Understanding the decline of deepwater sensitive species in Lake Tahoe: What is responsible, eutrophication or species invasions?

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Introduction

A comparison of historical and contemporary benthic surveys has shown a large decline in bottom invertebrate and macrophyte occurrence in the deepwater environment of Lake Tahoe (Caires et al. 2013). The decline in macrophytes and invertebrates may be directly or indirectly related to reductions in water clarity over the past 4 decades and invasive species introductions. Two of Lake Tahoe’s unique bottom species (the deepwater stonefly *Capnia lacustra* and the blind amphipod *Stygobromus* spp.), found nowhere else in the world, are at high risk given their significant reduction since the 1960s. We examine the spatial distribution of deepwater special status plants (*Chara* and moss) and invertebrates (*C. lacustra* and *Stygobromus* spp.). In doing so, we also gather information about the biology and ecology of these taxa. Specifically, we provide information about the life cycles of special status plants and invertebrates and feeding strategies of endemic invertebrates. We also measure the photosynthetic rates of *Chara*, a native deepwater macroalgae, at varying light conditions and relate observed photosynthetic responses to subsurface irradiance data from Lake Tahoe in order to understand the depths at which light is limiting for *Chara*. We describe important relationships between the distribution of native plants and endemic invertebrates and physical characteristics of the lake. Monitoring of relatively long-lived organisms such as aquatic plants and invertebrates provides an important biological indicator of the overall health of the system, one that has received little attention in Lake Tahoe.
Spatial Extent of Special Status Plants and Invertebrates

**Objective 1.** Increase our understanding of special status aquatic plant (stoneworts, liverworts, and mosses) and invertebrate (Tahoe stonefly and blind amphipod) communities in deepwater “hotspot” areas of Lake Tahoe and relate the distribution of these communities to depth, subsurface irradiance, substrate type, and availability of organic matter in sediments.

In March, May, and July 2013, we collected samples with a standard Ponar grab from the two areas from which endemic blind amphipods (*Stygobromus* spp.) and deepwater Tahoe stoneflies (*Capnia lacustra*) had been previously collected. We have named these areas the Camp Richardson and McKinney Bay sites. The Camp Richardson collections yielded an abundance of the native deepwater macroalgae, *Chara* and *C. lacustra* specimens, whereas it was difficult to find any endemic species at the McKinney Bay site. After exhaustively sampling the McKinney Bay area through July and finding very few individuals of *C. lacustra* and *Stygobromus* spp., we decided to discontinue sampling at McKinney Bay. Instead, we began sampling a small area of *Chara* and native moss in South Lake Tahoe, known as the South Shore Mound. The South Shore Mound collections yielded a greater abundance of *Stygobromus* spp. than had been found anywhere in the lake in recent years. Divers delineated the boundary of the Camp Richardson *Chara* bed and the South Shore Mound moss bed so that we could determine the extent of *C. lacustra* and *Stygobromus* spp. habitat in each area. At the deeper McKinney Bay site, the distribution of *C. lacustra* and *Stygobromus* spp. was determined from the contents of benthic samples collected with the Ponar.

Upon collection of a Ponar sample, plant material was removed and placed on ice for identification and determination of dry weight. Once plants were removed, samples were sieved through a 250 μm mesh bucket sieve. Material retained in the sieve was picked for live invertebrates. Relative percentages of different substrates (plant, silt, sand, clay, gravel, cobble, and organic matter) were estimated for each sample that was collected.
Current vs. Historical Extent of Endemic Communities

The three surveyed sites (Camp Richardson, McKinney Bay, and the South Shore Mound) are the only known sites in which *C. lacustra* and *Stygobromus* spp. occur in Lake Tahoe today (Fig. 1). Historically, *C. lacustra* and *Stygobromus* spp. occurred in southern and western areas of the lake where it is found currently, as well as eastern and northern parts of the lake, where it has not been found in recent collections (Fig. 1). Native mosses or *Chara* have been found at all of the sites from which *C. lacustra* and *Stygobromus* spp. have been collected recently. The Camp Richardson site is a dense bed of *Chara* (Fig. 2A) mixed with some moss. It occurs between a 30-35 m depth, and is approximately 0.105 km² in extent (Fig. 1). The McKinney Bay site is an area of small patches of moss between 55-85 m depth, covering a 0.011 km² area (Fig. 1). The South Shore Mound site is a small area at a depth of 50-55 m, surrounded on all sides by depths of >100 m. It is a mix of moss and *Chara* sp. (Figs. 2B, 2C) and is approximately 0.002 km² in extent (Fig. 1).

The greatest densities of *C. lacustra* occur at the Camp Richardson site, whereas the greatest densities of *Stygobromus* spp. are found at the South Shore Mound site (Table 1). Even when compared to historical densities from 1962-1963 in samples containing plants, the greatest densities of *C. lacustra* were found at the Camp Richardson site in 2013-2014 samples (Table 1). However, all historical sites contained greater densities of *C. lacustra* than those from recent collections at McKinney Bay and the South Shore Mound (Table 1). Densities of *Stygobromus* spp. in recent collections from the South Shore Mound are comparable to densities from historical collections in South Lake Tahoe in samples that had plants in them. Historical collections from Cave Rock contained the highest densities of *Stygobromus* spp. in samples that contained plants (Table 1). Historical densities of *Stygobromus* spp. from deeper samples that did
not contain plants were higher than any of the average *Stygobromus* spp. densities reported in Table 1 (see Caires et al. 2013). On average, both *C. lacustra* and *Stygobromus* spp. were found at deeper locations in the lake in the 1960s than they were in recent collections (Table 1).

**Plant-Invertebrate Associations**

*C. lacustra* was abundant in our samples that were dominated by *Chara* at the Camp Richardson site (Fig. 3). It has long been known that *C. lacustra* is associated with *Chara* beds in the lake (Jewett 1963, Frantz and Cordone 1996), and our findings support this habitat association. In addition to *C. lacustra* using *Chara* as habitat, it is also likely dependent on *Chara* for food, which will be discussed further in the following section on the biology and ecology of special status organisms. *Stygobromus* spp. was abundant in samples that were dominated by moss or peat (partially decomposed moss), suggesting that it may be using decomposed moss as a food source (Fig. 3). Most amphipods depend on the bacteria and fungi associated with decomposing organic matter for food (Covich and Thorp 2001) and thus the moss beds remaining in the lake would provide important hotspot areas of detritus for *Stygobromus* spp. colonization. The South Shore Mound is currently the only known such area and Camp Richardson is the only known area of high concentrations of *Chara* and *C. lacustra* assemblages. While it is clear that there is a strong association between *Chara* and *C. lacustra*, such a relationship between moss and *Stygobromus* spp. is not as evident. Historically, *Stygobromus* spp. occupied deeper areas without moss, suggesting that moss is not a limiting factor for its success in Lake Tahoe. Instead, it is possible that the combination of non-native mysid introductions and reduced organic matter due to fewer plant beds in the lake may be responsible for the blind amphipod’s severe population reduction since the 1960s.
**Substrate-Habitat Associations**

At the Camp Richardson site, *Chara* biomass was highest in samples containing silt as a dominant substrate, whereas moss biomass was highest in samples dominated by clay (Fig. 4A). Because the plant bed is relatively shallow (~30-40 m depth) at Camp Richardson, it is not likely that light is limiting at this site. Instead, growth of plants on the shallow end of the bed is probably limited by substrate type. Substrate around the Camp Richardson area in shallow water (<30 m) is dominated by sand, which appears to be a less suitable substrate than silt. At the deep end of the Camp Richardson plant bed, the morphology of the lakebed seems to limit plant growth. Divers described the north end of the bed as a steep drop off into deep water, in which they could not see the bottom. The Camp Richardson plant bed occurs on the shelf in that area between 30 and 40 m, which is dominated by silt substrate. The conditions that occur in this area must be ideal for growth of *Chara*, which forms a dense, nearly continuous bed for approximately 1/3 of a square mile.

At the South Shore Mound site, moss occurred in samples containing all substrate types, whereas *Chara* occurred mainly in samples containing silt (Fig. 4B). Moss dominates the South Shore Mound and does not appear to be limited by light at the site. *Chara* biomass per unit area is much less at the South Shore Mound than it is at Camp Richardson and it is less dense and displays less branching. It is possible that it is a different species, or a different ecotype of the same species adjusted to lower light intensity at the South Shore Mound. Both *Chara* and moss at the South Shore Mound site are limited to the small mound area, which drops off to deep water on all sides.
Fig. 1. Map showing the only 3 areas of Lake Tahoe in which *Capnia lacustra* and *Stygobromus* spp. have been found during recent sampling efforts (2013-2014) and a lakewide survey in 2008-09 (Caires et al. 2013). Arrows show transects in which *C. lacustra* and *Stygobromus* spp. were found in historic (1962-1963) surveys (Frantz and Cordone 1996). Bathymetric contour lines are given at 100 m intervals.
Fig. 2. Photographs of A) deepwater plant bed at Camp Richardson composed mostly of *Chara*, B) moss at the South Shore Mound site, and C) *Chara* at the South Shore Mound site. Photographs were taken by divers from New Millennium Dive Expeditions.
Table 1. Historical (1962-1963) vs. present (2013-2014) mean (±1 SE) densities (no./m$^2$) of *Capnia lacustra* and *Stygobromus* spp. in benthic samples containing plants (*Chara* sp. and moss) collected from various sites around Lake Tahoe (shown in Fig. 1). Mean (±SE) depths (m) of sample collection at each site are also given.

<table>
<thead>
<tr>
<th>Site description</th>
<th>n</th>
<th>C. lacustra density</th>
<th>Stygobromus density</th>
<th>Depth</th>
<th>Dominant plant type</th>
</tr>
</thead>
<tbody>
<tr>
<td>1962-1963</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cave Rock</td>
<td>24</td>
<td>100.5±32.8</td>
<td>638.6±220.8</td>
<td>83.6±4.5</td>
<td>Moss and <em>Chara</em></td>
</tr>
<tr>
<td>McKinney Bay</td>
<td>14</td>
<td>83.0±51.4</td>
<td>12.3±7.0</td>
<td>70.9±4.5</td>
<td>Moss and <em>Chara</em></td>
</tr>
<tr>
<td>South Tahoe</td>
<td>28</td>
<td>56.9±18.9</td>
<td>276.8±97.3</td>
<td>63.3±5.6</td>
<td>Moss and <em>Chara</em></td>
</tr>
<tr>
<td>Tahoe City</td>
<td>3</td>
<td>28.7±28.7</td>
<td>43.1±43.1</td>
<td>91.4±14.0</td>
<td>Moss and <em>Chara</em></td>
</tr>
<tr>
<td>2013-2014</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Camp Richardson</td>
<td>63</td>
<td>291.8±45.8</td>
<td>2.1±1.1</td>
<td>34.1±0.5</td>
<td><em>Chara</em></td>
</tr>
<tr>
<td>McKinney Bay</td>
<td>8</td>
<td>0.0±0.0</td>
<td>2.4±2.4</td>
<td>71.7±3.1</td>
<td>Moss</td>
</tr>
<tr>
<td>South Shore Mound</td>
<td>28</td>
<td>13.7±5.2</td>
<td>186.1±60.6</td>
<td>51.6±0.5</td>
<td>Moss and <em>Chara</em></td>
</tr>
</tbody>
</table>
Fig. 3. Mean (±SE) density of *Capnia lacustra* and *Stygbromus* spp. shown for each type of substrate or macrophyte that dominated the sample. Graphs are shown for the three sampled sites, A) Camp Richardson, B) McKinney Bay, and C) South Shore Mound.
Fig. 4. Dry weight (mean±SE g/m$^2$) of *Chara* and moss in collected from 19 Jun 13 to 7 May 14 at the A) Camp Richardson and B) South Shore Mound site in Lake Tahoe. Differences in dry weight by dominant substrate type are shown.

---

**A) Camp Richardson**

- **Chara**
  - Silt: $n = 21$
  - Sand: $n = 18$
  - Silt and Sand: $n = 14$
- **Moss**
  - Clay: $n = 4$
  - Peat: $n = 0$

**B) South Shore Mound**

- **Chara**
  - Silt: $n = 6$
  - Sand: $n = 4$
  - Silt and Sand: $n = 7$
- **Moss**
  - Clay: $n = 5$
  - Peat: $n = 3$
Objective 2. Increase our understanding of the biology and ecology of special status deepwater plants and invertebrates through seasonal tracking of populations, developmental state, and diet using stable isotopes.

The Camp Richardson and South Shore Mound sites were chosen for temporal analysis because of the large number of endemic invertebrates collected from the sites. Benthic grabs were collected with a Petite Ponar every other month from March 2013-March 2014 at Camp Richardson and July 2013-March 2014 at the South Shore Mound Site. Samples were processed as described for plant bed spatial extent determination. Temperatures just above the surface of the plant bed at the Camp Richardson site were measured using a YSI model 85 handheld probe on several occasions (June, August, September, and October 2013; and January, March, and April 2014).

Chara and Moss Biology and Ecology

Plant material was separated from other material in each sample for determination of dry weight and chlorophyll $a$ and $b$ concentrations. Dried plant material was also retained for stable isotope analysis. There did not appear to be seasonal variability in Chara or moss biomass at the Camp Richardson or South Shore Mound sites (Figs. 5A, 5B). The Camp Richardson site had high biomass of Chara and little moss throughout the year. Conversely, the South Shore Mound site had high biomass of moss with little Chara throughout the year.

We also investigated growth rates of Chara by placing 10 potted plants on the bottom of the lake at the Camp Richardson site on 27 June 2013 and retrieving them on 8 September 2013. When plants were collected in September, we found that 7 of 10 of the plants had been broken.
off, thus not allowing us to determine growth rates. For the 3 plants that remained in tact, growth was 111.1±25.6 mg for the duration of the 78 day incubation in the lake.

Chlorophyll content of Chara varied by season at the Camp Richardson site, where chlorophyll a was highest in the early summer and chlorophyll b was highest in the winter (Figs. 6A, 6C), although there were no significant differences among the total (a + b) chlorophyll content among the seasons. Too few moss samples were collected at the Camp Richardson site to evaluate a trend; however, chlorophyll a and b content of moss was similar at the site in early summer (Figs. 6A, 6C). At the South Shore Mound site, few chlorophyll samples were run on Chara and thus no seasonal trends can be evaluated. Chlorophyll a and b values were within the same range for Chara and moss at the South Shore Mound as they were at Camp Richardson (Figs. 6B, 6D). At the Camp Richardson site, the ratio of chl a/b in Chara decreased from early summer to winter, and then increased again in the spring (Fig. 7A). Moss also showed a high chl a/b ratio in the early summer and low ratio in the winter, though sample sizes were small (Fig. 7A). At the South Shore Mound site, Chara had relatively high chl a/b ratios in the winter and spring (extremely small sample sizes), and moss had a low chl a/b ratio in the fall (Fig. 7B). Decreases in chl a/b ratios can show that plants are producing relatively more pigment (chl b) for their light harvesting complexes (Andrews et al. 1984), and are thus allocating more energy towards obtaining light. The seasonal differences in relative concentrations of pigments in the Camp Richardson Chara population and the South Shore Mound moss population, therefore, may reflect seasonal decreases in light availability due to decreases in day length and light intensity, as well as increased shading from epiphytic growth that appears to increase in the late summer and fall.
Fig. 5. Dry weight (mean±SE g/m$^2$) of Chara and moss in collected from 19 Jun 13 to 7 May 14 at the A) Camp Richardson and B) South Shore Mound site in Lake Tahoe. Differences in dry weight by season are shown.
Fig. 6. Chl a (mean±SE mg/gDW) of Chara and moss at A) Camp Richardson and B) South Shore Mound, and chl b (mean±SE mg/gDW) of Chara and moss at C) Camp Richardson and D) South Shore Mound in Lake Tahoe.
Fig. 7. The ratio of chl \(a/\text{chl } b\) for Chara and moss at A) Camp Richardson and B) South Shore Mound.
**Capnia lacustra Biology and Ecology**

All *C. lacustra* individuals were enumerated and measured to the nearest 0.1 mm using an ocular micrometer. The number of individuals collected at each site allowed only for life history analysis to be conducted on *C. lacustra* from Camp Richardson. Individuals of *C. lacustra* ≥ 3 mm were sexed, while others were considered immature. All eggs found inside of *C. lacustra* were enumerated.

Female *C. lacustra* containing eggs from Camp Richardson collections in November 2013, March 2014, and May 2014 were kept alive in containers of water in an incubator for monitoring. Water temperatures were increased from 6 to 9°C over the course of incubation in the spring and were maintained at approximately 10°C in the fall. A small piece of *Chara* and a small piece of moss were provided for each female in each container. The incubator was placed on a light/dark cycle simulating the natural light/dark cycle for the time of year in which individuals were held in the laboratory. Water was refreshed every other day and *Chara* and moss were replaced every week. Females in incubators were checked twice daily in November/December and every other day from March-June for condition and egg movement and development. Pictures were taken of females whose eggs appeared mature. If hatched eggs were found inside of a female, observations of the female were made intermittently under the microscope over the course of several hours.

Length frequency diagrams indicated the presence of 2 asynchronous cohorts of *C. lacustra* with developmental times of approximately one year at the Camp Richardson site (Fig. 8). Eggs observed within collected females confirmed the presence of 2 cohorts, with egg development occurring in the spring (March-May) and fall (November) (Fig. 8).
Sex ratios in the Camp Richardson population varied throughout the year (Fig. 9). During months in which females were found with eggs (May and November 2013, and March and May 2014), few males were found and mature females were the dominant specimens (Fig. 9). Four months after females were found with eggs, immature specimens were abundant in our collections (Fig. 9).

Eggs and developing nymphs were readily apparent in many of the females from our spring and fall collections. Eggs were distributed throughout the abdomen, thorax, and occasionally the head of females (Fig. 10A). Gravid females from collections on 6 November 2013 that were kept in the incubator in our laboratory were observed with hatched nymphs in their containers between the period of 22 November-16 December 2013. Prior to the hatching of nymphs, eggs within the females exhibited movement and were well developed, with nymphal features evident (Fig. 10B). On several occasions, hatched nymphs were observed inside of live females, attempting to move towards the posterior end of the abdomen (Fig. 10C). Females appeared agitated prior to hatching of nymphs within their bodies, and lethargic during hatching. A large incubation pouch, spanning the entire length of the female’s abdomen and thorax, pulsed as eggs and hatched larvae were pushed toward the posterior section of the abdomen. On one occasion, live birth was observed in a female (Fig. 10D), but the process was slow and the female died after several hours, possibly due to the manipulation and temperature changes during microscope observation. Undisturbed, birthing of the nymphs took < 7 h, as determined from daily checks of the incubator. On several occasions, the final nymph to exit the abdomen became caught halfway out of the female and would stay alive this way for several days. Females normally died soon after giving birth (< 24 h), but sometimes lived up to a week after birth.
Gravid females from collections on 19 March and 7 May 2014 that were kept alive in the incubator were observed with unhatched eggs in their containers between the period of 26 May-26 June 2014 (Table 2). No hatching of nymphs within females was observed during this time period. Two of the 3 females from the March collection died before depositing eggs. All of the eggs deposited by females hatched between 3 and 21 d after being dropped. Eyespot development and movement of eggs occurred after egg deposition rather than within female body cavities. Females normally died soon after depositing eggs (< 24 h), but occasionally lived up to 4 d after egg release.

Gravid females from spring and fall collections had an average of 38-40 eggs (Table 2), and the average number of eggs in gravid females did not vary significantly among months (1-way ANOVA, $F_{3,39} = 0.11, p = 0.95$). Gravid females in November 2013 had a significantly greater body length than did gravid females in May 2013 (1-way ANOVA, $F_{2,35} = 6.26, p < 0.01$; Table 2).
Fig. 8. Size frequency distribution of *Capnia lacustra* at the Camp Richardson site during the study period (March 2013-May 2014). Males are represented on the left side of y-axes and females are shown on the right. Individuals < 3.0 mm were too immature for sex determination, thus the total number of individuals was divided equally between males and females. Width of horizontal bars indicates the proportional contribution of each size class and the sum of all bars in any given month is equal to 100%. Black bars represent the cohort that gave live birth between November and December and grey bars represent the cohort that deposited eggs between May and June. Arrows indicate the approximate timing of live birth/egg deposition.

Fig. 9. Number of *Capnia lacustra* females (grey), males (black), and immature nymphs (white) collected during 8 sampling periods in Lake Tahoe from March 2013-May 2014. Sampling effort differed for each collection period.
Fig. 10. Photographs of *C. lacustra* A) with eggs throughout abdomen, thorax and head, B) developing eggs, C) hatched nymph inside of adult female, and D) nymph exiting female abdomen.
Table 2. Reproductive characteristics of *Capnia lacustra* in Lake Tahoe. Mean (±1 SE) number of eggs per female and mean (±1 SE) total length (mm) of egg-bearing females from each collection period are given. Females from March and May collections are grouped as the spring reproductive cohort and females from November collections are considered the fall reproductive cohort. Data on live birth/egg deposition were gathered from a subset of the egg-bearing females from each collection period.

<table>
<thead>
<tr>
<th>Collection Date</th>
<th>No. eggs</th>
<th>Total length</th>
<th>n</th>
<th>Live birth/egg deposition</th>
<th>Period of egg live birth/egg deposition</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 May 2013</td>
<td>38±1.8</td>
<td>5.0±0.1</td>
<td>17</td>
<td>No data</td>
<td>No data</td>
</tr>
<tr>
<td>6 Nov 2013</td>
<td>39.9±3.3</td>
<td>5.7±0.2</td>
<td>17</td>
<td>Live birth</td>
<td>22 Nov-16 Dec 2013</td>
</tr>
<tr>
<td>19 Mar 2014</td>
<td>37.8±2.6</td>
<td>5.5±0.1</td>
<td>5</td>
<td>Egg deposition</td>
<td>13 Jun 2014</td>
</tr>
<tr>
<td>7 May 2014</td>
<td>37.8±9.3</td>
<td>No data</td>
<td>4</td>
<td>Egg deposition</td>
<td>26 May-26 Jun 2014</td>
</tr>
</tbody>
</table>
Our finding that *C. lacustra* develops 2 asynchronous reproductive cohorts simultaneously in Lake Tahoe further distinguishes this already-unique taxon from other stonefly taxa. Most stoneflies exhibit simple univoltine or semivoltine emergence patterns, which are dictated by temperature, photoperiod or nutrition (Hynes 1976, Anderson and Cummins 1979). A few stonefly taxa (also in the suborder Euholognatha) have displayed emergence of asynchronous cohorts in spring streams in which temperature is stable (López-Rodríguez and Tierno de Figueroa 2012, Bottová et al. 2013). *C. lacustra* lives in an environment in which temperatures are relatively constant, fluctuating by only about 6° C each year. It is possible that year-round suitable temperatures have allowed for the simultaneous development of fall and spring cohorts, with each cohort completing its life cycle in one year.

Other rare characteristics of *C. lacustra* are the development of eggs throughout the body and its ability to give birth to already-hatched nymphs. We could find no other accounts of viviparity (live birth) in Plecoptera, with the exception of a brief mention of the observation of nymphs hatched within the body of *Allocapnia vivipara* by Claassen (1924), a finding that was later discounted by Frison (1929) after he was unable to replicate the phenomenon in the laboratory. The presence of eggs throughout the abdomen, thorax, and even the occipital region of the head has also been observed in two other capniid stoneflies from Lake Baikal, *Baikaloperla elongata* and *Baikaloperla kozhovi* (Zhiltsova and Zapekina-Dulkeit 1977, Zapekina-Dulkeit and Zhiltsova 1973). These capniids display ovoviviparity, in which eggs develop within the adult female and hatch soon after oviposition. Ovoviviparity also exists in several other capniid and gripopterygid stonefly species (Clifford 1966, Hynes 1976, Michaelis 1984, Elliott 1986). Because *C. lacustra* deposited unhatched eggs in the spring and gave birth to live nymphs in the fall, it can be considered facultatively viviparous.
It is possible that *C. lacustra* can exhibit ovoviviparity or viviparity, depending on environmental conditions (i.e., food availability, temperature). It is unclear what role food availability plays in influencing *C. lacustra*’s life history characteristics. Females bearing well-developed eggs were observed to have material in their guts during monitoring in the laboratory, suggesting that food availability and quality is important for egg development within adult females. Seasonal peaks in diatom growth on *Chara* likely influence the growth and egg development of *C. lacustra*.

Temperature is also undoubtedly important in affecting the life history of *C. lacustra*. It is possible that the fall cohort always exhibits viviparity, while the spring cohort exhibits ovoviviparity. In the fall, leading up to the live birth of nymphs in November and December 2013, temperatures were the highest of all measured temperatures over the course of the study period (12.3° C in October). Warmer temperatures may have allowed for faster development of eggs, which resulted in the hatching of nymphs within female bodies. Conversely, in the spring, temperatures were the lowest of all measured temperatures (6.0° C and 6.2° C in March and April, respectively). It is possible that, because of the cooler temperatures, egg development was relatively slow and thus nymphs did not hatch within female bodies during the spring. The average body length of *C. lacustra* females with eggs in May 2013 was significantly less than the average body length of females with eggs in November 2013, suggesting that the fall cohort may have greater growth potential and thus greater egg development potential. Facultative viviparity has been documented for many species of Diptera, where seasonality and climatic conditions determine whether females deposit eggs or larvae (Meier et al. 1999).

Ovoviviparity has been thought to be a mechanism to provide eggs with a suitable environment during otherwise unfavorable environmental conditions, such as extreme cold or
drought (Hynes 1976, Michaelis 1984, Elliott 1986). *C. lacustra*, however, is not directly exposed to extreme cold or drought at the bottom of Lake Tahoe. It is possible that viviparity is a life history characteristic related to *C. lacustra*’s dependence on plants. When females deposit nymphs instead of eggs, the nymphs can immediately cling to the desired substrate (*Chara*), whereas eggs dropped on the lake bottom may be at risk of being carried away by the current. In the laboratory, newly hatched nymphs attached immediately to the stems of *Chara*, which would be advantageous in an environment in which they depend on these plants for habitat and food.

**Stygobromus spp. Biology and Ecology**

All *Stygobromus* spp. individuals were enumerated and measured to the nearest 0.1 mm using an ocular micrometer. There were only a sufficient number of *Stygobromus* spp. specimens collected from the South Shore Mound site for life history analysis. Any eggs that were encountered inside of *Stygobromus* spp. specimens were noted.

Size frequency plots of *Stygobromus* spp. did not reveal distinct size or age cohorts (Fig. 11). Instead, large and small specimens were collected during every month in which samples were collected. It should be noted that males and females were not distinguished in our samples and females are larger than males. Females with eggs were found during all collection periods except for November. In historical collections, females with eggs were also found throughout the year (May, July, August, September, October, and December; Holsinger 1974). It is likely that eggs were not found in our November collections only because of the low sample size. Therefore, it appears that reproduction and egg development occur throughout the year. This is consistent with the life history of another stygobromid species found in a groundwater system near in the Sierra Nevada (*Stygobromus sheldoni*), which are though to breed throughout the year as well.
Both *Stygbromus tahoensis* and *Stygbromus lacicolus* are present in Lake Tahoe. *S. tahoensis* was found to be much more abundant in the lake than *S. lacicolus* in the 1960s (Holsinger 1974), and in our more recent (2008-09) collections, we found 3 specimens of *S. tahoensis* and 2 potential, but damaged specimens of *S. lacicolus* in the McKinney Bay area (Caires et al. 2013). We did not distinguish between the species for this study and therefore do not know whether one or both species inhabit the South Shore Mound.
Fig. 11. Size frequency distribution of *Stygobromus* spp. at the South Shore Mound site during the study period (July 2013-March 2014). Width of horizontal bars indicates the proportional contribution of each size class and the sum of all bars in any given month is equal to 100%.
Stable Isotopes and Diet of C. lacustra and Stygobromus spp.

Tissue from C. lacustra, Chara, moss, filamentous algae, sediment, crayfish and mysids collected at the Camp Richardson site and Stygobromus spp., Chara, moss, sediment, and mysids collected from the South Shore Mound site were dried and prepared for stable isotope analysis. Isotopic trophic niche widths at two locations in Lake Tahoe were determined from $\delta^{15}$N and $\delta^{13}$C values in Program R. C. lacustra and Stygobromus spp. diets were evaluated using a dietary proportions analysis with Bayesian stable isotope mixing-models in Program R.

There was little overlap in trophic niches at the Camp Richardson (Fig. 12) and South Shore Mound sites (Fig. 13), suggesting that the taxa analyzed for isotopes do not share similar resources. According to the dietary proportions analysis, the diet of C. lacustra at the Camp Richardson site consisted mainly of Chara (53%) and algae (23%) (Fig. 14). The low sample size and thus large standard deviation of stable isotope data for Stygobromus spp. at the South Shore Mound site did not allow for dietary proportions analysis.

A side experiment was also conducted to investigate C. lacustra’s diet preference. In two incubators set to two different temperature treatments (8-10° C and 12-14° C), C. lacustra ($n = 5$ for each treatment) were placed in individual cups of lake water and were offered 4 foods (Chara, Rhizoclonium (a nearshore filamentous algae), moss, and a diatom felt collected from the nearshore). Controls were also run in which C. lacustra were not given any food. The experiment ran for 28 days and C. lacustra were weighed before and after the experiment to monitor growth. Growth was negative for all treatments except for the Rhizoclonium warm treatment and the diatom felt treatment, in which growth was positive and significantly greater than the control for both temperature treatments (Fig. 15). The results of this short-term experiment suggest that C. lacustra shows greater growth consuming diatoms (the epiphytic growth on Chara) than other
available food sources such as Chara, filamentous algae, and moss. However, the findings from the stable isotope analysis suggest that C. lacustra’s diet is made up mostly of Chara. We did not run diatoms for stable isotope analysis; however, it is possible that epiphytic diatoms were on the Chara that was run for isotope analysis and the isotopic signature of “Chara” shown in Figs. 12-14 instead represented the signature of Chara and epiphytic diatoms combined.
Fig. 12. Trophic niche model based on stable isotopes taken from benthic components of the Camp Richardson Chara bed. Overlap of polygons represent resource sharing between components. Polygons represent the total area of niches and ovals represent standard ellipse areas from $\delta^{15}$N and $\delta^{13}$C values.

Fig. 13. Trophic niche model based on stable isotopes taken from benthic components of the South Shore Mound moss bed. Overlap of polygons represent resource sharing between components. Polygons represent the total area of niches and ovals represent standard ellipse areas from $\delta^{15}$N and $\delta^{13}$C values.
Fig. 14. Dietary proportions of *C. lacustra* collected from the Camp Richardson site. Dark grey bars = 50\textsuperscript{th} percentiles, lighter grey = 75\textsuperscript{th} percentiles, and light grey = 95\textsuperscript{th} percentiles.

Fig. 15. Mean±SE *C. lacustra* growth rate shown for 4 diet treatments and a control. Rhizo. = *Rhizoclonium*, a filamentous algae. The diatom treatment was the only treatment that was significantly greater than the control, as indicated by a 2-way ANOVA (*p*=0.046; *F*=2.164) followed by a Tukey’s HSD test.
**Objective 3.** Develop a P-I curve for commonly encountered deepwater plants in Lake Tahoe and link the curve to historical light data to determine how plant communities have responded to changes in clarity and how they may respond to future changes in water clarity.

*Chara* was collected from the Camp Richardson site during August, October (2013), January, and April (2014) for development of seasonal photosynthesis-irradiance (P-I) curves. Measurements of subsurface irradiance in the field at the time of collections were taken with a Li-Cor underwater PAR sensor. *Chara* was kept in lake water at temperatures equivalent to those just above the plant bed during transportation to the laboratory. Upon arrival at the laboratory, a small amount of *Chara* (approximately 1 g each) was placed into glass BOD bottles filled with lake water. Each bottle was immediately placed in an incubator. Incubator shelves were exposed to varying light levels, and three replicate bottles were placed at each light level. Three replicates were also placed in dark bottles for respiration estimates. Because lake water was nearly saturated with oxygen, which would not allow accurate measurement of increased oxygen from photosynthesis, nitrogen gas was bubbled into the water to reduce oxygen levels. Oxygen was measured with a YSI BOD probe in each bottle before and after incubation. Bottles were incubated from 4-8 hours, or long enough to obtain a sufficient increase in oxygen for determination of photosynthetic rate.

**Seasonal Photosynthesis-Irradiance Relationships of Chara**

In August, *Chara* sp. displayed the highest maximum photosynthetic rate ($P_{\text{max}}$), which occurred at 57 µmol m$^{-2}$ s$^{-1}$ ($I_k$) (Fig. 16, Table 3), an irradiance level that occurred at a 43 m depth in Lake Tahoe during August when plants were collected. The irradiance level in which net photosynthesis was zero ($I_c$) in August was 21µmol m$^{-2}$ s$^{-1}$, which occurred at a 55 m depth at the field site at the time plants were collected for the experiment. This suggests that, in August,
*Chara* could potentially grow to a 55 m depth, but would show maximum growth at depths equal to or shallower than 43 m. In a plant bed as dense as the Camp Richardson site, plant self-shading would also affect the bed, likely reducing the maximum depth of photosynthesis (Sand-Jensen et al. 2007).

Despite warm temperatures in the field, the $P_{\text{max}}$ of *Chara* obtained in the October P-I experiment was lower than the $P_{\text{max}}$ from the other PI experiments (Figure 16, Table 3), likely due to the degraded state of the *Chara* that was collected for the experiments, which was both visually apparent and could be seen in the lower chlorophyll values from *Chara* collected for the October experiment (Table 4). $I_k$ occurred at 31 m and $I_c$ occurred at 46 m at the field site at the time plants were collected for the experiment. It is possible that *Chara* degrades seasonally in the fall, due to shading by epiphytic algal growth, which was evident on *Chara* filaments collected in the field or low availability of nutrients in the water column prior to lake mixing. $P_{\text{max}}$ values from the January and April experiments were similar to each other, as were the $I_k$ and $I_c$ values (Fig. 16, Table 3). In January, the $I_k$ and $I_c$ values in the field at the time of collection were found at 41 m and 72 m, respectively and in April $I_k$ and $I_c$ occurred at 56 and 83 m, respectively. The lower $I_k$ and $I_c$ values during colder parts of the year in which less light is available suggests that *Chara* adjusts seasonally to photosynthesize at lower light levels.

Overall, *Chara* showed maximum growth during August, and this growth rate was achieved at an irradiance value that would be obtained at 43 m or shallower. $I_c$ values in August indicate that *Chara* could survive as deep as 55 m, but at the expense of reduced photosynthetic rates. The dense *Chara* bed at Camp Richardson occurs between 30-40 m and a more sparsely colonized bed of *Chara* exists at the South Shore Mound between 50-55 m. The different morphology of the bed at the South Shore Mound (less dense with filaments spread further apart) probably
reflects the inability for Chara to grow at 50-55 m at densities that would result in self-shading. The P-I experiment data corroborate the current depth extent of known Chara beds at the bottom of Lake Tahoe. Historical data (1962-63) suggest that Chara beds existed much deeper (mainly from 45 to 90 m) than they are found today (Frantz and Cordone 1967), presumably due to greater clarity and light availability at the bottom of the lake in the 1960s.

Low-Light Adaptation Responses of Chara

In order to test the ability of Chara to adapt to lower light conditions, we conducted a low light adaptation experiment in which we exposed Chara to irradiance levels of 3 µmol m\(^{-2}\) s\(^{-1}\) for 4 weeks. After the low-light exposure period, we ran a P-I experiment to determine if Chara adjusted its \(I_k\) and \(I_c\) values (Fig. 17, Table 3). Values from this low-light experiment can be compared to values obtained during the January P-I experiment. Chara reached a similar \(P_{\text{max}}\) value in the low light adaptation experiment, while dropping its \(I_k\) and \(I_c\) values substantially (Table 3). The \(I_k\) and \(I_c\) values would have occurred at 53 m and 101 m in the field at the time of collection in January. This suggests that, in the short-term, Chara can adapt to low light conditions and maintain growth in extremely deep areas of the lake.

It remains unclear whether the reduction in depth distribution of Chara today is a result of reduced light availability or other factors (herbivory by non-native species, nutrient availability, substrate availability). It appears that Chara would be able to adapt to lower light conditions in the short term, but it cannot be assumed that this growth would be maintained over a longer period of exposure to low light conditions. We therefore cannot yet tie Chara growth potential to TMDL scenarios or changes in benthic light availability. Changes to Chara’s depth distribution, density, and P-I responses as clarity changes occur would provide further insight into long-term responses of Chara to light availability at the bottom of the lake.
Fig. 16. Photosynthesis-irradiance (P-I) relationships for Chara collected from Lake Tahoe during four seasons. Associated values are also provided in Table 3.
Table 3. Data from 5 photosynthesis-irradiance (P-I) experiments conducted on *Chara* sp. collected from Lake Tahoe. The shaded row shows the low-light adaptation P-I experiment in which plants were exposed to low light conditions (3 µmol m\(^{-2}\) s\(^{-1}\)) for 4 weeks prior to the experiment. All other experiments occurred immediately after *Chara* sp. was brought back to the laboratory.

<table>
<thead>
<tr>
<th>Date</th>
<th>Temperature</th>
<th>Incubation Period</th>
<th>(P_{\text{max}})</th>
<th>(I_k)</th>
<th>(I_c)</th>
<th>(\alpha)</th>
<th>(R)</th>
</tr>
</thead>
<tbody>
<tr>
<td>14-Aug-2013</td>
<td>8.7</td>
<td>4.5 to 8.5</td>
<td>4.5 to 8.5</td>
<td>1.6E-04</td>
<td>57</td>
<td>21</td>
<td>2.9E-06</td>
</tr>
<tr>
<td>23-Oct-2013</td>
<td>12.3</td>
<td>4.5 to 5.0</td>
<td>4.5 to 5.0</td>
<td>4.6E-05</td>
<td>63</td>
<td>16</td>
<td>7.4E-07</td>
</tr>
<tr>
<td>8-Jan-2014</td>
<td>6.9</td>
<td>5.0 to 7.0</td>
<td>5.0 to 7.0</td>
<td>8.3E-05</td>
<td>27</td>
<td>2.2</td>
<td>3.1E-06</td>
</tr>
<tr>
<td>26-Feb-2014</td>
<td>N/A</td>
<td>6.0 to 9.0</td>
<td>6.0 to 9.0</td>
<td>8.5E-05</td>
<td>9.6</td>
<td>0.21</td>
<td>7.5E-05</td>
</tr>
<tr>
<td>23-Apr-2014</td>
<td>6.3</td>
<td>5.0 to 7.0</td>
<td>5.0 to 7.0</td>
<td>8.9E-05</td>
<td>31</td>
<td>4.9</td>
<td>2.9E-06</td>
</tr>
</tbody>
</table>

\(P_{\text{max}}\) = maximum rate of photosynthesis  
\(I_k\) = irradiance representing onset of photosaturation  
\(I_c\) = irradiance at which respiratory losses equal photosynthetic gains (net photosynthesis = 0)  
\(\alpha\) = initial slope of photosynthesis vs irradiance (light-limited region)  
R = respiration rates in dark bottles

Table 4. Mean (±SE) chlorophyll \(a\) and \(b\) concentrations in *Chara* used for photosynthesis-irradiance experiments. The shaded row shows the low-light adaptation P-I experiment in which plants were exposed to low light conditions (3 µmol m\(^{-2}\) s\(^{-1}\)) for 4 weeks prior to the experiment. All other experiments occurred immediately after *Chara* sp. was brought back to the laboratory.

<table>
<thead>
<tr>
<th>Experiment Date</th>
<th>Chl (a) (mg/g DW)</th>
<th>Chl (b) (mg/g DW)</th>
<th>(n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>20-Aug-2013</td>
<td>2.94±0.07</td>
<td>1.52±0.05</td>
<td>55</td>
</tr>
<tr>
<td>23-Oct-2013</td>
<td>1.80±0.07</td>
<td>1.00±0.04</td>
<td>33</td>
</tr>
<tr>
<td>8-Jan-2014</td>
<td>2.88±0.08</td>
<td>1.75±0.06</td>
<td>32</td>
</tr>
<tr>
<td>26-Feb-2014</td>
<td>2.89±0.10</td>
<td>1.54±0.05</td>
<td>33</td>
</tr>
</tbody>
</table>
Fig. 17. The photosynthesis-irradiance (P-I) relationship for Chara collected from Lake Tahoe for a low-light adaptation experiment in which plants were exposed to low light conditions (3 μmol m$^{-2}$ s$^{-1}$) for 4 weeks prior to the experiment. Associated values are also provided in Table 3. The upper and lower graphs display identical data using different x-axes (the upper graph can be compared to graphics provided in Figure 16 and the lower graph shows the spread of data on a smaller scale.)
Mysid and Crayfish Effects on Special Status Communities

Objective 4. Determine if non-native invertebrate species are affecting special status communities.

Mysid Effects on Special Status Communities

Non-native oppossum (mysid) shrimp (*Mysis relicta*) were collected in benthic samples collected with a Petite Ponar from McKinney Bay and Camp Richardson in the spring (March and May 2013) between 10 am and 1 pm. A large number of mysids (80) were collected in McKinney Bay samples, whereas relatively few individuals (10) were collected in Camp Richardson samples. Mysids were collected from the South Shore Mound in July 2013 around noon, but only 3 individuals were found in samples. We did not expect to find mysids in our benthic samples, since they are thought of as a planktivorous taxon, but their presence in our benthic samples, as well as video taken by divers indicates that mysids spend a substantial amount of in the bottom environment of Lake Tahoe during the day, especially at the McKinney Bay and Camp Richardson sites. Mysids collected in benthic samples were preserved immediately. They were taken back to the laboratory where they were dissected and their stomachs were placed on a slide for gut content analysis. Some of the mysids were also dried and prepared for stable isotope analysis. Mysid dietary proportions from stable isotope results were analyzed in Program R.

Gut contents of mysids from the 3 sites varied substantially. At McKinney Bay, pollen dominated gut contents and organic matter, copepods, and rotifers were also present (Fig. 18). At Camp Richardson, 3 of the 5 mysids analyzed each contained parts of an adult stonefly as well as several stonefly eggs, suggesting that they had consumed a *C. lacustra* female full of eggs (Fig. 18). This suggests that mysids may target high-quality benthic food sources during certain times of the year, such as May when females full of eggs are abundant. At the South Shore Mound site,
1 of 3 of the mysids analyzed contained parts of an amphipod (Fig. 18), presumably *Stygobromus* spp., given that no other amphipod taxa are found at the South Shore Mound site. The other 2 mysids had pollen in their guts. The low sample size and thus large standard deviations of mysid data from Camp Richardson and the South Shore Mound did not allow for dietary proportions analysis using stable isotopes. Overall, however, the mysid gut content results suggest that mysids do utilize both *C. lacustra* and *Stygobromus* spp. as a benthic food source.

![Graph showing mean gut content proportions](image.png)

Fig. 18. Mean (±SE) diet proportions (number of items of one category divided by the total number of items found) in mysid gut contents from three sites in Lake Tahoe. Note substantial variation in sample size between sites: McKinney Bay (n = 80), Camp Richardson (n = 10) and South Shore Mound (n = 3).
Crayfish Effects on Special Status Communities

We attempted to collect crayfish on 14 and 21 June 2013 within the Camp Richardson site. We left traps out for several hours during the day, and did not find any crayfish in the traps upon collection, possibly because the traps were not able to penetrate the dense vegetation at the site. On 18 Sep 2013, we set 16 crayfish traps overnight just outside of the Camp Richardson site. A total of 23 crayfish were caught and were placed on dry ice immediately. In the laboratory, crayfish stomachs were removed and preserved for gut content analysis. The average total carapace length of the crayfish analyzed for gut contents was 46.0±1.3 mm. Crayfish tissue was also dried and prepared for stable isotope analysis. Crayfish dietary proportions were analyzed in Program R based on stable isotope results.

Moss, woody organic matter, and Chara were the dominant items in crayfish gut contents from Camp Richardson (Fig. 19). However, according to dietary proportions analysis based on stable isotopes, the main food item for crayfish collected from Camp Richardson was C. lacustra (61%), with only small dietary contributions by Chironomidae (8%) and Oligochaeta (13%) (Fig. 20). It is possible that crayfish in the Camp Richardson area consume plants and organic matter incidentally while searching for animal food sources such as (e.g., stoneflies, midges, and worms), which are assimilated more readily. Indeed, crayfish have been known to preferentially seek out animal protein, while consuming macrophytes in the process (Momot 1995). Additionally, gut content analyses often result in the assumption that plant and organic matter play a larger role in crayfish diets than is suggested by stable isotope analysis (e.g., Whitlege and Rabeni 1997, Hollows et al. 2002).

An experiment was also conducted to test for direct consumption of Chara and moss by crayfish. Crayfish were placed in aquaria with filaments of both Chara and moss for 48 hours.
The fresh weight of plants was recorded before and after the trials to determine crayfish consumption. The average total carapace length of the crayfish analyzed for gut contents was 41.0±0.3 mm.

After 48 hours, crayfish reduced moss and *Chara* biomass, but did not consume all of the material. An average of 51% of moss and 43% of *Chara* was consumed over the experimental period (Fig. 21). The amount of material consumed (193.3±40.3 mg fresh weight for *Chara* and 28.3±4.9 mg fresh weight for moss) within a 48 hour period was small when compared to macrophyte consumption rates for crayfish reported in the literature (e.g., Cronin et al. 2002, Nyström and Strand 1996). It is possible that consumption rates were reduced due to stress from laboratory conditions or that the incentive to consume plant material that did not contain invertebrates was reduced when compared to field conditions.

Our findings indicate that introduced mysids and crayfish in Lake Tahoe are using sensitive species as a direct food source. Additionally, crayfish appear to be modifying the habitat that sensitive species depend on by consuming moss and *Chara*. We have not quantified the effects of these non-native species on endemic invertebrate communities or the plants they rely on for habitat; however, it is clear that they are negatively impacting native plants and endemic invertebrates in the lake.
Fig. 19. Mean (±SE) diet proportions (estimated relative percentage) from gut contents of crayfish collected from the Camp Richardson area in September 2013.

Fig. 20. Dietary proportions of crayfish collected from the Camp Richardson site. Dark grey bars = 50th percentiles, lighter grey = 75th percentiles, and light grey = 95th percentiles.
Fig. 21. Mean (±SE) fresh weight of *Chara* and moss placed in aquaria with (experimental) and without (control) crayfish before and after a 48 hour period.
Conclusion

There is a tight association between Chara and C. lacustra in Lake Tahoe and it is likely that C. lacustra would not be able to maintain populations in the lake without Chara. There also is an association between the moss bed at the South Shore Mound and what is perhaps the only remaining extensive population of Stygobromus spp. in the lake. The Camp Richardson Chara bed and the South Shore Mound moss bed are currently the only known areas that harbor healthy populations of endemic invertebrates.

Although the biology of many of the endemic invertebrate taxa in Lake Tahoe remains unknown, we have made some interesting discoveries about the life history of C. lacustra. Few stoneflies develop two reproductive cohorts simultaneously, as does C. lacustra, and it is the only stonefly that is known to give live birth. We postulate that the ability for C. lacustra to give birth to live nymphs is related to the direct dependence on Chara beds for habitat and food. Recently-born nymphs are able to cling to the substrate (Chara) which will protect and sustain them.

Because of the decline in deepwater plant beds in Lake Tahoe, C. lacustra has had its habitat severely restricted (Caires et al. 2013). Although the stonefly was placed on a list of endangered invertebrates by the International Union for the Conservation of Nature in 1983, was listed as a Species of Concern by the US Fish and Wildlife Service, and was described as “critically imperiled” by the Nevada National Heritage Program in 1998, it has remained difficult to get recognition for this small species that lives at depth in Lake Tahoe. Although we have made headway in understanding the life history of C. lacustra, it has now become clear that we must also understand the ecology and biology of the plants that these endemic invertebrates depend on.
Light may be limiting for the growth of dense beds of *Chara* in Lake Tahoe. Although we found that, in the short-term, *Chara* can adapt to very low-light conditions, we are unsure whether *Chara* can sustain longer-term growth at these light levels. We also know that measurements made in the laboratory do not account for self-shading that would occur in a *Chara* bed as dense as the Camp Richardson bed. *Chara* growing on the South Shore Mound appears that it may be at its depth limit in the lake, given its distinct morphology and thinly dispersed and scattered growth pattern. We did not run photosynthesis-irradiance experiments on moss and thus do not know what its depth limit may be. Moss and *Chara* beds occurred much deeper historically; therefore, it is likely that areas of suitable habitat with silt-dominated substrate that were colonized by plants in the past are now in areas of the lake in which light levels are not sufficient for the growth of extensive plant beds.

Although we found evidence that non-native species (mysids and crayfish) are directly consuming both *Chara* and *C. lacustra*, it is unclear how much of a negative effect they are having on these plant-invertebrate assemblages. The Camp Richardson *Chara* bed maintains an extremely high density of both *Chara* and *C. lacustra*, despite non-native species predation. The negative effects of non-native species on *Stygobromus* spp. populations, however, may be more severe. *Stygobromus* spp. was one of the most abundant invertebrates in the lake in the 1960s and now it can hardly be found. In the 1960s, its densities were highest in deep areas of the lake in which plants did not grow. This suggests that non-native mysids and crayfish could have affected *Stygobromus* spp. either directly through predation (as we found evidence for in this study) or indirectly through consumption of organic matter the amphipod was dependent upon.

The unique nature of endemic invertebrate and plant populations in Lake Tahoe certainly warrants further attention and study. We suggest the delineation of conservation zones in the area
of the South Shore Mound and Camp Richardson plant beds. Further study is needed to investigate the potential for plant growth in deeper areas of the lake and to monitor plant bed growth during periods of increasing water clarity. Finally, a diver’s survey of the lake in areas potentially colonized by *Chara* and moss (e.g., the 30-50 m depth zone) is suggested in order to search for other hotspots of biodiversity on the lake bottom.

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Literature Cited


