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## Feeding Ecology of Two Plecopterans in Low Order Andean-Patagonian Streams

*key words:* feeding habits, leaf litter, periphyton, trait plasticity, functional ecology

### Abstract

Feeding plasticity of the Andean plecopteran *Klapopteryx kuscheli* and *Notoperla archiplatae* larvae was assessed through a field experiment using enclosures. *K. kuscheli* has previously been described as a shredder and *N. archiplatae* as a scraper. Further information on gut contents from different populations supported those results. In the experiment, larvae of both species were exposed to contrasting food items: leaf litter and periphyton. Consumption, growth and the efficiency of food conversion were measured. *K. kuscheli* was able to feed on periphyton, though it did not grow. *N. archiplatae* failed to feed on leaf litter. While *K. kuscheli* might be considered a facultative shredder, *N. archiplatae* functions as a specialist scraper. The natural distribution and seasonal abundance in two small streams showed contrasting habitat use of both species. *N. archiplatae* inhabited high velocity runs and riffles underneath large substrates while *K. kuscheli* presented a higher habitat plasticity. Implications of those results for ecosystem function are discussed.

### 1. Introduction

The study of feeding habit and food resource assimilation of aquatic invertebrates contributes to the understanding of stream processes by linking basic food categories to higher trophic levels (MIHUC, 1997). This functional approach involves two ecological aspects: one is associated to food intake, and the other concerns matter and energy transfer through food webs. Morpho-behavioral adaptations in invertebrate food acquisition systems are expected to be less selective than biochemically-based assimilation systems (CUMMINS and MERRITT, 1996). But available food for invertebrates is just a fraction of the potential food resource present in a system to which organisms can access. We understand feeding plasticity of an organism as its capability of displaying a wide range of food acquisition among available resources. True availability means no interference (e.g. mechanical) is involved that automatically excludes some of the materials without any morpho-behavioral specialization of the species (CUMMINS, 1973). Although omnivory in benthic invertebrates is more common than previously thought (FRIBERG and JACOBSEN, 1999; LEDGER and HILDREW, 2001), not all food items ingested are likely to be transformed into body tissue. Growth measurement and efficiency of food conversion estimates indicate to what extent organisms match contrasting food resources. Species that show significant body growth (MIHUC and MINSHALL, 1995) or significant assimilation efficiency (CUMMINS and MERRITT, 1996) when

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fed on more than one food category are referred to as generalists, facultative generalists or opportunistic feeders.

Functional classification of aquatic organisms by their feeding relationships (e.g. PALMER *et al.*, 1993a and b) enhances our knowledge of trophic dynamics in streams because they simplify the community into trophic subunits, interpreting process oriented ecosystem questions (CUMMINS, 1974; MERRIT *et al.*, 2002; BLONDEL, 2003). The method assumes the association between a limited set of feeding adaptations found in freshwater invertebrates and their basic nutritional resource categories (CUMMINS, 1996). The studies of McSHAFFREY and McCAFFERTY (1988), PALMER and O'KEEFE (1992) and PALMER *et al.* (1993a and b) concluded that feeding behavior, gut content and mouthpart morphology (or additional body parts) analyses provide the most certain FFG (functional feeding group) designations. On the other hand, MIHUC and MIHUC (1995) stressed that food habits do not necessarily reflect an accurate assessment of functional trophic relationships for benthic invertebrates since most taxa may function as generalists even when reported as specialists. Recently, the stable isotopes technique has been applied to feeding ecology studies as subrogate of resource assimilation; however, the technique is only applicable when food resource isotopic signatures contrast to each other and gut content analysis are still needed to link resources to consumers (FÜREDER *et al.*, 2003).

The feeding ecology of Andean stream invertebrates has not yet been intensively addressed. Most of the references to feeding relationship were based on studies that have basically analyzed gut content and mouthpart morphology of invertebrates (WAIS and BONETTO, 1988; WAIS, 1990; DE CABO and WAIS, 1991; DÍAZ VILLANUEVA and ALBARIÑO, 1999; MISERENDINO, 2000; ALBARIÑO and BALSEIRO, 2002; VELÁZQUEZ and MISERENDINO, 2003). The study of feeding behavior has recently commenced in order to enhance functional designations and provide the ecological roles of invertebrates in freshwaters (ALBARIÑO and VALVERDE, 1998; ALBARIÑO, 2001; DÍAZ VILLANUEVA *et al.*, 2004). The food habits of the Andean plecopteran *Klapopteryx kuscheli* (ILLIES, 1960) and *Notoperla archiplatae* (ILLIES, 1958) have been previously assessed (DÍAZ VILLANUEVA and ALBARIÑO, 1999; ALBARIÑO, 2001). Although their mouthpart morphologies suited the Type I (detritivore-herbivore condition) *sensu* STEWART and STARK (1988), both species displayed contrasting trophic habits in nature. *K. kuscheli* was identified as detritivore-shredder while *N. archiplatae* as periphyton brusher-scraper.

The main goal of this study was to test to what extent *K. kuscheli* and *N. archiplatae* were capable of exploiting and assimilating different food resources. We experimentally tested whether such taxa previously assigned to one FFG were able to display a generalist or facultative strategy. We checked previous gut content field data analyzing a set of larval gut contents of both species from other populations (i.e. different catchments). Additionally, we studied their distribution and seasonal abundance by surveying the benthic assemblage in two low order streams.

## 2. Materials and Methods

### 2.1. Study Area

The study was carried out in two low order stream tributaries of the Ñireco basin belonging to Nahuel Huapi lake catchment (41°13' S; 71°20' W). The streams were called A1 and A2 and physical characteristics mainly differ in basin area (Table 1). They drain a Southern mountain beech (*Nothofagus pumilio*) forest. In Patagonian Andes, this deciduous beech constitutes the highest mountain belt of the temperate forest, up to the timberline (HILDEBRAND-VOGEL *et al.*, 1990). In the area, human activity above 1000 m a.s.l. is restricted to hiking. Peak discharges are in fall-winter (rain) and spring (snowmelt) and the lowest values late in summer.

Table 1. Characteristics of the studied streams in Ñireco basin.

Streams	A1	A2
Catchment Area (ha)	25.6	69.8
Total Length (m)	828	1252
Outlet Altitude (m a.s.l.)	1083	1065
Water Temperature (°C)	Min	3.0
	Max	16.0
Conductivity ( $\mu\text{S cm}^{-1}$ )	49	55
Stream Width (m)	0.4–2.7	1.6–4.2
Dominant Substrate	boulder-cobble	
Dominant Morphology	riffles alternating with step pools	

Absolute and relative abundance of *K. kuscheli* and *N. archiplatae* in the macroinvertebrate community of these streams were studied in July (winter) and November (spring) 1996, January (summer) and April (autumn) 1997. Six replicates were taken at each sampling site (ca. 1100 m a.s.l.) with a Surber sampler (0.09 m<sup>2</sup>) and samples fixed *in situ* with 5% formalin. In the laboratory, the different taxa were identified, counted and dried at 80 °C for 48 h to obtain dry mass.

## 2.2. The Invertebrates

Both taxa belong to endemic Patagonian genera and are common species in low order streams of the South Andes and the Coast Ranges (Argentina and Chile). *K. kuscheli* spans from near 38° to 51° S, where it has been found in piedmont streams at low altitude (ca. 300 m a.s.l., MCLELLAN, 2001; CAMPOS *et al.*, 1984). However, at low latitude it is limited to low order reaches (WAIS, 1987; ALBARIÑO, 1997; MISERENDINO, 2000; ALBARIÑO, pers. obs.). *N. archiplatae* has a more restricted latitudinal distribution (33° to 41° S) (ILLIES, 1963) as well as narrower habitat preferences. For instance, *N. archiplatae* is absent in low reaches of piedmont streams and rivers (ILLIES, 1963; WAIS, 1990; ALBARIÑO, 1997; MISERENDINO, 2000). While the taxonomy of the genus *Klapopteryx* (Austroperlidae) has recently been revised (MCLELLAN, 2001), the genus *Notoperla* (Gripopterygidae) is currently under study (IAN MCLELLAN, pers. comm.).

## 2.3. Experimental Design

The experiment to compare feeding plasticity of the plecopterans *K. kuscheli* and *N. archiplatae* was conducted in A2 stream, as both species were found there. Macroinvertebrates were collected in a stream reach below the experimentation site by picking up stones and dislodging from the underside or collecting larvae within leaf litter in a white tray. All larvae were measured in the field under a stereo-microscope with an eyepiece micrometer to estimate initial body mass. Total body length and head width were measured to the nearest 0.05 and 0.02 mm, respectively. Larvae of *K. kuscheli* ( $23.6 \pm 0.3$  mm; mean  $\pm$  SE) were almost twice the size of *N. archiplatae* individuals ( $11.9 \pm 0.2$  mm), and were in the range of intermediate to late instars.

The trial consisted in offering two single food resources to both plecopterans in order to measure their capability of feeding just on a single food resource and also their ability to survive and grow under these conditions. Two food sources were performed: 1) periphyton and 2) leaf litter. These were selected because they are the most representatives in moderate-fast flowing habitats where these species live: periphyton being more abundant on exposed stone surfaces and plant leaves being retained between and below stone interstices. Experimental units in periphyton treatment consisted of an 8  $\times$  8 cm unglazed ceramic tile, which served as biofilm substrate and to study the grazing activity of chosen species. Each tile was enclosed lengthwise in a transparent plastic tube (10 cm length and 8 cm in diameter). The experimental chambers in the leaf litter treatment consisted of plastic tubes of the same size, but opaque (to avoid natural biofilm development) supplied with leaf litter. Both ends of the chambers were closed

with 1 mm mesh size net tied with elastic bands and placed horizontally on the stream bottom with one end facing the current.

In each leaf litter chamber, we introduced 203 mg dry mass of non-pre-dried senescent undamaged *Nothofagus pumilio* leaves that were pre-hydrated to permit handling without damaging. They were held by the petioles with a plastic strand and fixed at the centre of the chamber to avoid flow obstruction inside. Leaves were collected from the forest soil at abscission time, in the stream surroundings. In the laboratory, a leaf litter fresh mass (FM) – dry mass (DM) regression was established to estimate initial dry mass of leaf packs. Fresh and dry masses of leaves were obtained by drying material at 40 °C for 24 h or at 105 °C for 48 h, respectively.

Each feeding chamber was replicated 10 times for each plecopteran species. Controls for periphyton growth and leaf litter decay were run without larvae ( $n = 6$ ). In addition, opaque chambers without food addition served as larval growth Control ( $n = 6$ ).

All experimental units were introduced in the stream on March 3<sup>rd</sup> 2000. All units were randomly attached to three metal frames placed along a 60 m reach at three riffle sites. Chambers were left 25 days prior to the inclusion of invertebrates for periphyton colonisation and leaf litter conditioning. On March 28<sup>th</sup> larvae were introduced, one in each chamber. Simultaneously, six experimental units of each treatment were removed as initial Controls. They were taken to the laboratory in individual plastic containers, thermally isolated and in total darkness. During the experiment, periphyton, sediments and fine particulate organic matter accumulation at the screens on both ends were removed every third day with a nylon brush to avoid clogging. After 44 days of experimentation, all experimental units were taken to the laboratory to measure changes in periphyton, leaf mass and larval size.

## 2.4. Laboratory Methods

### 2.4.1. Resource Consumption

In the laboratory, leaf material from the feeding chambers was examined for shredding signs by naked eye. Then it was dried at 105 °C for 48 h to obtain dry mass and incinerated at 550 °C to obtain the final ash free dry mass (AFDM).

Periphyton was scraped from each tile with a razor blade and washed with distilled water. Periphyton biomass was determined in terms of AFDM and expressed per square centimetre. Each sample was filtered onto pre-weighed Whatman GF/C filter and dried at 80 °C for 24 h. The filters were weighed and incinerated (APHA, 1989). To estimate the relative importance of the photosynthetic fraction in the community, the Chl *a*/OM index was calculated as the percentage of Chlorophyll *a* concentration per mg organic matter (Chl *a* × 100/OM) (WEBER and MCFARLAND, 1969). Chlorophyll *a* was determined following NUSCH (1980). Spectrophotometric measurements were carried out at 665 nm and 750 nm. Corrections for phaeophytin were performed by acidification with HCl. Samples for species composition were preserved in 4% formalin. The percentage of diatoms and non-diatom algae was calculated based on direct counts in a Sedgwick-Rafter chamber of 18 µl under a microscope at 400 × of magnification. Diatom identification and counting were performed on permanent slides. Samples were treated with hydrogen peroxide to oxidise organic matter, and then mounted in Naphrax<sup>®</sup> and examined at 1000×. A minimum of 800 valves was counted in each sample.

### 2.4.2. Larval Growth

Final body length and head width of each larva were measured to estimate final body mass. For each larval species, we obtained a regression for body ash free dry mass (BAFDM) in mg against body size from a set of larvae collected previously. Larvae were dried at 80 °C for 24 h to obtain their dry mass and then combusted at 550 °C for 1 h for AFDM. The power equations that best fitted our data were:

$$\text{BAFDM} = -0.84 + 2.83 \text{ POW}^{1.92} \text{ (for } K. \textit{kuscheli}; r_{\text{adj}}^2 = 0.84; \text{SE} = 4.46, n = 110),$$

and

$$\text{BAFDM} = 1.54 + 0.03 \text{ BL}^{2.35} \text{ (for } N. \textit{archiplatae}; r_{\text{adj}}^2 = 0.79; \text{SE} = 4.39, n = 68),$$

where POW and BL were post-ocular width (mm) and body length (cerci excluded) (mm), respectively. Larval growth was calculated as the difference between initial and final biomass.

### 2.4.3. Calculations

Larval instantaneous rates for consumption and growth were referred to initial body mass for comparison. Thus, relative consumption rate (RCR) and relative growth rate (RGR) were expressed as percentage per day (% d<sup>-1</sup>). Net values were obtained by subtracting mean controls from each experimental replicate. Efficiency of conversion of ingested food (ECI) was defined as the ratio between net food intake and net body growth expressed in percentage (ARSUFFI and SUBERKROPP, 1986):

$$ECI = \frac{FBc - FBt}{IBt - IBs} \times 100$$

where FBc is food biomass in controls and FBt in treatments, IBt is the insect's biomass in treatments and IBs is the starved insect's biomass.

Data were tested for significant differences by using one or two-way ANOVA. Multiple comparisons were tested with *a posteriori* methods (Student-Neuman-Keuls; S-N-K). Significant differences were detected at 95% of confidence.

### 2.4.4. Larval Diet

In addition to previous work on the food habits of *K. kuscheli* and *N. archiplatae* (DÍAZ VILLANUEVA and ALBARIÑO, 1999; ALBARIÑO, 2001), we also investigated gut contents of individuals of both species collected in different streams: Pescadero stream, a western sub-catchment of Nahuel Huapi Lake, 5 km far from the Ñireco sub-basin and Catedral stream in a different catchment located 100 km from the Ñireco basin. Seven mature larvae of *N. archiplatae* from a previous sampling in November 1992 at Ñireco stream (main channel) were also analysed because they represented the only late instars of this species. Larvae from Pescadero were collected at 850 m a.s.l. in a well-shaded reach in June 2004. Those from Catedral stream were collected at 1000 m a.s.l. in a partially canopied reach at different dates (in April, December 2004 and February 2005). Specimens were immediately fixed with 5% formalin and transported to the laboratory. Twenty five individuals of each species were dissected and foregut content mounted on a slide, dispersed with distilled water and observed under direct microscope at 400× (or 1000 × if required). Four categories were assessed: a) periphyton including amorphous organic matter, inorganic particles and algae, b) coarse leaf fragments (≥100 μm), c) fine leaf fragments (<100 μm), and d) invertebrate remains. The percentage of each food item and the percentage of larvae containing each item were calculated.

## 3. Results

### 3.1. Species Distribution in Ñireco Basin

We found a different spatial distribution for these two plecopterans at the catchment scale. While in stream A2 they coexisted, in A1 only *K. kuscheli* was present. Furthermore they showed contrasting standing stocks throughout the study. In both streams, *K. kuscheli* reached its maximum density in summer and maximum biomass in autumn-winter (Table 2). In contrast, *N. archiplatae* density and biomass were higher in winter and spring, respectively (Table 2). Biomass of both species peaked at the time larger specimens were collected (i.e. late instars). Although a relative abundance in terms of density was low for both species in Ñireco basin, their relative biomass represented over 20–40% of total community biomass (Fig. 1), thus dominating the assemblage. *K. kuscheli* subpopulation of A1 stream was larger both in density and biomass than the subpopulation of A2 (Table 2).

Table 2. Total density (ind. m<sup>-2</sup>) and biomass (mg m<sup>-2</sup>) of *Klapopteryx kuscheli* and *Notoperla archiplatae* in A1 and A2 streams (Nireco basin) in the four seasonal samplings (mean ± SE).

		A1		A2	
		<i>Klapopteryx kuscheli</i>	<i>Klapopteryx kuscheli</i>	<i>Notoperla archiplatae</i>	
Winter	density	181 ± 49	15 ± 9	131 ± 42	
	biomass	860 ± 303	117 ± 73	490 ± 190	
Spring	density	189 ± 79	22 ± 11	15 ± 9	
	biomass	395 ± 108	2 ± 0.2	603 ± 40	
Summer	density	541 ± 182	52 ± 23	0 ± 0	
	biomass	654 ± 176	14 ± 10	0 ± 0	
Autumn	density	237 ± 46	28 ± 16	20 ± 9	
	biomass	585 ± 271	98 ± 81	18 ± 8	

Spatial distribution was also markedly different at the reach scale within A2. *N. archiplatae* was usually found in turbulent habitats underneath large substrates (i.e. cobble-boulders). Although *K. kuscheli* was occasionally found cohabitating the same substrates with *N. archiplatae*, it was more abundant in low to moderate velocity habitats associated to substrate size ranging from pebble to boulder. Moreover, *K. kuscheli* was found in A1 at very low discharge (late summer) when certain pools remained connected with wet channel sections.

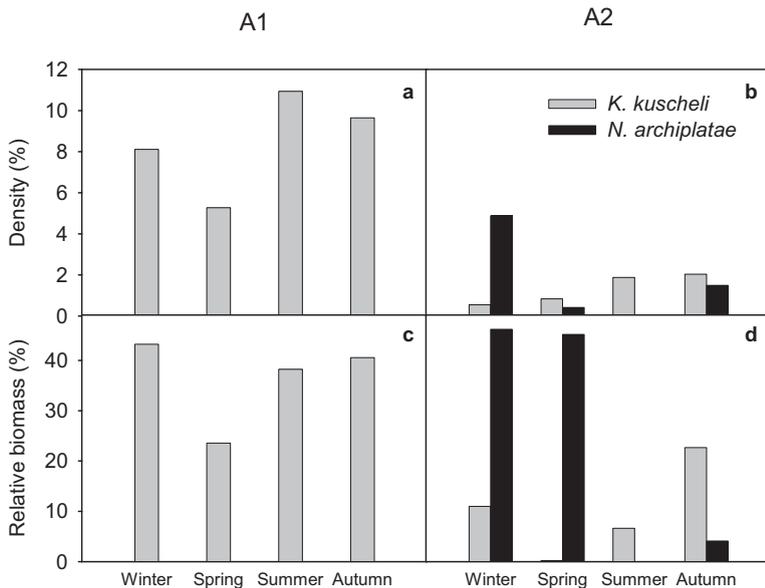


Figure 1. Seasonal relative abundance of *Klapopteryx kuscheli* and *Notoperla archiplatae*, density (a, b) and biomass (c, d) in A1 and A2 streams.

### 3.2. Gut Content Analysis

Gut contents of larvae from the different streams showed contrasting results between species. Except for three larvae with empty guts, all *N. archiplatae* presented periphyton (algal cells, fine inorganic particles and amorphous matter) in their guts (Table 3). Diatoms were the dominant algae (mainly represented by *Gomphonema*, *Gomphoneis* and *Fragilaria* species), but filamentous green algae composed the main diet of one specimen and a mucilaginous colony of cells represented 30% of another individual. On the other hand, all but four *K. kuscheli* guts contained 83.9% coarse plant fragments (basically from *Nothofagus* deciduous species) (Table 3). Sixty percent of *K. kuscheli* larvae were late instars and the remaining were intermediate instars. In the case of *N. archiplatae*, 28% were late instars. No clear differences in diet were observed between sexes. In general, the diet of *N. archiplatae* was more rigid in terms of the food items observed and their proportions than that of *K. kuscheli*.

Table 3. Mean percentage of food items in gut contents and the percentage of individuals containing each item for *Klapopteryx kuscheli* and *Notoperla archiplatae* in three different streams.

	<i>Klapopteryx kuscheli</i>		<i>Notoperla archiplatae</i>	
Stream Source				
Catedral	13		6	
Pescadero	12		12	
Ñireco	0		7	
Total guts inspected	25		25	
Empty guts	4		3	
Intermediate instar specimens <sup>1</sup>	10 (13.7–20.3)		18 (9.0–11.2)	
Late instar specimens <sup>1,2</sup>	15 (22.2–31.7)		7 (16.5–29.8)	
Female : male ratio	8 : 7		4 : 3	
Mean item abundance (%) <sup>3</sup>				
Periphyton	9.0	(9)	91.6	(100)
Leaf Fragments ≥100 µm	83.9	(90)	0.1	(4)
Leaf Fragments <100 µm	4.5	(14)	4.1	(13)
Invertebrate remains	2.6	(9)	4.2	(4)

<sup>1</sup> Number of larvae inspected and body size range (mm) in brackets.

<sup>2</sup> Late instar larvae were those where sex was identified.

<sup>3</sup> In brackets, percentage of total individuals inspected that contained the specific food item.

### 3.3 Field Experiment

#### 3.3.1. Resource Consumption

Visual observation of leaves from *K. kuscheli* chambers showed abundant shredding signs while they were absent in Control leaves. On the other hand, feeding signs by *N. archiplatae* were scarcely found. These larvae did not bite but slightly abraded leaves. However, their feeding activity was not enough to cause a significant reduction of leaf mass, thus relative consumption rate was null (ANOVA,  $P > 0.05$ ) (Fig. 2). Leaves ingested by *K. kuscheli* represented 4.8% of its initial body mass *per day* (Fig. 2). On the other hand, *N. archiplatae* exposed to periphyton showed a daily consumption rate of 10.0% of its initial body mass and they consumed 62% more periphyton than *K. kuscheli* (S-N-K's test,  $P < 0.05$ ) (Fig. 2).

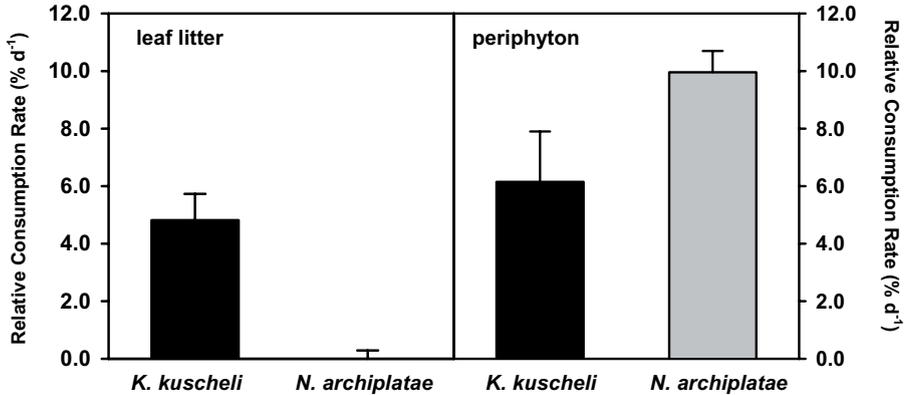


Figure 2. Relative consumption rate of *Klapopteryx kuscheli* and *Notoperla archiplatae*, expressed as percentage of AFDM consumed food (leaf litter and periphyton) by initial individual larval AFDM per day. Error bars = 1 SE.

Algal consumption by *K. kuscheli* on periphyton treatment was confirmed by the observation of gut contents, which showed the presence of the filamentous *Melosira varians* AGARDH.

After 25 days of colonization, when the larvae were introduced, periphyton consisted of 98% diatoms. The most abundant species in terms of cell number was *Gomphonema angustum* AGARDH (25% relative abundance), while in terms of biovolume *M. varians* comprised 75%. The effect of both plecopterans on periphyton composition was similar, as they reduced the abundance of filamentous algae while abundance of algae adhered to the substratum was favoured (Fig. 3). At the end of the experiment, *M. varians* was the most abundant species in control, not only in terms of biovolume (87%) but also in terms of cell counts (24%). This species was strongly reduced, to 2% and 4% relative abundance in terms of

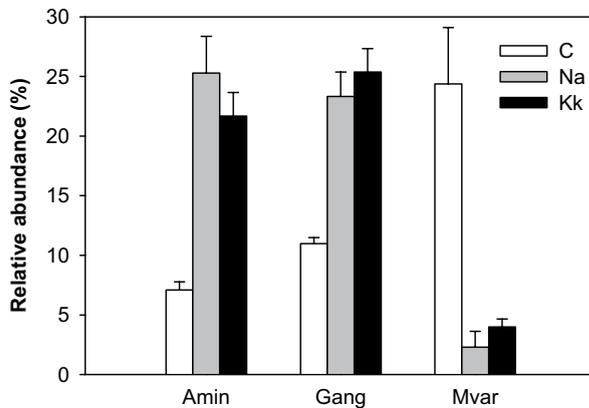


Figure 3. Relative abundance of the most abundant species (>10%), in terms of cell counts, in the periphyton of Control (C), *Klapopteryx kuscheli* (Kk) and *Notoperla archiplatae* (Na) treatments. Reference names: Amin: *Achnanthydium minutissimum*, Gang: *Gomphonema angustum*, Mvar: *Melosira varians*. Error bars = 1 SE.

cell counts, in *N. archiplatae* and *K. kuscheli* treatments, respectively (Fig. 3). On the other hand, *Achnanthidium minutissimum* (KÜTZ.) CZARNECKI and *G. angustum* relative abundances increased compared with final control, from 11% to 25% (*N. archiplatae* treatment) and 23% (*K. kuscheli* treatment) (Fig. 3). However, the higher Chl *a*/OM ratio in *N. archiplatae* treatment indicates that the grazing effect of both species differed. In *N. archiplatae* trial the ratio Chl *a*/OM remained significantly higher ( $4.1 \pm 0.5$ ) than in Control and *K. kuscheli* treatment ( $2.7 \pm 0.5$  and  $2.9 \pm 0.4$ , respectively). This result suggests that *N. archiplatae* maintained a more autotrophic epilithic layer and prevented the development of a senescent deep layer in the periphyton. In contrast, *K. kuscheli* dislodged the whole periphytic matrix.

### 3.3.2. Larval Survival, Growth and ECI

After 44 days of experimentation, more than 50% of larvae were still alive in treatments and control. The highest larval survivorship was observed for *N. archiplatae* fed on periphyton (90%). However, in control chambers (with no food addition) 66% of *N. archiplatae* and 80% of *K. kuscheli* larvae survived, indicating that the former was more susceptible to starving conditions. Fed on leaf litter, *N. archiplatae* survived as it did in controls (60%). In contrast, *K. kuscheli* showed similar survivorship on both food items and in control chambers (80%).

Larval growth differed between both species and it was significant only for *N. archiplatae* fed on periphyton and for *K. kuscheli* on leaf litter (S-N-K's test,  $P < 0.05$ ) (Fig. 4). Overall relative growth rate on leaves was 6.5 times higher for *K. kuscheli* than for *N. archiplatae* ( $P < 0.05$ ) (Fig. 4). On the other hand, the RGR on periphyton for *N. archiplatae* was significantly higher than for *K. kuscheli* which yielded a negative value ( $P < 0.05$ ) (Fig. 4). The efficiency of food conversion into body biomass (ECI) was calculated only for *K. kuscheli* fed on leaf litter and for *N. archiplatae* on periphyton because the other conditions achieved negative values of consumption and/or growth. *K. kuscheli* converted  $14 \pm 4\%$  (mean  $\pm$  SE) of the consumed CPOM (coarse particulate organic matter) into body mass while the ECI for *N. archiplatae* on periphyton was  $5 \pm 1\%$ .

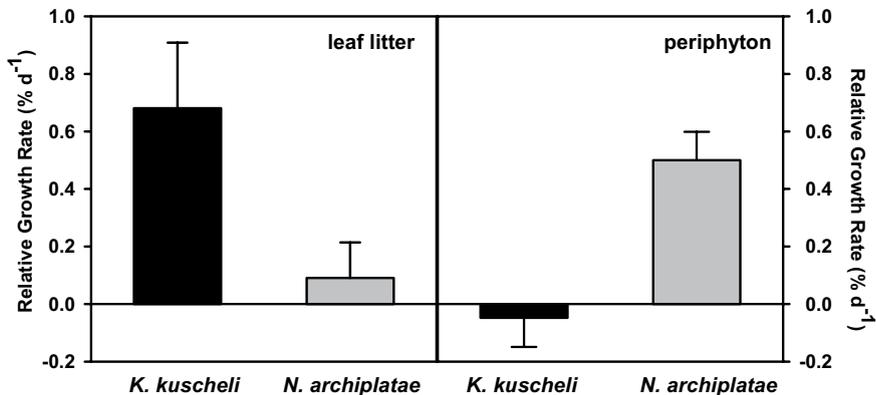


Figure 4. Relative growth rate of *Klapopteryx kuscheli* and *Notoperla archiplatae*, expressed as percentage of body AFDM gained by initial individual larval AFDM per day. Error bars = 1 SE.

## 4. Discussion

### 4.1. Feeding Relationships

Both studied species differed in their capabilities of exploiting contrasting food resources. Although larvae from different catchments showed dietary preference (i.e. the dominant food type in foreguts) either for leaf litter (*K. kuscheli*) or for periphyton (*N. archiplatae*), our results demonstrated that *K. kuscheli* was able to access both periphyton and leaves, while *N. archiplatae* was not. In our experiments, *K. kuscheli* was very efficient in feeding on CPOM but also it was able to consume algae. It seems possible that the laciniae with apical pointed teeth and concave inner margin (ALBARIÑO, 2001) allowed collecting loose algae as confirmed by the high consumption rates. The main algal species found in foreguts from *K. kuscheli* used in the experiment was the filamentous *Melosira*; consequently it likely performed browsing activity instead of scraping. In this sense, *K. kuscheli* might be a facultative shredder as it displayed a wider range of food acquisition than it was thought (ALBARIÑO, 2001). MIHUC (1997) and FRIBERG and JACOBSEN (1999) pointed out that feeding plasticity in the detritivore-shredder functional group might be more common than traditionally reported. Particularly, FRIBERG and JACOBSEN (1994) found that two shredders had almost the same consumption rate for two contrasting food types (the filamentous alga *Microspora* and leaf litter), and one of those species presented the highest growth rate on *Microspora* compared to leaf litter of several species. In contrast, in our one-month field experiment *K. kuscheli* larval growth was achieved on leaf litter but not on algae.

In general, aquatic insects would be expected to ingest the food that is most efficiently harvested or assimilated (CUMMINS and MERRITT, 1996). This contention agrees with the diet data reported for both plecopterans in this and previous studies (DÍAZ VILLANUEVA and ALBARIÑO, 1999; ALBARIÑO, 2001) and seems to show a "natural" preference experiment where each larval species fed on the food they found profitable. On the contrary, when there is no choice, feeding should occur on the single item available. *N. archiplatae* was not able to feed on leaf material, indicating feeding restriction. These larvae did not bite but slightly abraded leaves probably as a result of persistent browsing or scraping activities to obtain the biofilm developed on leaf surfaces. Although other grazers can feed and even grow on leaf litter using their brushing or scraping-adapted mouthparts (MIHUC and MINSHALL, 1995; CUMMINS and MERRITT, 1996), *N. archiplatae* had negligible results. Therefore, we suggest a mismatch of its morpho-behavioral adaptations to exploit leaf litter, e.g. bristles on the tip of laciniae cannot be used in strong shredding (ARENS, 1990). Furthermore, while MISE-RENDINO (2000) reported that late instars of *N. archiplatae* switch to predation in a population from Esquel stream (200 km further south), our gut content samples from late instars did not show evidence of a diet switch.

Despite its high consumption rate on periphyton, *K. kuscheli* lost weight. This might imply that assimilation efficiency was low for this item or that the energy gained by food assimilation was expended in handling and ingestion. The efficiency with which ingested food is converted to growth (ECI) depends on the digestive capabilities (i.e., the assimilation system). Moreover, when food quality is low, larval consumption should increase to balance for its lower nutritional value (CUMMINS and KLUG, 1979). Perhaps *K. kuscheli* tried to compensate for such poor food by ingesting periphyton at almost the same rate as for leaves. This compensatory strategy has been shown for *K. kuscheli* fed upon low quality leaf litter (ALBARIÑO and BALSEIRO, 2001). On the other hand, the method to measure periphyton consumption is prone to overestimating net food intake. It is recognized that invertebrate movement on the substrate detaches biofilm, producing overestimated periphyton consumption rates (LAMBERTI *et al.*, 1987; CATTANEO and MOUSSEAU, 1995) and it could have happened in *Klapopteryx* trials.

The ECI indexes of *K. kuscheli* on leaf detritus and *N. archiplatae* on periphyton were significantly positive. The efficiency of *K. kuscheli* (14%) was higher than the ECI calculated for the same species and food item (*N. pumilio* leaf litter) (ECI: 4%) in a side-channel experiment (ALBARIÑO and BALSEIRO, 1998). It is likely that differences in experimental designs led to dissimilar values. Shredding produces small leaf fragments that may enter the Fine Particulate Organic Matter pathway, and net leaf litter consumption may also be overestimated, consequently producing lower ECI values. ALBARIÑO (2001) found that FPOM production through mechanical breakage by *K. kuscheli* represented 12% of net consumed leaf litter. In the case of *N. archiplatae*, it is the first time that an ECI has been calculated and it was done in its environment, as this species is very vulnerable to handling. In contrast to our expectations, this species showed a relative low efficiency on periphyton (5%) compared to other herbivores (e.g. 20% for the grazer *Baetis* sp.; CUMMINS and KLUG, 1979). Similarly to the reasons explained above for *K. kuscheli*, the ECI for *N. archiplatae* is likely to have been underestimated.

Mouthpart classification divides North American plecopterans into two types (I: detritivore-herbivores, II: predators, STEWART and STARK, 1988). In South American gripopterygids, the potential for omnivory within detritivores and herbivores is apparent on account of the similarity of their mouthparts (ILLIES, 1963; SEPTON and HYNES, 1983). For instance, differences across families may also be minor, as between *N. archiplatae* and *K. kuscheli* (DÍAZ VILLANUEVA and ALBARIÑO, 1999; ALBARIÑO, 2001). Thus, the single analysis of mouthpart morphology and gut content is not entirely enough to infer food habits (CUMMINS and MERRITT, 1996). A combination of those approaches with feeding experiments and behavior observations has demonstrated to be more suitable for an accurate assignment to FFGs (MCSHAFFREY and MCCAFFERTY, 1988; PALMER *et al.*, 1993a and b). However, unless the methodology for calculating ECI values considered corrections for biofilm dislodgment or leaf litter comminution losses from net food consumption, the functional grouping of organisms should only be based on growth measurements.

Herbivore and detritivore specialists are expected to outcompete generalists as the former achieve higher efficiency of food conversion (CUMMINS and MERRITT, 1996). However, a generalist strategy would be better adapted to unpredictable changes in environmental conditions when particular resources may become unavailable. Watershed alteration that imposes changes to the input timing, standing crop or quality of basic primary resources could strongly affect the studied populations, particularly those of *N. archiplatae*. Perhaps the feeding plasticity of *K. kuscheli* confers acceptable survivorship during periods of leaf litter shortage during the year. The species has probably two to three years of larval growth before emergence and the moderate-low ECI obtained on leaf litter and the absence of development on periphyton help to explain its long larval life.

#### 4.2. *Notoperla* and *Klapopteryx* in Benthic Assemblages of Low Order Streams

Habitat use of both species contrasted during the field study. *N. archiplatae* was usually found in high velocity runs and riffles underneath large substrates (i.e. large cobbles to boulders). They are also frequent in stepped fast flowing habitats, not surveyed quantitatively in this study. Scrapers typically have morpho-behavioral adaptations for maintaining their position on exposed surfaces in rapidly flowing water (CUMMINS and KLUG, 1979). The absence of *N. archiplatae* in A1 may be associated to the lack of hydraulic microhabitats still common in A2 during low discharge periods. In contrast, *K. kuscheli* was found in low to high velocity sites in A1 and A2, suggesting high habitat plasticity. For instance, during the summer – early autumn 1996, A1 had a very low discharge and pools became more or less isolated. At this time, larvae of different body sizes could be found living both in pool margins and in the wetted rocky bottom (underneath coarse substrates). It is likely that hydraulic con-

ditions and food availability were responsible for a higher density and biomass of *K. kuscheli* in A1 compared to A2. In particular, A1 only attained higher leaf litter standing stock than A2 in winter, after complete leaf falling and retention into the streambed (2.75 vs. 0.86 g m<sup>-2</sup>, respectively). This suggests that food availability and stream retentiveness (resulting from hydraulic features) are suitable habitat traits to this species. It is possible that our observed similarities in leaf litter standing stock along the rest of the year (A1: 0.90 g m<sup>-2</sup> vs. A2: 0.69 g m<sup>-2</sup>) may have resulted from higher shredding activity in A1 with a larger population of *K. kuscheli*. In fact, this species represented 52% of shredder abundance in leaf litter bags of an experiment previously run in A1. In such occasion, *N. pumilio* leaves decayed at 0.0074 d<sup>-1</sup> (*k*) and leaf litter loss by large shredders was estimated in 22% (ALBARIÑO and BALSEIRO, 2002).

In Northern Patagonia, *N. archiplatae* would have a synchronized univoltine cycle with the earlier instars occurring during summer and the later ones in late spring (DÍAZ VILLANUEVA and ALBARIÑO, 1999; MISERENDINO, 2000). Its density in A2 was very low in summer when we expected to find a numerous population formed by early instars. It is likely that larvae were in a different microhabitat other than the one sampled or they had not hatched at the time sampling occurred.

*Klapopteryx kuscheli* and *N. archiplatae* are frequent invertebrates in low order streams in Patagonia (CAMPOS *et al.*, 1984; MISERENDINO, 1996; ALBARIÑO, 1997). Although our studied populations were not numerically abundant compared to other invertebrates, they dominated the assemblage in biomass (Fig. 1). In systems where top predators are visual hunters (e.g. exotic salmonids), prey size is relevant and those species might be vulnerable items. Introduced salmonids have become the most widespread top predators in Patagonia freshwaters and their impact on streams is currently under study (L. BURIA unpublished data). Therefore the studied species may represent an important link between the base of the food webs and top levels. Furthermore, their roles as primary processors of allochthonous and autochthonous matter appear significant to Andean stream functioning.

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## 6. References

- ALBARIÑO, R. J., 1997: Spatial distribution of Plecoptera from an Andean-patagonic lotic environment in Argentina. – *Rev. Brasil. Biol.* **57**: 629–636.
- ALBARIÑO, R. J. and E. G. BALSEIRO, 1998: Larval size and leaf conditioning in the breakdown of *Nothofagus pumilio* leaves by *Klapopteryx kuscheli* (Insecta, Plecoptera) in a South Andean stream. – *Internat. Rev. Hydrobiol.* **83**: 397–404.
- ALBARIÑO, R. J. and A. C. VALVERDE, 1998: Hábito alimentario del estado larval de *Parasericostoma cristatum* (Trichoptera: Sericostomatidae). – *Rev. Soc. Entomol. Argent.* **57**: 131–135.
- ALBARIÑO, R. J., 2001: The Food Habits and Mouthpart Morphology of a South Andes Population of *Klapopteryx kuscheli* (Plecoptera: Austroperlidae). – *Aquatic Insects* **23**: 171–182.
- ALBARIÑO, R. J. and E. G. BALSEIRO, 2001: Food quality, larval consumption, and growth of *Klapopteryx kuscheli* (Plecoptera: Austroperlidae) from a south Andes stream. – *J. Freshwater Ecol.* **16**: 517–526.
- ALBARIÑO, R. J. and E. G. BALSEIRO, 2002: Leaf litter breakdown in Patagonian streams: native versus exotic trees and the effect of invertebrate size. – *Aquat. Conserv.* **12**: 181–192.
- AMERICAN PUBLIC HEALTH ASSOCIATION, 1989: Standard methods for the examination of water, sewage, and wastewater. American Public Health Association. 715 pp. Washington D.C

- ARENS, W., 1990: Wear and tear of mouthparts: a critical problem in stream animals feeding on epilithic algae. – *Can. J. Zool.* **68**: 1896–1914.
- ARSUFFI, T. L. and K. SUBERKROPP, 1986: Growth of two stream caddisflies (Trichoptera) on leaves colonized by different fungal species. – *J. N. Am. Benthol. Soc.* **5**: 297–305.
- BLONDEL, J., 2003: Guilds or functional groups: does it matter? – *Oikos* **100**: 223–231.
- CAMPOS, H., J. ARENAS, C. JARA, T. GONSER and R. PRINS, 1984: Macrozoobentos y fauna íctica de las aguas limnéticas de Chiloe y Aysen Continentales (Chile). – *Medio Ambiente* **7**: 52–64.
- CATTANEO, A. and B. MOUSSEAU, 1995: Empirical analysis of the removal rate of periphyton by grazers. – *Oecologia* **103**: 249–254.
- CUMMINS, K. W., 1973: Trophic relations of aquatic insects. – *Ann. Rev. Entomol.* **18**: 183–206.
- CUMMINS, K. W., 1974: Structure and function of stream ecosystems. – *BioScience* **24**: 631–641.
- CUMMINS, K. W. and M. I. KLUG, 1979: Feeding ecology of stream invertebrates. – *Ann. Rev. Ecol. System.* **10**: 147–172.
- CUMMINS, K. W., 1996: Invertebrates. – *In*: G. PETTS and P. CALOW (eds.): *River Biota*. – Blackwell Science Ltd. pp. 75–91. Oxford.
- CUMMINS, K. W. and R. W. MERRITT, 1996: Ecology and distribution of aquatic insects. – *In*: R. W. MERRITT and K. W. CUMMINS (eds): *Aquatic insects of North America*. p. 74–86. – Kendall/Hunt. Iowa.
- DE CABO, L. I. and I. R. WAIS, 1991: Macrozoobenthos prospection in central Neuquen streams, Patagonia, Argentina. – *Verh. Internat. Verein. Limnol.* **24**: 2091–2094.
- DÍAZ VILLANUEVA, V. and R. J. ALBARIÑO, 1999: Feeding habit of *Notoperla archiplatae* (Plecoptera) larvae in a North Patagonia Andean stream, Argentina. – *Hydrobiologia* **412**: 43–52.
- DÍAZ VILLANUEVA, V., R. J. ALBARIÑO and B. MODENUTTI, 2004: Grazing impact of two aquatic invertebrates on periphyton from an Andean-Patagonian stream. – *Arch. Hydrobiol.* **159**: 455–471.
- FRIBERG, N. and D. JACOBSEN, 1994: Feeding plasticity of two detritivore-shredders. – *Freshwat. Biol.* **32**: 133–142.
- FRIBERG, N. and D. JACOBSEN, 1999: Variation in growth of the detritivore-shredder *Sericostoma personatum* (Trichoptera). – *Freshwat. Biol.* **42**: 625–635.
- FÜREDER L., C. WELTER and J. K. JACKSON, 2003: Dietary and stable isotope ( $^{13}\text{C}$ ,  $^{15}\text{N}$ ) analyses in alpine stream insects. – *Internat. Rev. Hydrobiol.* **88**: 314 – 331.
- HILDEBRAND-VOGEL, R., R. GODOY and A. VOGEL, 1990: Subantarctic-Andean *Nothofagus pumilio* forests. Distribution area and systematic overview, vegetation and soils as demonstrated by an example of a South Chilean stand. – *Vegetation* **89**: 55–68.
- ILLIES, J., 1963: Revision der südamerikanischen Gripopterygidae (Plecoptera). – *Mitt. schweiz. entomol. Ges.* **34**: 145–248.
- LAMBERTI, G. A., L. R. ASHKENAS, S. V. GREGORY and A. D. STEINMAN, 1987: Effects of three herbivores on periphyton communities in laboratory streams. – *J.N. Am. Benthol. Soc.* **6**: 92–104.
- LEDGER, M. E. and A. G. HILDREW, 2001: Growth of an acid-tolerant stonefly on epilithic biofilms from streams of contrasting pH. – *Freshwat. Biol.* **46**: 1457–1470.
- MCLELLAN, I. D., 2001: A revision of South American Austroperlidae (Plecoptera). – *Aquatic Insects* **23**: 233–251.
- MC SHAFFREY, W. and P. MCCAFFERTY, 1988: Feeding behavior of *Rhithrogena pellucida* (Ephemeroptera: Heptageniidae). – *J. N. Am. Benthol. Soc.* **7**: 87–99.
- MERRIT, R. W., K. W. CUMMINS, M. B. BERG, J. A. NOVAK, M. J. HIGGINS, K. J. WESSELL and J. L. LESSARD, 2002: Development and application of a macroinvertebrate functional-group approach in the bioassessment of remnant river oxbows in southwest Florida. – *J.N. Am. Benthol. Soc.* **21**: 290–310.
- MIHUC, T. B. and J. R. MIHUC, 1995: Trophic ecology of five shredders in a Rocky Mountain stream. – *J. Freshwat. Ecol.* **10**: 209–216.
- MIHUC, T. B. and G. W. MINSHALL, 1995: Trophic generalist vs. trophic specialist: implications for food web dynamics in port-fire streams. – *Ecology* **76**: 2361–2372.
- MIHUC, T. B., 1997: The functional trophic role of lotic primary consumers: generalist versus specialist strategies. – *Freshwat. Biol.* **37**: 455–462.
- MISERENDINO, M. L., 1996: Ninfas de Plecoptera de ambientes lóticos del noroeste del Chubut. – *Naturalia patagónica, Ciencias Biológicas* **4**: 75–83.
- MISERENDINO, M. L., 2000: Distribución y variación estacional de Plecoptera en un sistema fluvial de la cordillera patagónica. – *Rev. Soc. Entomol. Argent.* **59**: 147–158.

- NUSCH, E. A., 1980: Comparison of different methods for chlorophyll and phaeopigment determination. – *Arch. Hydrobiol. Beih.* **14**: 14–36.
- PALMER, C. and J. O'KEEFFE, 1992: Feeding patterns of four macroinvertebrate taxa in the headwaters of the Buffalo River, Eastern Cape. – *Hydrobiologia* **228**: 157–173.
- PALMER, C., J. O'KEEFFE, A. PALMER, T. DUNNE and S. RADLOFF, 1993a: Macroinvertebrate functional feeding groups in the middle and lower reaches of the Buffalo River, eastern Cape, South Africa. I. Dietary variability. – *Freshwat. Biol.* **29**: 441–53.
- PALMER, C., J. O'KEEFFE and A. PALMER, 1993b: Macroinvertebrate functional feeding groups in the middle and lower reaches of the Buffalo River Eastern Cape, South Africa. II. Functional morphology and behaviour. – *Freshwat. Biol.* **29**: 455–62.
- SEPTON, D. H. and H. B. N. HYNES, 1983: Food and mouthpart morphology of the nymphs of several Australian Plecoptera. – *Aust. J. Mar. Freshw. Res.* **34**: 893–908.
- STEWART, K. W. and B. P. STARK, 1988: Nymphs of North American stonefly genera (Plecoptera). – *Thomas Say Found. Ser. Ent. Soc. Am.* **12**: 1–460.
- VELÁSQUEZ, S. M. and M. L. MISERENDINO, 2003: Habitat type and macroinvertebrate assemblages in low order Patagonian streams. – *Arch. Hydrobiol.* **158**: 461–483.
- WAIS, I. R., 1987: Macrozoobenthos of Negro River Basin, Argentine, Patagonia. – *Studies of Neotropical Fauna and Environment* **22**: 73–91.
- WAIS, I. R. and A. A. BONETTO, 1988: Analysis of the allochthonous organic matter and associated macroinvertebrates in some streams of Patagonia (Argentina). – *Verh. Internat. Verein. Limnol.* **23**: 1455–1459.
- WAIS, I. R., 1990: A checklist of the benthic macroinvertebrates of the Negro River Basin, Patagonia, Argentina, including an approach to their functional feeding groups. – *Acta Limnol. Brasil.* **III**: 829–845.
- WEBER, C. I. and B. H. MCFARLAND, 1969: Periphyton biomass-chlorophyll ratio as an index of surface waters and effluents. – *Environmental Monitoring Series EPA-670/4-73-001*. United States Environmental Protection Agency, Cincinnati, Ohio.

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