PLANT LITTER FEEDING PREFERENCES OF TROPICAL ISLAND STREAM INVERTEBRATES

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Abstract. Stream ecosystems have a unique food web consisting of autochthonous and allocthonous inputs and in tropical island streams, allocthonous input in the form of decomposing plant litter are a more important food source than in temperate streams. This is an understudied area, especially non-leaf plant litter such as fruit and flowers. This study looked at the preference of the shrimp *Caradina weberi, Atyoida pilipes* and *Machrobrachium lar*, as well *Thiarid* snails, on Moorea, French Polynesia for different types of leaf litter. It also investigated whether they prefer fruit and flowers to leaves. There was also a field component looking into the distribution of invertebrate and fish species in stream pools. Invertebrates preferred leaves with soft tissue, second to those of *Angiopteris evecta. Hibiscus tiliaceus* flowers were preferred over leaves, and the role of fruit could not be established. Field surveys showed that species composition of stream pools varies with altitude.

Key words: stream invertebrate; plant litter; feeding preference; Moorea, French Polynesia

INTRODUCTION

Stream food webs are unique in that they have two main types of energy sources: autochthonous (generated within the stream) in the form of primary production of algae, or allocthonous (originating elsewhere), in the form of plant litter, terrestrial insects etc. (Davies et al. 2008). Plant litter is decomposed by microorganisms, which in turn are targeted by 'shredders', invertebrates that consume the litter (Cummins et al. 1989). Tropical and temperate streams differ in many ways, including in this regard. Plant detritus is only present in temperate streams during brief periods of the year and 'shredders' are present mostly in the form of aquatic insects, which make up a major portion of the stream biomass (Davies et al. 2008). In tropical streams primary production is at least an order of magnitude higher. Another difference is that insects play a less significant role and contribute very little to the biomass, especially in tropical island streams, as few insects are able to reach and colonize them (Resh and de Szalay 1995 and Wantzen et al. 2008). On the other hand, plant litter is present in streams year round, and there is a lot of variety in the rate of breakdown of litter from different species (Wantzen et al. 2008). In tropical island streams, the 'shredder' role is filled by macro invertebrates like crustaceans and gastropods, which are marine-derived diadromous organisms, along with some omnivorous fish (Resh et al. 1990).

FIG. 1. Location of sites on Moorea. Base map courtesy of the Geospatial Innovation Facility, University of California, Berkeley.

The feeding behavior of aquatic organisms and general community dynamics of tropical streams is understudied, especially when compared to temperate streams. Studies from Puerto Rico and Australia have shown that even though aquatic shrimp are considered generalists, in lab tests, they show definite preferences for leaves of certain species (Wright and Covich 2005 and Bastian et al. 2007). It has also been established that shredders prefer leaves that have been conditioned for a longer period i.e. have

#	Litter Type	Species	Conditioning Time (days)	*C. weberi*	*M. lar*	*A. pilipes*	*Thiaridae*
1	Leaves	*B. asiatica, M. calvescens, I. fagifer, S. campanulata*	8	11	7	0	13
2	Leaves	*S. campanulata, H. tiliaceus*	4	13	2	2	3
3	Leaves	*M. calvescens, I. fagifer*	4	17	2	0	3
4	Flowers	*H. tiliaceus*	1	18	5	4	5
5	Fruit	*I. fagifer*	2	15	3	2	3
6	Fruit	*A. comosus*	0	17	3	3	3
7	Leaves, Flowers	*H. tiliaceus*	4,1	15	3	0	2
8	Leaves	*H. tiliaceus, A. evecta*	4	17	2	0	3

TABLE. 1. Feeding trials

undergone more microbial activity (Bastian et al. 2007). Another element of plant litter in tropical streams is fruit. Tropical plant leaves are generally tough for predation defense against herbivores, and so often decompose very slowly in streams. Fruits on the other hand, are soft and fleshy, and so decompose significantly faster than leaves, and are present year round in tropical streams (Wantzen et al. 2008). However, the role of fruit in tropical stream ecosystems has received almost no attention. One study on Hawaii showed that guavas and mangoes, the two fruit species present in highest abundance, were fed on by the various shrimp species (Larned et al. 2001), but did not test whether leaves or fruit are preferred. The role of flowers, which decompose even more quickly, has been poorly studied also.

The island of Moorea in French Polynesia has been the site of several stream studies. One study compared the rates of decomposition of *Hibiscus tiliaceus*, *Inocarpus fagifer*, and *Miconia calvescens*, which are a native, an ancient introduction and a recent invasive, respectively (Gade, 1993). *H. tiliaceu*s and *I. fagifer* make up a large portion of the riparian vegetation, whereas *M. calvascens* is rapidly invading the forests (Meyer and Florence 1996). The study found that all three species decompose more rapidly when not excluded from macro invertebrates, showing that they are fed upon. It also established that of the three, *H. tiliaceus* was broken down the quickest, followed by *M.* calvescens. The fact that the tough lignified *I. calvescens* leaves were eaten the least suggests that softer leaf are preferred. Another study that looked at the feeding ecology of the shrimp *Caradina weberi* found that the shrimp preferred *H. tiliaceus* leaves to those of *I. fagifer*, and in general preferred leaves to moss (Williams 1999).

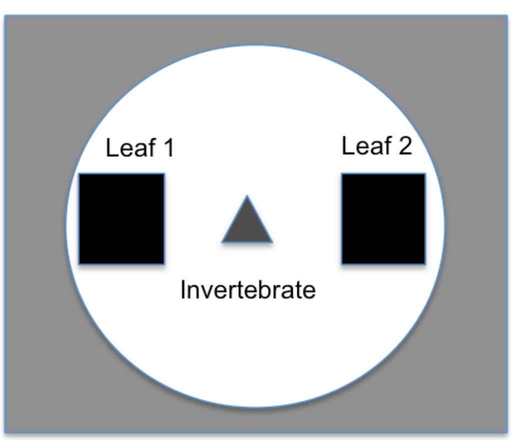

FIG. 2. Experimental set-up.

My study built on this previous work, and addressed some broader questions in the same stream system on Moorea. I carried out macro invertebrate and fish richness surveys of several pools at two altitudes, while recording proportion of leaf cover, pool depth and flow rate. I also examined the leaf litter from each pool to understand the species composition of the litter. I did lab preference tests tinvertebrates used for the lab feeding tests were three species of shrimp (*Caridina weberi*, *Machrobrachium lar* and *Atyoida pilipes)*, and *Thiarid* snails (Resh et al. 1990).

I hypothesized that when offered a choice between different types of litter, shredders would prefer certain types of leaves over others, specifically those with softer tissue, and would prefer flowers and fruit over leaves. I also thought that 'conditioned' leaves, with a microbial film, would be preferred over leaves that had not been decomposed. I also hypothesized that faunal communities would more diverse in pools with greater leaf cover, and would also vary with altitude.

METHODS

Study site

The field components of this study, i.e. the surveys as well as the collection of invertebrates and plant matter, were conducted at two sites along the same stream in the Opunohu watershed on the island of Moorea, French Polynesia (Fig. 1). Site 1 (17˚ 31' 33.33" S and 149˚ 50' 12.15" W) is at an elevation of roughly 30m and Site 2 (17˚ 32' 11.85" S and 149˚ 49' 46.25" W) is at an elevation of roughly 152m.

Feeding Experiments

The first step was determining what plants should be used in the feeding experiments. The litter from 5 pools at each site was surveyed, by using a 6-inch aquarium net to scoop up leaf litter from three random points in each pool. The different species were identified by Dr. Brent Mishler, and the leaves, flowers and fruit from the woody plants that could be easily collected were used in the experiments. The leaves were plucked from the trees, and relatively undamaged flowers and fruit were picked from the ground, as they grew too high up on the trees to pick fresh. They were then soaked in plastic boxes of stream water for a few days; the exact number for each experiment can be seen in Table 1.

Four types of invertebrates were used in the experiments:

- The shrimp *Caradina weberi*, which is the most common shrimp in the middle reaches of Moorean streams (pers. obs.).
- *Atyoida pilipes*, a filter feeding shrimp (Resh et al. 1990).
- *Machrobrachium lar*, a species of large, omnivorous prawns (Marquet 1991).
- Snails from the family *Thiaridae*.

Invertebrates were collected from pools at both sites, using a 6-inch aquarium net. The net was used to scoop up leaf litter, and the invertebrates were sorted out of it. They were transported to the lab in small tubs of stream water, and were then kept in tap water. The *C. weberi* and *Thiarid* snails were kept in the same tub, while each individual *A. pilipes* and *M. lar* was kept in a separate plastic cup. The water in all the containers was changed everyday, and each one had oxygen bubbled through it for at least 15 minutes a day.

Each of the 8 experiments included a total of 20-32 invertebrates. The proportion of individuals of each species varied a lot based on how many could be caught, and the exact numbers of individuals is shown in Table 1. In each experiment, every individual invertebrate was placed in a separate cup with water and the different types of litter (Fig. 2). Each treatment had a control in the form of 3 cups with the different kinds of litter, but no invertebrates. The experiments were checked every 12 hours and data was collected when all the cups showed signs of feeding, or at the end of 48 hours. The data was recorded as 0s and 1s for each litter type (1 = eaten, 0 = non-eaten). The types of litter tested in each experiment are detailed in Table 1. Experiment 2 included trials A and B, with leaves conditioned in stream and tap water, respectively. This was done to confirm that the invertebrates were targeting the microbial film on the leaves and not the leaf tissue; the leaves conditioned in stream water would have significantly more microbial growth than those conditioned in tap water.

Field Surveys

The second part of the study was a series of surveys of macro invertebrates and vertebrates conducted on 8 pools at Site 1 and 4 pools at Site 2, between November 10th and 20th, 2013. Each pool survey consisted of a 2-minute observation period followed by 5 minutes of sifting through leaf litter. The four types of invertebrate from the experiments were included, along with the fish *Poecilia reticulata* and those from the family *Gobiodei.* The presence and absence of different species was recorded, but number of individuals was

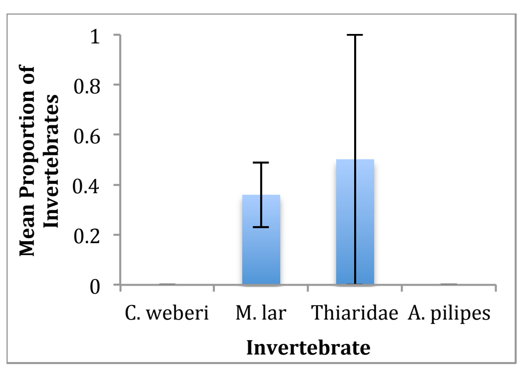

FIG. 3. Feeding preference of invertebrates for *S. campanulata* leaves conditioned in stream water in experiment 2.

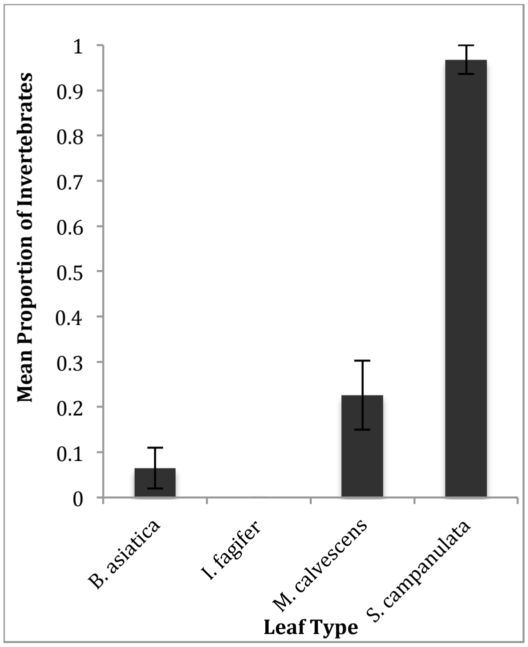

FIG. 4. Experiment 1 - Proportion of invertebrates that consumed leaves of *B. asiatica*, *I. fagifer*, *M. calvescens*, *S. campanulata* (χ^2=97.68, df=3, p<0.01). *S. campanulata* preferred.

not, as it was very difficult to accurately estimate. The percent leaf cover, flow rate (slow, medium or fast) and depth of each pool were recorded based on a visual estimate.

RESULTS

Leaf Litter Survey

5 pools at each site were surveyed for leaf

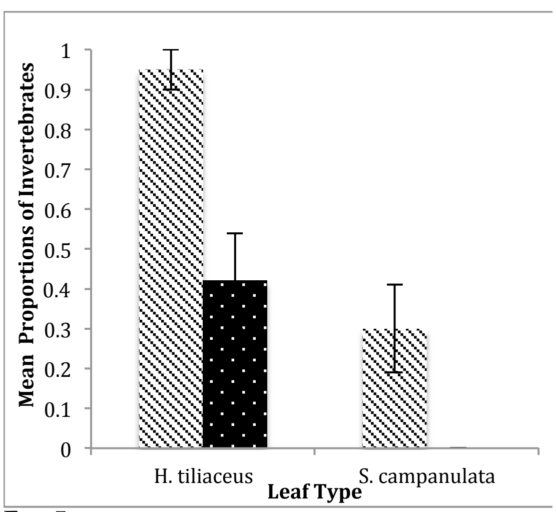

FIG. 5. Experiment 2 - Proportion of invertebrates that ate leaves of *S. campanulata* and *H. tiliaceus* conditioned in tap water (right) and stream water (left) (leaf type χ^2=34.53, df=1, p<0.01; water type χ^2=23.8, df=1, p<0.01.

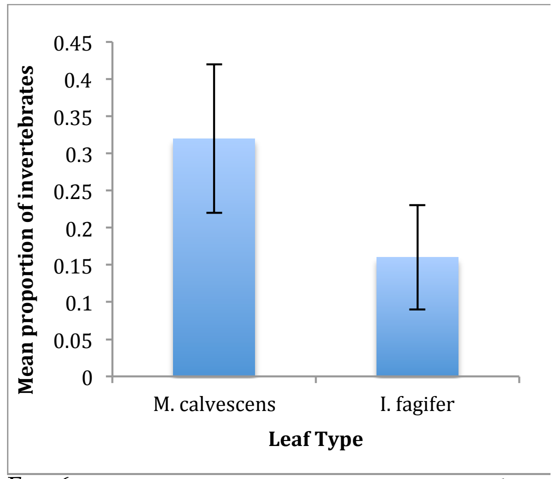

FIG. 6. Experiment 3 – Proportion of invertebrates that ate leaves of *M. calvescens* and *I. fagifer* (χ^2=1.83, df=1, p=0.18). Neither preferred.

litter composition. The species of woody plants whose leaves were found at each site are listed below:

Marae site – *Inocarpus fagifer*, *Miconia calvescens*, *Hibiscus tiliaceus* and *Angiopteris evecta*

Opunohu crossing site - *Inocarpus fagifer*, *Miconia calvescens*, *Hibiscus tiliaceus*, *Barringtonia asiatica* and *Spathodea campanulata*

The leaves from these species were used in preference experiments. Flowers from *Hibiscus tiliaceus* and fruit from *Inocarpus fagifer* were also commonly found, and hence were used in experiments.

Feeding Experiments

Invertebrates did not have statistically significant food preference differences between each other (see Fig 3 for feeding on *Spathodea campanulata* leaf conditioned in stream water in experiment 2). The p-value of each experiment was above 0.5, and thus not statistically significant.

No data was collected from experiments 5 and 6 because of issues with the experimental set up (explained in Discussion).

Experiment 1 showed that *S. campanulata* was consumed by 4 times as many organisms as *M. calvescens* and 8 times as many organisms as *B. asiatica* (Fig. 4).

In experiment 2, *H. tiliaceus* leaves were eaten twice as much as *S. campanulata* leaves, and leaves conditioned in stream water were eaten twice as much as leaves conditioned in tap water (Fig. 5).

In experiment 3, neither *M. calvescens* nor *I. fagifer* was preferred (Fig. 6).

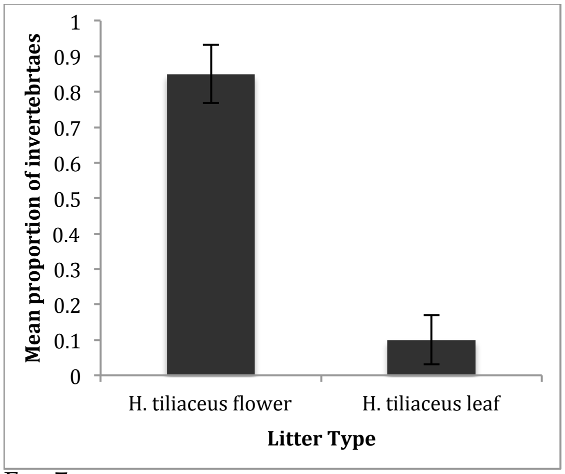

FIG. 7. Experiment 7 – Proportion of invertebrates that ate leaves and flowers of *H. tiliaceus* (χ^2=25.5, df=1, p<0.01). Flowers preferred.

In experiment 4, 80% of organisms ate *H. tiliaceus* leaves.

In experiment 7, the flowers of *H. tiliaceus* were eaten 8 times more than the leaves (Fig. 7).

In experiment 8, the leaves of *A. evecta* were eaten 8 times more than those of *H. tiliaceus* (Fig. 8).

Field Surveys

In the field survey, altitude was the only factor that affected distribution of organisms. *P. reticulata* was only found at Site 1 and *Gobiid* fish were only found at Site 2. *M. lar* was found in twice as many pools at Site 2 than at Site 1 (Fig. 9).

DISCUSSION

The results from the feeding experiments were very informative and agreed with some of the study's hypotheses. The significant

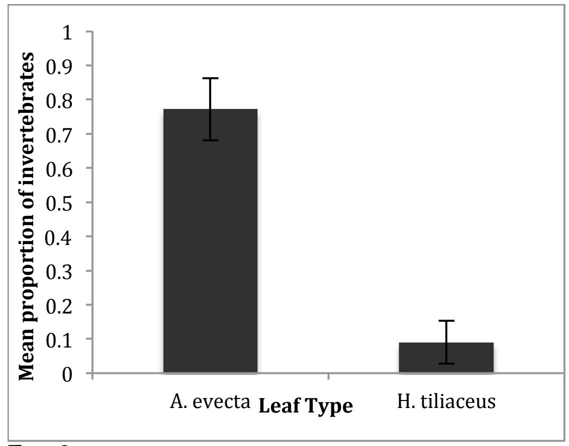

FIG. 8. Experiment 8 – Proportion of invertebrates that ate leaves of *A. evecta* and *H. tiliaceus* (χ^2=23.42, df=1, p<0.01). *A. evecta* preferred.

preference for *Spathodea campanulata* in experiment 1 and *Hibiscus tiliaceus* showed that the stream invertebrates do prefer feeding on leaves with softer tissue, which decompose rapidly. The preference in experiment 2 for leaves conditioned in stream water confirms the idea that stream invertebrates target the microbial film growing on decomposing leaves, and not the actual leaf tissue. The fact that *Miconia calvescens* was the second most preferred leaf type in experiment 1 could be important from a conservation standpoint, as *M. calvescens* has taken over more than two-thirds of Tahiti, and is also expanding on Moorea (Meyer and Florence 1996). If it does eventually take over more of Moorea's forests, it would be reassuring to know that the stream invertebrates will eat M. *calvescens* leaves if other more preferred options are not available. However, in experiment 3, a significant number of *M. calvescens* leaves were not eaten. This could be because in this experiment, the leaves were only conditioned

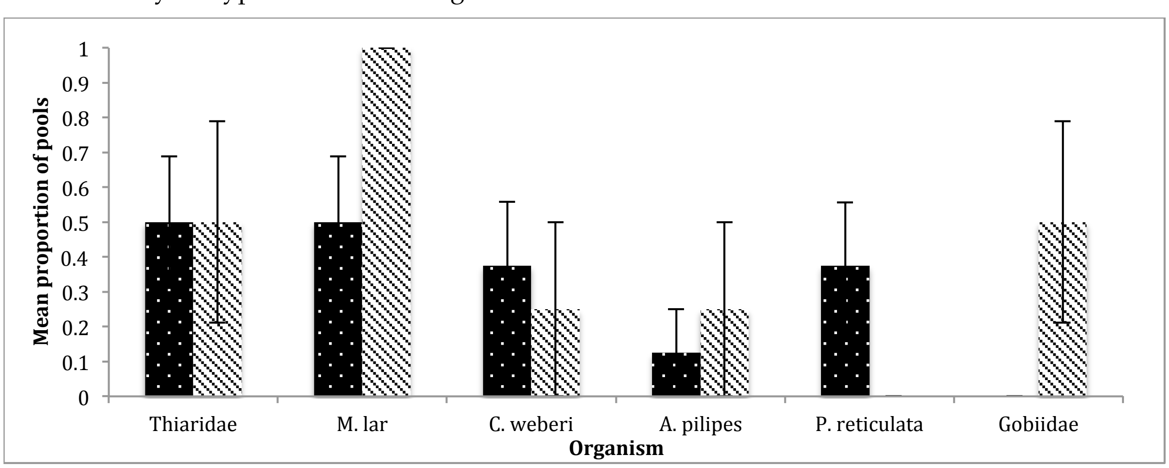

FIG. 9. Field Survey – Mean proportion of pools in which each type of organism was found at Site 1 (left) and Site 2 (right) (χ^2=69.5, df=10, p=0.01).

for 4 days, and this may not have been long enough for the relatively tough *M. calvescens* leaves to be sufficiently conditioned. Further research should be conducted into whether *M. calvescens* is a viable food source for Moorean stream invertebrates.

The fact that the *H. tiliaceus* flowers were preferred over the leaves of the same species shows that flowers, which are present in tropical stream litter year round, may actually be an important food source for stream invertebrates. The control cups for experiment 4, which contained only *H. tiliaceus* petals, had a large accumulation of green biofilm that was absent in all the experiments, indicating that the flowers stimulate a lot of microbial growth, which would explain why they are preferred over leaves. The role of flowers as a food source in streams has not been studied at all, and future studies could look at whether other types of flowers are also preferred over leaves. Field manipulations should also be carried out to verify that flowers in stream litter are consumed in nature, and not just in the lab.

The preference of *Angiopteris evecta* leaves over those of *H. tiliaceus* in experiment 8 was entirely unexpected. The *A. evecta* were tougher than those of *H. tiliaceus* and decomposed less quickly. More importantly, fern leaves are consumed by significantly fewer insects than the leaves of flowering plants and conifers, and are thought to contain compounds that deter insects from eating them (Copper-Driver 1978 and Gerson 1979). The combination of these factors suggests that *A. evecta* leaves must have some specific quality that makes them a preferred food source for stream invertebrates. This phenomenon is definitely an avenue for future research that could shed some light on the characteristics of ferns, and their relationship with insects and other invertebrates.

Unfortunately, experiments 5 and 6, which looked at the consumption of fruit, were both unsuccessful. In experiment 5, the *Inocarpus fagifer* fruit, being very tough and hard to break, was too damaged during preparation to detect signs of feeding. In experiment 6, the presence of acidic *Ananas comosus* in a small cup of water led to the death of most of the invertebrates. Although these experiments failed, the question they were addressing should be investigated further. Previous studies have shown that fruit can be an important component of the diet of tropical stream invertebrates, but have not examined whether sugary fruit is preferred over the microbial film that grown on decomposing leaves. (Larned et al. 2001).

The results of the field survey show that *Poecilia reticulate* was only found at Site 1, and the *Gobiid* fish were only found at Site 2, indicating that species composition of the pools varies with altitude. Physical factors such as flow rate and depth did not have an affect on the presence of macro invertebrates and vertebrates in the pools. However, as these factors can be changed substantially by even a single rainstorm (pers. obs.), it follows that the organisms living in the streams have adapted to the changing conditions. It is possible that these physical factors have an affect on the number of individuals of the different species.

Previous studies have found that the presence of 'shredder' invertebrates in tropical island streams may or may not have an affect on the rate of decomposition of plant litter (Crowl et al. 2001 and Rosemond et al. 1998). However, the experiments conducted in this study confirm that shrimp and snails in tropical island streams eat decomposing plant matter. It is likely that as these organisms have very little biomass compared to the amount of litter in the pools (pers. obs.), the amount of litter that they eat does not have an obvious effect on the overall breakdown of litter. However, while *Cararydina weberi* is a known shredder, and was always found in leaf packs, *Machrobrachium lar* is known as an omnivore, and *Atyoida pilipes* as a filter feeder (Resh et al. 1990 and Marquet 1991). It is possible that individuals of these species opportunistically ate leaves in the lab, but do not eat them in nature unless necessary. Field based studies looking at the feeding habits of these tropical stream invertebrates would contribute to understanding the importance of plant litter as a food source.

In conclusion, the results of these experiments confirmed some things that were previously known about the feeding habits of tropical stream invertebrates, while also finding new preferences, such as those for flowers and *A. evecta* leaves. There are many directions in which this study could be followed up, in order to further shed light on the unique but understudied tropical island stream system.

ACKNOWLEDGMENTS

I would like to thank my professors Brent Mishler, Vincent Resh, Stephanie Carlson, George Roderick and Jonathon Stillman for their advice and guidance. I would also to thank my GSIs, Christopher DiVittorio, Lindsey Dougherty and Julie Hopper for their help with study design and statistical analysis, as well as endless support and encouragement. Lastly, I am extremely grateful to my classmates for their help in the field, especially Caylee Falvo for sharing her shrimp with me.

LITERATURE CITED

Bastian, M., Boyero, L., Jackes, B.R. and Pearson, R.G. 2007. Leaf Litter Diversity and Shredder Preferences in an Australian Tropical Rain-Forest Stream. Journal of Tropical Ecology **23** (2): 219-229.

Cooper-Driver, G. A. 1978. Insect-fern associations. Entomol Exp. Appl. **24**: 310-316.

Cummins, K.W., Wilzbach, M.A., Gates, D.M., Perry, J.B. and W.B. Taliaferro. 1989. Shredders and Riparian Vegetation. Bioscience **39** (1): 24-30.

Davies, P.M., Bunn, S.E. and Hamilton S.K. 2008. Primary Production in Tropical Streams and Rivers. 23-42 *in* D. Dudgeon, editor. Tropical Stream Ecology. Academic Press.

Gade, K.J., 1993. Comparison of Leaf Tissue Losses by Riparian Trees and an Invading Plant in a Tropical Island Stream. UC Berkeley, Moorea Student Research Papers **2**: 342-359.

Gerson, U. 1979. The associations between Pteridophytes and arthropods. Fern Gaz. **12**: 29-45.

Google. 2013. Google Earth, Version 7. Retrieved from http://www.google.com/earth/.

Larned, S.T., Chong, C.T. and N. Punewai. 2001. Detrital Fruit Processing in a Hawaiian Stream Ecosystem. Biotropica. **33** (2): 241-248.

Marquet, G. 1991. Freshwater crustaceans of French Polynesia: taxanomy, distribution and biomass (Decapoda). Crustaceana **61**: 125-140.

Meyer, J. and J. Florence. 1996. Tahiti's native flora endangered by the invasion of *Miconia calvescens* DC. (Melastomataceae). Journal of Biogeography **23**: 775-781.

R Development Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna Austria. Retrieved from http://www.R-project.org/.

Resh V.H. and F.A. de Szalay. 1995. Streams and Rivers of Oceania. Pages 717-736 *in* C.E. Cushing, K.W. Cummins and G.W. Minshall, editors. River and Stream Ecosystems. Elsevier, New York, New York.

Resh, V.H., Barnes, J.R. and D.A. Craig. 1990. Distribution and Ecology of Benthic Macro Invertebrates in the Opunohu River Catchment, Moorea, French Polynesia. Annales de Limnologie **26**: 195-214.

Wantzen, K.M., Yule, C.M., Mathooko, J.M. and C.M. Pringle. 2008. Organic Matter Processing in Tropical Streams. Pages 43-61 *in* D. Dudgeon, editor. Tropical Stream Ecology. Academic Press.

Williams, J.L. 1999. The Feeding Ecology and Effects of the Tropical Freshwater Shrimp, *Caridina weberi (Atyidae).* UC Berkeley, Moorea Student Research Papers **8**: 183-190.

Wright, M.S. and A.P. Covich. 2005. Relative Importance of Bacteria and Fungi in a Tropical Headwater Stream: Leaf Decomposition and Invertebrate Feeding Preference. Microbial Ecology **49** (4): 536-546.