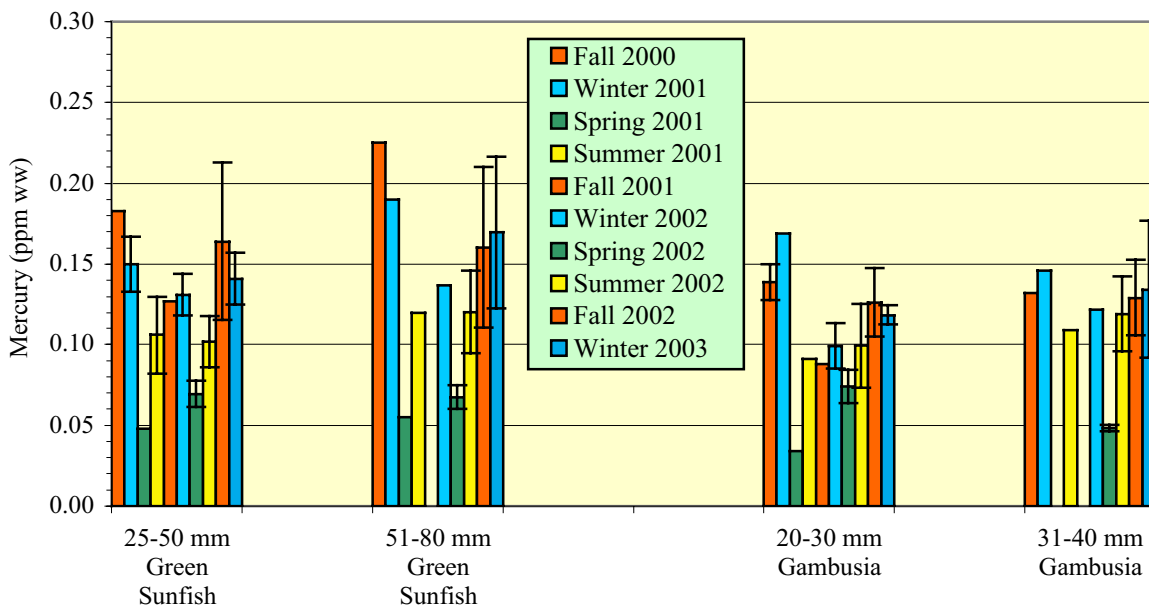




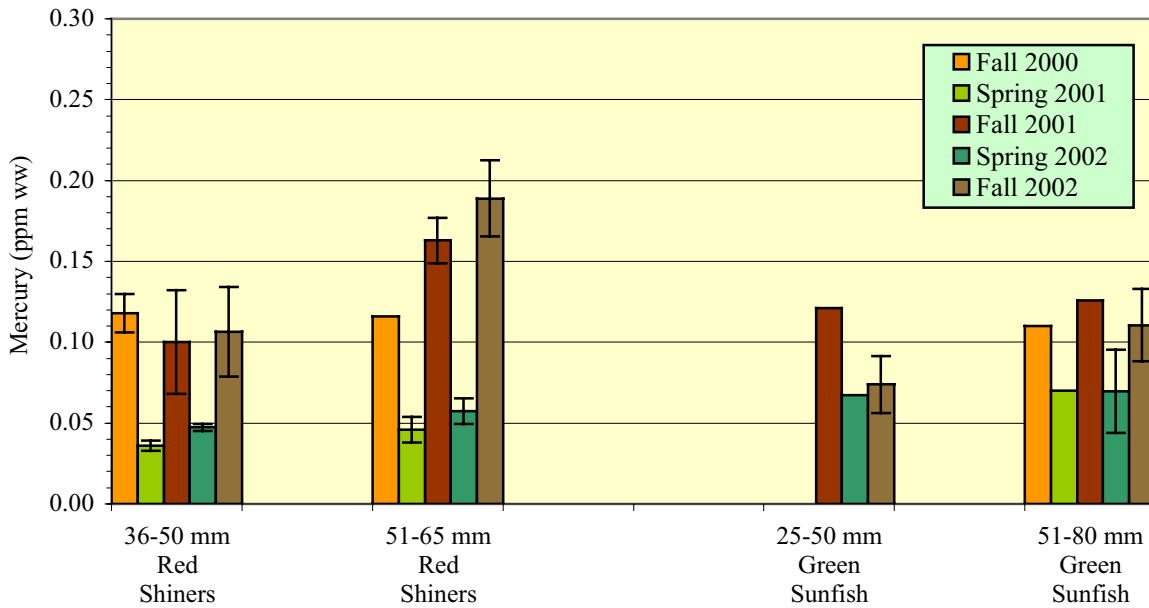
**6(c). Nature Preserve: Red Shiners and Fathead Minnows.**



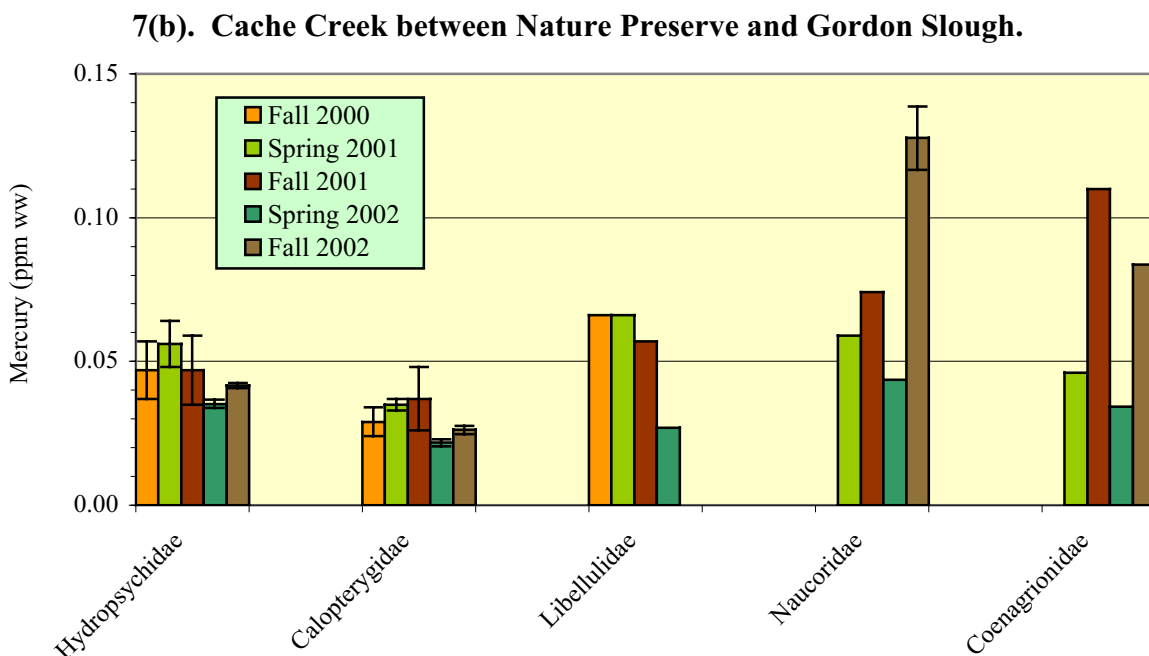
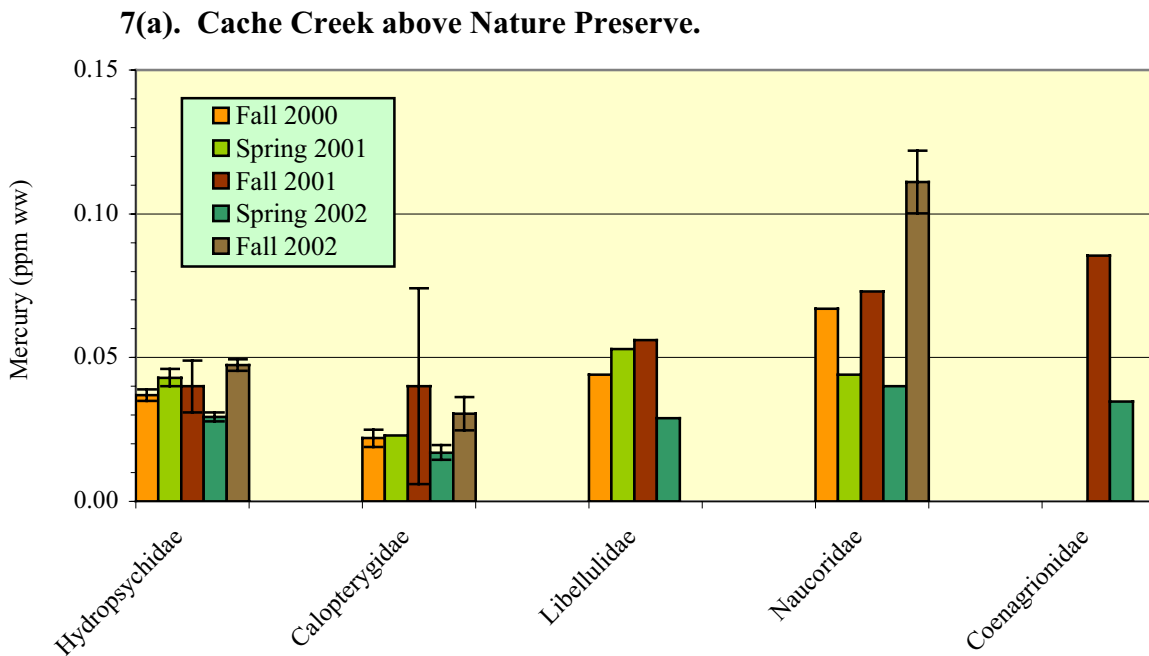
**6(d). Nature Preserve: Green Sunfish and Mosquitofish.**

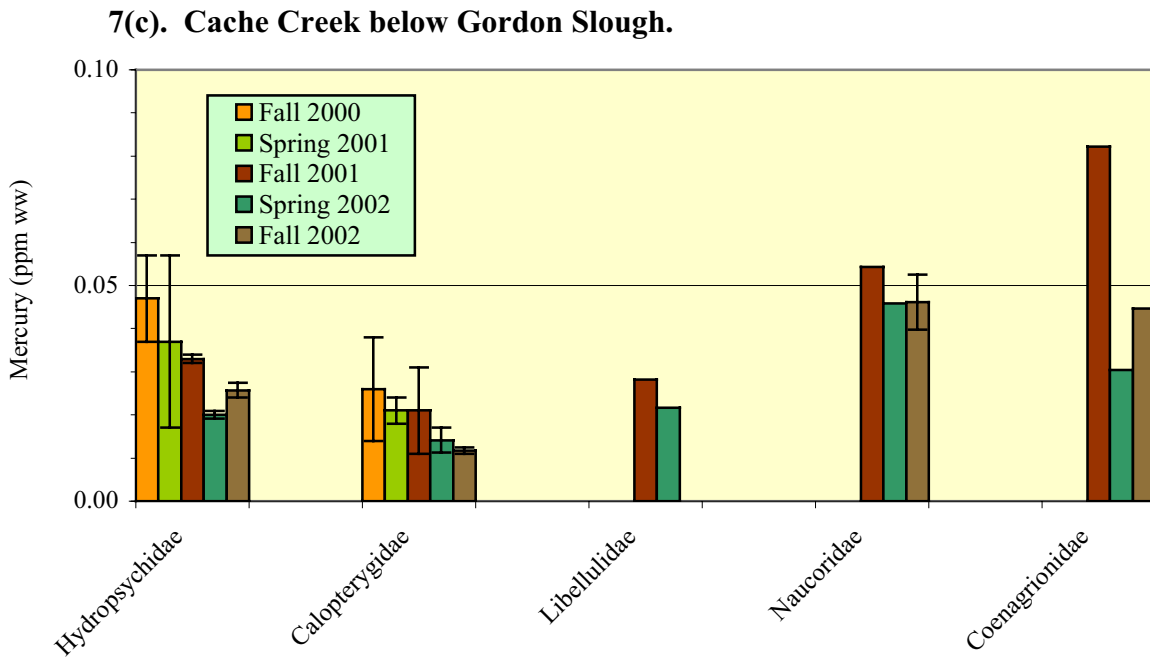


**6(e). Cache Creek below Gordon Slough.**



**Fig. 7 (a-c). INVERTEBRATE Composite Hg VS SEASON**  
(mean values plotted for replicate composites)  
(95% confidence intervals shown for samples with  $\geq 3$  replicates)





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### **Organic (Methyl) Mercury vs. Total Mercury in 2002/2003 Biotic Samples**

As already noted, the methylmercury data closely followed the corresponding total mercury values in nearly all paired comparisons, with the methylmercury concentration almost uniformly a high percentage proportion of the total mercury. Among the 213 fish samples analyzed for both parameters, methylmercury ranged from 74 to 111% of total mercury, with a mean of  $93\% \pm 7\%$  (standard deviation). The 38 invertebrate composites demonstrated methylmercury proportions that ranged from 67 to 97%, with a mean percentage of  $84\% \pm 9\%$ . Thus, the total mercury for these samples was primarily organic. The trends and conclusions to be drawn from these data would continue to be very similar using either parameter as a monitoring tool. However, we felt that it would be valuable to develop a baseline database of methyl vs total mercury information in bioindicator organisms for these types of wetland/creek systems.

### **CONCLUSIONS--FIFTH SEMI-ANNUAL REPORT**

With the completion of two and one half years of seasonal samplings in this three-year mercury monitoring program, we have accumulated a sizeable database of inter-comparable mercury bioaccumulation information from the Cache Creek Nature Preserve and related adjacent sites. With some difficulty, it has been possible to collect a series of closely corresponding fish samples from the Preserve and three additional fish locations. No corresponding aquatic insects were obtainable among all of the sites, although three types (Hydropsychid caddisfly larvae, Calopterygid damselfly nymphs, and Naucorid predaceous water bugs) were taken in excellent numbers at a series of three Cache Creek locations.

The following points can be made about the data collected in the past semi-annual period:

- Of the sites sampled throughout the project area, Gordon Slough continued to exhibit the lowest levels of mercury bioaccumulation in fish.
- Fish taken within the Nature Preserve contained mercury at levels similar to those seen at the upstream Cache Creek site, which was typically the most elevated. These levels, while similar to those from the Creek, represented a substantial increase over conditions in Gordon Slough, which provides the source water for the Nature Preserve.
- Fall season invertebrates collected at the intermediate Cache Creek site, located between the Nature Preserve outflow and Gordon Slough, did not demonstrate a consistent trend in localized mercury elevation, relative to the upstream Cache Creek site, as seen in some previous collections. Some of the data were somewhat lower at this site, some somewhat higher. None of the differences were statistically significant.

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It is notable that no water was released from the Nature Preserve wetlands in 2002 following the brief flushing conducted in April.

- The phenomenon of occasional increases in Cache Creek biotic mercury downstream of the Preserve continued to be highly localized. Throughout the project to date, Cache Creek below Gordon Slough contained biotic mercury at levels similar to and in most cases lower than corresponding samples from Cache Creek above the Preserve.
- Mercury in small fish bioindicators continued to exhibit a strong seasonal pattern, with maximum concentrations in the fall and minimum concentrations in the spring. We believe that this actually represents a delayed measure of underlying cycles of mercury methylation and methylmercury bioavailability, which likely peak in the summer and reach low points in the winter. Invertebrate bioindicators, as sampled, did not consistently demonstrate this pattern; related data from other research suggests that this may be due to different accumulation dynamics in the invertebrates (more rapid equilibration) and not to a different underlying trend in seasonal biological mercury exposure. The invertebrate bioindicators, however, can be used as highly accurate measures of fine scale spatial differences in relative mercury exposure, as well as short-term changes over time.
- The organic (methyl) mercury analyses conducted during this most recent semi-annual period again indicate that methylmercury constituted the majority of the mercury measured in fish ( $93\pm 7\%$ ) and invertebrates ( $84\pm 9\%$ ) from the project region. The methylmercury data generally mirrored the total mercury results, at a high proportion of the corresponding total mercury levels. At this point in the analytical cross-comparison, identical or very similar trends and conclusions would be supported by either parameter.
- The experimental use of multiple whole individual fish samples rather than composites of numerous individuals resulted in less precise statistical confidence intervals. We conclude that greater than 10 individual analyses are required to achieve the precision of 3-5 replicate composite samples, each containing multiple individuals. This is important information. In some situations, small numbers of available specimens would preclude the composite technique. For the remainder of this project, we will try to collect replicate multi-individual composites when possible.

## LITERATURE CITED

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Rudd, J.W.M. 1995. Sources of methyl mercury to freshwater ecosystems: A review. *Water, Air and Soil Pollution*, 80:697-713.

Slotton, D.G., S.M. Ayers, J.E. Reuter, and C.R. Goldman. 1997. Cache Creek watershed preliminary mercury assessment, using benthic macro-invertebrates. *Final Report for the Central Valley Regional Water Quality Control Board and the National Science Foundation*, June 1997, 36 pp.



**Table 2a. Laboratory QA/QC Summary for UC Davis Total Mercury Analyses Used in This Data Report.**

*Method Detection Limit (MDL) = 0.005 µg THg/g (ppm)*

	Std Curve R <sup>2</sup>	Lab Split RPD	Spike Recoveries	Lab Cont. Std. Recoveries	Cont. Calibration Validation
Ideal Recovery	1.000	(0%)	(100%)	(100%)	(100%)
Control Range (%)	≥0.975	≤25%	75-125%	75-125%	75-125%
Tracking Method		Control Chart	Control Chart	Control Chart	Control Chart
Recoveries (%)	0.9993-0.9999	0.3%-7.4%	92%-99%	96%-102%	97%-102%
(n)	n=8	n=12	n=24	n=18	n=26
Mean Recoveries (%)	0.9996	2.5%	95.7%	100.0%	99.0%

Standard Reference Materials

	TORT-2 Lobster	DOLT-2 Dogfish
Certified Level (ppm THg)	0.27±0.02	2.14±0.28
Ideal Recovery	(100%)	(100%)
Control Range (%)	75-125%	75-125%
Tracking Method	Control Chart	Control Chart
Control Range (ppm)	0.20-0.34	1.61-2.68
Recoveries (%)	95%-111%	95%-103%
Recoveries (ppm)	0.26-0.30	2.03-2.20
(n)	n=18	n=8
Mean Recoveries (%)	100.5%	98.9%
Mean Recoveries (ppm)	0.27	2.12

**Table 2b. Laboratory QA/QC Summary for UC Davis Methyl Mercury Analyses Used in This Data Report.**

*Method Detection Limit (MDL) = 0.005 µg THg/g (ppm)*

	Std Curve R <sup>2</sup>	Lab Split RPD	Spike Recoveries	Lab Cont. Std. Recoveries	Cont. Calibration Validation
Ideal Recovery	1.000	(0%)	(100%)	(100%)	(100%)
Control Range (%)	≥0.975	≤25%	75-125%	75-125%	75-125%
Tracking Method		Control Chart	Control Chart	Control Chart	Control Chart
Recoveries (%)	0.9993-0.9999	0.7%-13.9%	91%-115%	96%-102%	93%-104%
(n)	n=8	n=12	n=24	n=18	n=26
Mean Recoveries (%)	0.9996	3.8%	102.0%	100.0%	99.3%

Standard Reference Materials

	TORT-2 Lobster	DOLT-2 Dogfish
Certified Level (ppm THg)	0.152±0.013	0.693±0.053
Ideal Recovery	(100%)	(100%)
Control Range (%)	75-125%	75-125%
Tracking Method	Control Chart	Control Chart
Control Range (ppm)	0.114-0.190	0.520-0.866
Recoveries (%)	93%-105%	97%-105%
Recoveries (ppm)	0.14-0.16	0.67-0.73
(n)	n=18	n=8
Mean Recoveries (%)	99.0%	102.0%
Mean Recoveries (ppm)	0.15	0.71

**Table 3. November 2002 Fish Mercury:  
RED SHINER (*Notropis lutrensis*).**  
(Mean values of multiple (n) whole body individual analyses)  
(wet weight = fresh weight,  $\mu\text{g}$  mercury per gram = parts per million)

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>		<u><math>\mu\text{g/g}</math> METHYL Hg</u>	<u><math>\mu\text{g/g}</math> TOTAL Hg</u>	<u>% Methyl Hg</u>	
		<u>wt (g)</u>	<u>lgth (mm)</u>	<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>	
<u>Cache Creek Above Preserve</u>							
<b>2</b>	(36-50 mm)	6	0.69	44	<b>Means: 0.154</b>	<b>0.164</b>	<b>94%</b>
					<i>SD:</i> 0.064	0.064	5%
					<i>95% Conf. Intervals:</i> $\pm 0.067$	$\pm 0.067$	$\pm 6\%$
<b>3</b>	(51-65 mm)	6	1.77	58	<b>Means: 0.338</b>	<b>0.363</b>	<b>93%</b>
					<i>SD:</i> 0.068	0.058	5%
					<i>95% Conf. Intervals:</i> $\pm 0.072$	$\pm 0.061$	$\pm 5\%$
<u>Gordon Slough</u>							
<b>2</b>	(36-50 mm)	8	0.75	43	<b>Means: 0.084</b>	<b>0.085</b>	<b>98%</b>
					<i>SD:</i> 0.022	0.022	7%
					<i>95% Conf. Intervals:</i> $\pm 0.019$	$\pm 0.018$	$\pm 6\%$
<b>3</b>	(51-65 mm)	8	1.59	57	<b>Means: 0.165</b>	<b>0.176</b>	<b>96%</b>
					<i>SD:</i> 0.130	0.145	4%
					<i>95% Conf. Intervals:</i> $\pm 0.108$	$\pm 0.121$	$\pm 3\%$

(continued)

**Table 3. (Continued).**

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>			<u>µg/g METHYL Hg</u>	<u>µg/g TOTAL Hg</u>	<u>% Methyl Hg</u>	
		<u>wt (g)</u>	<u>lgth (mm)</u>		<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>	
<u>Nature Preserve Wetlands</u>								
<b>2</b>	(36-50 mm)	10	0.83	46	<b>Means:</b>	<b>0.164</b>	<b>0.165</b>	<b>99%</b>
					<i>SD:</i>	0.032	0.032	5%
					<i>95% Conf. Intervals:</i>	± 0.023	± 0.023	±4%
<b>3</b>	(51-65 mm)	10	1.39	55	<b>Means:</b>	<b>0.185</b>	<b>0.189</b>	<b>98%</b>
					<i>SD:</i>	0.035	0.033	5%
					<i>95% Conf. Intervals:</i>	± 0.025	± 0.024	±3%
<u>Cache Creek Below Gordon Slough</u>								
<b>2</b>	(36-50 mm)	6	0.83	46	<b>Means:</b>	<b>0.105</b>	<b>0.106</b>	<b>98%</b>
					<i>SD:</i>	0.028	0.026	4%
					<i>95% Conf. Intervals:</i>	± 0.030	± 0.028	±4%
<b>3</b>	(51-65 mm)	6	1.97	59	<b>Means:</b>	<b>0.181</b>	<b>0.189</b>	<b>96%</b>
					<i>SD:</i>	0.024	0.022	2%
					<i>95% Conf. Intervals:</i>	± 0.025	± 0.023	±3%

**Table 4. November 2002 Fish Mercury:  
 FATHEAD MINNOW (*Pimephales promelas*).  
 (Mean values of multiple (n) whole body individual analyses)  
 (wet weight = fresh weight,  $\mu\text{g}$  mercury per gram = parts per million)**

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>		<u><math>\mu\text{g/g}</math> METHYL Hg</u>	<u><math>\mu\text{g/g}</math> TOTAL Hg</u>	<u>% Methyl Hg</u>
		<u>wt (g)</u>	<u>lgth (mm)</u>	<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>
<u>Nature Preserve Wetlands</u>						
<b>2</b> (36-50 mm)	6	0.85	48	<b>Means:</b> <b>0.108</b>	<b>0.125</b>	<b>86%</b>
				<i>SD:</i> 0.027	0.030	4%
				<i>95% Conf. Intervals:</i> $\pm 0.028$	$\pm 0.032$	$\pm 4\%$
<b>3</b> (51-70 mm)	6	1.84	59	<b>Means:</b> <b>0.085</b>	<b>0.095</b>	<b>90%</b>
				<i>SD:</i> 0.017	0.021	3%
				<i>95% Conf. Intervals:</i> $\pm 0.018$	$\pm 0.022$	$\pm 3\%$

**Table 5. November 2002 Fish Mercury:  
GREEN SUNFISH (*Lepomis cyanellus*).**  
(Mean values of multiple (n) whole body individual analyses)  
(wet weight = fresh weight,  $\mu\text{g}$  mercury per gram = parts per million)

Size Class	n	Mean Fish Size			$\mu\text{g/g}$ METHYL Hg	$\mu\text{g/g}$ TOTAL Hg	% Methyl Hg
		wt (g)	lgth (mm)		in WET Sample	in WET Sample	of Total Hg
<u>Cache Creek Above Preserve</u>							
1 (25-50 mm)	3	2.03	50	<b>Means:</b>	<b>0.138</b>	<b>0.152</b>	<b>91%</b>
				SD:	0.003	0.003	3%
				95% Conf. Intervals:	$\pm 0.008$	$\pm 0.008$	$\pm 8\%$
2 (51-80 mm)	6	5.97	70	<b>Means:</b>	<b>0.203</b>	<b>0.217</b>	<b>94%</b>
				SD:	0.054	0.060	4%
				95% Conf. Intervals:	$\pm 0.057$	$\pm 0.063$	$\pm 4\%$
<u>Gordon Slough</u>							
1 (25-50 mm)	1	1.20	43		<b>0.032</b>	<b>0.035</b>	<b>91%</b>
2 (51-80 mm)	4	6.45	72	<b>Means:</b>	<b>0.078</b>	<b>0.086</b>	<b>92%</b>
				SD:	0.004	0.008	6%
				95% Conf. Intervals:	$\pm 0.006$	$\pm 0.012$	$\pm 10\%$

(continued)

**Table 5. (Continued).**

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>			<u>µg/g METHYL Hg</u>	<u>µg/g TOTAL Hg</u>	<u>% Methyl Hg</u>
		<u>wt (g)</u>	<u>lgth (mm)</u>		<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>
<u>Nature Preserve Wetlands</u>							
<b>1</b> (25-50 mm)	6	1.31	43	<b>Means:</b>	<b>0.148</b>	<b>0.164</b>	<b>91%</b>
				<i>SD:</i>	0.039	0.047	6%
				<i>95% Conf. Intervals:</i>	± 0.040	± 0.049	±7%
<b>2</b> (51-80 mm)	6	4.44	66	<b>Means:</b>	<b>0.151</b>	<b>0.160</b>	<b>95%</b>
				<i>SD:</i>	0.042	0.047	4%
				<i>95% Conf. Intervals:</i>	± 0.044	± 0.050	±4%
<u>Cache Creek Below Gordon Slough</u>							
<b>1</b> (25-50 mm)	4	1.77	48	<b>Means:</b>	<b>0.077</b>	<b>0.074</b>	<b>105%</b>
				<i>SD:</i>	0.007	0.011	8%
				<i>95% Conf. Intervals:</i>	± 0.011	± 0.018	±12%
<b>2</b> (51-80 mm)	6	5.62	68	<b>Means:</b>	<b>0.102</b>	<b>0.111</b>	<b>93%</b>
				<i>SD:</i>	0.018	0.021	2%
				<i>95% Conf. Intervals:</i>	± 0.019	± 0.022	±2%

**Table 6. November 2002 Fish Mercury:  
MOSQUITOFISH (*Gambusia affinis*).**  
(*Gambusia* SC 1&2 are composite samples of (n) whole individuals)  
(Class 3: Mean values of multiple (n) whole body individual analyses)  
(wet weight = fresh weight,  $\mu\text{g}$  mercury per gram = parts per million)

Size Class	n	Mean Fish Size		$\mu\text{g/g}$ METHYL Hg	$\mu\text{g/g}$ TOTAL Hg	% Methyl Hg
		wt (g)	lgth (mm)	in WET Sample	in WET Sample	of Total Hg
<u>Cache Creek Above Preserve</u>						
1 (20-30 mm)	(15)	0.17	26	0.130	0.142	92%
1 (20-30 mm)	(15)	0.17	26	0.140	0.156	90%
1 (20-30 mm)	(15)	0.17	26	0.145	0.162	89%
1 (20-30 mm)	(15)	0.16	26	0.124	0.137	91%
<b>Means:</b>				<b>0.135</b>	<b>0.149</b>	<b>90%</b>
95% Conf. Intervals:				$\pm 0.015$	$\pm 0.019$	$\pm 2\%$
2 (31-40 mm)	(4)	0.40	34	0.146	0.169	86%
2 (31-40 mm)	(4)	0.38	34	0.137	0.176	78%
2 (31-40 mm)	(4)	0.44	34	0.140	0.179	78%
2 (31-40 mm)	(4)	0.43	34	0.152	0.177	86%
<b>Means:</b>				<b>0.143</b>	<b>0.175</b>	<b>82%</b>
95% Conf. Intervals:				$\pm 0.011$	$\pm 0.007$	$\pm 7\%$
3 (41-50 mm)	3	1.10	47	<b>Means:</b>	<b>0.234</b>	<b>0.273</b>
				SD:	0.033	0.042
				95% Conf. Intervals:	$\pm 0.082$	$\pm 0.105$
						86%
						4%
						$\pm 9\%$
<u>Gordon Slough</u>						
1 (20-30 mm)	(15)	0.19	26	0.059	0.062	95%
1 (20-30 mm)	(15)	0.18	26	0.057	0.059	97%
1 (20-30 mm)	(15)	0.19	26	0.063	0.065	96%
1 (20-30 mm)	(15)	0.18	26	0.051	0.055	93%
<b>Means:</b>				<b>0.058</b>	<b>0.060</b>	<b>95%</b>
95% Conf. Intervals:				$\pm 0.008$	$\pm 0.007$	$\pm 3\%$
2 (31-40 mm)	(5)	0.37	34	0.078	0.094	83%
2 (31-40 mm)	(5)	0.42	34	0.061	0.073	83%
2 (31-40 mm)	(5)	0.38	34	0.056	0.066	85%
2 (31-40 mm)	(5)	0.35	34	0.066	0.076	87%
<b>Means:</b>				<b>0.065</b>	<b>0.077</b>	<b>85%</b>
95% Conf. Intervals:				$\pm 0.015$	$\pm 0.019$	$\pm 3\%$
3 (41-50 mm)	3	0.88	48	<b>Means:</b>	<b>0.126</b>	<b>0.148</b>
				SD:	0.017	0.028
				95% Conf. Intervals:	$\pm 0.043$	$\pm 0.070$
						86%
						5%
						$\pm 12\%$



**Table 6. (Continued).**

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>		<u>µg/g METHYL Hg</u>	<u>µg/g TOTAL Hg</u>	<u>% Methyl Hg</u>	
		<u>wt (g)</u>	<u>lgth (mm)</u>	<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>	
<u>Nature Preserve Wetlands</u>							
1 (20-30 mm)	(10)	0.18	26	0.108	0.111	97%	
1 (20-30 mm)	(10)	0.18	26	0.133	0.144	93%	
1 (20-30 mm)	(10)	0.18	26	0.123	0.122	101%	
1 (20-30 mm)	(10)	0.19	26	0.122	0.127	96%	
				<b>Means:</b>	<b>0.122</b>	<b>0.126</b>	<b>97%</b>
				<i>95% Conf. Intervals:</i>	± 0.016	± 0.021	±5%
2 (31-40 mm)	(5)	0.42	34	0.096	0.118	81%	
2 (31-40 mm)	(5)	0.41	34	0.110	0.136	81%	
2 (31-40 mm)	(5)	0.38	34	0.097	0.116	83%	
2 (31-40 mm)	(5)	0.44	34	0.124	0.147	84%	
				<b>Means:</b>	<b>0.107</b>	<b>0.129</b>	<b>82%</b>
				<i>95% Conf. Intervals:</i>	± 0.021	± 0.024	±3%
3 (41-50 mm)	3	0.92	48	<b>Means:</b>	<b>0.115</b>	<b>0.135</b>	<b>86%</b>
				<i>SD:</i>	0.008	0.014	3%
				<i>95% Conf. Intervals:</i>	± 0.020	± 0.035	±8%
<u>Cache Creek Below Gordon Slough</u>							
1 (20-30 mm)	(15)	0.17	26	0.079	0.079	100%	
1 (20-30 mm)	(15)	0.18	26	0.066	0.069	96%	
1 (20-30 mm)	(15)	0.19	26	0.069	0.071	98%	
1 (20-30 mm)	(15)	0.19	26	0.069	0.073	95%	
				<b>Means:</b>	<b>0.071</b>	<b>0.073</b>	<b>97%</b>
				<i>95% Conf. Intervals:</i>	± 0.009	± 0.007	±4%
2 (31-40 mm)	(5)	0.48	35	0.067	0.079	85%	
2 (31-40 mm)	(5)	0.42	35	0.071	0.095	74%	
2 (31-40 mm)	(5)	0.44	35	0.085	0.105	81%	
2 (31-40 mm)	(5)	0.45	35	0.068	0.086	79%	
				<b>Means:</b>	<b>0.073</b>	<b>0.091</b>	<b>80%</b>
				<i>95% Conf. Intervals:</i>	± 0.014	± 0.018	±7%
3 (41-50 mm)	4	1.04	47	<b>Means:</b>	<b>0.124</b>	<b>0.135</b>	<b>92%</b>
				<i>SD:</i>	0.019	0.024	3%
				<i>95% Conf. Intervals:</i>	± 0.031	± 0.038	±4%

**Table 7. November 2002 Fish Mercury:**  
**Other fish from Nature Preserve Wetlands**  
(Mean values of multiple (n) whole body individual analyses)  
(wet weight = fresh weight,  $\mu\text{g}$  mercury per gram = parts per million)

<u>Size Class</u>	<u>n</u>	<u>Mean Fish Size</u>		<u><math>\mu\text{g/g}</math> METHYL Hg</u>	<u><math>\mu\text{g/g}</math> TOTAL Hg</u>	<u>% Methyl Hg</u>
		<u>wt (g)</u>	<u>lgth (mm)</u>	<u>in WET Sample</u>	<u>in WET Sample</u>	<u>of Total Hg</u>
<u>INLAND SILVERSIDE (<i>Menidia beryllina</i>)</u>						
<b>2</b> (51-65 mm)	8	0.96	60	<b>Means:</b> <b>0.167</b>	<b>0.185</b>	<b>90%</b>
				SD: 0.016	0.014	6%
				95% CI $\pm 0.013$	$\pm 0.012$	$\pm 5\%$
<b>3</b> (66-80 mm)	3	1.71	71	<b>Means:</b> <b>0.236</b>	<b>0.262</b>	<b>91%</b>
				SD: 0.068	0.092	7%
				95% CI $\pm 0.170$	$\pm 0.229$	$\pm 18\%$
<u>BLUEGILL (<i>Lepomis macrochirus</i>)</u>						
<b>1</b> (31-50 mm)	6	1.43	48	<b>Means:</b> <b>0.123</b>	<b>0.135</b>	<b>92%</b>
				SD: 0.018	0.023	5%
				95% Conf. Intervals: $\pm 0.019$	$\pm 0.025$	$\pm 6\%$
<b>2</b> (51-80 mm)	5	6.44	75	<b>Means:</b> <b>0.128</b>	<b>0.126</b>	<b>102%</b>
				SD: 0.026	0.025	7%
				95% Conf. Intervals: $\pm 0.032$	$\pm 0.031$	$\pm 9\%$
<u>BLACK CRAPPIE (<i>Pomoxis nigromaculatus</i>)</u>						
<b>3</b> (70-100 mm)	6	6.85	85	<b>Means:</b> <b>0.138</b>	<b>0.151</b>	<b>91%</b>
				SD: 0.061	0.064	5%
				95% Conf. Intervals: $\pm 0.064$	$\pm 0.067$	$\pm 5\%$

**Table 8. November 2002 Invertebrate Mercury:  
Primary Indicator Samples From Cache Creek Transect;  
HYDROPSYCHIDAE (Caddisfly Larvae, Omnivores).**  
*(composite samples of multiple (n) whole individuals)*  
*(WET weight  $\mu\text{g}$  mercury per gram = parts per million)*

<u>(n)</u>	<u>Size</u> <u>(mean mm)</u>	<u><math>\mu\text{g/g}</math> METHYL Hg</u> <u>in WET Sample</u>	<u><math>\mu\text{g/g}</math> TOTAL Hg</u> <u>in WET Sample</u>	<u>% Methyl Hg</u> <u>of Total Hg</u>
<u>Cache Creek Above Preserve</u>				
(70)	12	0.038	0.049	78%
(70)	12	0.037	0.047	79%
(70)	12	0.037	0.048	77%
(70)	12	<u>0.038</u>	<u>0.046</u>	<u>83%</u>
	<b>Means:</b>	<b>0.038</b>	<b>0.047</b>	<b>79%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.001$	$\pm 0.002$	$\pm 5\%$
<u>Cache Creek Between Nature Preserve and Gordon Slough</u>				
(70)	12	0.031	0.042	74%
(70)	12	0.030	0.042	72%
(70)	12	0.030	0.041	73%
(70)	12	<u>0.030</u>	<u>0.042</u>	<u>72%</u>
	<b>Means:</b>	<b>0.030</b>	<b>0.042</b>	<b>73%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.001$	$\pm 0.001$	$\pm 2\%$
<u>Cache Creek Below Gordon Slough</u>				
(40)	12	0.018	0.027	67%
(40)	12	0.017	0.025	69%
(40)	12	0.017	0.025	68%
(40)	12	<u>0.018</u>	<u>0.025</u>	<u>71%</u>
	<b>Means:</b>	<b>0.018</b>	<b>0.026</b>	<b>69%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.001$	$\pm 0.002$	$\pm 3\%$

**Table 9. November 2002 Invertebrate Mercury:  
Primary Indicator Samples From Cache Creek Transect;  
CALOPTERYGIDAE (Damselfly Nymphs, Omnivorous Type).  
(composite samples of multiple (n) whole individuals)  
(WET weight  $\mu\text{g}$  mercury per gram = parts per million)**

(n)	Size (mean mm)	$\mu\text{g/g}$ METHYL Hg in WET Sample	$\mu\text{g/g}$ TOTAL Hg in WET Sample	% Methyl Hg of Total Hg
<u>Cache Creek Above Preserve</u>				
(16)	23	0.031	0.035	88%
(16)	23	0.027	0.030	91%
(16)	23	0.024	0.027	91%
(16)	23	<u>0.027</u>	<u>0.029</u>	<u>91%</u>
	<b>Means:</b>	<b>0.027</b>	<b>0.030</b>	<b>90%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.005$	$\pm 0.006$	$\pm 2\%$
<u>Cache Creek Between Nature Preserve and Gordon Slough</u>				
(22)	22	0.024	0.027	88%
(22)	22	0.023	0.026	90%
(22)	22	0.025	0.027	91%
(22)	22	<u>0.024</u>	<u>0.025</u>	<u>93%</u>
	<b>Means:</b>	<b>0.024</b>	<b>0.026</b>	<b>91%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.001$	$\pm 0.001$	$\pm 3\%$
<u>Cache Creek Below Gordon Slough</u>				
(28)	23	0.011	0.012	88%
(28)	23	0.010	0.012	88%
(28)	23	0.010	0.012	82%
(28)	23	<u>0.010</u>	<u>0.011</u>	<u>94%</u>
	<b>Means:</b>	<b>0.010</b>	<b>0.012</b>	<b>88%</b>
	<i>95% Conf. Intervals:</i>	$\pm 0.001$	$\pm 0.001$	$\pm 8\%$

**Table 10. November 2002 Invertebrate Mercury:  
Additional Samples from Cache Creek Sites.**  
*(composite samples of multiple (n) whole individuals)*  
*(WET weight  $\mu\text{g}$  mercury per gram = parts per million)*

<u>Insect Family</u>	<u>Description</u>	<u>Trophic Level</u>	<u>(n)</u>	<u>Size (mm)</u>	<u>Wet ppm Hg</u>		<u>Percent Methyl</u>
					<u>METHYL</u>	<u>TOTAL</u>	
<u>Cache Creek Above Preserve</u>							
Naucoridae	water bug	lg predator	(18)	10	0.095	0.112	85%
			(18)	10	0.101	0.120	84%
			(18)	10	0.084	0.106	80%
			(18)	10	<u>0.099</u>	<u>0.106</u>	<u>93%</u>
<b>Means:</b>					<b>0.095</b>	<b>0.111</b>	<b>85%</b>
<i>95% Conf. Intervals:</i>					$\pm 0.012$	$\pm 0.011$	$\pm 9\%$
<u>Cache Creek Between Nature Preserve and Gordon Slough</u>							
Coenagrionidae	damsfly nymph	predator	(15)	17	0.080	0.084	96%
Naucoridae	water bug	lg predator	(9)	10	0.123	0.136	90%
			(9)	10	0.097	0.121	80%
			(9)	10	0.115	0.131	88%
			(9)	10	<u>0.104</u>	<u>0.123</u>	<u>84%</u>
<b>Means:</b>					<b>0.110</b>	<b>0.128</b>	<b>86%</b>
<i>95% Conf. Intervals:</i>					$\pm 0.019$	$\pm 0.011$	$\pm 7\%$
<u>Cache Creek Below Gordon Slough</u>							
Coenagrionidae	damsfly nymph	predator	(8)	15	0.043	0.045	96%
Naucoridae	water bug	lg predator	(21)	11	0.047	0.048	97%
			(21)	11	0.041	0.048	84%
			(21)	11	0.036	0.040	89%
			(21)	11	<u>0.040</u>	<u>0.048</u>	<u>84%</u>
<b>Means:</b>					<b>0.041</b>	<b>0.046</b>	<b>89%</b>
<i>95% Conf. Intervals:</i>					$\pm 0.007$	$\pm 0.006$	$\pm 10\%$

**Table 11. Winter 2003 Fish Mercury:  
Nature Preserve Wetlands Only**  
(Mean values of multiple (n) whole body individual analyses)  
(Gambusia SC 1&2 are composite samples of (n) whole individuals)  
(wet weight = fresh weight, µg mercury per gram = parts per million)

Size Class	n	Mean Fish Size			µg/g METHYL Hg	µg/g TOTAL Hg	% Methyl Hg
		wt (g)	lgth (mm)		in WET Sample	in WET Sample	of Total Hg
<u>FATHEAD MINNOW (<i>Pimephales promelas</i> )</u>							
2 (36-50 mm)	6	0.86	47	<b>Means:</b>	<b>0.083</b>	<b>0.091</b>	<b>91%</b>
				<i>SD:</i>	0.018	0.016	5%
				<i>95% Conf. Intervals:</i>	± 0.019	± 0.017	±5%
3 (51-70 mm)	6	2.26	59	<b>Means:</b>	<b>0.066</b>	<b>0.075</b>	<b>87%</b>
				<i>SD:</i>	0.017	0.018	2%
				<i>95% Conf. Intervals:</i>	± 0.018	± 0.019	±2%
<u>RED SHINER (<i>Notropis lutrensis</i> )</u>							
2 (36-50 mm)	1	1.30	52		<b>0.165</b>	<b>0.172</b>	<b>96%</b>
3 (51-70 mm)	1	2.59	67		<b>0.158</b>	<b>0.169</b>	<b>94%</b>
<u>GREEN SUNFISH (<i>Lepomis cyanellus</i> )</u>							
1 (25-50 mm)	6	1.26	43	<b>Means:</b>	<b>0.140</b>	<b>0.141</b>	<b>99%</b>
				<i>SD:</i>	0.011	0.015	4%
				<i>95% Conf. Intervals:</i>	± 0.012	± 0.016	±4%
2 (51-80 mm)	3	3.56	61	<b>Means:</b>	<b>0.158</b>	<b>0.170</b>	<b>93%</b>
				<i>SD:</i>	0.021	0.019	2%
				<i>95% Conf. Intervals:</i>	± 0.051	± 0.047	±6%

(continued)

Table 11. (Continued).

Size Class	n	Mean Fish Size		$\mu\text{g/g}$ METHYL Hg	$\mu\text{g/g}$ TOTAL Hg	% Methyl Hg	
		wt (g)	lgth (mm)	in WET Sample	in WET Sample	of Total Hg	
<u>MOSQUITOFISH (<i>Gambusia affinis</i> )</u>							
1 (20-30 mm)	(15)	0.16	24	0.120	0.123	98%	
1 (20-30 mm)	(15)	0.19	24	0.110	0.114	97%	
1 (20-30 mm)	(15)	0.17	24	0.113	0.118	96%	
1 (20-30 mm)	(15)	0.18	24	0.112	0.118	95%	
				<b>Means:</b>	<b>0.114</b>	<b>0.118</b>	<b>96%</b>
				95% Conf. Intervals:	$\pm 0.007$	$\pm 0.006$	$\pm 2\%$
2 (31-40 mm)	(4)	0.76	44	0.125	0.151	83%	
2 (31-40 mm)	(4)	0.88	50	0.101	0.117	86%	
2 (31-40 mm)	(4)	1.00	50	0.122	0.135	90%	
				<b>Means:</b>	<b>0.116</b>	<b>0.134</b>	<b>86%</b>
				95% Conf. Intervals:	$\pm 0.033$	$\pm 0.042$	$\pm 9\%$
3 (41-50 mm)	3	0.88	48	<b>Means:</b>	<b>0.126</b>	<b>0.148</b>	<b>86%</b>
				SD:	0.017	0.028	5%
				95% Conf. Intervals:	$\pm 0.043$	$\pm 0.070$	$\pm 12\%$
<u>INLAND SILVERSIDE (<i>Menidia beryllina</i> )</u>							
3 (66-80 mm)	2	1.57	71	<b>Means:</b>	<b>0.217</b>	<b>0.217</b>	<b>100%</b>