

Biometry of neotropical invertebrates inhabiting floodplain rivers: unraveling bionomy

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Received 26 February 2016

Accepted 6 January 2017

DOI: 10.1590/1678-4766e2017014

ABSTRACT. Currently, it is widely recognized that invertebrates play key roles in neotropical floodplains and in many other environments worldwide. However, little information has been published concerning their biometry, in spite that it represents an essential tool for many different studies. Here, we provided length-mass and length-length relationships by fitting the linearized model ($\log_{10} Y = \log_{10} a + b \log_{10} X$) and several mean biomass ratios \pm SE for bivalves, gastropods, quironomids, ephemeropterans, oligochaetes and hirudineans. We measured, weighed, oven dried and incinerated to ashes specimens collected from 2005 to 2014 in the Paraná River, Argentina. The lineal equations had fit levels higher than 75% in most of the significant regressions. Hence, when slopes were compared, differences raised from ontogeny and phylogeny of taxa. Additionally, slopes resulted different from constants of other regions, types of environments and climates. In addition, organic matter ratios resulted significantly different among invertebrates according to their feeding types. The equations and ratios that we provided will facilitate future research on life history, productivity and energy transference in the food webs of invertebrates inhabiting floodplain wetlands and can be used as tools for planning management strategies and in restoration projects of aquatic environments.

KEYWORDS. Allometry, organic matter, Paraná River, wetlands, benthos and pleuston.

RESUMEN. **Biometría de invertebrados de ríos de llanura: desentrañando su bionomía.** Actualmente, es muy reconocido que los invertebrados tienen un rol clave en los ambientes de llanura aluvial neotropicales así como en otros ambientes alrededor del mundo. Sin embargo, se cuenta con muy poca información publicada en relación con su biometría, a pesar de ser una herramienta esencial para muchos tipos de estudios. En este trabajo se obtuvieron relaciones longitud-masa ajustadas por el modelo linealizado ($\log_{10} Y = \log_{10} a + b \log_{10} X$) y diferentes proporciones de materia orgánica promedio \pm ES para bivalvos, gastrópodos, quironómidos, efemerópteros, oligoquetos e hirudíneos. Medimos, pesamos, secamos e incineramos hasta cenizas especímenes colectados entre 2005 y 2014 en el río Paraná, Argentina. Las ecuaciones lineales tuvieron valores de ajuste mayores al 75% en la mayoría de las regresiones significativas. Al comparar las pendientes, se encontraron diferencias relacionadas con la ontogenia y filogenia de las taxa. Asimismo, las pendientes resultaron diferentes de las constantes para otras regiones, tipos de ambientes y climas. Asimismo, las proporciones de materia orgánica fueron significativamente diferentes entre invertebrados de acuerdo a su tipo de alimentación. Las ecuaciones y proporciones que obtuvimos facilitarán futuras investigaciones sobre historia de vida, productividad y transferencia de energía en las tramas tróficas de invertebrados en humedales de llanura aluvial y pueden ser empleadas para planificar estrategias de manejo y en proyectos de restauración de ambientes acuáticos.

PALABRAS-CLAVE. Alometría, materia orgánica, río Paraná, humedales, bentos y pleuston.

Having a thoughtful knowledge of aquatic ecosystems requires an evaluation of the structure and functioning of its components. Thus, as macroinvertebrates play an important role in food webs and matter cycling in most aquatic systems, it is essential to count with tools to study their biomass and productivity. Actually, biomass is a key variable in quantifying a variety of ecological processes at any of the organization levels in either aquatic or terrestrial environment, ranging from individual consumption and bioenergetics to the spatial transfer of energy between habitats (BENKE *et al.*, 1999).

However, direct determination of fresh or preserved mass is not feasible in many cases such as when estimation of biomass of small and abundant animals is too laborious and time consuming and when rare specimens cannot be destroyed. Moreover, differences on live mass of aquatic

organisms might reflect not only differences in biomass, but differences on water retention, thus direct determination of dry mass is recommended in order to obtain reliable estimates of biomass (DONALD & PATERSON, 1977).

Length-dry or wet mass conversion is one of the most used techniques for estimating biomass from samples when direct measurement is neither possible nor practical (BENKE *et al.*, 1999). Indeed, this method is usually considered better to others (i. e. biovolume determinations or direct weighing) primarily because results faster and more precise (ROGERS *et al.*, 1976; BURGHER & MEYER, 1997). However, length-mass relationships obtained for invertebrates in a specific region or type of environment are not generally transferable to other study sites (MEYER, 1989). The length-mass analyses have been widely used and tested for mass estimations of

terrestrial, benthic and planktonic invertebrates from other biogeographic regions (mostly compiled in SMOCK, 1980; BENKE *et al.*, 1999 and JOHNSTON & CUNJAK, 1999 for North America, TOWERS *et al.*, 1994 for New Zealand, MEYER, 1989, BURGHERR & MEYER, 1997 for Europe). Even though the importance of biometry, few data are available just for aquatic as well as terrestrial neotropical insects (CRESSA & LEWIS JR., 1984; CRESSA, 1986, 1999; LÓPEZ *et al.*, 1997; ZILLI *et al.*, 2008, 2009), whereas most data were obtained for the other macroinvertebrate groups, of streams in cold climates (ALBARIÑO & BALSEIRO, 1998; MISERENDINO, 2001; GUALDONI *et al.*, 2013). Furthermore, there are scarce studies on biometry and available information on such relationships for most aquatic invertebrate groups inhabiting floodplain wetlands.

Many invertebrates inhabiting the Paraná River floodplain – such as worms, leeches, midges, mayflies, bivalves and snails- are widespread across similar rivers of tropical regions. In wetlands of the Paraná River they are benthic or associated to macrophytes, with high abundance and richness (POI DE NEIFF & NEIFF, 2006; EZCURRA DE DRAGO *et al.*, 2007). In addition, they can reach high biomass and secondary production (ZILLI, 2012, 2013; ZILLI & PAGGI, 2013) and link basal sources with higher trophic levels (GALIZZI *et al.*, 2012; SAIGO *et al.*, 2015).

In spite of their ecological importance for rivers functioning, there is still a lack of published information concerning biometry of neotropical macroinvertebrates. Moreover, in the frame of increased river perturbations worldwide in a global warming scenario, it becomes essential to obtain information concerning such relationships in relatively unaltered systems such as the Middle Paraná River, prior to management and conservation strategies development. In this frame, our objective was to provide length-mass relationships and to assess the ratios of organic (ash free) mass for some of the common groups of macroinvertebrates inhabiting large rivers and to enable ecologist to count with biomass estimates that are generally highly suitable for ecosystem functioning studies.

MATERIALS AND METHODS

The biometric relationships were obtained for mollusks, midges, aquatic worms, mayflies and leeches collected in the frame of different studies from 2005 through 2014 from ponds, streams, rivers, and temporary wetlands very closely located to 2 RAMSAR sites (31°37'52"S - 60°30'45"W and 31°53'34"S - 60°40'36"W, Paraná River floodplain, Argentina, Table I). All these taxa comprise the most frequent and abundant invertebrates found in the Paraná River and other neotropical large rivers. Invertebrates were collected with dredges and nets from benthos and pleuston respectively, fixed with 10% formalin and preserved in 70% ethanol solution.

Measurements, biomass an organic matter obtention. Total body length or some other dimension (Tab. I) was measured using a optic microscope supported with

a micrometric scale (accurate to 0.001 mm) for the smaller invertebrates and a digital caliper for the higher organisms (Schwiz, Electronic Digital Caliper, accurate to 0.01 mm). For all the taxa we aimed to ensure that, a wide size range was included in the measurements.

The wet weight, hereafter named as WW (in mg), was obtained after soaking the invertebrates for ten minutes in water and drying in a piece of blotting paper for some seconds. The dry weight, hereafter called DW (in mg) was obtained after oven dry at 60°C for 72 hs to constant, and then cooling in a desiccator. The WW and DW estimations only approximate organic matter if the weights of calcareous skeletons and/ or body moisture are eliminated (STIRN, 1981), biomass for mollusks is presented both with and without shells, except in the case of Planorbidae and Sphaeriidae, as it was impossible to separate soft form hard parts.

It has been widely studied that preservatives such as alcohol, cause changes in the weight of preserved animals, which may affect weight estimates. Therefore, in the present study, compensations were made for changes in weight by using the factors reported by LEUVEN *et al.* (1985).

The ash free dry weight biomass, hereafter AFDW (mg), was obtained for invertebrates. Some mollusks were weighed individually, whereas other invertebrates were pooled into groups before weighing. After dry weight was obtained as described above, the samples were muffle furnace at 500°C for 3 hs (LAPPALAINEN & KANGAS, 1975). In every case, before weighing, the animals were cooled down to room temperature in a desiccator equipped with absorbent packets of silica gel. The AFDW was calculated by subtracting the ash-weight from the DW.

The biomass measurements were obtained by using either an OHAUS balance (Model ANALYTICAL Plus) or Mettler Toledo (model AB204-S/FACT) balance (Accuracy: 0.01 and 0.1 mg respectively), according to the sample and balance accuracy.

Data analysis. The lineal regression describes Y as function of X within the range of observed values of X and, therefore, generally it is an unsafe procedure to predict Yi values from Xi values outside the observed range of Xi (ZAR, 1996). Hence, when estimations should be performed out of the ranges of regressions, then proportions may represent a useful tool. Therefore, in the present paper, several mean ratios \pm SE are presented.

Prior to run the statistical analysis, data were explored for outliers, linearity and homoscedasticity (Box-plots, QQ-plots and Breusch-Pagan test). The lineal coefficients that better fit to the relationships among metrics and biomass data, the R² and significance values were obtained. Data were $\log_{10} x+1$ transformed to better fit the lineal equations in the form: $\log_{10} Y = \log_{10} a + b \log_{10} X$.

Comparisons were made for fit values and slopes of dimensions-mass and dimension-dimension regressions and organic matter proportions, either with Kruskal-Wallis, Mann-Whitney and Wilcoxon tests. The statistical analyses were run by using the R VERSION 3.1.2 and PAST version 2.17c (HAMMER *et al.*, 2001). Additionally, the ratios among different metrics were calculated.

Tab. I. Habitat and measurements made for different invertebrates of the Parana River floodplain, Argentina. The structures measured on each group together with the abbreviations which will be utilized are described below.

Taxa; abreviations	Habitat	Description of linear metrics measured
Hirudinea (Leeches); Hir	Associated both, to macrophytes and benthos	Body contraction due to fixation may conduct to highly biased measurements Thus, lineal measurements were not included in this study
Oligochaeta (Aquatic worms); Oli Opistocystidae (<i>Opistocysta</i> spp. and <i>Trieminentia corderoi</i> Harman, 1969); Op Tubificininae (<i>Aulodrilus pigueti</i> Kowalewski, 1914; Ap, <i>Limnodrilus</i> spp.; Lim)	Associated to macrophytes, but mainly to benthos	L: Total length measured from the anterior edge of the prostomium to the posterior edge of the last segment W: average of 3 measurements made in the first, mid and last segments, perpendicularly to the main body axis
Bivalvia (Clams); Biv Corbiculidae (mainly the non-native <i>Corbicula fluminea</i> Müller, 1774); Cor	Infaunal benthic clams	L: in a lateral view, measured from the anterior to the posterior border of the shell H: in a lateral view, from the top of umbo to the ventral border of the shell. W: in a dorsal view, measured perpendicularly to H in the wider section of each specimen
Sphaeriidae (mainly <i>Musculium</i> spp. and <i>Pisidium</i> spp.); Sph	Associated to macrophytes, but mainly to benthos	
Hyriidae (<i>Castalia ambigua</i> Lamarck 1819 and <i>Diplodon</i> spp., mainly <i>D. parallelopedon</i> Lea, 1834); Hyr	Infaunal benthic clams	
Gastropoda; Gas Ampullariidae (Apple snails, mainly <i>Pomacea canaliculata</i> Lamarck 1828, and <i>Asolene</i> spp.); Amp	Benthic, but mainly associated to macrophytes Amphibious	H= from the tip of the apex to the further border of the outer lip. D1: In a ventral view, measured at the wider section of the body whorl D2: measured, in a 90° angle to D1.
Planorbidae (Ramshorn snails, <i>Biomphalaria</i> spp., <i>Drepanotrema</i> spp.); Pla		D: in a lateral view, measured across the wider section
Ephemeroptera (Mayflies); Eph Polymitarcidae, mainly <i>Campsurus violaceus</i> Needham & Murphy, 1924	Nymphs mainly from benthos, but also from pleuston	L: from the anterior edge of the head to the posterior edge of the last abdominal tergite Tibia: measured in the fore leg tibia from the articulation with the femur to the end of the tibia projection. Interoc: in a dorsal view of the head, measured from eye to eye
Chironomidae (Midges); Ch Chironominae: <i>Chironomus</i> spp., <i>Cladopelma</i> spp., <i>Saetheria</i> spp., <i>Harnischia</i> spp.; Chir Tanypodinae: <i>Coelotanypus</i> spp., <i>Ablabesmyia</i> spp., <i>Tanypus</i> spp.; Tany	Aquatic larvae inhabiting benthos and pleuston	L: in a dorsal view, measured across the body from the tip of the labrum to the end of the last abdominal segment cW: measured perpendicularly to L at the maximum width of the cephalic capsule

RESULTS AND DISCUSSION

Invertebrate biometry by lineal models. We present new Lineal to DW and Lineal to Lineal dimension regressions based on either total body length or shorter dimensions (e.g., head width) in Appendices I and II. Additionally, detailed information is showed concerning data (n), significance level (p), metric ranges, fit level ($=R^2$) and lineal coefficients for each equation.

Most of the significant regressions that we obtained explained more than 75% of the DW variation due to increments in the lineal metrics, with just a few ranging from 25% to 50% fit (Fig. 1). This high fit level was also obtained for several regressions by other authors (SMOCK, 1980; BURGHERR & MEYER, 1997; BENKE *et al.*, 1999; CRESSA, 1999; MISERENDINO, 2001; GUALDONI *et al.*, 2013).

Furthermore, also other models were tested. Indeed, L-DW relationships fitted best to polynomial regressions even after $\log_{10}(x+1)$ transformations of data. However, in most of the cases, differences of R^2 among one or higher order power regressions were of just 1% of the DW variability explanation (i. e. R^2 were relatively higher in smooth curves

than more lineal ones), except for midges and mayflies that had a 12% - 25% and 2% differences. Indeed, discontinuous growth by moulting is most compatible with sharp breaks in allometry. Thus, when freshwater arthropods molt, they suddenly increase their body volume without increasing their DM (the cast exuviae actually represents a loss of mass). This point may introduce variability into length-mass regressions as animals of the same body dimensions may vary considerably in their DM. This is probably the case, mainly for holometabolous insects such as midges, which larvae grow sharply between instars.

On the other hand, individual growth is usually estimated from a mass sample of individuals in various growth stages and thus any sudden spurt occurring at varying sizes in different individuals will be damped to a more gently curved transition in the mass data regression (GOULD, 1966). This compound allometry were also found in other studies for mayflies and it had been explained in terms of the nymphs passing from immature to mature stages, when the ratios of specific growth rates changed due to an increasing rate of development of imaginal structures (CLIFFORD, 1970). Conversely, from a large number of relationships investigated

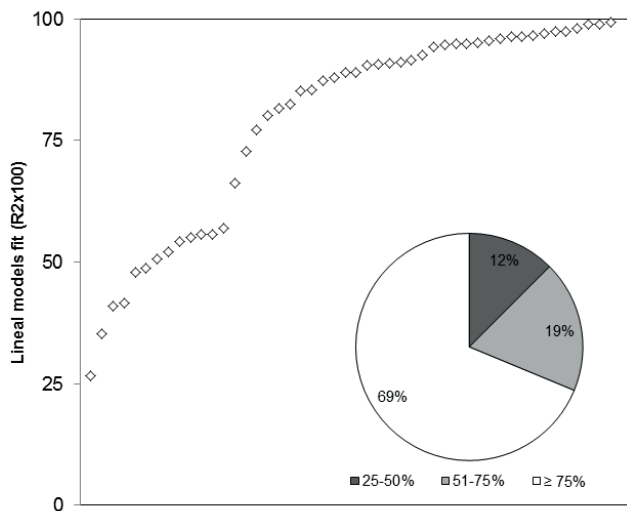


Fig. 1. Lower to higher fit level (R^2) for the lineal models on metric-DW variation of invertebrates. The pie-chart shows the proportion (%) of values that fall within each R^2 category. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

for mayflies, only 32% cases of isometry were found as, in most cases, the change in the allometric relationship late in nymphal life did not occur at the same total body length for all of the dimensions investigated (CAMPBELL, 1991). Actually, in our study, isometry was not obtained for mayflies (Appendix II) and thus sharp changes in biomass as nymphs grow might be related to differences in allometry within each instar.

Additionally, closer fits can always be obtained by adding further terms to a power series (VON BERTALANFFY, 1964). However, choosing a reasonably simple functional expression involving the minimum of non-interpretable parameters it would be essential to increase biological understanding (SCHOLL, 1954) and thus, the lineal function has enjoyed almost exclusive use in allometric studies.

Generally, length was the metric that best fitted to biomass changes for all the taxa and thus, \log_{10} plots are showed on Figures 2-5. However, width and mass were also highly correlated for mollusks and mayflies. Conversely, lower fit levels were obtained for worms and for midges in contrast with results reported for these insects by other authors either for terrestrial (ROGERS *et al.*, 1976; GOWING & RECHER, 1985) or aquatic species (SMOCK, 1980; WENZEL *et al.*, 1990; MEYER, 1989; BURGHERR & MEYER, 1997; CRESSA, 1999).

Growth and allometry: assessing invertebrates bionomy. Most of the regressions showed higher fit levels for mollusks and mayflies than worms and midges (Fig. 6), with even some regressions that didn't even fit such as width-DW for *Aulodrilus pigueti*, Opistocystidae (both oligochaetes) and *Coelotanypus* (midges). Moreover, fit values for L-DW and W-DW regressions were statistically different for taxa (Wilcoxon signed rank test, $p < 0.05$), with higher values for the former. This might be related to the fact that length usually represents a better measurement of ecological links as reflect resources acquisition (BENKE *et*

al., 1999). The comparisons showed stepped differences for worms but mainly for midges. Indeed, many authors reported that body length performed as a better predictor of biomass than head capsule width for most insects (SMOCK, 1980; MEYER, 1989; BURGHERR & MEYER, 1997). Conversely, the trend was not such clear for mayflies and mollusks that showed also high fit levels for the other dimensions. These results might be directly related to differences in growth type. Indeed, from the data obtained, worms and midges grew more in length than in width, while metrics of mayflies but mainly mollusks showed a higher isometry as it can be observed from slopes (=b) in the Appendix II. Moreover, among clams, the Hyriidae showed lower fit levels, probably because individuals of largely different shaped genera (*Diplodon*, mainly *D. parallelpipeton* Lea, 1834 and *Castalia ambigua* Lamarck 1819) were pooled within the data set, while Sphaeriidae and Corbiculidae resulted more homogenous.

In the case of insects, differences in allometry could be related to the type of preimaginal molts. To accomplish the large increase in mass after molting, much of the cuticle of holometabolous insects remains unsclerotized and undifferentiated. For example, when larval caddisflies molt, the initially soft cuticle of their heads rapidly reaches its instar-specific width and quickly hardens, but the undifferentiated cuticle of their abdomen is somewhat extensible and facilitates more continuous growth and as much as a tripling of mass (BENKE *et al.*, 1999). Indeed, larvae belonging to the holometabolous orders generally have greater within-instar variability in DM related to CW than those in the hemimetabolous orders, because the former tend to have fewer instars and thus increase more in linear dimensions (and mass) from 1 instar to the next than the Hemimetabola (COLE, 1980; BUTLER, 1984). Actually, cephalic width is a common metric used to estimate larval instar among midges (DYAR, 1890).

The DW variability of mollusks, was highly explained not only when the whole individuals but also when only soft tissues were considered in every lineal combination ($R^2 \times 100 \geq 80\%$, Appendix II) with no significant differences between fit levels for both (Wilcoxon signed rank test, $p > 0.05$). However, soft tissues accounted for less than 1% in clams (Hyriidae and Corbiculidae respectively) and 22% of total weight in apple snails (Appendix III). Thus, as shells are mainly constituted by inorganic materials (on average $95 \pm 2\%$ is represented by ashes) care must be taken when matter and energy transference are intended. Hence, changes in DW of bivalves as metrics increased were more pronounced considering the whole individuals than only soft tissue, with almost a unit difference for Corbiculidae and less than 0.15 but still positive for Hyriidae (Fig.7). Conversely, for apple snail slopes were higher when DW of soft tissues was regressed against metrics than when the entire specimens were considered. This might probably be explained by the varying calcium intake of clams at different stages of their life cycle, while apple snails might invest more energy on soft tissue.

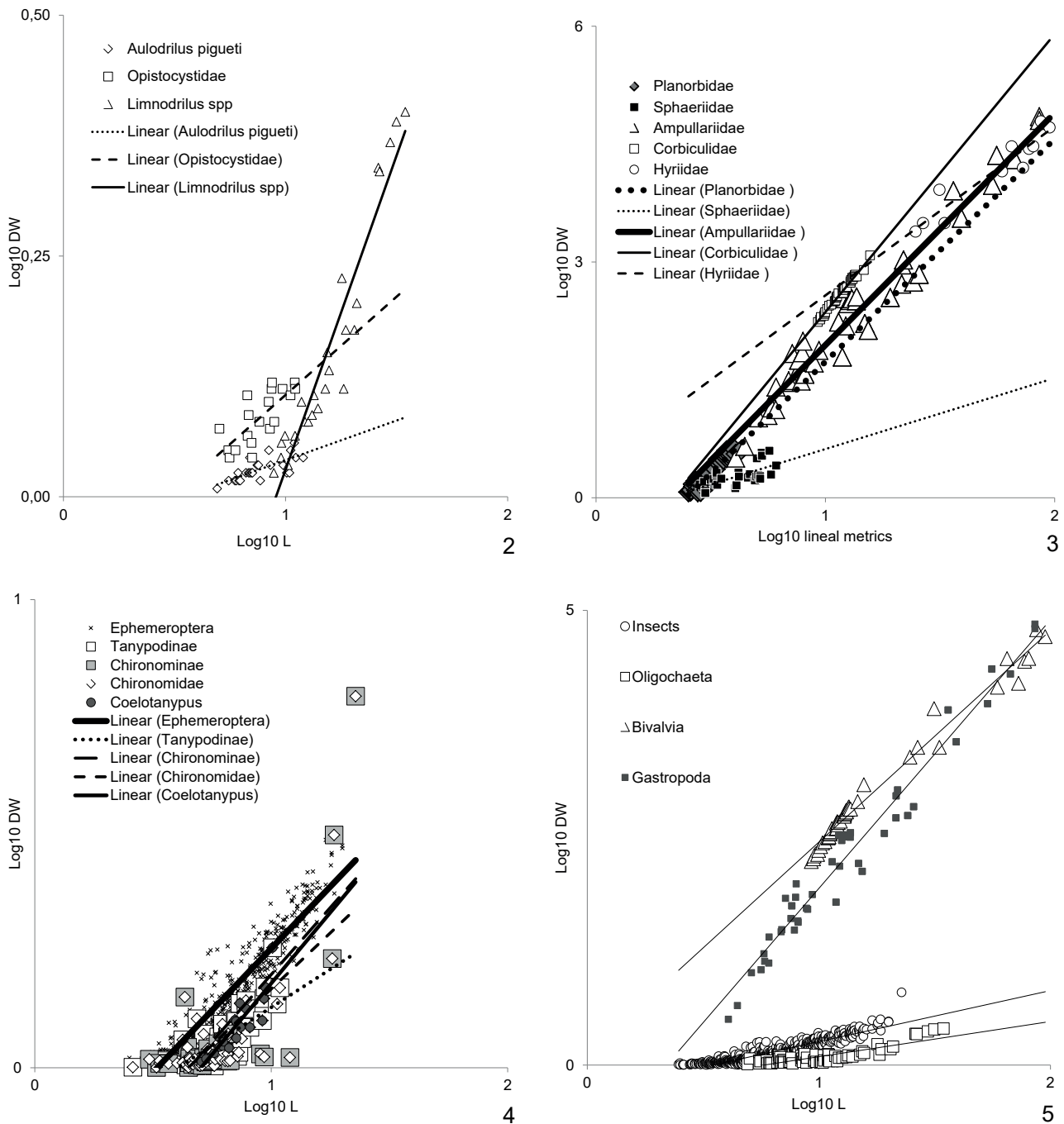


Fig. 2-5. Dispersion plots and linear curves on L-DW regressions for (2) oligochaetes, (3) mollusks, (4) insects and (5) all invertebrates together. For Planorbidae L=D and for Ampullariidae L= H. In Fig. 5 only apple snails H-DW were considered within “Gastropoda” and only Corbiculidae and Hyriidae (and not Sphaeriidae) were considered within “Bivalvia”. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

Furthermore, the predictive capability of species-specific equations is higher when using equations based on data grouped at higher taxonomic levels (BENKE *et al.*, 1999). In the present study, such trend was obtained for *Limnodrilus* spp. (oligochaetes) with higher adjusts than all worms pooled, for mayflies in relation to all insects pooled, for midges and when considering all macroinvertebrates pooled. Additionally, differences in slopes among equations would suggest that it is better to use the closest level to avoid errors associated with substitutions.

In most studies, it is supposed that animals grow allometrically and for most of the aquatic invertebrates groups the constant b is often close to a value of 3 even for length and width (BENKE *et al.*, 1999). The underlying assumption for this value is that body mass is more influenced by surface than volume (ENGELMANN, 1961). In our study, 41% of the obtained constants were 3 or very close while 45% were ≤ 3 and only 14% were >3 (Fig. 8).

Shape and specific gravity must remain exactly the same throughout development for the expected value of b to

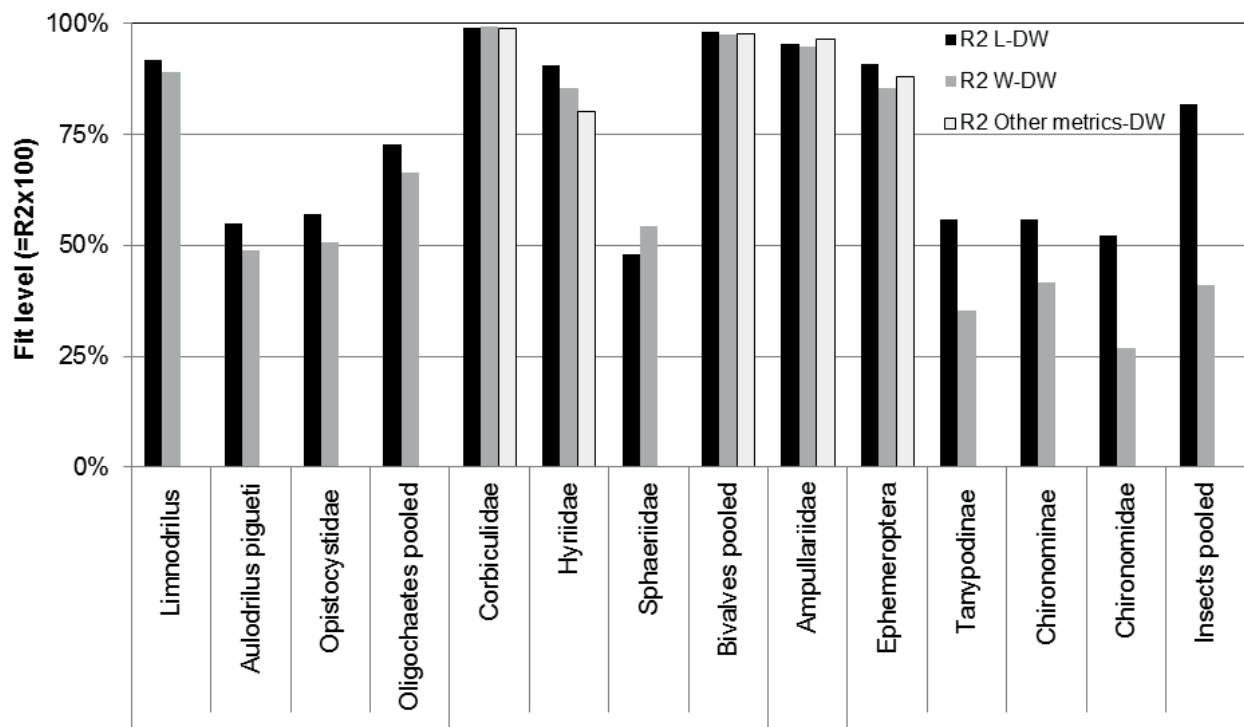


Fig. 6. Fit levels obtained from freshwater invertebrates metric-DW lineal regressions. L= length, W= width and other metrics= "W" for clams, "D2" for apple snails and "Tibia" for mayflies. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

be exactly 3 (BENKE *et al.*, 1999). Indeed, our findings for mollusks showed that slopes were very close to 3 when full specimens and only soft parts were regressed against metrics. However, displacements from the perfect cubic relationship were explored. Thus, in our study, mollusks showed an isometric growth of all the metrics along development, with slightly lower slopes just for Hyriidae that must be related to differences in shape changes of the genera involved (Appendix II). Conversely, differences in slopes for both, apple snails and clams, showed that displacement from the perfect cubic relationship could be explained by growth changes in specific gravity. Thus, for clams there was a higher slope when the entire specimens were considered mainly for Corbiculidae (Fig. 7). This seems to be related to a variable rate of material addition to valves along their growth cycle. Opposite, apple snails showed the inverse trend (Fig. 7). This could be related to changes in specific gravity of soft parts due to investment on structures related to reproduction such as penis in males and albumen glands that increase considerably when the female is about to produce eggs. In the case of Sphaeriidae, the equations obtained might have been affected by calcium lost due to fixation and preservation. Therefore, use of these equations should be cautiously made.

Moreover, in our study, values lower than 3 were mostly obtained for worms and insects. For insects, the mean value of *b* for length-mass equations is actually somewhat <3 for all orders, families and individual taxa (compiled in SMOCK, 1980; MEYER, 1989; TOWERS *et al.*,

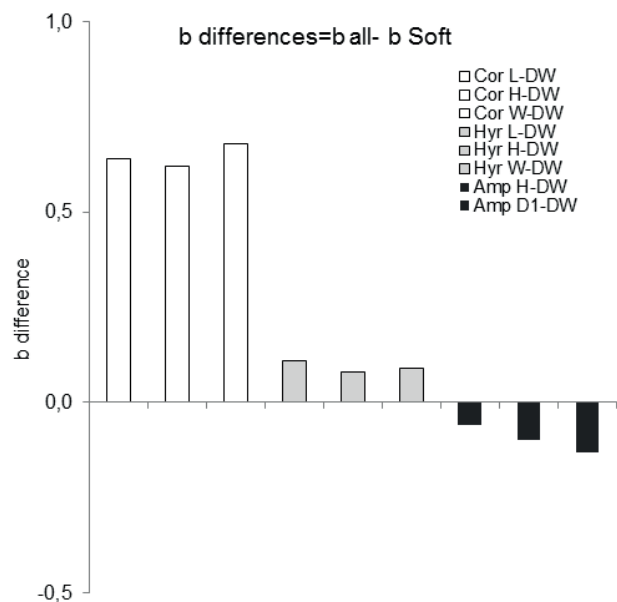


Fig. 7. Differences in slopes obtained for lineal regressions of mollusks with and without shells. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

1994; BURGHERR & MEYER, 1997). On the other hand, several authors reported values <3 for several invertebrates across different environments of the neotropics (CRESSA, 1999; BECKER *et al.*, 2009; GUALDONI *et al.*, 2013). The reasons why the mean value of *b* would be consistently <3 for insects and oligochaetes are either that these invertebrates

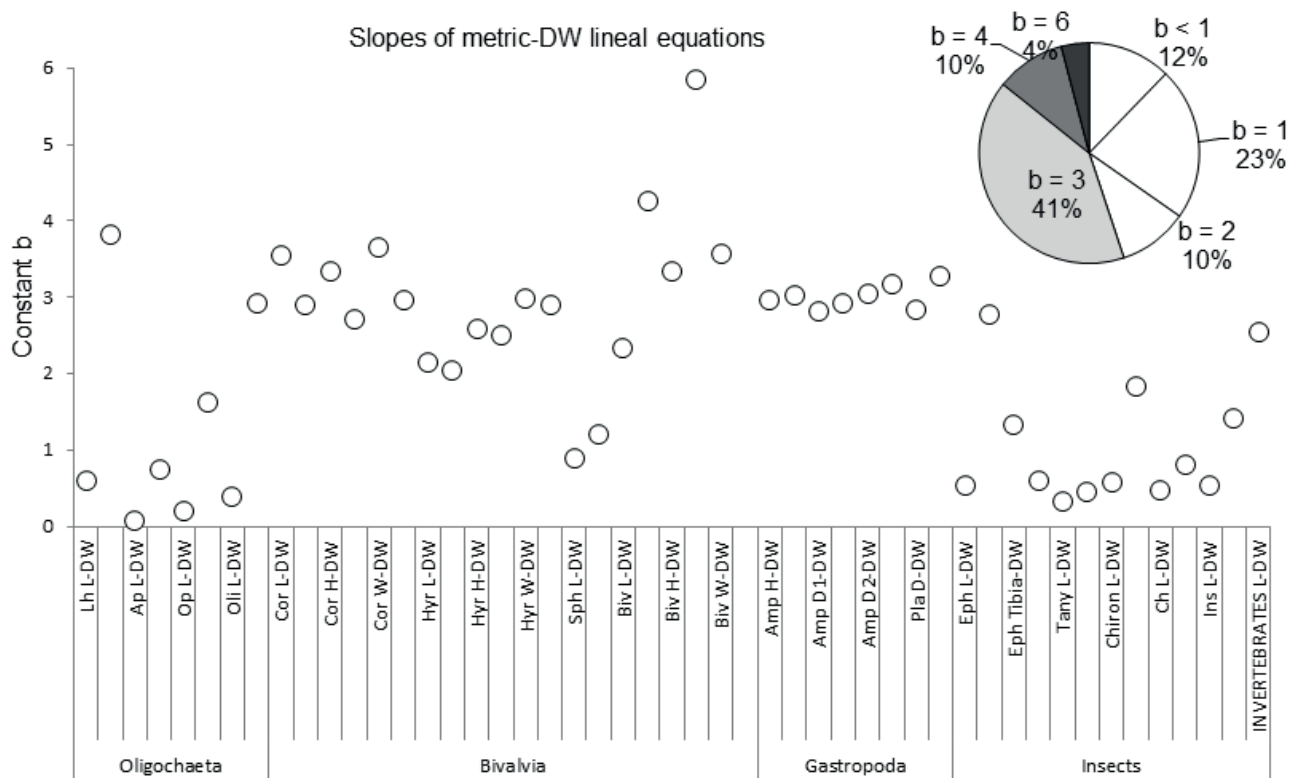


Fig. 8. Values of slopes (=b) of regression equations obtained for metrics-DW relationships. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

become proportionately narrower as body length increases, or that specific gravity declines with size (BENKE *et al.*, 1999). Certainly, slopes were much lower than 1 for all L-W regression equations (Appendix II).

Conversely, several equations for temperate or cold climate insects showed slopes relatively higher than ours, whereas they were relatively similar to the ones obtained for mollusks (several can be found in SMOCK, 1980; BURGHERR & MEYER, 1997; BENKE *et al.*, 1999). This is probably related to the fact that some tropical groups of insects have longer and thinner bodies than temperate ones (GOWING & RECHER, 1985). Moreover, slopes were relatively lower for regressions of dipterans than the ones obtained in cold environments of Patagonia (Argentina) whereas being relatively similar for *Limnodrilus hoffmiesiteri* (MISERENDINO, 2001).

In relation to insects, Interoc-DW was close to 3 for mayflies, whereas for dipterans cW-DW was relatively close to 1. A reason for head-width regressions having $b < 3$ (e.g., Diptera), is that head width may be proportionately smaller in early instars than in later instar (BENKE *et al.*, 1999).

When comparisons were assessed within our data set, slopes were all significantly higher for W-DW than for L-DW (Wilcoxon signed rank test, $p < 0.05$). When assessed within each taxa, we found that these differences were significant for worms and insects but not for mollusks (Kruskall-Wallis test $p < 0.05$, Mann-Whitney pairwise comparisons test $p < 0.05$). Additionally, slopes were significantly different for overall metrics-DW combined, among insects and worms

with bivalves and gastropods that had the lower and higher slopes respectively (Kruskall-Wallis test $p < 0.05$, Mann-Whitney pairwise comparisons test $p < 0.05$).

Biomass and organic matter ratios. Allometric regressions are empirically determined and apply only to size ranges represented by actual data. Thus, when inferences are intended but data are out of range, is better to have proportions available. Therefore, several DW-L mean ratios \pm SE are presented on Appendix III. Additionally, also organic matter and biomass data (AFDW, WW and DW) mean ratios \pm SE are provided. WATERS (1977) approximated a value of 10% ash (or 90% organic matter) estimates for benthos and zooplankton. In our study, just some values for soft tissues of clams were around 90% organic matter, whereas leeches had percentages that fall below and worms and insects above it. For mollusks, the higher ash contents were obtained for the shells of clams, Sphaeriidae and planorbids due to exoskeletons composition.

The AFDW/DW ratios were statistically different among taxa (Kruskall-Wallis test, $p < 0.01$) with significant differences mainly for leeches and all the other groups (Fig. 9). Indeed, predatory leeches showed a higher average organic matter proportion in relation to the other groups that feed mostly on basal resources such as apple snails that feed mostly on macrophytes and the other groups on detritus. Predators invest high part of their time in seeking for preys, whereas basal resources feeders can benefit from largely available and low-cost pursuing resources. However, predators consume a resource of high nutritious value (CUMMINS & KLUG, 1979)

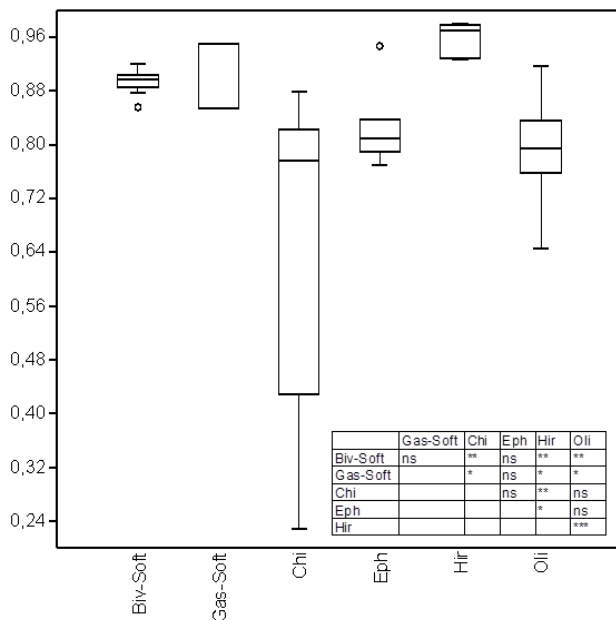


Fig. 9. Box-plot of invertebrates AFDW/DW. Significance values (p) of Mann-Whitney pairwise comparisons are shown on the lower-right side. P-values: * p= 0.01-0.05; ** p< 0.01-0.001; *** p<0.001. Invertebrates were collected in the Paraná River floodplain from 2005 through 2014.

and the assimilation rates of invertebrates are widely variable ranging from less than 5% for detritivores to almost 90% for predators (BENKE & WALLACE, 1980). Therefore, our findings may support the idea that organic matter content of invertebrates is closely related to trade-offs involved in the type of feeding by invertebrates.

Final remarks. Simple lineal models proved to be very well adjusted for biomass estimations of invertebrates living in floodplain habitats of the Paraná River. Differences raised from variable growth types and phylogeny of invertebrates. Additionally, slopes differed from constants, mainly for insects obtained for other regions, climates and type of environments. Thus, we encourage other researchers to select equations from places as similar as possible in region, climatic zone and habitat characteristics to the communities being studied and from the closest taxonomic level to avoid errors associated with substitutions. Additionally, we consider that measuring any body structure of an invertebrate is less time-consuming than to weight all the organisms collected and thus we encourage to use the information we provided whenever possible. When research is carried out on size ranges out of actual data, then we recommend to use proportions we provided. However, also significant differences were obtained mainly related to feeding types of different groups and thus we suggest to consider this issue when using proportions in either biodiversity or functional approaches.

Acknowledgements. This research was partially financially supported by PRESTAMO BID PICT 2012 N° 2791, FONCyT, ANPCyT and PIP 11220130100318, CONICET, both MinCyT, Argentina.

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Appendix I. Lineal coefficients obtained for each equation of the linear regression for the conversion of lineal metrics (mm) into dry mass (mg, DW) for the Paraná River floodplain (Argentina) macroinvertebrates. Data (n), metric ranges and R² values calculated are showed. Significance level (p), was always < 0.01.

Taxa	x-y measurements used in the calculations (Log ₁₀)	Regression constants					
		log10a	b	p	R ²	n	Metric range (mm)
<i>Limnodrilus</i> spp.	L-DW	0.62±0.05	0.65±0.04	***	0.916	25	2.70-33.58
	W-DW	-0.23±0.02	3.83±0.21	***	0.890	41	0.11-0.58
<i>Aulodrilus pigueti</i>	L-DW	-0.04±0.01	0.08±0.02	***	0.550	25	2.30-11.00
	W-DW	-0.02±0.01	0.75±0.15	***	0.488	29	0.13-0.35
Opistocystidae	L-DW	-0.10±0.04	0.20±0.04	***	0.570	19	4.04-10.05
	W-DW	-0.10±0.05	1.63±0.47	**	0.506	17	0.20-0.34
Oligochaetes combined	L-DW	-0.30±0.03	0.39±0.03	***	0.728	69	2.30-33.58
	W-DW	-0.18±0.02	2.93±0.23	***	0.662	87	0.11-0.58
Corbiculidae	L-DW	-1.18±0.06	3.54±0.06	***	0.989	40	8.28-14.66
	L-DWSoft	-2.03±0.09	2.90±0.08	***	0.970	42	8.28-14.66
	H-DW	-0.88±0.05	3.34±0.05	***	0.993	41	7.51-14.35
	H-DWSoft	-1.76±0.10	2.72±0.10	***	0.948	43	7.51-14.35
	W-DW	-0.74±0.06	3.65±0.06	***	0.989	40	5.49-10.24
Hyriidae	W-DWSoft	-1.64±0.09	2.97±0.10	***	0.955	43	5.49-10.24
	L-DW	0.42±0.41	2.15±0.23	***	0.905	11	23.75-93.96
	L-DWSoft	0.58±0.42	2.04±0.24	***	0.890	11	23.75-93.96
	H-DW	0.30±0.53	2.58±0.36	***	0.854	11	16.01-57.29
	H-DWSoft	0.39±0.47	2.50±0.32	***	0.873	11	16.01-57.29
Sphaeriidae* ¹	W-DW	0.16±0.66	2.98±0.49	***	0.802	11	11.01-38.99
	W-DWSoft	0.24±0.60	2.89±0.45	***	0.824	11	11.01-38.99
	L-DW	-0.28±0.11	0.90±0.18	***	0.480	29	1.83-5.11
	H-DW	-0.36±0.11	1.21±0.21	***	0.542	29	1.20-3.88
	L-DW	0.11±0.06	2.34±0.05	***	0.981	51	8.28-93.96
Bivalves combined* ²	L-DWSoft	-3.43±0.15	4.26±0.12	***	0.959	53	8.28-93.96
	H-DW	-0.86±0.09	3.33±0.08	***	0.975	51	7.51-57.29
	H-DWSoft	-4.95±0.30	5.85±0.26	***	0.907	54	7.51-57.29
	W-DW	-0.65±0.80	3.56±0.08	***	0.975	51	3.18-38.99
	W-DWSoft	-4.65±0.26	6.33±0.25	***	0.925	54	4.12-38.99
	H-DW	-1.01±0.10	2.96±0.08	***	0.952	69	1.55-84.73
Ampullariidae	H-DWSoft	-1.76±0.20	3.02±0.15	***	0.949	24	6.65-84.73
	D1-DW	0.59±0.09	2.82±0.08	***	0.947	69	1.47-78.02
	D1-DWSoft	-1.39±0.20	2.92±0.15	***	0.942	24	5.39-78.02
	D2-DW	-0.71±0.08	3.05±0.07	***	0.963	69	1.30-59.45
	D2-DWSoft	-1.54±0.16	3.18±0.13	***	0.963	24	4.7-59.45
	D-DW	-1.11±0.07	2.84±0.14	***	0.912	44	1.53-3.33
Planorbidae	D* ³ -DW	-1.18±0.06	3.28±0.06	***	0.965	113	1.41-78.02
Gastropoda combined	L-DW	-0.27±0.007	0.53±0.01	***	0.908	503	1.48-19
	Interoc-DW	-0.11±0.005	2.78±0.05	***	0.853	474	0.07-0.55
Ephemeroptera	Tibia-DW	-0.12±0.005	1.33±0.02	***	0.879	474	0.16-1.63
	Chironomidae						
<i>Coelotanytus</i> spp.	L-DW	-0.43±0.07	0.61±0.08	***	0.772	20	4.08-9.13
Tanypodinae combined	L-DW	-0.20±0.03	0.33±0.04	***	0.557	56	1.6-9.88
	cW-DW	-0.11±0.03	1.01±0.21	***	0.352	46	0.23-0.73
Chironominae combined	L-DW	-0.37±0.07	0.57±0.09	***	0.557	33	2.05-21.75
	cW-DW	-0.14±0.05	1.84±0.40	***	0.416	32	0.11-0.78
Chironomidae combined	L-DW	-0.31±0.04	0.48±0.05	***	0.521	89	1.6-21.75
	cW-DW	-0.10±0.03	1.14±0.22	***	0.267	78	0.11-0.73
Insects combined	L-DW	-0.28±0.008	0.53±0.01	***	0.817	592	1.48-21.75
	Width* ⁴ -DW	-0.04±0.009	1.69±0.09	***	0.410	552	0.07-0.73
Invertebrates combined	L-DW	-1.70±0.09	2.55±0.10	***	0.477	775	0.39-93.96

*1 Soft tissues and valves weren't separated

*2 Regressions were calculating by pooling data from Hyriidae and Corbiculidae

*3 D refers to D1 for Ampullariidae.

*4 Width represented by cW for midges and Interoc for mayflies

Appendix II. Lineal coefficients obtained for each equation of the linear regression for the conversion of lineal metrics (mm) into other linear metrics (mm) for the Paraná River floodplain (Argentina) macroinvertebrates. Data (n), metric ranges and R² values calculated are showed. Significance level (p), was always < 0.01.

Taxa	x-y measurements used in the calculations (Log ₁₀)	Regression constants		p	R ²	n	Metric range (mm)
		log10a	b				
<i>Limnodrilus</i> spp.	L-W	-0.01±0.01	0.10±0.01	***	0.561	53	2.70-33.58
Oligochaetes combined	L-W	0.04±0.01	0.06±0.01	***	0.228	85	2.30-33.58
Corbiculidae	L-H	-0.098±0.02	1.07 ± 0.02	***	0.982	42	6.33-14.66
	L-W	-0.12±0.02	0.97±0.02	***	0.988	42	
	H-W	-0.04±0.02	0.91±0.02	***	0.989	43	5.50-14.35
Hyriidae	L-H	0.33±0.26	0.67±0.15	**	0.682	11	23.75-93.96
	L-W	0.47±0.27	0.50±0.15	**	0.545	11	
Sphaeriidae	L-H	0.03±0.04	0.80±0.07	***	0.84	30	1.83-5.11
	L-H	0.03±0.02	0.89±0.02	***	0.949	83	1.83-93.96
Bivalves combined	L-W	0.23±0.03	0.64±0.02	***	0.932	53	6.33-93.96
	H-W	-0.05±0.02	0.93±0.01	***	0.987	54	1.20-57.29
Ampullariidae	H-D1	-0.07±0.01	0.99±0.01	***	0.991	83	1.55-84.73
	H-D2	-0.07±0.01	0.95±0.01	***	0.993	83	
	D1-D2	0.01±0.01	0.96±0.01	***	0.993	83	
Ephemeroptera	L-Tibia	-0.09±0.004	0.37±0.005	***	0.932	474	1.48-16.88
	L-Interoc	-0.04±0.002	0.16±0.002	***	0.907	474	
	Tibia-Interoc	-0.0001±0.0007	0.46±0.003	***	0.977	474	0-16-1.63
Tanypodinae	L-cW	0.05±0.03	0.16±0.04	***	0.239	52	1.60-9.88
Chironominae	L-W	-0.07±0.02	0.24± 0.02	***	0.76	32	2.05-21.75
Chironomidae combined	L-W	-0.01± 0.02	0.21± 0.03	***	0.371	84	1.6-21.75
Insects combined	L-Width*1	-0.03±0.005	0.17±0.006	***	0.569	558	1.48-21.75

*1 Width represented by CW for midges and Interoc for mayflies

Appendix III. Mean weight ratios (\pm SE) of invertebrate taxa. N represent either individual (when not specified) or pools of them.

	Mean	\pm SE	N
Sph DW/WW	0.45	0.07	5 pools of \approx 110 individuals each
Sph AFDW/DW	0.82	0.03	"
Sph H/L	0.753	0.01	31
Sph DW/L (mg/mm)	0.302	0.03	32
Cor DW/WW	0.69	0.002	40
Cor Soft DW/WW	0.17	0.004	44
Cor Soft AFDW/DW	0.89	0.005	3 pools of \approx 32 individuals each
Cor Shell AFDW/DW	0.03	0.003	4 pools of \approx 53 valves each
Cor DW Soft/all	0.03	0.0005	41
Cor DW Soft/Shell	0.03	0.0005	"
Cor DW Soft/L (mg/mm)	1.065	0.06	45
Cor W/L	0.671	0.003	44
Cor H/L	0.939	0.004	"
Cor W/H	0.715	0.002	45
Hyr DW/WW	0.66	0.04	11
Hyr Soft DW/WW	0.76	0.01	"
Hyr Soft AFDW/DW	0.89	0.01	7
Hyr Shell AFDW/DW	0.05	0.00	"
Hyr DW Soft/all	0.92	0.03	"
Hyr DW Soft/L (mg/mm)	287.0	51.18	11
Hyr W/L	0.4	0.05	"
Hyr H/L	0.6	0.05	"
Hyr W/H	0.7	0.03	"
Amp DW/WW	0.42	0.01	69
Amp Soft DW/WW	0.31	0.02	4
Amp Soft AFDW/DW	0.89	0.02	"
Amp Soft AFDW/WW	0.27	0.02	"
Amp Shell AFDW/DW	0.05	0.01	3
Amp DW Soft/all	0.22	0.01	24
Amp DW/H	65.01	18.27	67
Amp D1/D2	1.13	0.01	83
Amp H/D1	1.24	0.01	"
Amp H/D2	1.40	0.01	"
Pla DW/WW	0.32	0.01	48
Pla AFDW/DW	0.27	x	2
Plano DW/D	0.50	0.05	48
Ch DW/WW	0.47	0.17	3 pools of \approx 68 individuals each
Ch AFDW/DW	0.63	0.10	"
Tany ccW/L	0.09	0.01	56
Tany DW/L	0.02	0.003	"
Chir ccW/L	0.06	0.003	31
Chir DW/L	0.01	0.002	30
Eph DW/WW	0.49	x	2 pools of \approx 19 individuals each
Eph AFDW/DW	0.83	0.03	"
Eph DW/L	0.161	0.004	503
Eph Tibia/L	0.243	0.002	474
Eph interoc/L	0.113	0.001	"
Hir DW/WW	0.42	0.07	7 pools of \approx 41 individuals each
Hir AFDW/DW	0.96	0.01	"
Oli DW/WW	0.50	0.05	13 pools of \approx 163 individuals each
Oli AFDW/DW	0.79	0.02	"
Ap W/L	0.03	0.002	33
Ap DW/L	0.01	0.001	34
Op W/L	0.04	0.004	21
Op DW/L	0.03	0.002	"
Lim W/L	0.02	0.001	52
Lim DW/L	0.02	0.002	51