2020

Leaf Choice by Salmonfly Nymphs (Pteronarcys californica) in Western Montana

Emily L. Hamant
eh107306@umconnect.umt.edu

Art Woods
art.woods@mso.umt.edu

James Frakes
jameson.frakes@umontana.edu

Recommended Citation

Hamant, Emily L.; Woods, Art; and Frakes, James, "Leaf Choice by Salmonfly Nymphs (Pteronarcys californica) in Western Montana" (2020). *Undergraduate Theses and Professional Papers*. 276.
https://scholarworks.umt.edu/utpp/276

Leaf Choice by Salmonfly Nymphs (*Pteronarcys californica*) in Western Montana

Emily Hamant, University of Montana

W.A. Franke College of Forestry and Conservation

Jameson Frakes, Graduate Student, University of Montana

Art Woods, Faculty Mentor, University of Montana

Abstract

The giant salmonfly (*Pteronarcys californica*), the largest species of stonefly in the world, is vital to streams in western North America. Understanding their diet may shed light on interactions between invertebrates and common riparian trees such as alder (*Alnus* spp.), cottonwood (*Populus* spp.), chokecherry (*Prunus virginiana*), dogwood (*Cornus sericea*), and willow (*Salix* spp.). *P. californica* consume primarily fungal and microbial colonies on decomposing 'conditioned' leaves in streams. The rate and timing of leaf decomposition varies due to the toughness and chemistry of the leaves and the communities of microbes that colonize them. There are multiple species of leaves in the stream at any time, but they decompose with different dynamics. Leaves that decompose quickly disappear from the streams, leaving only tougher leaves during winter and the following spring and summer seasons. Because of this variability, *P. californica* may prefer leaves that decompose faster, such as chokecherry and alder, under short conditioning periods, but may prefer tough leaves, such as cottonwoods, under longer conditioning periods. We first asked whether stoneflies choose strongly among leaf type (based on either leaf species or conditioning time). We then assessed the consequences of those choices by measuring stonefly growth in no-choice assays (individuals given single leaf types). We also quantified leaf mass area for each leaf species as a proxy for toughness. This study showcases the complex interactions between riparian species and invertebrate growth. With the results of this study, restoration efforts on western streams may include planting a variety of riparian tree species to improve growth and winter survival of *P. californica*, and thus improve fisheries of western streams.

Introduction

In low-order headwater streams, the primary sources of energy for aquatic-based organisms are allochthonous (Oertli 1993), meaning that they arrive from primary production that occurs outside the stream. Leaf decomposition allocates these inputs into the stream through a three-step process: abiotic leaching, microbial colonization, and shedder consumption. Abiotic leaching releases hydrophilic compounds from the leaves into the stream (Cummins et al. 1989, Marks 2019, Schulze and Walker 1997). The rate of leaching varies due to the susceptibility of the compounds within the leaves (Campbell and Fuchshuber 1995, Swain 1979). Microbial colonization occurs when fungi and bacteria colonize the leaves (Cummins et al. 1989). Microbial colonization varies due to variation in the physical (e.g., leaf toughness) and chemical (e.g., polyphenolic concentrations) characteristics of riparian species (Anderson and Sedell 1979, Arsuffi and Suberkropp 1989, Canhoto and Graca 1999, Cummins 1989, Driebe and Whitman 2000). In turn, variation in colonization influences shredder consumption rates and preference of the leaves (Barlocher and Kendrick 1975, Kaushik and Hynes 1971, Schulze and Walker 1997). Shredders can clearly discriminate leaf types and prefer more conditioned leaves (Arsuffi and Suberkropp 1989, Bastian et al. 2007, Bueler 1984, Casotti et al. 2019, Golloday et al. 1983, Graca et al. 2001, Irons et al. 1988). Preference of shredders also causes variation in leaf decomposition due to varying rates of consumption and fragmentation (Swan and Palmer 2006), but rates of weight loss in leaves throughout the decomposition process are highly variable (Anderson and Sedell 1979, Casotti et al. 2019, Li et al. 2008). Studies show variation in shredder leaf preference is due to a combination of factors, such as microbial and fungal colonization, leaf type, and conditioning period (Bastian et al. 2007, Cummins and Klug 1979 and 1989, Graca et al. 1993). Choices by shredders may have direct consequences on aspects of fitness, including how fast they grow and how long they spend in stream. Few studies attempt to quantify the consequences of preference (Casotti et al. 2019, Connolly and Pearson 2013, Graca 2001), and none have demonstrated leaf preference for *Pteronarcys californica.*

P. californica, an herbivorous stonefly with a four-year instream life cycle (Townsend and Pritchard 1998), lives in large streams and rivers with swift currents and loose substrates throughout western North America (Baumann et al. 1977, Elder and Gaufin 1973, Stewart and Stark 2002). Stonefly life histories are linked to nutrient pulses (Anderson and Grafius 1979, Townsend and Pritchard 1998), and varying rates of leaf decomposition facilitates overwinter survival (Irons et al. 1988). Shredders experience resource bottlenecks due to few leaves falling into the stream in the spring, but leaves that decompose slowly may alleviate food shortages even if they do not contain high levels of nutrients (Muto et al. 2011). However, few studies quantify whether shredders prefer different leaf species as conditioning periods lengthen (Muto et al. 2011), and no studies showcase fitness consequences throughout Pacific Northwest or Mountain stream ecosystems.

Rock Creek, located near Clinton, Montana, is a relatively pristine mountain watershed ecosystem, with five main riparian species in its lowest reaches: willow (*Salix* spp.), alder (*Alnus* spp.), chokecherry (*Prunus virginiana*), dogwood (*Cornus sericea*), and cottonwood (*Populus* spp.) dominating litter inputs. Leaves of *Populus* spp., an especially dominant species through this lower reach, can be found in the benthic zone of Rock Creek throughout the year (*personal observation). P. californica* is abundant throughout this stream system. In this study, I measured the relative preference of *P. californica* for different species of instream leaves, whether those preferences depend on conditioning time, and what the consequences of the choices are for insect growth. I conducted trials in which leaves under different conditioning periods are given to *P. californica* to quantify how leaf conditioning affects leaf preference and growth. I also determined leaf toughness for each species using leaf mass area under each conditioning period to explain the underlying causes of preference for *P. californica.* I hypothesized that *P. californica* will prefer softer leaves under little conditioning times and tougher leaves under longer conditioning periods.

Methods

Leaf Conditioning

We collected leaves of five riparian species: *Salix* spp., *Alnus* spp., *P. virginiana, C. sericea,* and *Populus* spp., from the last three miles of Rock Creek before the confluence with the Clark Fork River, either from the ground or from trees in late January. We conditioned leaves in a five-gallon bucket in separate mesh bags in a Percival incubator, model 166LL, at 12°C for 7 and 25 days with water collected from Rock Creek, and changed the water every ten days.

Growth of P. californica

We weighed (grams) and placed three individuals of *P. californica* in 9.5-ounce rectangular Tupperware with excess leaves of each riparian species. We placed the containers in a double door Percival incubator, model 166LL, for fourteen days at 12°C under a 12-hour diurnal cycle. We inserted a Dannex air pump manifold system tube into the water to aerate each container. We changed the water in each container every 7 days. Each leaf species treatment had four replicates under two leaf conditioning times: 7 days and 25 days.

Leaf Preference by P. californica

We conditioned leaves of each riparian species in mesh bags for 7 and 25 days (see Leaf Conditioning). We measured leaf preference in a series of two-choice experiments. Three individuals of *P. californica* were placed in an eight-ounce Surefresh circular Tupperware container with two types of leaves, and held in a Percival incubator, 166LL, at 12^{0}C in a 12-hour diurnal cycle. We aerated each container with a manifold tube connected to a Dannex air pump. For 10 days, each morning we noted which leaf type *P. californica* rested on. We had four replicates of each two-leaf choice experiment, each compared one riparian species against *Populus* spp. After 10 days, we removed *P. californica*.

Leaf Mass Area

Leaf Mass Area, used as a proxy for toughness, is computed as area divided by dry mass. We photographed and analyzed area for each leaf sample in ImageJ. We dried each sample at 60^{o}C for 24 hours and then immediately weighed each leaf.

Consumption

We estimated how much leaf area each stonefly ate by measuring the difference in leaf area between the beginning and end of each preference experiment. Before the start of each preference experiment, we imaged each leaf to measure leaf area using ImageJ. After 10 days, we took photos of the remaining leaves and measured remaining leaf area using ImageJ.

Analysis

We measured leaf consumption in ImageJ as a function of leaf area lost from initial leaf area for each leaf species in a two-leaf choice experiment and tested for a difference using Student's t-test. We completed all analyses in R version 3.4.1 (R Core Team 2017). We analyzed leaf preference using a chi-square test expecting random location for each individual. We analyzed growth using an ANOVA test for a difference in group means. We analyzed growth against initial weight using a linear model.

Results

Growth

Under seven days of conditioning, we found no evidence of a difference in growth in *P. californica* between leaf species (ANOVA, F=0.455, and P=0.768, Fig. 1). On average, individuals gained 0.0192g when consuming *Alnus* spp. leaves, 0.0091g when consuming *P. virginiana* leaves, 0.0104 g when consuming *Populus* spp., 0.0058g when consuming *C. sericea* leaves, and 0.0137g when consuming *Salix* spp. There is no evidence of an interaction between treatment type and initial weight, therefore, treatment type did not affect growth rate of individuals (F= 1.21, P=0.1633 on 9 and 50 df, Table 1, Fig. 2). Individually, both *P. virginiana* and *C. sericea* negatively affected growth (P=0.0625 and 0.0287, respectively). There was no difference in growth between large (greater than 0.37 g) and small individuals (ANOVA, P=0.2179 with 58 df).

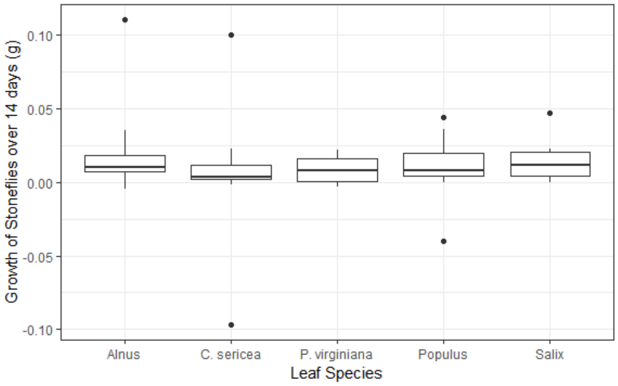

Figure 1. 14-day Growth difference (g) of P. californica given excess of 7-day conditioned leaves

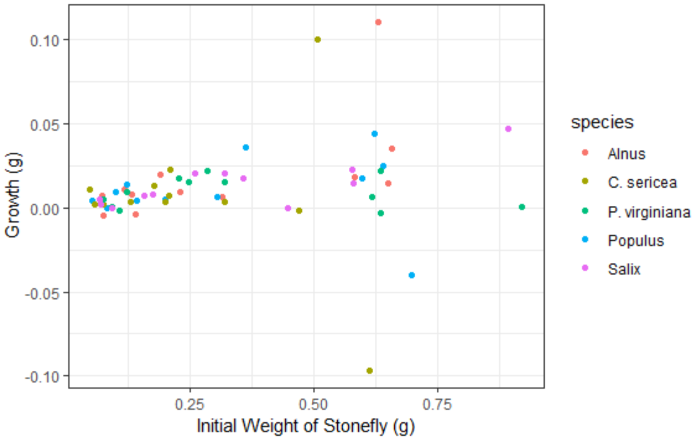

Figure 2. Initial weight (g) v. Growth (g) of P. californica given excess of 7-day conditioned leaves

Table 1. ANOVA Table summarizing main effects and interaction from the linear model fitted to data shown in Figure 2.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Initial mass	1	0.001475	0.00147539	2.2957	0.1360
Species	4	0.001139	0.00028476	0.4431	0.7769
initial:species	4	0.004387	0.00109665	1.7064	0.1633
Residuals	50	0.032134	0.00064268		

Likewise, under 25 days of conditioning, we also found no evidence of a difference in average growth between leaf treatment types (ANOVA, F=0.2203, P=0.926, Fig. 3). On average, individuals grew 0.0007g when consuming *Alnus* spp., 0.0023g when consuming *P*. virginiana, 0.0032g when consuming *Populus* spp. leaves, and 0.0059g when consuming *C. sericea* leaves. On average, individuals shrunk by 0.0003g when consuming *Salix* spp. We also found no difference in growth between individuals consuming leaves that had been conditioned for 25 days and leaves that had been conditioned for 7 days (F=1.1023, P=0.4091). We found that initial weight is a strong predictor of growth of *P. californica* when feeding on 25-day conditioned leaves (ANOVA, P=0.00120, Table 2), regardless of the type of leaf consumed by *P. californica* (ANOVA, P=0.497, Table 2). One *Salix* spp. treatment had an aeration tube malfunction on day 13. Conditioning time was a strong predictor of growth (P=0.01702, Table 3). There was also a significant interaction between initial weight of the stoneflies and the conditioning period, indicating that initial weight influences growth of *P. californica* differently, depending on conditioning time (ANOVA, P=0.00202, Table 3). During this experiment, two individuals molted in a *Populus* spp. treatment, and one individual molted in a *P. virginiana* treatment.

Table 2. ANOVA Table summarizing main effects and interaction from the linear model fitted to data shown in Figure 4.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Initial mass	1	0.0031496	0.003150	11.8045	0.001198
Species	4	0.0004037	0.000101	0.3783	0.823064
Initial mass: species	4	0.0009131	0.000228	0.8556	0.497068
Residuals	50	0.0133408	0.000267		

Table 3. ANOVA table summarizing main effects and interaction from the linear model fitted to data shown in Figure 5.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Initial mass	1	0.000051	0.000051	0.1131	0.73720
Conditioning	1	0.002644	0.002644	5.8626	0.01702
Initial mass: Conditioning	1	0.004500	0.004501	9.9786	0.00202
Residuals	116	0.052317	0.000451		

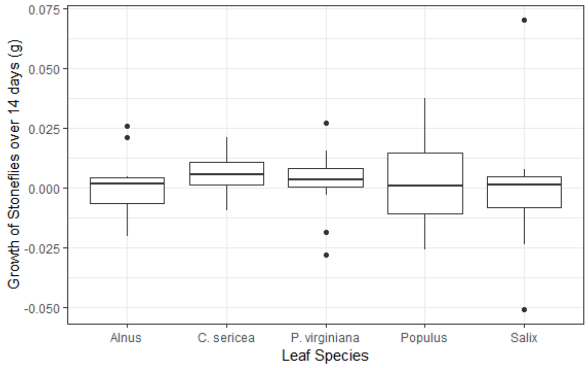

Figure 3. 14-day Growth difference (g) of P. californica given excess of 25-day conditioned leaves

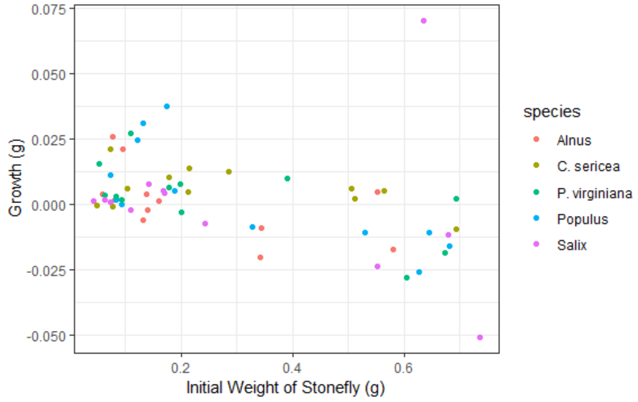

Figure 4. Initial weight (g) v. Growth (g) of P. californica given excess of 25-day conditioned leaves

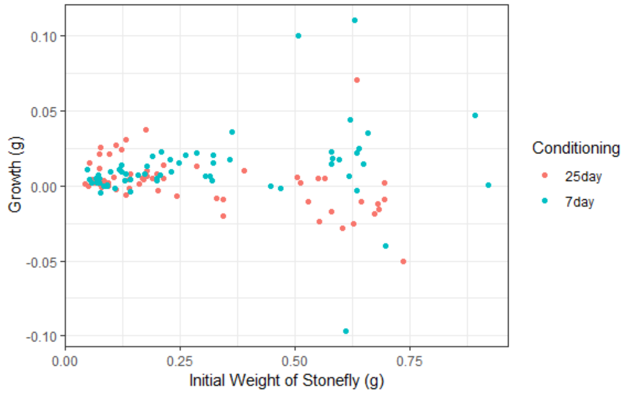

Figure 5. Growth (g) v. Initial Weight (g) of P. californica, grouped by period of leaf conditioning

Preference

Preference was measured as daily location of individuals on each leaf type. Under seven days of conditioning, *P. californica* preferred *Alnus* spp. over *Populus* spp. (Chi-square test, P<0.0001 under 1 df, Table 5), but preferred *Populus* spp. over *P. virginiana*, *C. sericea*, and *Salix* spp. (Chi-square test, P<0.0001, P=0.1025, and P<0.00001, respectively, under 1 df, Tables 6-8).

Under twenty-five days of conditioning, there was no difference in preference of *P. californica* between *Alnus* spp. and *Populus* spp. (Chi-square test, P<0.4652 under 1 df, Table 9). *P. californica* preferred *C. sericea* over *Populus* spp. (Chi-square test, P=0.068 under 1 df, Table 10). *P. californica* continued to prefer *Populus* spp. over *P. virginiana* and *Salix* spp. (Chi-square test, P<0.00001 for both treatments under 1 df, Table 11 and 12, respectively). Under both conditioning periods, there is evidence of a difference in preferred location of *P. californica*, where individuals prefer *Populus* spp. over other species, excluding *Alnus* spp.

Table 5. Chi-square table of location of P. californica *given two 7-day conditioned leaf types for* Alnus *spp.*

Species	*Alnus* spp	*Populus* spp.	
Probability	0.5	0.5	
Observed	84	36	
Expected	60	60	
X^2 Statistic			19.2

Table 6. Chi-square table of location of P. californica *given two 7-day conditioned leaf types for* P. virginiana

Species	*Prunus virginiana*	*Populus* spp.	
Probability	0.5	0.5	
Observed	6	33	
Expected	19.5	19.5	
X^2 Statistic			18.69

Table 7. Chi-square table of location of P. californica *given two 7-day conditioned leaf types for* C. sericea

Species	*C. sericea*	*Populus* spp.	
Probability	0.5	0.5	
Observed	21	33	
Expected	27	27	
X^2 Statistic			2.67

Table 8. Chi-square table of location of P. californica *given two 7-day conditioned leaf types for* Salix *spp.*

Species	*Salix* spp.	*Populus* spp.	
Probability	0.5	0.5	
Observed	7	68	
Expected	37.5	37.5	
X^2 Statistic			49.61

Table 9. Chi-square table of location of P. californica *given two 25-day conditioned leaf types for* Alnus *spp.*

Species	*Alnus* spp.	*Populus* spp.	
Probability	0.5	0.5	
Observed	64	56	
Expected	60	60	
X^2 Statistic			.533

Table 10. Chi-square table of location of P. californica *given two 25-day conditioned leaf types for* C. sericea

Species	*C. sericea*	*Populus* spp.	
Probability	0.5	0.5	
Observed	70	50	
Expected	60	60	
X^2 Statistic			3.333

Table 11. Chi-square table of location of P. californica *given two 25-day conditioned leaf types for* Prunus virginiana

Species	*Prunus virginiana*	*Populus* spp.	
Probability	0.5	0.5	
Observed	18	102	
Expected	60	60	
X^2 Statistic			58.8

Table 12. Chi-square table of location of P. californica *given two 25-day conditioned leaf types for* Salix *spp.*

Species	*Salix* spp.	*Populus* spp.	
Probability	0.5	0.5	
Observed	6	104	
Expected	55	55	
X^2 Statistic			87.309

Consumption

Under seven days of conditioning, there is moderately strong evidence of a difference in leaf consumption by *P californica* (ANOVA, F=2.821, P=0.0271, Figure 6). Between *Alnus* spp. and *Populus* spp., *P. californica* consumed more *Alnus* spp. leaves (average of 35.0 cm^2) than *Populus* spp. leaves (average of 13.4 cm^2) (t-test, P= 0.03619 under 6 df). In *P. virginiana* treatments, *P. californica* consumed an average of 28.4 cm^2 of *Populus* spp. leaves, compared to 20.5 cm^2 of *P. virginiana* leaves. Individuals consumed more *Populus* spp. than *P. virginiana* (t-test, P=0.05463 under 6 df), and more *Populus* spp. than *Salix* spp. (t-test, P=0.00135 under 6 df), where individuals consumed 32.8 cm^2 of *Populus* spp. leaves compared to 15.1 cm^2 of *Salix* spp. leaves. *P. californica* consumed 29.2 cm^2 of *Populus* spp. leaves in *C. sericea* treatments, and 26.2 cm^2 of *C. sericea*, and there was no evidence of preference in this choice treatment (t-test, P=0.3439 under 6 df). All *C. sericea* and all *P. virginiana* leaves were consumed, on average, in 3.88 days, and all *Salix* spp. leaves were consumed by 6 days.

Under 25 days of conditioning, there is suggestive evidence of a difference in consumption by *P. californica* between each leaf species (ANOVA, P=0.108, Fig. 7). In *Alnus* spp. treatments, *P. californica* consumed an average of 28.0 cm^2 of *Populus* spp. leaves, and an average of 31.2 cm^2 of *Alnus* spp. leaves, and there was no evidence of a difference in consumption between leaf species (Welch's t-test, P= 0.3808 under 3.3777 df). There was no difference in consumption in *P. virginiana* choice treatments (Welch's t-test, P=0.5068 under 4.7044 df), where *P. californica* consumed an average of 21.1 cm^2 of *P. virginiana* leaves and an average of 20.8 cm^2 of *Populus* spp. leaves, nor a difference in *C. sericea* choice treatments (Welch's t-test, P=0.4178 under 5.864 df), where individuals consumed and average of 9.0 cm^2 and 7.7 cm^2 of *Populus* spp. and *C. sericea* leaves, respectively. In *Salix* spp. treatments, there was strong evidence of a preference for *Salix* spp. (Welch's t-test, P=0.008445, under 5.8083 df), where individuals consumed an average of 13.0 cm^2 of *Populus* spp. compared to 20.5 cm^2 of *Salix* spp.

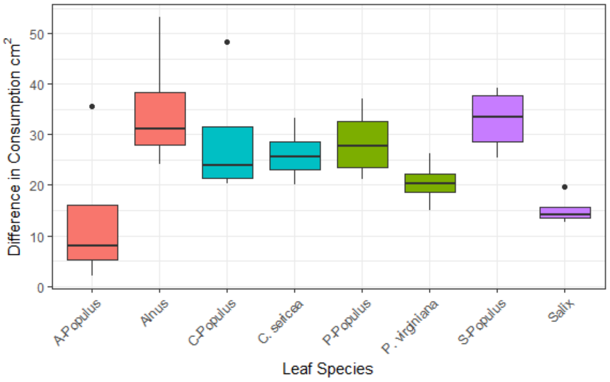

Figure 6. Area Consumed (cm^2) of Each Leaf Type after 7 days of conditioning.

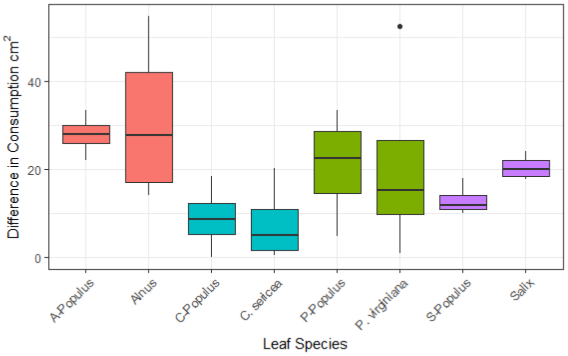

Figure 7. Area Consumed (cm^2) of Each Leaf Type after 25 days of conditioning.

Leaf Mass Area

Under 7 days of conditioning, leaf mass area, a proxy for toughness, ranged from 4.9×10^{-5} g/cm^2 to 9.9×10^{-5} g/cm^2. *Salix* spp. had the highest leaf mass area, followed by *P. virginiana*, *Alnus* spp., *C. sericea*, and *Populus* spp. Under 25 days of conditioning, leaf mass area ranged from 3.8×10^{-5} g/cm^2 to 8.6×10^{-5} g/cm^2. *Alnus* spp. had the highest leaf mass area, followed by *C. sericea*, *Populus* spp., *Salix* spp., and *P. virginiana*. Leaf mass area increased for both *Alnus* spp. and *Populus* spp., indicating these leaves became more tough as conditioning period lengthened.

Table 13. Leaf Mass Area for Leaf Species after 7 and 25 days of conditioning

Species	LMA 7 days (g/cm^2)	LMA 25 days (g/cm^2)
Salix spp.	9.9×10^{-5}	4.8×10^{-5}
Alnus spp.	8.2×10^{-5}	8.6×10^{-5}
P. virginiana	9.3×10^{-5}	3.8×10^{-5}
Populus spp.	4.9×10^{-5}	5.4×10^{-5}
C. sericea	7.4×10^{-5}	7.1×10^{-5}

Discussion

P. californica shifts its preference in consumption of leaf species as conditioning period lengthens. This shift does not result in a growth advantage, nor is it caused by a shift in leaf toughness, indicating there are other factors dictating this change of preference by *P. californica*. Since there were no differences in growth between leaf types, *P. californica* could require a varied diet to grow and complete its life cycle. This experiment showcased a need for future experiments examining how other factors, such as leaf compounds and microbial colonization during leaf decomposition, affects leaf preference by *P. californica.*

As shown by these data, *P. californica* prefers to generally consume non *Populus* spp. leaves over *Populus* spp. leaves, excluding *Alnus* spp., when the leaves have been conditioned for a short period of time. This difference in consumption homogenizes as conditioning period lengthens. This shows that *P. californica* shifts its leaf preference depending on the length of conditioning the leaves have undergone. This change in preference as conditioning period lengthens does not follow the change in leaf toughness, indicating that other factors during the leaf decomposition process could drive this change in consumption, such as a change in microbial community composition or a change in the concentration of leaf compounds as conditioning continues.

These differences in preference are the opposite of what we hypothesized – that *P. californica* will prefer softer leaves as conditioning times lengthens. This difference could be because leaf toughness is not indicative of the types of compounds found within the leaves. Other compounds, such as lignins and tannins, could drive preference of shredders more so than leaf toughness (Leite-Rossi et al. 2016). These compounds stay within leaves for at least 30 days (Casotti et al. 2019), and therefore a difference in leaf preference due to a change in lignin and tannin concentrations would not be observed during this experiment.

P. californica preferred to rest on larger leaves, such as *Populus* spp. and *Alnus* spp. during all experiments, although they did not consume these species as rapidly. The difference between location preference and food preference shows that the location of *P. californica* cannot be used as a proxy for what they consume. *P. californica* could prefer to rest on larger leaves because these leaves could decrease chances of dislodgement and could provide more protection from predators. *P. californica* has the highest chances of predation during dislodgement since they live in swift moving streams, which provides them little opportunity to reattach to debris once dislodged.

P. californica showed no advantage in growth due to their preference in consumption. These insects did not show large increases in growth after consuming a specific leaf type, which indicates that growth is not a fitness consequence to preference. In fact, larger nymphs decreased in body mass after consuming leaves that conditioned for a longer period. This could be a relic of *P. californica*'s life history, as this trend, where smaller individuals steadily gain weight and larger individuals are more variable, has been seen in other experiments (*pers. comm.)*. Due to their four year life cycle within the stream, smaller nymphs could focus more on growth during the winter months, whereas larger nymphs could be entering a phase of maturation and focus more on building adult body parts. However, 0.37 grams still is relatively small (many late stage nymphs weigh more than 1 gram). This could indicate that strictly one leaf species does not constitute an adequate diet for *P. californica*.

This study shows that *P. californica* could require a varied diet to grow, instead of a diet consisting of one species. This is vital information as managers improve riparian habitats through restoration and bioremediation efforts. A more biodiverse riparian corridor with high species richness improves not only terrestrial ecosystem functions, but also has potential positive consequences within the aquatic system.

References

Anderson, N.H., J.R. Sedell. (1979). Detritus Processing by Macroinvertebrates in Stream Ecosystems. *Annual Review of Entomology*. **24:**351-377.

Anderson, N.H., E. Grafius. (1979). Population Dynamics, Bioenergetics, and Role of Lepidostoma Quercina Ross (Trichoptera:Lepidostomatidae) in an Oregon Woodland Stream. *Ecology*. **60(2):** 433-441.

Arsuffi, T.L., K. Suberkropp. (1989). Selective Feeding by Shredders on Leaf-Colonizing Stream Fungi: Comparison of Macroinvertebrate Taxa. *Oecologia.* **79(1):** 30-37.

Barlocher, F., B. Kendrick. (1975). Leaf-Conditioning by Microorganisms. *Oecologia*. **20(4):** 359-362.

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

Baumann, R.W., A.R. Gaufin, R.F. Surdick. 1977. The stoneflies (Plecoptera) of the Rocky Mountains. *Memoirs of the American Entomological Society*. **31**: 1–208.

Bueler, C.M. (1984). Feeding Preference of Pteronarcys pictetii (Plecoptera: Insecta) from a Small, Acidic,Woodland Stream. *The Florida Entomologist*. **67(3):** 393-401.

Canhoto, C. M.A.S. Graca. (1999). Leaf Barriers to Fungal Colonization and Shredders (*Tipula lateralis*) Consumption of Decomposing *Eucalyptus globulus. Microbial Ecology.* **37:** 163-172.

Casotti, C.G., et. al. (2019). The longer the conditioning, the better the quality? The effects of leaf conditioning time on aquatic hyphomycetes and performance of shredders in a tropical stream. *Aquatic Ecology.* **53:**163-178.

Connolly, N.M., R.G. Pearson. (2013). Nutrient enrichment of a heterotrophic stream alters leaf litter nutritional quality and shredder physiological condition via the microbial pathway**.** *Hydrobiologia*. **718**:85–92.

Cummins, K.W., M.J. Klug. (1979). Feeding Ecology of Stream Invertebrates. *Annual Review of Ecologic Systems.* **10:** 147-172.

Cummins, K.W., et. al. (1989). Shredders and Riparian Vegetation. *Bioscience.* **39(1):**24-30.

Driebe, E.M., T.G. Whitham. (2000). Cottonwood Hybridization Affects Tannin and Nitrogen Content of Leaf Litter and Alters Decomposition. *Oecologia*. **123(1):** 99-107.

Elder, J.A., A.R. Gaufin. (1973). Notes on the occurrence and distribution of *Pteronarcys californica* Newport (Plecoptera) within streams. *Great Basin Naturalist*. **33**: 218–220.

Graca, M.A.S., L. Maltby, P. Calow. (1993). Importance of fungi in the diet of *Gammarus pulex* and *Asellus aquaticus* I: feeding strategies. *Oecologia.* **93**: 139-144.

Graca, M.A.S., et. al. (2001). Food quality, feeding preferences, survival and growth of shredders from temperate and tropical streams. *Freshwater Biology.* **46:** 947-957.

Irons et al. (1988). Consumption of leaf detritus by a stream shredder: Influence of tree species and nutrient status. *Hydrobiologia.* **160**: 53-61.

Golladay, S.W., J.R. Webster, E.F. Benfield. (1983). Factors Affecting Food Utilization by a Leaf Shredding Aquatic Insect: Leaf Species and Conditioning Time. *Holarctic Ecology*. **6(2):** 157-162.

Kaushik, N.K., and H. B. N. Hynes. 1971. The fate of the dead leaves that fall into streams. Arch. Hydrobiol. 68: 465-515.

Li, Aggie O.Y., Lily C.Y. Ng, D. Dudgeon. (2008). Effects of Leaf Toughness and Nitrogen Content on Litter Breakdown and Macroinvertebrates in a Tropical Stream. *Aquatic Sciences.* **71:** 80-93.

Leite-Rossi, L.A., Saito, V.S., Cunha-Santino, M.B. *et al.* (2016). How does leaf litter chemistry influence its decomposition and colonization by shredder Chironomidae (Diptera) larvae in a tropical stream?. *Hydrobiologia.* **771:**119–130.

Muto, E.A., D.P. Kreutzweiser, P.K. Sibley. Over-winter decomposition and associated macroinvertebrate communities of three deciduous leaf species in forest streams on the Canadian Boreal Shield. *Hydrobiologia*. **658**:111–126.

Oertli, B. (1993). Leaf litter processing and energy flow through macroinvertebrates in a woodland pond (Switzerland). *Oecologia*. **96:**466-477.

R Core Team (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Schulze, D.J., K.F. Walker. (1997). Riparian Eucalypts and Willows and their Significance for Aquatic Invertebrates in the River Murray, South Australia. *Regul. Rivers: Res. Mgmt.* **13:** 557–577.

Stewart, K.W., B.P. Stark. 2002. Nymphs of North American stonefly genera (Plecoptera). *Caddis Press.* Columbus, OH.

Swan, C.M., M.A. Palmer. (2005). Preferential feeding by an aquatic consumer mediates non-additive decomposition of speciose leaf litter. *Oecologia*. **149(1):**107-114.

Townsend, G.D., G. Pritchard. (1998). Larval growth and development of the stonefly *Pteronarcys californica* (Insecta: Plecoptera) in the Crowsnest River, Alberta. *Canadian Journal of Zoology*. **76(12):** 2274-2280.