Mediterranean Climate Flows and In-Stream Fauna: Mosquitofish and Invertebrate Abundances in Grayson Creek, California

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ABSTRACT

Gambusia affinis is a fish used worldwide in mosquito control programs. Although designated an invasive species in California, it is currently unknown how well stocked fish persist after high winter flows of Mediterranean climates. California mosquito control relies on stocking mosquitofish that have to be restocked intermittently from losses. In Northern California, from November-February 2021, I inventoried habitat characteristics, sampled aquatic habitats for macroinvertebrates and banksides for terrestrial insects, and counted stocked mosquitofish in Grayson Creek, in 3 sampling sites (11 sub-sites). Using sticky traps, I sampled nearly 2,300 terrestrial insects. *Sciaridae*, *Psychodidae*, and *Chironomidae* dominated the terrestrial samples and increased in abundance after heavy rains to levels greater than those just before the wet season. I collected roughly 600 in-stream fauna using drift nets at each site, and abundance decreased immediately after flows and then recovered, except at the furthest downstream location, where macroinvertebrate abundance continuously decreased. Mosquitofish abundance via visual survey counts peaked at 69 individuals before the wet season and also decreased during periods of high flow until disappearing altogether by mid-February. Insect abundance recovered after heavy flows removed the mosquitofish, and *Culicidae* were rarely present throughout the entire sampling period. Thus, it is unclear if abundance is attributed to the mosquitofish or other ecosystem characteristics.

KEYWORDS

invasive species, urban stream syndrome, mosquito control, benthic macroinvertebrates, terrestrial insects

INTRODUCTION

The continuing increase in human development worldwide has severely degraded urban streams, and, in turn, urban streams can cause problems for development. Paved surfaces increase runoff (drainage) rates, and runoff washes pollutants and litter from roads into the water (Dunne and Leopold 1978). Increased runoff from storms can trigger flash floods, and altered channels increase erosion rates, which can be a serious risk for development on steep banks (Booth 1990). Biologically, urban streams have reduced biodiversity and are populated by more tolerant species that can withstand poor water and impaired habitat quality (Paul and Meyer 2001, Meyer et al. 2005). These problems can be described under the umbrella term "urban stream syndrome" (Walsh et al. 2005), which describes conditions of degraded urban streams around the world.

The presence of mosquitoes is a characteristic of degraded streams (Walsh et al. 2005, Oliver and Brooke 2018, Glunt et al. 2018). Mosquitoes are perceived as pests and are also vectors for diseases such as malaria, West Nile Virus, and dengue, among many others (Bohart and Washino 1978, Becker et al. 2010, Porse et al. 2015). Consequently, this makes controlling mosquito populations a major public health issue (Becker et al. 2010); in California, copious amounts of money have been spent to study and control mosquito populations due to the state's diversity of habitats and intensive water use (Bohart and Washino 1978). Mosquitoes are very tolerant, however, and quickly adapt to pollution, insecticides, or other habitat variables (i.e. temperature) (Oliver and Brooke 2018, Glunt et al. 2018). Their persistence has driven many managers to consider adding Western mosquitofish (*Gambusia affinis*) into the water to feed on mosquito larvae as a method of biological control. Although this approach is largely immune to mosquito adaptation, there are still implications for adding *Gambusia* into bodies of water. *Gambusia affinis* is native to the southern and eastern United States (Turlock Mosquito Abatement District n.d.), but *Gambusia* is used worldwide to control mosquito populations (Pyke 2005, Cote et al. 2010), and its impacts as an invasive species in the United States and elsewhere are innumerable (Alcaraz and García-Berthou 2007).

Contra Costa County, located in the San Francisco Bay Area, has been stocking *Gambusia* in creeks and residential areas within its boundaries to control mosquito populations since 1997 (Contra Costa County Vector and Control District n.d.). In the Bay Area's Mediterranean climate, flash floods typically punctuate the end of the dry season in late fall (Bonada et al. 2006). Many organisms in Mediterranean streams are adapted to the highly variable water flows and are accustomed to seasonal cycles of flooding and drying (Gasith and Resh 1999), but mosquitofish are not. *Gambusia* is fairly tolerant to water pollution, but its small size prevents it from swimming against a strong current (Pyke 2005); winter storm flows that characterize the Mediterranean climate in California may completely eliminate *Gambusia* populations in Grayson Creek, located in Contra Costa County, so that they need to be restocked annually to continue controlling mosquito populations. Thus, the extent of the effects of *Gambusia* on the greater macroinvertebrate community in Grayson Creek is still a mystery since the *Gambusia* "disappear" after increased flow each winter and need to be restocked (Wexler 2020).

To study the relationship between stocked *Gambusia affinis* and the stream habitat of Grayson Creek, I examined variations in *Gambusia* abundance across different microhabitats in the semi-restored stretch of Grayson Creek. More specifically, I examined the precipitation events and water discharge rates, benthic macroinvertebrate populations, and emerging aquatic and terrestrial insect populations. I then compared them all to *Gambusia* population sizes to see the extent to which the factors above are related to *Gambusia* populations. Understanding these relationships can offer insight into how to increase the efficiency of *Gambusia* for controlling mosquitoes and *Gambusia*'s greater impacts on Grayson Creek.

METHODS

Study site

Grayson Creek is a Mediterranean climate stream situated in Contra Costa County, California. Originating in Briones Regional Park (37.976984, -122.068359), Grayson Creek flows northeast, merges with Walnut Creek under California State Route 4 and then flows into Pacheco Slough before spilling into Suisun Bay (Figure 1). Parts of Grayson Creek have been transformed into vertical concrete channels or moved underground, and the entire stream has undergone significant alterations and restoration (Walnut Creek Watershed Council and Restoration Design Group 2013).

I chose my three study sites (Figure 1) based on accessibility and the presence of restored (reconstructed earth) banks (Contra Costa Watershed Forum 2003), and I further divided them into 11 sub-sites, each 10 m from the others. Site 1 was characterized predominantly by narrow channels with heavy vegetation on both sides. General morphology was consistent for the first and third sub-sites, however, the second sub-site, located under a bridge, was much wider with sparser vegetation on one side, and a concrete wall on the other. Site 2, was also heavily vegetated, but the first sub-site was two times wider than the other sub-sites, the second sub-site had eroded banks revealing some of the slag that was used to reconstruct the channel, and the third sub-site had mats of aquatic plants that were not found at any other sites. There were also two pools at Site 2 that connected to the main channel (and each other) when the water level surpassed the strip of land separating them, and these were the fourth and fifth sub-sites. Site 3 was located under a wide bridge and was uniform in channel width. The banks were entirely made of slag, except for the vegetation that grew at the end of the first and third sub-sites, on either side of the bridge.

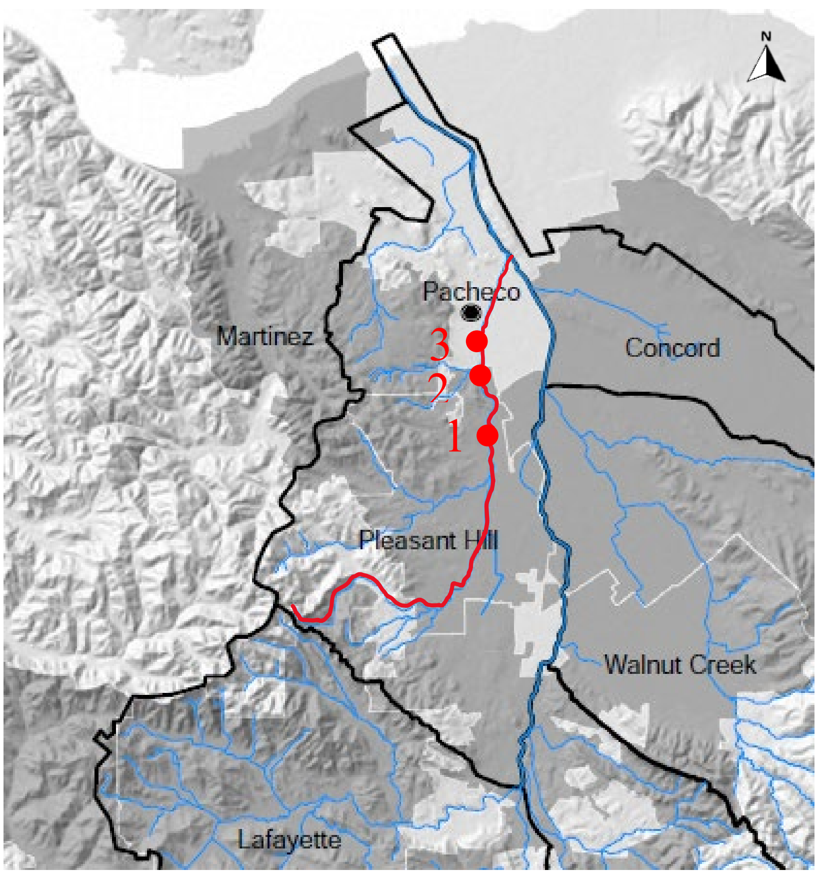

Figure 1. Map of Grayson Creek with sampling sites labelled. Map obtained from Walnut Creek Council and Restoration Design Group.

I sampled from November 2020 through February 2021 with a period from mid-November to mid-December where I was unable to go collect data. Otherwise, I collected data at most once every week with each sampling event at least four days apart, weather permitting. I sampled once at each sub-site at all three sites.

Physical habitat and discharge

To characterize the stream channel, I recorded bankside vegetation, channel cover, and substrate and noted any significant changes over my sampling period at each sub-site. I measured the velocity of water flow (distance per unit time) by timing how long it took for a leaf to drift down three meters of the creek. To calculate discharge, I measured the cross-sectional area of the channel (channel width x channel depth) using a tape measure. I then multiplied the two values together (velocity x cross-sectional area). I repeated this entire process at each sub-site, moving downstream roughly 10 m each time. I also obtained supplemental rain tip and stage height data for Grayson Creek from March 2020 to March 2021 from two sampling stations that are part of the California Data Exchange Center (https://cdec.water.ca.gov/): Grayson Creek at City Yard (GCC) and Grayson Creek at Center Avenue (GCA). The rain tip data was collected every hour, and the stage height data was collected every 15 minutes. I converted the rain tip data into a precipitation graph and graphed the stage height data as a proxy for discharge to visualize the wet season and aid in looking at differences within sampling sites.

Mosquitofish

To determine the abundance of *Gambusia* at the three sites, I conducted visual counts. At each sub-site, I searched for mosquitofish from the banks. Once I sighted the first fish, I started my timer for 30 seconds and counted as many fish as I could see in that time from my current location. If I did not find any fish after 2 minutes, then I determined there were not any fish at the sampling point and moved downstream.

In-stream benthic macroinvertebrates

Because mosquitofish feed at all levels of the water column (Gambusia affinis n.d.), I quantified drifting macroinvertebrates by setting up a drift net made of a 5-gallon paint strainer sewed onto a 30 cm x 15 cm PVC frame and secured using 2-foot long rebar stakes pounded into the stream bed (Figure 2) at the downstream end of each sampling site. After 24 hours, I collected the nets and moved any organisms into vials of 75% ethyl alcohol. I then identified the specimens under a microscope (Table 1).

Figure 2. Drift net set up at Site 2.

Figure 3. Sticky trap set up at Site 1.

Emerging aquatic and terrestrial insects

To quantify emerging aquatic and terrestrial insects in the streamside vegetation, I also set up three sticky traps at each sampling sub-site for a total of 11 sticky trap collections per sampling event, alongside where I counted mosquitofish and measured water flow rates. I constructed the sticky traps out of transparent 20 cm x 13 cm marking flags with Tanglefoot® Tangle-Trap® applied to one side. I placed the flags either at the edge of the water or up to 1 m up the bank depending on the conditions of each sub-site and positioned the flags roughly 18 cm above the surface of the water or ground with the sticky side facing the water (Figure 3). I collected the flags after 72 hours in transparent sandwich bags and then scanned them at 2400 dpi for identification using the methods of Mendez et al. (2018) (Table 1).

Table 1. List of identification keys used.

Title	Author/Website
Adult Diptera Identification Key	https://shire.science.uq.edu.au/bb/parasitology/diptera/diptera-key1.html
An Introduction to the Aquatic Insects of North America, 3rd edition	R. W. Merritt and K. W. Cummins
Dichotomous Key to Common Aquatic Invertebrates	G. Mathias Kondolf, adapted from *Guide to the Aquatic Invertebrates of the Upper Midwest* by the University of Minnesota's Chironomidea Research Group and *Save Our Streams* by the Izaak Walton League of America
Macroinvertebrate Identification Key	https://www.macroinvertebrates.org/
Methods in Stream Ecology, Appendix 21.1	F. Richard Hauer and Gary A. Lamberti
Zurqui All-Diptera Biodiversity Inventory	http://phorid.net/zadbi/education/how-to-identify-flies/how-to-identify-flies-antennae-shape/

Data analysis

As a consequence of the seasonal patterns of Mediterranean streams (Gasith and Resh 1999), I sampled only during the winter storm flow months from November to February. I compiled the collected data so that it could be read in R and examined trends over the sampling period for mosquitofish, drifting macroinvertebrates, and insects across all three sampling sites, at each site, and at each sub-site. To observe how the drifting macroinvertebrate and insect assemblages changed over time, I used non-metric multidimensional scaling (NMDS) and then compared the ordination to discharge periods.

RESULTS

Precipitation and flow data

Of the two CDEC sampling stations in Grayson Creek, only GCC, located roughly 500 m upstream from my first sampling site, had rain tip data available. The GCC sampling site precipitation events ranged from 0.25 to 1.76 inches/day (Table 2). The wet season started with an event of 0.25 inches on Nov. 17, 2020, just after I started sampling, followed by more frequent rain events in December (12/12/2020, 12/18/2020) to total 0.78 inches and 0.34 inches, respectively. The most intense event occurred on Jan. 25, 2021, with 1.76 inches of rain. Two more smaller precipitation events occurred in February (2/2/2021, 2/11/2021) with a total of 0.42 and 0.27 inches of rain, respectively. The tipping bucket meter is emptied every July to reset the precipitation measurement (Figure 4A).

GCC and GCA sampling stations both had stage height data available. Each rain event appears as a spike on both hydrographs (Figures 4B and C) despite GCA being located 2.7 km downstream from GCC, located between the second and third sampling sites. The spikes coincide with the dates and magnitudes from the storm events visible in the precipitation data (Table 1): at GCC, the first precipitation event on November 17, 2020, increased the stage height to 0.40 ft; the December 12 and 18, 2020, events reached 0.82 and 0.94 ft, respectively; the larger storm on January 25, 2021, reached 1.85 ft; and the two smaller precipitation events on February 02 and 11, 2021, increased the stage height to 0.75 and 0.61 ft, respectively. This pattern was mirrored at GCA, which saw the stage height increase to 1.55 ft on November 17, 3.02 ft and 3.08 ft on December 12 and 18, 4.90 ft on January 25, and then 2.38 ft and 2.01 ft on February 02 and 11.

The shapes of the two hydrographs are nearly identical, with the most prominent difference between the two graphs being that the stage height at GCA is consistently at least double that of GCC. This is most likely due to GCA being located in the upstream stretch of Grayson Creek that is still concrete channel; the concrete channel is substantially wider than the channels in the downstream stretch of reconstructed earth. Because there was no discharge data available from CDEC, I used the stage height data as a proxy. The stage height at GCA was triple that of GCC during the sampling time frame, as opposed to GCA's stage height being only double that of GCC the prior rain season as well as during the dry season. Each of these rain events were enough to flood the stream, so it appears that the stream reaches bankfull width around the 2 ft marker of the GCA flood stage gauge.

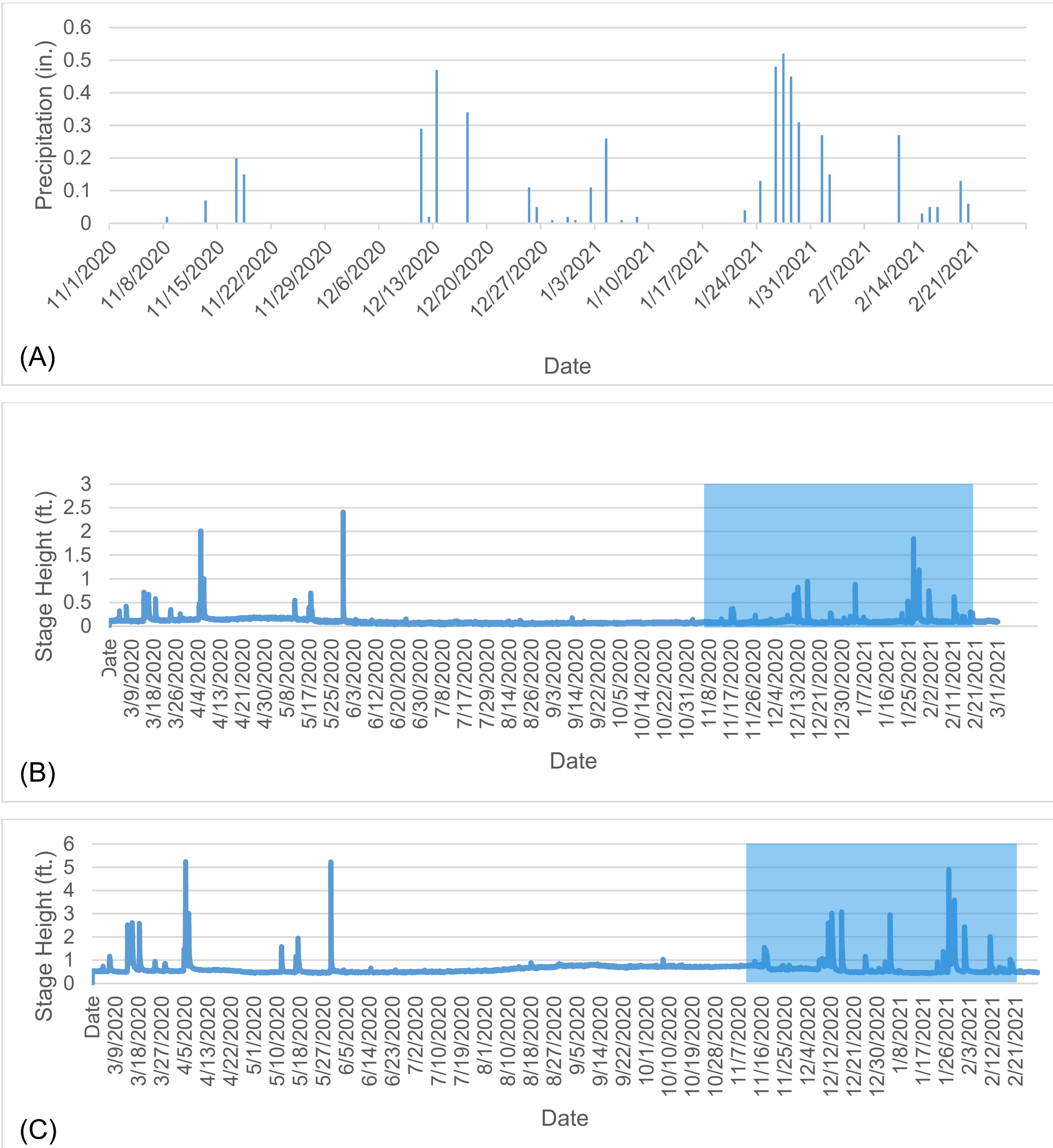

Figure 4. Precipitation and flow events. (A) Precipitation from November 2020 to March 2021 at the GCC sampling station (upstream from sampling sites). Raintip data obtained from CDEC and converted into precipitation data. (B) GCC stage height from March 2020 to March 2021. (C) GCA stage height from March 2020 to March 2021. This particular sampling station is located between Site 2 and 3. Sampling period is highlighted in light blue. Data obtained from CDEC.

Table 2. Rain events during sampling period (Nov. 14 to Feb. 24). Data obtained from CDEC.

Date (mm/dd/year)	Precipitation Event Duration (hrs)	Total Precipitation over Event (in)	Flow Event Duration (hrs)	Max Stage Height (ft) (GCC/GCA)
11/17/2020	48	0.25	48	0.40/1.55
12/12/2020	72	0.78	72	0.82/3.02
12/17/2020	18	0.34	18	0.94/3.08
01/04/2021	18	0.26	18	0.89/2.95
01/25/2021	96	1.76	96	1.85/4.90
02/02/2021	48	0.42	24	0.75/2.38
02/11/2021	24	0.27	24	0.60/2.01

Mosquitofish

As a result of variations in microhabitats, mosquitofish were only present in certain sub-sites, but each site had at least one sub-site where mosquitofish were present. Mosquitofish were most abundant before the larger precipitation events from December-onward, totaling 67 fish on Nov. 24, 2020 (Figure 5A). However, this trend in fish abundance was only true for Site 2 and 3 (Figures 5C and D) since mosquitofish were most abundance in mid-January at Site 1 (Figure 5B). I counted nearly 10 times more mosquitofish present at Site 3 than at Site 1 and 2. Only three mosquitofish remained at Site 2 after the two precipitation events in December. I was unable to locate any at Site 1 and 3. Mosquitofish populations at all three sites began to recover during the brief period without winter rains from Jan. 5-25, 2021, but the large storm from Jan. 25-28, 2021, once again reduced their populations to only a few individuals. Any surviving fish were washed away in the ensuing smaller storms. I was unable to locate any more mosquitofish after Feb. 12, 2021, and ended the monitoring part of this study.

In-stream benthic macroinvertebrates

The in-stream benthic macroinvertebrate assemblages varied across all sites. In general, however, macroinvertebrates were most abundant before major storm events, decreased immediately after the event, and recovered by the next event. Sometimes their abundances did not

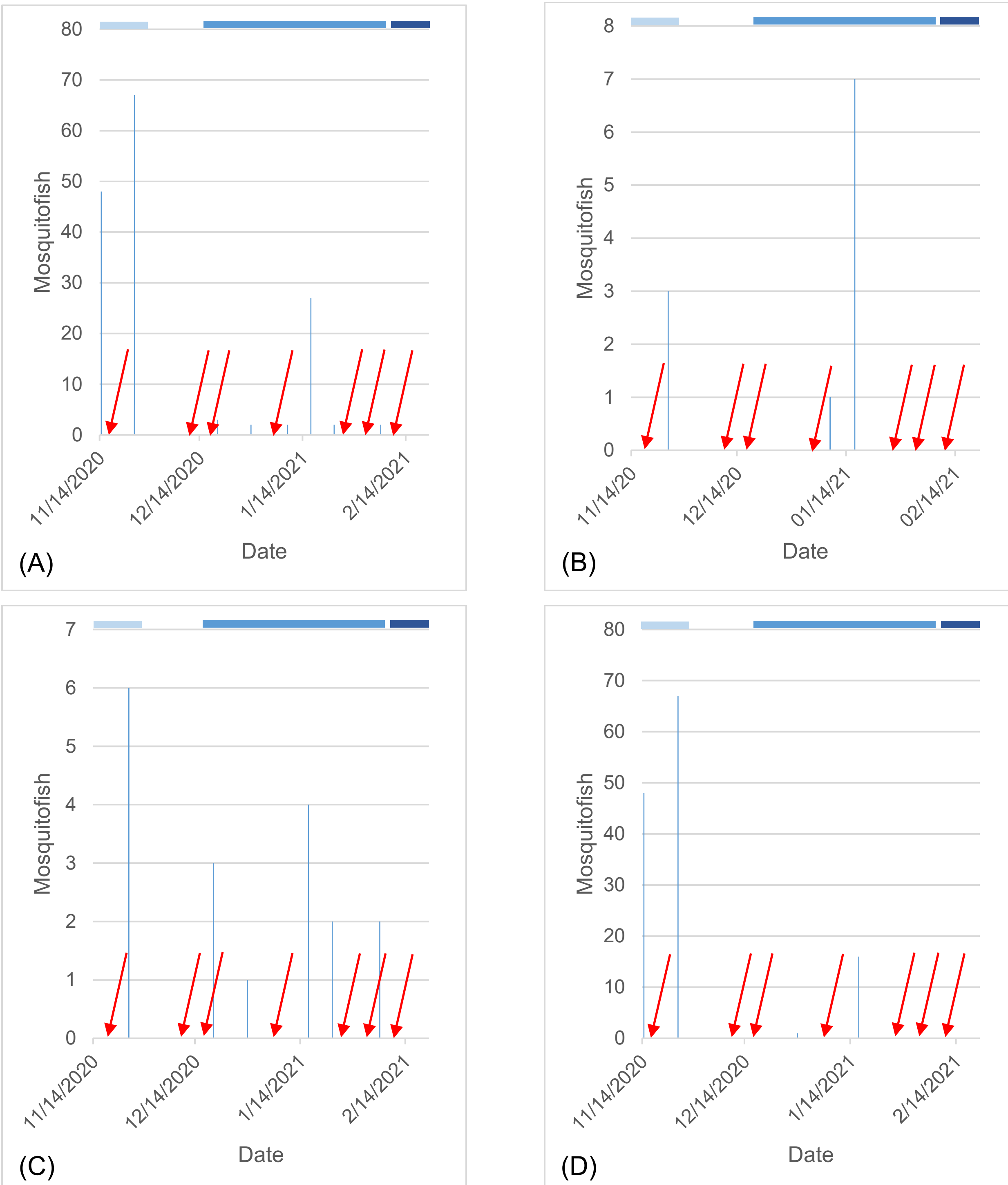

Figure 5. Mosquitofish abundances over time. (A) Aggregate mosquitofish counts throughout sampling period. (B) Mosquitofish counts at Site 1 throughout sampling period. (C) Mosquitofish counts at Site 2 throughout sampling period. (D) Mosquitofish counts at Site 3 throughout sampling period. Red arrows indicate a major storm event. The light blue bar on top indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

change after these storm events (Figure 6). Benthic macroinvertebrate abundance peaked on Nov. 24, 2020, at 119 individuals, and abundance decreased to 6 on Dec. 19, 2020. At the end of the sampling period, the macroinvertebrate community recovered to 99 and 58 counts on Feb. 14 and 20, 2021, respectively. Site 3 was the only site that decreased in overall benthic macroinvertebrate abundance, reducing from 34 macroinvertebrates on Nov. 24, 2020, to 6 on Feb. 20, 2021.

The NMDS ordination (Figure 8) further displays the differences in dominant taxa before, during, and after the wet season: simuliid larvae from Site 1 and juvenile corixids from Site 3 made up the bulk of the pre-rain population; baetids from Site 1 dominated during the wet season; and chironomids from Sites 1 and 2 and scuds from Sites 2 and 3 took over after the rain. As suggested, each site had its own unique macroinvertebrate community dominated by different species at different times. Site 1 had the greatest abundance of drifting macroinvertebrates across the entire sampling period, so much so that the counts from this site make up much of the aggregate sample, and it also had the highest richness compared to the other two sites as well with 12 taxa (Figure 7).

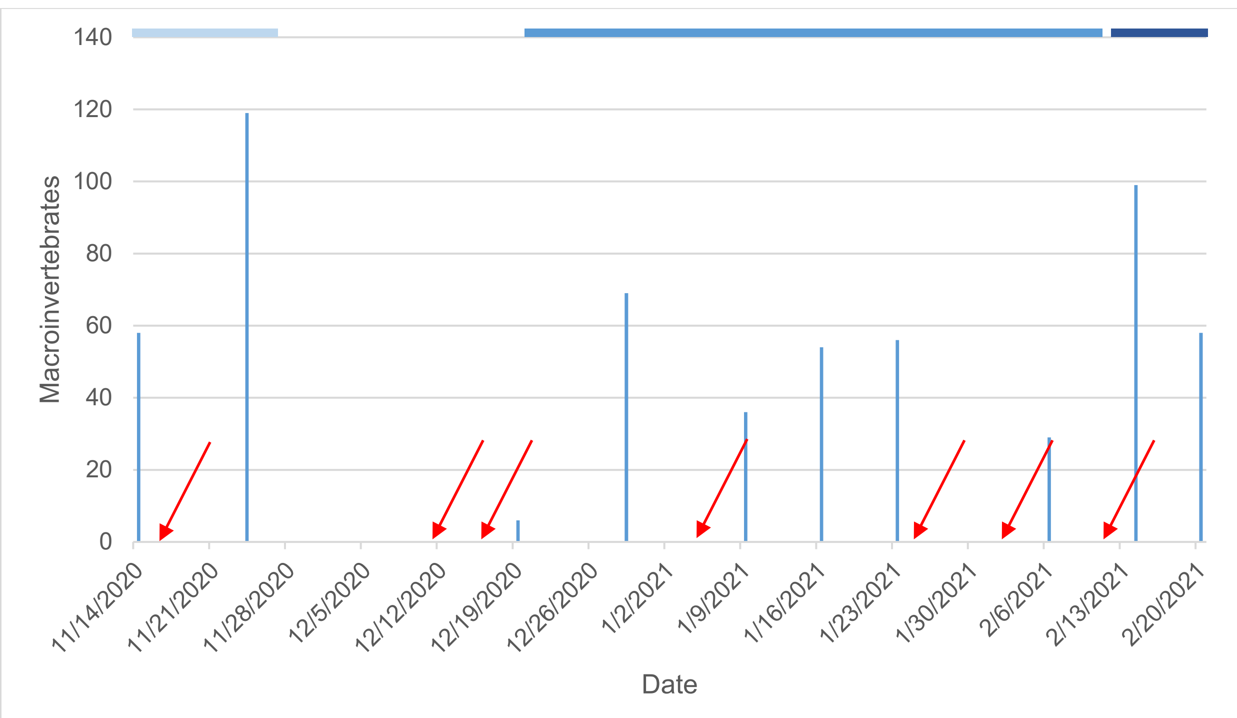

Figure 6. Aggregate benthic macroinvertebrate counts throughout sampling period. Red arrows indicate a major storm event. The light blue bar on top indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

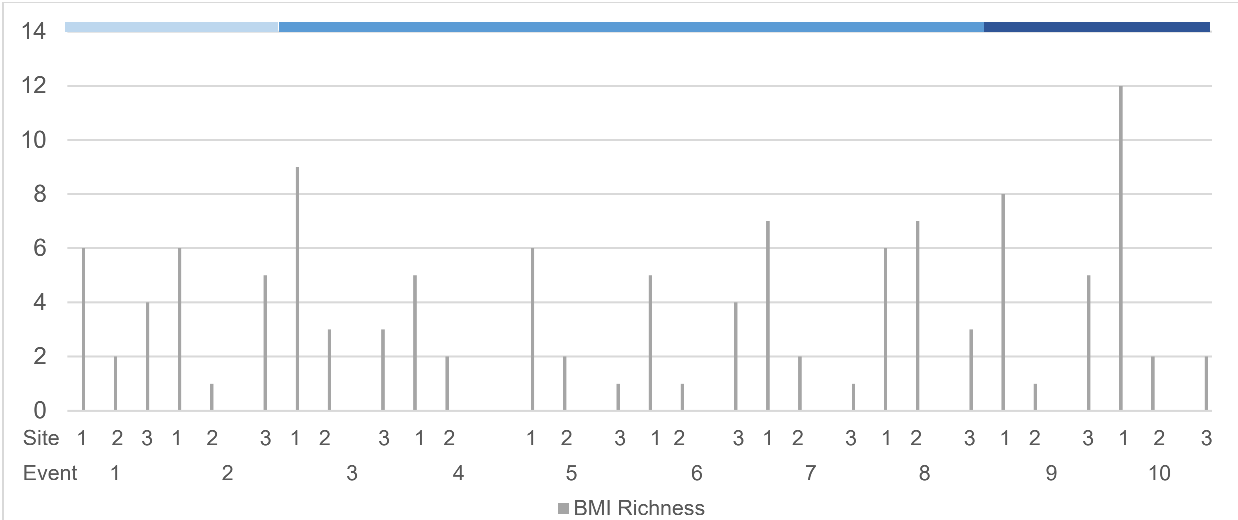

Figure 7. Benthic macroinvertebrate richness throughout sampling period. The light blue bar indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

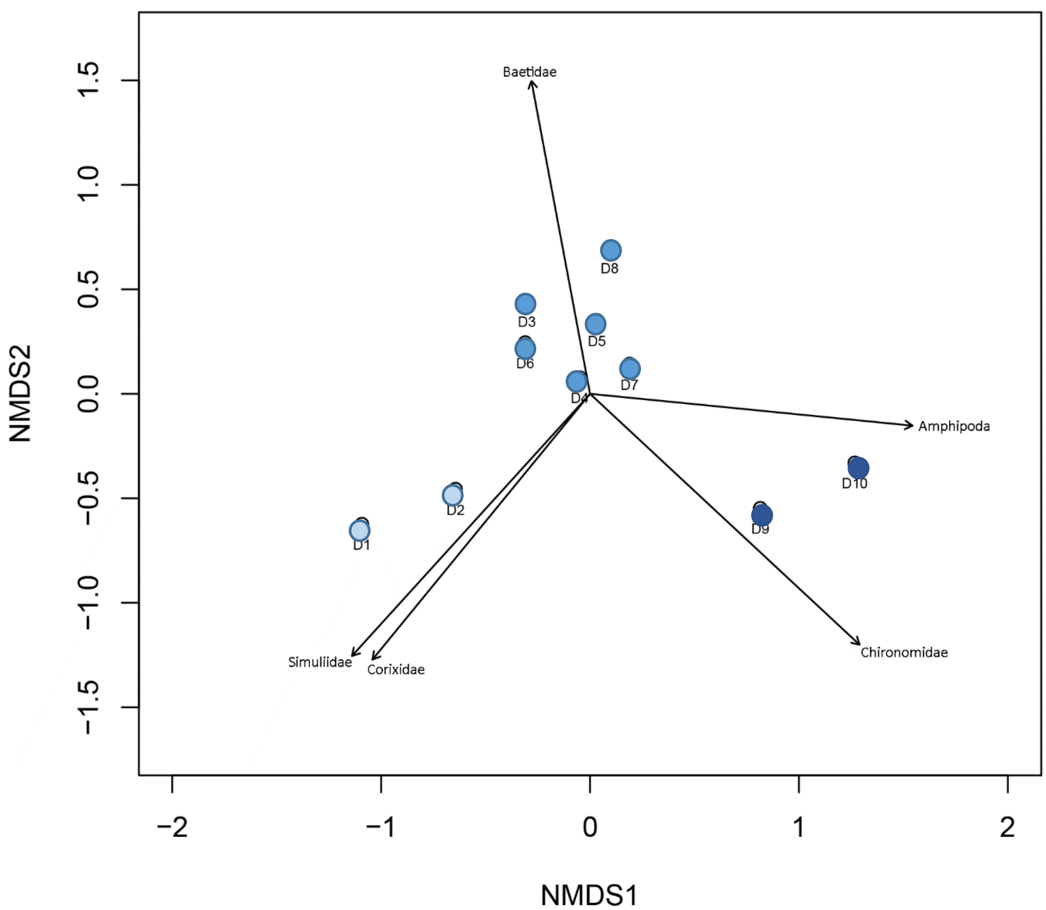

Figure 8. Aggregate benthic macroinvertebrate NMDS. D1 to D10 is each sampling event in chronological order. The light blue dots indicate sampling events prior to major precipitation events, medium blue dots indicate sampling events during the wet season, and dark blue dots indicate sampling events after major precipitation events. $R^2 = 0.993$.

Emerging aquatic and terrestrial insects

Unlike the macroinvertebrates and mosquitofish, emerging aquatic and terrestrial insects captured on sticky traps were most abundant after the wet season (Figure 14); the flying insect population increased from 196 individuals on Nov. 14, 2020, to 718 on Feb. 20, 2021. Both aquatic and terrestrial taxa increased in number during this time, but terrestrial insects proliferated more after the rain; of the 718 insects counted at the end of the sampling period, 545 were terrestrial insects and 166 were aquatic insects (7 were unidentifiable). However, the insect abundances had greater richness prior to the wet season than the macroinvertebrates with 15 taxa (Figure 15). It was difficult to determine more site-specific differences in abundance, however, certain taxa were dominant at different times across all sites. Most notably, terrestrial flies in the families *Sciaridae* and *Bibionidae* began to proliferate later in the wet season (Figure 16), corresponding to grass growing and providing ideal habitat for reproduction; unsurprisingly, they were most abundant at Sites 1 and 2, which had more vegetation on the banks. In addition, psychodids were the only aquatic fly species to dominate toward the end of the sampling period. They were present throughout the sampling period at all sites while other aquatic species were no longer present after storm events.

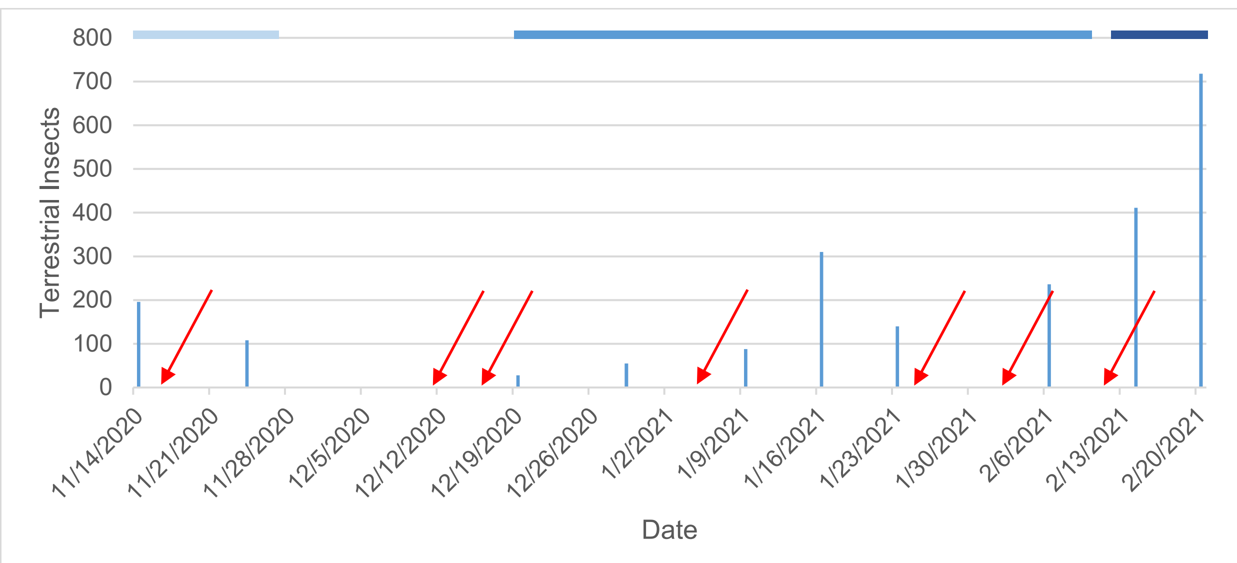

Figure 14. Aggregate emerging aquatic and terrestrial insect counts throughout sampling period. Red arrows indicate a major storm event. The light blue bar on top indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

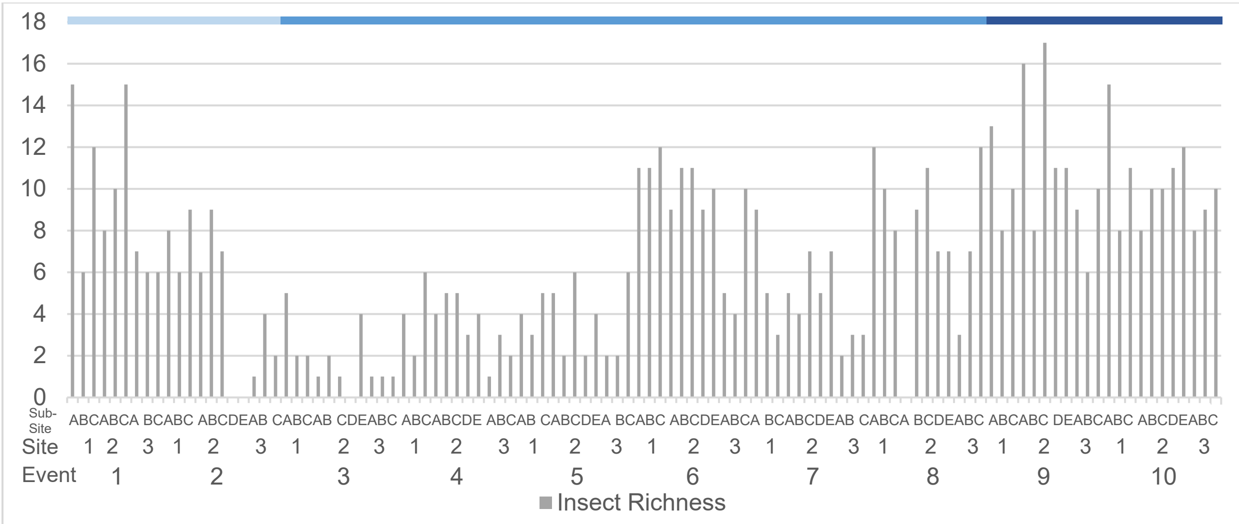

Figure 15. Emerging aquatic and terrestrial insect richness. The light blue bar indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

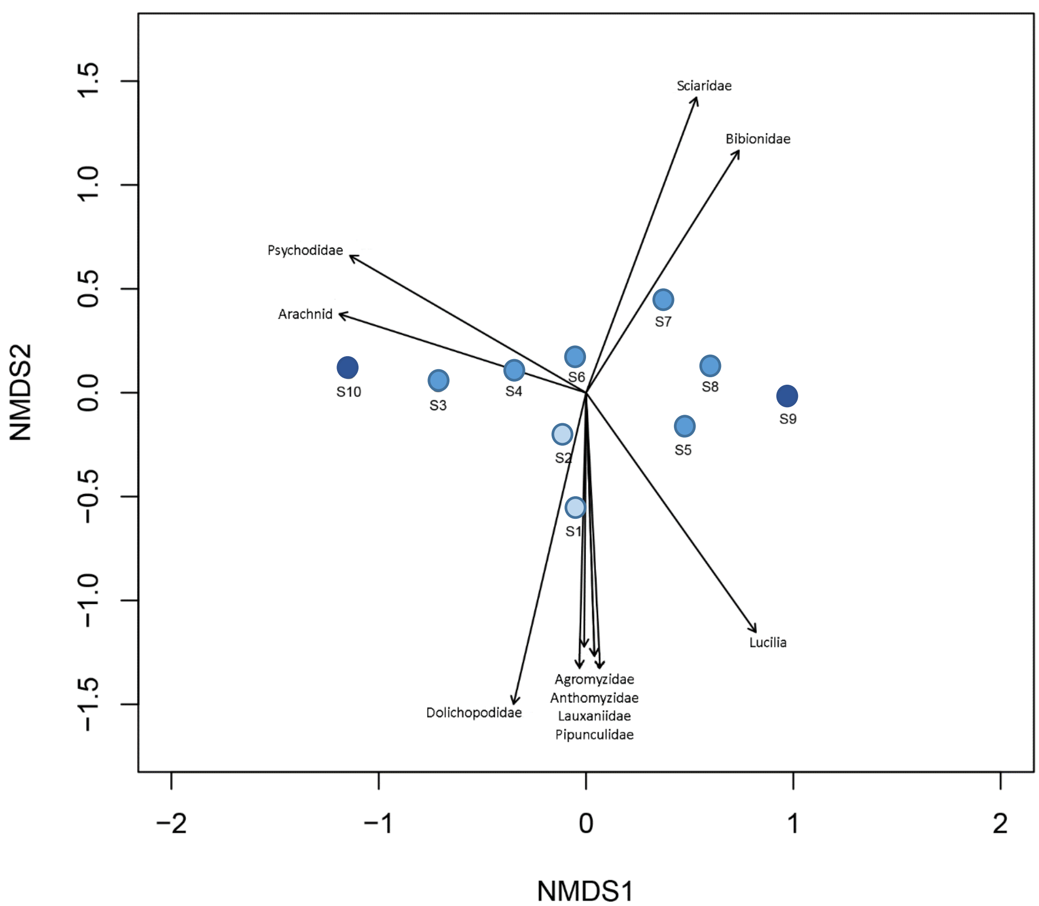

Figure 16. Aggregate emerging aquatic and terrestrial insect NMDS. S1 to S10 is each sampling event in chronological order. The light blue dots indicate sampling events prior to major precipitation events, medium blue dots indicate sampling events during the wet season, and dark blue dots indicate sampling events after major precipitation events. $R^2 = 0.995$.

DISCUSSION

In Grayson Creek, winter storm flows and conditions resulting from urban stream syndrome heavily impact mosquitofish populations, an otherwise tolerant species non-native to California and not adapted to Mediterranean climates. Potential food sources for mosquitofish are not as impacted by winter storm flow events and persist through the wet season, whereas mosquitofish need to be manually restocked by the Contra Costa Mosquito Vector and Control District each spring (or summer if it is a wet year). This pattern is likely persistent throughout urban streams in California, but the mosquitofish still holds much potential as a biological alternative to chemical insecticides for mosquito control in California.

Mosquitofish persistence and invasive potential

Mediterranean winter storm flows compounded with flash flooding due to urban stream syndrome ensured that no mosquitoes made it to the next spring. Because mosquitofish are otherwise a tolerant species and because benthic macroinvertebrates and terrestrial insects were persistent throughout the sampling period, I concluded that flow, not water quality or food availability limited mosquitofish persistence. Even though the mosquitofish were most abundant *after* the first precipitation event of the wet season, suggesting that there is a flow threshold that mosquitofish can withstand, larger storms still washed them away. Thus, vector control districts must manually restock mosquitofish annually.

Contrary to expectations, urban stream syndrome, which typically allows for more tolerant and/or invasive species (like mosquitofish) to proliferate in urban streams (Walsh et al. 2005), seems to hinder mosquitofish's ability to establish themselves as an invasive species (Harmon and Smith 2020) in Grayson Creek. Whether this phenomenon is limited to Grayson Creek or all streams in Contra Costa County, which are all similarly degraded, or all of California is unclear. However, a study conducted by Crivelli and Boy (1987) examining the diets of mosquitofish in the wetlands of southern France, which has a similar climate to the Bay Area, made no mention of disappearing mosquitofish and had collected data throughout an entire year. In addition, in an essay detailing the history of mosquito control in California from 1915 to 2015, Wekesa (2015) only briefly mentioned mosquitofish a few times throughout the essay, merely citing them as having been implemented in mosquito control programs by various vector control districts and not going into any detail about their effectiveness. Thus, it is very likely that mosquitofish being unable to persist through the wet season is a phenomenon limited to urban/heavily altered streams in California.

Benthic macroinvertebrates as food resources

Benthic macroinvertebrates collected from my sampling sites were largely characterized by tolerant taxa, which is one of the symptoms of urban stream syndrome mentioned earlier. Winter storm flows affected benthic macroinvertebrate communities to some extent; the dominant species characterized the three distinct benthic macroinvertebrate communities at different times of the wet season (before, during, and after). These shifts may imply that the macroinvertebrates are either adapted to high flow conditions (*Baetidae* in particular) (Buss and Salles 2007, Elliott 1972) or time their life cycles to avoid the storms (Bonada et al. 2007). The overall abundance of macroinvertebrates stayed roughly the same, ensuring that any voracious mosquitofish remaining during the storms had adequate food throughout the sampling period. However, there was only one mosquito larvae out of the 600 individuals caught in the drift nets.

Emerging aquatic and terrestrial insects as food resources

Emerging aquatic and terrestrial insect abundance increased substantially toward the end of the sampling period; while both groups increased in number, terrestrial insects were more than three times as abundant as aquatic insects by the end of the sampling period (Figure 17). Thus, insects that fall into the stream or enter the water to oviposit are also potential food resources for mosquitofish. Urban stream syndrome does not seem to impact aquatic insects much, but the proliferation of terrestrial insects, particularly *Sciaridae*, can possibly be explained by increasing habitat complexity, as grasses grew rapidly in between precipitation events. The shifts in insect community were also less defined than those of the benthic macroinvertebrates, and while the overall richness decreased, there was more of a gradient in community type throughout the sampling period.

Of the 2,300 individuals caught in the sticky traps, only 22 were mosquitoes. The lack of mosquitoes during the sampling period is unsurprising given that mosquitoes are least active during the winter months in the Bay Area (Reisen et al. 2008); the few that I did find are possibly older mosquitoes (with the exception of the one larva in the drift net) that did not enter diapause when the temperature cooled (Reisen 2012, Nelms et al. 2016). I also did not employ any strategies specifically designed to capture mosquitoes (i.e. CO_2 or light traps) (Reisen et al. 1999, Nelms et al. 2016), which may also explain why there were so few mosquitoes. The 22 mosquitoes were spread almost equally throughout the sampling period, so it is unlikely that flow or mosquitofish presence impacted their numbers during the wet season.

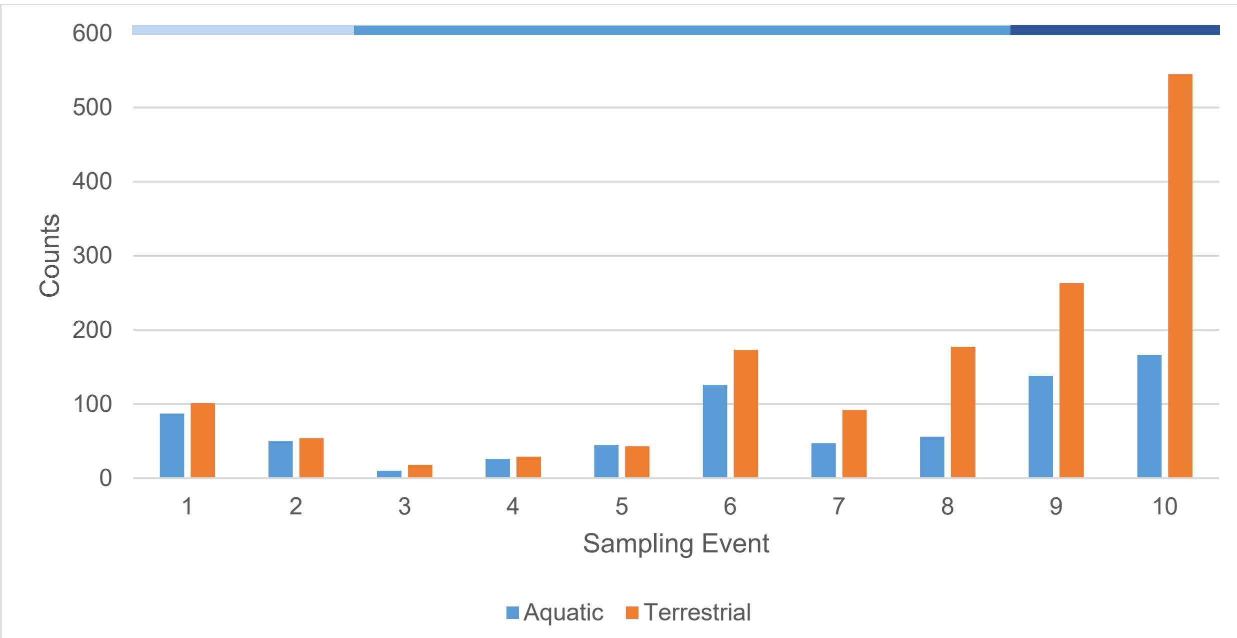

Figure 17. Breakdown of aggregate aquatic and terrestrial insect counts by habitat during each sampling event. The light blue bar on top indicates sampling events prior to major precipitation events, medium blue bar indicates sampling events during the wet season, and dark blue bar indicates sampling events after major precipitation events.

Flow events impacting mosquitofish persistence

High flows during storm events is the primary limiting factor for mosquitofish in Grayson Creek; urban stream syndrome conditions (primarily flashier floods) compounded with Mediterranean climate winter storm flows result in high discharge events that mosquitofish cannot withstand. Consequently, vector control districts that use mosquitofish in urban and Mediterranean streams must restock them annually after the wet season. It is unclear if this is a more labor- and/or cost-intensive endeavor than applying chemical insecticides, which have much more detrimental impacts on stream fauna and aquatic ecosystems. If mosquitofish are actually effective at controlling mosquito populations, then it would be worth the time and money to utilize this method as opposed to chemical control or no control. In addition, mosquitoes developing resistance to insecticide is one of the major issues with mosquito control today (Wekesa Ph.D. 2015), and, of course, not doing anything to control mosquito populations would lead to increased transmission of deadly diseases (Becker et al. 2010, Porse et al. 2015, Oliver and Brooke 2018). Mosquitofish being unable to establish themselves as an invasive species in Californian urban streams is a bonus perk that makes them even more of a viable solution for mosquito control because it limits their ability to be a successful and persistent invasive species.

Limitations and future directions

As mentioned, many female mosquitoes will enter diapause from October to April, and they are most abundant from June to September in California (Reisen et al. 2008). Mosquitofish were also essentially nonexistent toward the end of the sampling period, but both populations being low in number was likely unrelated to one another. We did not sample during the summer, when both populations are more abundant, or observe interactions between mosquitofish and mosquitoes. Comparison of mosquito abundance before and after mosquitofish restocking events are required to get a better understanding the impact and effectiveness of mosquitofish on mosquito populations. Further studies can also compare mosquito abundance in Grayson Creek with mosquito abundance in streams without mosquitofish.

Management implications

The timing of mosquitofish persistence and mosquito life cycles in Grayson Creek works out almost perfectly; there are few mosquitoes when there are few or no mosquitofish. Using temperature data and historical weather patterns, vector control districts can anticipate when mosquitoes will proliferate (spring versus summer) (Reisen et al. 2008, Nelms et al. 2016) and restock mosquitofish in waterways accordingly. Thus, to effectively use mosquitofish to control mosquito populations, it well worth conducting more research to get a better understanding of how mosquitofish persist and how effective they really are in urban Mediterranean streams like Grayson Creek.

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REFERENCES

Alcaraz, C., and E. García-Berthou. 2007. Life history variation of invasive mosquitofish (Gambusia holbrooki) along a salinity gradient. *Biological Conservation* 139(1):83–92.

Becker, N., D. Petric, M. Zgomba, C. Boase, M. Madon, C. Dahl, and A. Kaiser. 2010. *Mosquitoes and Their Control*. Springer Science & Business Media.

Bohart, R. M., and R. K. Washino. 1978. *Mosquitoes of California*. University of California, Agriculture and Natural Resources.

Bonada, N., M. Rieradevall, N. Prat, and V. H. Resh. 2006. Benthic macroinvertebrate assemblages and macrohabitat connectivity in Mediterranean-climate streams of northern California. *Journal of the North American Benthological Society* 25(1):32–43.

Bonada, N., M. Rieradevall, and N. Prat. 2007. Macroinvertebrate community structure and biological traits related to flow permanence in a Mediterranean river network. *Hydrobiologia* 589(1):91–106.

Booth, D. B. 1990. Stream-Channel Incision Following Drainage-Basin Urbanization1. *JAWRA Journal of the American Water Resources Association* 26(3):407–417.

Buss, D. F., and F. F. Salles. 2007. Using Baetidae Species as Biological Indicators of Environmental Degradation in a Brazilian River Basin. *Environmental Monitoring and Assessment* 130(1–3):365–372.

Cote, J., S. Fogarty, K. Weinersmith, T. Brodin, and A. Sih. 2010. Personality traits and dispersal tendency in the invasive mosquitofish (Gambusia affinis). *Proceedings of the Royal Society B: Biological Sciences* 277(1687):1571–1579.

Crivelli, A. J., and V. Boy. 1987. The diet of the mosquitofish *Gambusia affinis* (Baird & Girard) (*Poeciliidae*) in Mediterranean France. *Revue d'Ecologie (La Terre et la Vie)* 42:421–435.

Dunne, T., and L. B. Leopold. 1978. *Water in Environmental Planning*. Macmillan.

Elliott, J. M. 1972. Effect of Temperature on the Time of Hatching in *Baëtis rhodani* (Ephemeroptera: Baëtidae). *Oecologia* 9:47-51.

Gasith, A., and V. H. Resh. 1999. Streams in Mediterranean Climate Regions: Abiotic Influences and Biotic Responses to Predictable Seasonal Events. *Annual Review of Ecology and Systematics* 30(1):51–81.

Glunt, K. D., S. V. Oliver, R. H. Hunt, and K. P. Paaijmans. 2018. The impact of temperature on insecticide toxicity against the malaria vectors Anopheles arabiensis and Anopheles funestus. *Malaria Journal* 17(1):131.

Harmon, J. J., and G. R. Smith. 2020. Invasive fish (Gambusia affinis) as an ecological filter for macroinvertebrate colonization of experimental ponds. *Freshwater Science* 40(1):151–161.

Hauer, F. R., and G. A. Lamberti. 1996. *Methods in Stream Ecology.* Academic Press.

Kondolf, G. M. n.d. *Dichotomous key to common aquatic invertebrates*. Adapted from *Guide to the Aquatic Invertebrates of the Upper Midwest* by the University of Minnesota's Chironomidea Research Group and *Save Our Streams* by the Izaak Walton League of America.

Learning to See, Seeing to Learn. n.d. *Macroinvertebrate Identification Key* [Online]. Available at: https://www.macroinvertebrates.org/key/ (Accessed: 6 October 2020)

Mendez, P. K., S. Lee, and C. E. Venter. 2018. Imaging natural history museum collections from the bottom up: 3D print technology facilitates imaging of fluid-stored arthropods with flatbed scanners. *ZooKeys* 795:49–65.

Merritt, R. T., and K. W. Cummins. 1996. *An Introduction to the Aquatic Insects of North America*, 3rd edition. Kendall/Hunt Publishing Company.

Meyer, J. L., M. J. Paul, and W. K. Taulbee. 2005. Stream ecosystem function in urbanizing landscapes. *Journal of the North American Benthological Society* 24(3):602–612.

Nelms, B. M., T. C. Thiemann, D. N. Bridges, A. E. Williams, M. L. Koschik, B. M. Ryan, and J. J. Scott. 2016. Bionomics and Vector Potential of Culex thriambus (Diptera: Culicidae) Mosquitoes in Lake County, California. *Journal of Medical Entomology* 53(6):1473–1481.

Oliver, S. V., and B. D. Brooke. 2018. The effect of metal pollution on the life history and insecticide resistance phenotype of the major malaria vector Anopheles arabiensis (Diptera: Culicidae). *PLOS ONE* 13(2):e0192551.

Paul, M. J., and J. L. Meyer. 2001. Streams in the Urban Landscape:35.

Porse, C. C., V. Kramer, M. H. Yoshimizu, M. Metzger, R. Hu, K. Padgett, and D. J. Vugia. 2015. Public Health Response to Aedes aegypti and Ae. albopictus Mosquitoes Invading California, USA. *Emerging Infectious Diseases* 21(10):1827–1829.

Pyke, G. H. 2005. A Review of the Biology of Gambusia affinis and G. holbrooki. *Reviews in Fish Biology and Fisheries* 15(4):339–365.

Reisen, W., K. Boyce, R. Cummings, O. Delgado, A. Gutierrez, R. Meyer, and T. Scott. 1999. Comparative effectiveness of three adult mosquito sampling methods in habitats representative of four different biomes of California. *Journal of the American Mosquito Control Association* 15:24–31.

Reisen, W. K. 2012. The Contrasting Bionomics of Culex Mosquitoes in Western North America. *Journal of the American Mosquito Control Association* 28(4):82–91.

Reisen, W. K., D. Cayan, M. Tyree, C. M. Barker, B. Eldridge, and M. Dettinger. 2008. Impact of climate variation on mosquito abundance in California. *Journal of Vector Ecology* 33(1):89–98.

Turlock Mosquito Abatement District. (n.d.) *Mosquitofish* [Online]. Available at: http://www.mosquitoturlock.com/mosquitofish.shtm (Accessed: 18 October 2020)

The University of Queensland. (2015) *Adult Diptera Identification Key* [Online]. Available at: https://shire.science.uq.edu.au/bb/parasitology/diptera/diptera-key1.html (Accessed: 9 December 2020)

Walsh, C. J., A. H. Roy, J. W. Feminella, P. D. Cottingham, P. M. Groffman, and R. P. Morgan. 2005. The urban stream syndrome: current knowledge and the search for a cure. *Journal of the North American Benthological Society* 24(3):706–723.

Wekesa Ph.D., J. W. 2015. A Century of Mosquito Control in California: 1915 - 2015 26:33–46.

Zurqui All-Diptera Biodiversity Inventory. (2013) *Hot to Identify Flies – Antennae Shape* [Online]. Available at: http://phorid.net/zadbi/education/how-to-identify-flies/how-to-identify-flies-antennae-shape/ (Accessed: 9 December 2020)