

# Species diversity and community structure of crustacean zooplankton in the highland small waterbodies in Northwest Yunnan, China

Xing Chen<sup>1,2</sup>, Qinghua Cai<sup>1</sup>, Lu Tan<sup>1</sup>, Shuoran Liu<sup>3</sup>, Wen Xiao<sup>3</sup>, Lin Ye<sup>Corresp. 1</sup>

<sup>1</sup> State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, the Chinese Academy of Sciences, Wuhan, Hubei, China

<sup>2</sup> University of Chinese Academy of Sciences, Beijing, China

<sup>3</sup> Institute of Eastern-Himalaya Biodiversity Research, Dali University, Dali, Yunnan, China

Corresponding Author: Lin Ye

Email address: yelin@ihb.ac.cn

Small waterbodies are a unique aquatic ecosystem with an increasing recognition for their important role in maintaining regional biodiversity and delivering ecosystem services. However, small waterbodies in Northwest Yunnan, one of the most concerned global biodiversity hot-spots, remain largely unknown. Here, we investigated the community structure of crustacean zooplankton and their relationships with limnological, morphometric and spatial variables in the highland small waterbodies in Northwest Yunnan in both the dry (October 2015) and rainy (June 2016) seasons. A total of 38 species of crustacean zooplankton were identified in our study, which is significantly higher than many other reported waterbodies in the Yunnan-Guizhou plateau as well as in the Yangtze River basin. This suggests that the highland small waterbodies are critical in maintaining regional zooplankton diversity in Northwest Yunnan. Meanwhile, we found limnological variables could explain most variation of crustacean zooplankton community, comparing to the morphometric and spatial variables in both the rainy and dry seasons. Our study revealed the diversity and community structure of crustacean zooplankton in the highland small waterbodies in Northwest Yunnan and highlighted the importance of small waterbodies in maintaining regional biodiversity.

**Title:** Species diversity and community structure of crustacean zooplankton in the highland small waterbodies in Northwest Yunnan, China

**Author:** Xing Chen<sup>1,2</sup>, Qinghua Cai<sup>1</sup>, Lu Tan<sup>1</sup>, Shuoran Liu<sup>3</sup>, Wen Xiao<sup>3</sup>, Lin Ye<sup>1,\*</sup>

1. State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, the Chinese Academy of Sciences, Wuhan, Hubei, P. R. China

2. University of Chinese Academy of Sciences, Beijing, P. R. China

3. Institute of Eastern-Himalaya Biodiversity Research, Dali University, Dali, Yunnan, P. R. China

\* Corresponding author:

Lin Ye

No. 7 Donghu South Road, Wuhan, Hubei, 430072, P. R. China

Email address: [yelin@ihb.ac.cn](mailto:yelin@ihb.ac.cn)

# Abstract

Small waterbodies are a unique aquatic ecosystem with an increasing recognition for their important role in maintaining regional biodiversity and delivering ecosystem services. However, small waterbodies in Northwest Yunnan, one of the most concerned global biodiversity hotspots, remain largely unknown. Here, we investigated the community structure of crustacean zooplankton and their relationships with limnological, morphometric and spatial variables in the highland small waterbodies in Northwest Yunnan in both the dry (October 2015) and rainy (June 2016) seasons. A total of 38 species of crustacean zooplankton were identified in our study, which is significantly higher than many other waterbodies in ~~the many other reported~~ waterbodies in the Yunnan-Guizhou plateau as well as in the Yangtze River basin. This suggests that ~~the~~ highland small waterbodies are critical in maintaining regional zooplankton diversity in Northwest Yunnan. ~~Meanwhile, we found~~ limnological variables ~~could~~ explain most variation of crustacean zooplankton community, comparing to the morphometric and spatial variables in both the rainy and dry seasons. Our study revealed the diversity and community structure of crustacean zooplankton in the highland small waterbodies in Northwest Yunnan and highlighted the importance of small waterbodies in maintaining regional biodiversity.

**Keywords:** Northwest Yunnan; Small waterbodies; Biodiversity; Crustacean zooplankton

# Introduction

Small waterbodies are critical for regional biodiversity and are increasingly recognized for their essential role in maintaining biodiversity and providing ecosystem services (*Williams et al., 2004; Biggs, von Fumetti & Kelly-Quinn, 2017; Kuczyńska-Kippen, 2020*). Small waterbodies with low ~~or even absence of~~ fish and abundant submerged vegetation ~~could~~ support high biodiversity of aquatic organisms and contributed a large proportion of rare or endemic species to local freshwater habitats (*Williams et al., 2004; Oertli et al., 2005; Scheffer et al., 2006*). ~~Meanwhile,~~ small waterbodies have important ecological functions (*Céréghino et al., 2014; Biggs, von Fumetti & Kelly-Quinn, 2017*). Small waterbodies can significantly reduce nutrient concentrations and protect downstream waters (*Cheng & Basu, 2017*). On the other hand, small waterbodies are ~~sensitive and vulnerable because of their small size and will respond directly to~~ environmental changes (*Biggs, von Fumetti & Kelly-Quinn, 2017*).

Crustacean zooplankton is an important group in freshwater ecosystems because they occupy central positions in aquatic food webs, transferring energy to higher trophic levels (*Sommer et al., 1986; Fussmann, 1996*). In addition, crustacean zooplankton is sensitive to climate and environmental change (*Keller & Conlon, 1994; Jones & Gilbert, 2016*). For quite a

long time, the research on crustacean zooplankton in freshwater ecosystems ~~are~~ mainly focused on lakes (*Barbiero et al., 2019*) and reservoirs (*Liu et al., 2020*). Yet, the ecology of crustacean zooplankton in highland small waterbodies remains seldom addressed.

Northwest Yunnan, located in Southwest China, has been designated as a global biodiversity “hot-spot” by World Wildlife Fund (WWF) and International Union for Conservation of Nature (IUCN) because of its rich biodiversity, unique and diverse highland landscape (*Mackinnon et al., 1996; Xu & Wilkes, 2004; Trizzino et al., 2014*). This region is in the upper stream of the Yangtze (Jinsha) River, the Mekong (Lancang) River, the Salween (Nujiang) River, and the Irrawaddy (Dulongjiang) River, attracting extensive attention of local and international communities (*Xu & Wilkes, 2004; Ao et al., 2021*). Currently, most ecology and biodiversity related studies in this region focus on the terrestrial vegetation and endangered wild animals (*Xu & Wilkes, 2004; Li et al., 2014*), yet still few studies addressed the aquatic ecosystems, especially for small waterbodies ecosystems ~~in this region~~.

In this study, we focus on the community structure and species diversity of crustacean zooplankton in highland small waterbodies in Northwest Yunnan, China. ~~As far as our knowledge, the ecology of crustacean zooplankton in highland small waterbodies of Northwest Yunnan remains largely unknown.~~ Besides the limnological variables (e.g. water temperature, nutrients), previous studies have reported that morphometric variables (e.g. surface area, depth) and spatial variables (e.g. distance) also have critical effects on zooplankton diversity and community composition (*Dodson, 1992; MacLeod, Beisner, et al, 2006, Keller & Paterson, 2018*). Here, we hypothesized that ~~the~~ crustacean zooplankton in the small waterbodies are co-determined by limnological, morphometric and spatial variables. Specifically, the main aims of our study are to understand: i) the diversity and community structure of crustacean zooplankton in highland small waterbodies in Northwest Yunnan, ii) the difference of community structure ~~of crustacean zooplankton~~ in rainy and dry seasons ~~in this region~~, iii) how the limnological, morphometric and spatial variables ~~determining~~ the spatiotemporal variations of diversity and community structure ~~of the crustacean zooplankton in highland small waterbodies in Northwest Yunnan~~.

## Materials & Methods

### Study sites and field sampling

The study sites were distributed on the east and west sides of a high mountain ridge located in Gong-shan Country, Yunnan province, China (Fig. 1). ~~The altitude of the ridge is~~ 3700 m above

~~sea level~~. The average annual temperature and precipitation (from August 1, 2014 to July 31 2015) was 7.7 °C and 2515 mm respectively (*Liu et al., 2018*). ~~The east side of the ridge was defined as “area E” and the west side was defined as “area W”~~. There was a disused road lying across the “area E”, which separated this area into Upstream (EU) and Downstream (ED) subgroups. The average elevation and area of small waterbodies are 3131 m and 9.9 m<sup>2</sup> for the area W, 3328 m and 13 m<sup>2</sup> for the area EU, and 3274 m and 41 m<sup>2</sup> for the area ED, respectively. In addition, these ~~temporal~~ small waterbodies ~~were characterized by the absence of fish and~~ abundant macrophytes.

Two ~~field~~ samplings were carried out in the dry (October 2015) and rainy (June 2016) seasons reflecting different hydrological regimes. A total of 32 and 30 small waterbodies were sampled in the dry and rainy seasons, respectively. For each small waterbody, the morphometric and spatial variables, including the water depth, water surface area, coordinates and altitude (using Garmin eTrex20, China) were measured *in-situ*. The physical parameters, including ~~the~~ conductivity (Cond), dissolved oxygen (DO), pH, and water temperature (WT), were measured by a portable multi-parameter device (YSI Professional plus, USA) *in-situ*. Water samples for chemical analysis were collected from the center ~~surface~~ of each small waterbodies using a 350 ml plastic bottle. Ammonia nitrogen (NH<sub>3</sub>N), nitrate nitrogen (NO<sub>3</sub>N), total nitrogen (TN), phosphate (PO<sub>4</sub>P), total phosphorus (TP), dissolved silicate (DSi), and dissolved organic carbon (DOC) were analyzed by ~~the~~ segmented flow analyzer (Skalar SAN++, Netherlands), according to the user manual ~~of Skalar~~. ~~Meanwhile,~~ another 350 ml water sample was filtered through a micro-filter (~1.2 µm, GF/C Whatman) for the measurement of chlorophyll a (Chl-a). The concentration of Chl-a was measured with a spectrophotometer (Shimadzu UV-1800, Japan) with the standard method of APHA (*1999*).

Using 2 L buckets to collect crustacean zooplankton samples from the surface of open water by filtering 20 L water sample with a plankton net (64 µm in mesh size). All crustacean zooplankton samples were preserved in 5% formalin ~~solution~~.

### **Zooplankton counting and identification**

Crustacean zooplankton was counted and identified under the stereoscope (Zeiss Stereo Discovery V20, German). All crustacean zooplankton samples were identified to the species level as far as possible. Specifically, all samples were screened under the stereoscope because of the low density of the crustacean zooplankters. The major reference books for identification were Chiang and Du (*1979*), Shen (*1979*) and Błędzki and Rybak (*2016*).

## Statistical analysis

The rarefaction method was used to compare species richness and Shannon diversity between the rainy season and dry season because biodiversity was affected by sampling efforts, such as the number of sites and individual numbers (Chao *et al.*, 2014). Specifically, we calculated appropriate species richness and Shannon diversity index for the whole of the small waterbodies as the individual numbers were infinity (Chao *et al.*, 2014). Then, we plotted individual-based species rarefaction curves for crustacean zooplankton for each season to compare the differences of species richness and Shannon diversity index.

Further, the similarity percentage analysis (SIMPER) was conducted to investigate differences of crustacean zooplankton community composition between the rainy and dry seasons and to determine the contribution of each species to the observed similarities based on Bray–Curtis dissimilarities (Clarke, 1993). Meanwhile, a nonmetric multidimensional scaling (NMDS) was carried out to illustrate taxonomical and abundance similarity between the rainy and dry seasons.

In order to test our hypothesis, we conducted the variation partitioning with redundancy analysis (RDA) to compare crustacean zooplankton variation relationship with the limnological, morphometric and spatial variables. Spatial variables can represent the importance of migration and dispersal on the crustacean zooplankton communities' structure according to the metacommunity theory (Heino, *et al.*, 2014). To examine and quantify spatial patterns of crustacean zooplankton, we conducted principal coordinates of neighbor matrices analysis (PCNM) to obtain eigenvectors with positive eigenvalues from the distance matrix (Borcard & Legendre, 2002). First, longitude and latitude were converted into Cartesian coordinates (the unit is kilometer). Second, the Euclidian distance matrix among the small waterbodies was calculated. The eigenvectors with positive eigenvalues were used as spatial predictors. Then minimum variables were selected using the “forward selection” (Blanchet, Legendre & Borcard, 2008).

To reduce the weight of the abundance species, crustacean zooplankton abundance data were Hellinger transformed before the variation partitioning analyses (Legendre & Gallagher, 2001). To avoid collinearity, only the variables with the correlation coefficient below 0.7 were selected in the RDA (Dormann, *et al.*, 2013). Finally, TN, NO<sub>3</sub>N, NH<sub>3</sub>N, PO<sub>4</sub>P, DSi, DOC, Cond, WT, Chl-a, area, and depth were selected in RDA. All analyses were implemented with R statistical software (R Development Core Team, 2020). The rarefaction was carried out with “iNEXT” package (Hsieh *et al.*, 2016). PCNM and RDA variation partitioning were performed using “vegan” package (Oksanen *et al.*, 2019).

# Results

## Community composition

A total of 38 crustacean zooplankton taxa, including 20 Cladocera species and 18 Copepoda species, were identified in all sampled small waterbodies in northwest Yunnan (Table 1). In the rainy season, the most common species in this region were *Cyclops vicinus*, *Mesocyclops leuckarti*, *Alona affinis*, *Microclops varicaricans*, *Moina irrasa*, *Cyclops strenuuss*, *Ectocyclops phaleratus*, which occurred in more than 50% of the surveyed small waterbodies. In the dry season, *Chydorus ovalis*, *M. varicaricans*, *Tropocyclops prasinus*, *Ceriodaphnia laticaudata*, *Alonella exigua*, had a relative occurrence above 50% in this region (Table 1).

The species accumulation curves showed that we have sampled considerable individuals of crustacean zooplankton in both the rainy and dry seasons (Fig. 2). The observed value of species richness is almost same as the estimated values of species richness in both the rainy and dry seasons. And the species richness in the dry season is significantly higher than that in the rainy season (Fig. 2a). However, Shannon diversity index showed that an explicit overlapping of observed and estimated species richness for the rainy and dry seasons (Fig. 2b).

The results of the Similarity Percentage analysis (SIMPER) are summarized in Table 2. As shown in Table 2, the composition and abundance of crustacean zooplankton changed significantly between the rainy and dry seasons. *M. leuckarti*, *M. varicaricans*, *C. ovalis*, *C. vicinus*, *A. exigua* and *Sinodiaptomus sarsi* are most influential species based on cumulative contribution. Further NMDS showed that species compositions had significant differences between the rainy and dry seasons (Fig. 3).

## Crustacean zooplankton community variation partitioning

The variation explained by limnological variables, morphometric variables and spatial variables differed between the rainy and dry seasons (Fig. 4). In the dry season, the limnological, morphometric, and spatial variables totally explained 23.69% of the crustacean zooplankton community structure (Fig. 4a). The limnological variables explained the most variation of zooplankton community structure (7.01%), which is significantly higher than spatial variables (3.44%) and morphometric variables (1.70%). Variation partitioning revealed 7.31% of the shared variation between limnological variables and spatial variables. However, only 1.48% of the variation was shared between the morphometric and spatial variables.



In the rainy season, all predictors explained 26.65% of the crustacean zooplankton community structure (Fig. 4b), which was slightly higher than the dry season. The limnological variables ~~pure~~ explained 18.12% of the variation. And the spatial variables had a lower contribution (3.45%) and followed by morphometric variables (0.64%).

## Discussion

One interesting finding of our study is that the species richness ~~in the small waterbodies~~ in our study area is significantly higher than many other reported waterbodies in the Yunnan-Guizhou plateau as well as in the Yangtze River basin (Table 3). For example, Guo (2009) identified ~~a total of~~ 36 crustacean zooplankton species in 13 different lakes in the Yunnan-Guizhou plateau with areas ranged from 10.7 to 297.9 km<sup>2</sup>. Another similar research carried out in the plateau lake (Erhai Lake) in Yunnan province only recorded 11 crustacean zooplankton species for 12 field stations with one-year continuous monthly monitoring (Yang *et al.*, 2014). Comparing to the lakes and other waterbodies, small waterbodies usually have a high habitat heterogeneity which can support more diverse species and maintain a high diversity community (Williams *et al.*, 2004).

The absence of ~~predation~~ fish and ~~complicated~~ habitat with abundant macrophytes might explain high crustacean zooplankton diversity in the highland small waterbodies in our study. Fish more likely to be absent in small and isolated waterbodies because of high risks of extinction and low chances of colonization (Scheffer *et al.*, 2006). In our field survey, we did not observe fish in ~~all small~~ waterbodies. Presence of fish could ~~profound~~ impact ~~on~~ crustacean zooplankton community structure by reducing species richness and simplifying community composition, especially in small waterbodies (Scheffer *et al.*, 2006). The predation from fish is an important factor affecting crustacean zooplankton in small lakes (Pinel-Alloul & Mimouni, 2013). Meanwhile, some studies also suggested that macrophyte cover is important to maintain zooplankton diversity because of macrophyte provide good habitats for zooplankton (Celewicz-Goldyn & Kuczyńska-Kippen, 2017). These natural, temporal, and mountain small waterbodies have good water quality and high coverage of macrophytes (Kuczyńska-Kippen, 2020), providing ~~lots of available~~ ecological niches ~~and~~ for rare species (such as *Graptoleberis testudinaria* and *A. karua*) or endemic species (such as *T. hebereri* and *N. mariadvigae*).

~~We found~~ the species composition in the rainy and dry seasons are quite different in the highland small waterbodies in Northwest Yunnan, China. Specifically, *C. ovalis*, *M. varicaricans*, *T. prasinus*, *C. laticaudata*, *A. exigua* were the most common species in the dry season (~~Table 1~~). However, in the rainy season, the common species shifted to *C. vicinus*, *M. leuckarti*, *A. affinis*,



*M. varicicans*, *M. irrasa*, *C. strenuuss*, *E. phaleratus*. Among these species, we found 2 endemic species (*Tropodiptomus hebereri* and *Neutrodipodomus mariadvigae*) in the Yunnan-Guizhou plateau (Shen, 1979). Meanwhile, we also found 9 common species (e.g. *C. vicinus*, *M. leuckarti*, *A. affinis*, *M. irrasa*) in the Yangtze River basin (Chiang & Du, 1979).

We identified the limnological variables could explain the most variation of crustacean zooplankton community in both the rainy (NO<sub>3</sub>N, DSI, Cond, DO) and dry (NO<sub>3</sub>N and WT) seasons, comparing to the morphometric and spatial variables. This result is coherent with many other studies which also showed limnological variables are the most important factors in explaining variations of crustacean zooplankton compared to spatial variables. (Heino, et al., 2017; Lévesque et al., 2017; Brasil et al., 2020). Our finding suggests that environmental filter played a key role in community structure in the highland small waterbodies in Northwest Yunnan. It was possibly related to the environmental heterogeneity of small waterbodies in the Northwest of Yunnan. Previous experience showed that the environmental heterogeneity of small waterbodies in the Northwest of Yunnan depended on the watershed and precipitation (Liu et al., 2018).

We should add a caveat that not all potential limnological variables affecting the crustacean zooplankton communities were examined in our study due to the limited data. Some researches suggested that macrophytes cover is important to maintain zooplankton diversity because of macrophyte provide shelter and concealment from predators (Cazzanelli et al., 2008; Sagrario et al., 2009). In our study, we didn't address the effects of macrophytes. However, the zooplankton samples were collected in the open water area with no macrophytes, suggesting the direct effect of macrophytes on zooplankton samples we collected may weak. Future works in discovering the factor shaping zooplankton community in small waterbodies could focus on the effect of macrophyte which are probably important to affect zooplankton species assemblages (Celewicz-Goldyn et al., 2017).

## Conclusions

In this study, we reported the crustacean zooplankton community and their relationships with the limnological, morphometric and spatial variables in the highland small waterbodies in Northwest Yunnan for both the rainy and dry seasons. We identified a total of 38 species of crustacean zooplankton in the highland small waterbodies, which is significantly higher than many other waterbodies in the Yunnan-Guizhou plateau as well as in the Yangtze River basin. This suggests that small waterbodies are biodiversity hotspot and are important in maintaining regional zooplankton diversity in Northwest Yunnan. Limnological variables could explain the most

variation of crustacean zooplankton community, comparing to morphometric and spatial variables in both the rainy and dry seasons. This study improved our understanding of the diversity and community structure of crustacean zooplankton in the highland small waterbodies in Northwest Yunnan and highlighted the importance of small waterbodies for biodiversity conservation and research.

## Acknowledgements

We thank Jun Sun, Xiaoyang He and Wenshu Yang for their assistance during field samplings.

## References

- Ao SC, Chiu MC, Li XF, Tan L, Cai QH, Ye, L. 2021.** Watershed farmland area and instream water quality co-determine the stream primary producer in the central Hengduan Mountains, southwestern China. *Science of The Total Environment* 770: 145267. DOI: 10.1016/j.scitotenv.2021.145267.
- APHA. 1999.** *Standard Methods for the examination of waste water, 20th Edn.* American Public Health Association.
- Beisner, BE, Peres-Neto, PR, Lindström, ES, Barnett, A, Longhi, ML. 2006.** The role of environmental and spatial processes in structuring lake communities from bacteria to fish. *Ecology* 87: 2985-2991.
- Biggs J, von Fumetti S, Kelly-Quinn M. 2017.** The importance of small waterbodies for biodiversity and ecosystem services: implications for policy makers. *Hydrobiologia* 793:3–39. DOI: 10.1007/s10750-016-3007-0.
- Biggs J, Williams P, Whitfield M, Nicolet P, Brown C, Hollis J, Arnold D, Pepper T. 2007.** The freshwater biota of British agricultural landscapes and their sensitivity to pesticides. *Agriculture, Ecosystems and Environment* 122:137–148. DOI: 10.1016/j.agee.2006.11.013.
- Blanchet FG, Legendre P, Borcard D. 2008.** Forward selection of explanatory variables. *Ecology* 89:2623–2632. DOI: 10.1890/07-0986.1.
- Błędzki LA, Rybak JI. 2016.** *Freshwater crustacean zooplankton of Europe.* Berlin: Springer International Publishing. DOI: 10.1007/978-3-319-29871-9.
- Borcard D, Legendre P. 2002.** All-scale spatial analysis of ecological data by means of principal coordinates of neighbour matrices. *Ecological Modelling* 153:51–68. DOI: 10.1016/S0304-3800(01)00501-4.
- Brasil J, Santos JBO, Sousa W, Menezes RF, Huszar VLM, Attayde JL. 2020.** Rainfall leads to habitat homogenization and facilitates plankton dispersal in tropical semiarid lakes. *Aquatic Ecology* 54:225–241. DOI: 10.1007/s10452-019-09738-9.
- Cazzanelli M, Warming TP, Christoffersen KS. 2008.** Emergent and floating-leaved macrophytes as refuge for zooplankton in a eutrophic temperate lake without submerged vegetation. *Hydrobiologia* 605: 113–122.
- Celewicz-Goldyn S, Kuczynska-Kippen N. 2017.** Ecological value of macrophyte cover in creating habitat for microalgae (diatoms) and zooplankton (rotifers and crustaceans) in

- small field and forest water bodies. *PLoS One* 12:e0177317. DOI: 10.1371/journal.pone.0177317.
- Céréghino R, Boix D, Cauchie HM, Martens K, Oertli B. 2014.** The ecological role of ponds in a changing world. *Hydrobiologia* 723:1–6. DOI: 10.1007/s10750-013-1719-y.
- Chao A, Gotelli NJ, Hsieh TC., Sander EL, Ma KH, Colwell RK, EllisonAM. 2014.** Rarefaction and extrapolation with Hill numbers: a framework for sampling and estimation in species diversity studies. *Ecological monographs* 84:45-67. DOI: 10.1890/13-0133.1.
- Cheng FY, Basu NB. 2017.** Biogeochemical hotspots: Role of small water bodies in landscape nutrient processing. *Water Resource Research* 53:5038-5056. DOI:10.1002/2016WR020102.
- Chiang SC, Du NS. 1979.** *Fauna Sinica, Crustacean: Freshwater Cladocera*. Beijing: Science Press.
- Clarke, KR. 1993.** Non - parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology*. 18:117-143.
- Deng D, Xie P, Zhou Q, Yang H, Guo L, Geng H. 2008.** Field and experimental studies on the combined impacts of cyanobacterial blooms and small algae on crustacean zooplankton in a large, eutrophic, subtropical, Chinese lake. *Limnology* 9:1–11. DOI: 10.1007/s10201-007-0229-x.
- Dodson S. 1992.** Predicting crustacean zooplankton species richness. *Limnology and Oceanography*. DOI: 10.4319/lo.1992.37.4.0848.
- Dong Y, Wang Z. 2014.** Zooplankton community structure and its seasonal variation in the surface water of Lugu Lake. *Journal of Hydroecology* 35:38-45.
- Dormann, CF, Elith, J, Bacher, S, Buchmann, C, Carl, G, Carré, G, Marquéz, JRG, Gruber, B, Lafourcade, B, Leitão, PJ, Münkemüller, T, McClean, C, Osborne, PE., Reineking, B, Schröder, B, Skidmore, AK., Zurell, D, Lautenbach, S. 2013.** Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. *Ecography* 36: 27-46. DOI: 10.1111/j.1600-0587.2012.07348.x
- Fussmann G. 1996.** The importance of crustacean zooplankton in structuring rotifer and phytoplankton communities: An enclosure study. *Journal of Plankton Research* 18:1897–1915. DOI: 10.1093/plankt/18.10.1897.
- Gianuca AT, Engelen J, Brans KI, Hanashiro FTT, Vanhamel M, van den Berg EM, Souffreau C, Meester L De. 2018.** Taxonomic, functional and phylogenetic metacommunity ecology of cladoceran zooplankton along urbanization gradients. *Ecography*. DOI: 10.1111/ecog.02926.
- Guo N, Zhang M, Yu Y, Qian S, Li D, Kong F. 2009.** Crustacean zooplankton communities in 13 lakes of Yunnan-Guizhou plateau: Relationship between crustacean zooplankton biomass or size structure and trophic indicators after invasion by exotic fish. *Annales de Limnologie* 45:279–288. DOI: 10.1051/limn/2009022.

- 344 **Heino J, Soininen J, Alahuhta J, Lappalainen J, Virtanen R. 2017.** Metacommunity ecology  
345 meets biogeography: effects of geographical region, spatial dynamics and environmental  
346 filtering on community structure in aquatic organisms. *Oecologia* 183 121-137.
- 347 **Hsieh TC, Ma KH, Chao A. 2016.** iNEXT: an R package for rarefaction and extrapolation of  
348 species diversity (Hill numbers). *Methods in Ecology and Evolution* 7:1451–1456. DOI:  
349 10.1111/2041-210X.12613.
- 350 **Jones NT, Gilbert B. 2016.** Changing climate cues differentially alter zooplankton dormancy  
351 dynamics across latitudes. *Journal of Animal Ecology* 85:559–569. DOI: 10.1111/1365-  
352 2656.12474.
- 353 **Keller W, Conlon M. 1994.** Crustacean Zooplankton Communities and Lake Morphometry in  
354 Precambrian Shield Lakes. *Canadian Journal of Fisheries and Aquatic Sciences* 51:2424–  
355 2434. DOI: 10.1139/f94-242.
- 356 **Kuczyńska-Kippen N. 2020.** Biodiversity of Zooplankton in Polish Small Water Bodies. In:  
357 *Handbook of Environmental Chemistry*. 55–76. DOI: 10.1007/978-3-030-12139-6\_3.
- 358 **Legendre P, Gallagher ED. 2001.** Ecologically meaningful transformations for ordination of  
359 species data. *Oecologia* 129:271–280. DOI: 10.1007/s004420100716.
- 360 **Lévesque D, Pinel-Alloul B, Méthot G, Steedman R. 2017.** Effects of climate, limnological  
361 features and watershed clearcut logging on long-term variation in zooplankton communities  
362 of Boreal Shield lakes. *Water*. 9 733.
- 363 **Li Y, Li D, Ren B, Hu J, Li B, Krzton A, Li M. 2014.** Differences in the activity budgets of  
364 yunnan snub-nosed monkeys (*rhinopithecus bieti*) by age-sex class at xiangguqing in  
365 baimaxueshan nature reserve, China. *Folia Primatologica* 85:335–342. DOI:  
366 10.1159/000368831.
- 367 **Liu SR, He XY, Yang WS, Ren GP, Li YP, Zhou J, Cai QH, Xiao W. 2017.** Spatial  
368 distribution and significance of high mountain micro-waterbodies in northwestern Yunnan,  
369 China. *Journal of Hydroecology* 38:18–23.
- 370 **Liu S, Lu T, Yang D, Ren G, He X, Yang W, Cai Q, Xiao W. 2018.** Spatiotemporal  
371 Environmental Heterogeneity of Alpine Micro-Waterbodies. *Fresenius Environmental*  
372 *Bulletin* 27:8088–8095.
- 373 **Mackinnon J, Sha M, Cheung C, Carey G, Xiang Z, Melville D. 1996.** *A biodiversity review*  
374 *of China*. Hong Kong: WWF China Programme.
- 375 **MacLeod J, Keller W, Paterson AM. 2018.** Crustacean zooplankton in lakes of the far north of  
376 Ontario, Canada. *Polar Biology* 41:1257–1267. DOI: 10.1007/s00300-018-2282-9.
- 377 **Oertli B, Biggs J, Céréghino R, Grillas P, Joly P, Lachavanne JB. 2005.** Conservation and  
378 monitoring of pond biodiversity1: Introduction. In: *Aquatic Conservation: Marine and*  
379 *Freshwater Ecosystems*. 535–540. DOI: 10.1002/aqc.752.
- 380 **Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlinn D, Minchin PR,**  
381 **O’Hara RB, Simpson GL, Solymos P, Stevens MH. 2019.** vegan: Community Ecology  
382 Package. R package version 2.5-5.
- 383 **Pan J, Xiong F, Li W, Li A. 2009.** Community structure and spatial distribution of crustacean  
384 zooplankton in Lake Fuxian, Yunnan, China. *Journal of Lake Sciences*. 21:408-414.

- 385 **Pinel-Alloul B, Mimouni EA. 2013.** Are cladoceran diversity and community structure linked to  
386 spatial heterogeneity in urban landscapes and pond environments? *Hydrobiologia*. DOI:  
387 10.1007/s10750-013-1484-y.
- 388 **R Development Core Team. 2020.** R: A language and environment for statistical computing. R  
389 Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>  
390 (accessed 05 March 2020).
- 391 **Sagrario G, De Los Angeles M, Balseiro E, Ituarte R, Spivak E. 2009.** Macrophytes as refuge  
392 or risky area for zooplankton: a balance set by littoral predacious macroinvertebrates.  
393 *Freshwater Biology*. 2009; 54: 1042–1053.
- 394 **Scheffer M, Zimmer K, Jeppesen E, Søndergaard M, Butler MG, Hanson MA, Declerck S,  
395 Meester L De. 2006.** Small habitat size and isolation can promote species richness: second-  
396 order effects on biodiversity in shallow lakes and ponds. *Oikos* 112:227–231. DOI:  
397 10.1111/j.0030-1299.2006.14145.x.
- 398 **Shen JR. 1979.** *Fauna Sinica Crustacea Freshwater Copepoda*. Beijing: Science Press.
- 399 **Shurin JB, Winder M, Adrian R, Keller WB, Matthews B, Paterson AM, Paterson MJ,  
400 Pinel-Alloul B, Rusak JA, Yan ND. 2010.** Environmental stability and lake zooplankton  
401 diversity - contrasting effects of chemical and thermal variability. *Ecology Letters* 13:453–  
402 463. DOI: 10.1111/j.1461-0248.2009.01438.x.
- 403 **Sommer U, Gliwicz ZM, Lampert W, Duncan A. 1986.** The PEG-model of seasonal  
404 succession of planktonic events in fresh waters. *Archiv für Hydrobiologie* 4:433–471.
- 405 **Trizzino M, Bisi F, Maiorano L, Martinoli A, Petitta M, Preatoni DG, Audisio P. 2014.**  
406 Mapping biodiversity hotspots and conservation priorities for the Euro-Mediterranean  
407 headwater ecosystems, as inferred from diversity and distribution of a water beetle lineage.  
408 *Biodiversity and Conservation* 24:149–170. DOI: 10.1007/s10531-014-0798-z.
- 409 **Wei W, Chen R, Wang L, Fu L. 2017.** Spatial distribution of crustacean zooplankton in a large  
410 river-connected lake related to trophic status and fish. *Journal of Limnology* 76:546–554.  
411 DOI: 10.4081/jlimnol.2017.1622.
- 412 **Williams P, Whitfield M, Biggs J, Bray S, Fox G, Nicolet P, Sear D. 2004.** Comparative  
413 biodiversity of rivers, streams, ditches and ponds in an agricultural landscape in Southern  
414 England. *Biological Conservation* 115:329–341. DOI: 10.1016/S0006-3207(03)00153-8.
- 415 **Xu J, Wilkes A. 2004.** Biodiversity impact analysis in northwest Yunnan, southwest China.  
416 *Biodiversity and Conservation* 13:959–983. DOI: 10.1023/B:BIOC.0000014464.80847.02.
- 417 **Yang W, Deng D, Zhang S, Hu C. 2014.** Seasonal dynamics of crustacean zooplankton  
418 community structure in Erhai Lake, a plateau lake, with reference to phytoplankton and  
419 environmental factors. *Chinese Journal of Oceanology and Limnology* 32:1074–1082. DOI:  
420 10.1007/s00343-014-3204-5.

**Table 1**(on next page)

Relative occurrences of crustacean zooplankton species in all samples, samples in area E, and samples in area W in the rainy (32 samples) and dry (30 samples) seasons.

species	Rainy season			Dry season		
	% of all samples	% of E samples	% of W samples	% of all samples	% of E samples	% of W samples
<i>Alona affinis</i>	65.6	72.7	50.0	0.0	0.0	0.0
<i>Moina irrasa</i>	56.3	54.5	60.0	0.0	0.0	0.0
<i>Chydorus ovalis</i>	40.6	31.8	60.0	93.3	90.1	100.0
<i>Diaphanosoma sp.</i>	31.3	31.8	30.0	12.5	9.9	0.0
<i>Bosmina coregoni</i>	21.9	27.3	10.0	3.3	4.5	0.0
<i>Alona guttata</i>	12.5	18.2	0.0	3.3	4.5	0.0
<i>Ceriodaphnia laticaudata</i>	0.0	0.0	0.0	53.3	45.5	75.0
<i>Alonella exigua</i>	0.0	0.0	0.0	53.3	59.1	37.5
<i>Alona karua</i>	0.0	0.0	0.0	30.0	31.8	25.0
<i>Graptoleberis testudinaria</i>	0.0	0.0	0.0	20.0	18.2	25.0
<i>Alona rectangula</i>	0.0	0.0	0.0	23.3	22.7	25.0
<i>Moina rectirostris</i>	0.0	0.0	0.0	16.7	13.6	25.0
<i>Ceriodaphnia quadrangula</i>	0.0	0.0	0.0	20.0	9.1	50.0
<i>Alonella globulosa</i>	0.0	0.0	0.0	16.7	13.6	25.0
<i>Ceriodaphnia reticulata</i>	0.0	0.0	0.0	13.3	18.2	0.0
<i>Alona quadrangularis</i>	0.0	0.0	0.0	10.0	13.6	0.0
<i>Chydorus barroisi</i>	0.0	0.0	0.0	6.7	9.1	0.0
<i>Alonella sp.</i>	0.0	0.0	0.0	6.7	9.1	0.0
<i>Alona sp.</i>	0.0	0.0	0.0	3.3	0.0	12.5
<i>Alonella nana</i>	0.0	0.0	0.0	3.3	0.0	12.5
<i>Cyclops vicinus</i>	71.2	63.6	90.0	0.0	0.0	0.0



<i>Mesocyclops leuckarti</i>	71.2	63.6	90.0	0.0	0.0	0.0
<i>Microclops varicaricans</i>	62.5	50.0	90.0	90.0	86.4	100.0
<i>Ectocyclops phaleratus</i>	59.4	38.5	90.0	6.7	9.1	0.0
<i>Cyclops strenuuss</i>	56.3	45.5	80.0	3.3	0.0	12.5
<i>Limnoithona sinensis</i>	46.7	36.4	80.0	26.7	4.5	87.5
<i>Nitocra lacustri</i>	43.8	54.5	20.0	0	0	0
<i>Sinodiaptomus sarsi</i>	43.8	31.8	70.0	0	0	0
<i>Eucyclops serrulatus</i>	37.5	40.9	30.0	16.7	22.7	0.0
<i>Sinocalanus dorrii</i>	21.9	13.6	40.0	10.0	4.5	25.0
<i>Onychocamptus mohammed</i>	21.9	27.3	10.0	46.7	27.3	100.0
<i>Neutrodiaptomus mariadvigae</i>	15.6	13.6	20.0	10.0	0.0	37.5
<i>Bryocamptus sp.</i>	9.4	13.6	0.0	3.3	4.5	0.0
<i>Tropodiaptomus hebereri</i>	6.3	9.1	0.0	13.3	0.0	50.0
<i>Tropocyclops prasinus</i>	0.0	0.0	0.0	66.7	63.6	75.0
<i>Paracyclops fimbriatus</i>	0.0	0.0	0.0	16.7	18.2	12.5
<i>Paracyclops affinis</i>	0.0	0.0	0.0	10.0	9.1	12.5
<i>Schmackeria inopinus</i>	0.0	0.0	0.0	10.0	4.5	25.0

## **Table 2**(on next page)

Results of SIMPER analysis for species that accounted for the 90% of cumulative contribution.

species	Contribution	Standard deviation	Ratio	cumulative contribution	P
<i>M.leuckarti</i>	0.20879	0.20616	1.0127	0.2313	0.675
<i>M.varicaricans</i>	0.15750	0.16750	0.9403	0.4058	0.001
<i>C.ovalis</i>	0.10450	0.14320	0.7297	0.5216	0.001
<i>C.vicinus</i>	0.07331	0.09875	0.7424	0.6028	0.001
<i>A.exigua</i>	0.06395	0.14393	0.4443	0.6737	0.019
<i>S.sarsi</i>	0.04930	0.12002	0.4108	0.7283	0.091
<i>E.phaleratus</i>	0.02948	0.04346	0.6783	0.7610	0.006
<i>C.laticaudata</i>	0.02447	0.05694	0.4299	0.7881	0.016
<i>L.sinensis</i>	0.02324	0.04526	0.5136	0.8139	0.600
<i>C.strenuuss</i>	0.02290	0.04765	0.4806	0.8392	0.034
<i>M.irrasa</i>	0.01874	0.03545	0.5286	0.8600	0.008
<i>E.serrulatus</i>	0.01756	0.04665	0.3765	0.8794	0.464
<i>A.affinis</i>	0.01276	0.01787	0.7145	0.8936	0.001

# **Table 3**(on next page)

A comparison of the species richness in the small waterbodies in the highland small waterbodies in Northwest Yunnan with other waterbodies in Yunnan-Guizhou plateau and Yangtze River basin.

*n* indicating the number of total samples in the reported case.

1

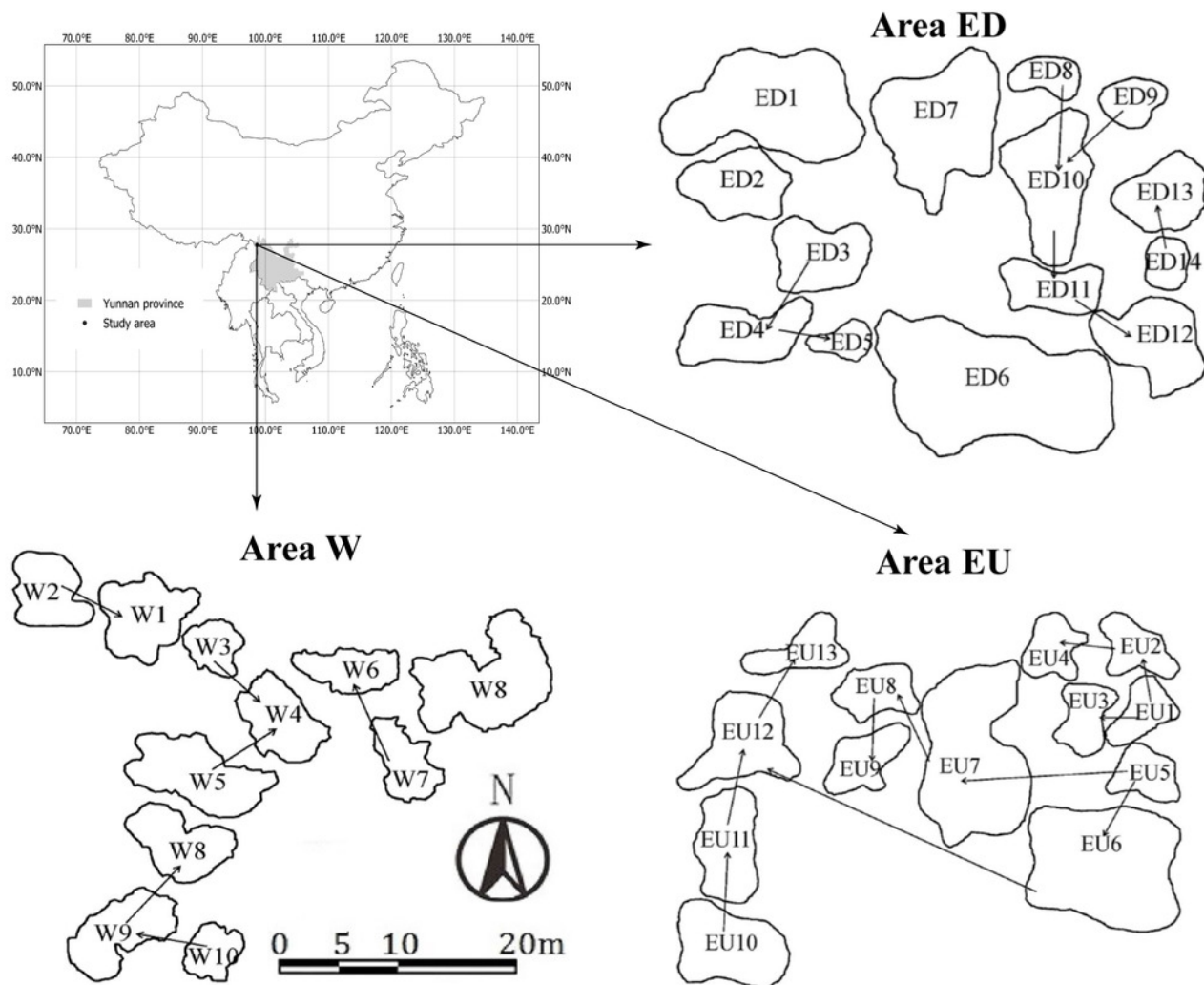
Study area	Province	Area(km <sup>2</sup> )	Species richness	Reference
Thirteen lakes in Yunnan and Guizhou ( <i>n</i> = 112)	Yunnan and Guizhou	10.7~297.9	36	<i>Guo et al., 2009</i>
Gaoyou Lake ( <i>n</i> = 26)	Jiangsu	674	26	<i>Wei et al., 2017</i>
Chaohu Lake ( <i>n</i> = 228)	Anhui	780	23	<i>Deng et al., 2008</i>
Lugu Lake ( <i>n</i> = 36)	Yunnan	57.7	23	<i>Dong et al., 2014</i>
Fuxian Lake ( <i>n</i> = 220)	Yunnan	211	8	<i>Pan et al., 2009</i>
Erhai ( <i>n</i> = 144)	Yunnan	249	11	<i>Yang et al., 2014</i>
Our study ( <i>n</i> = 62)	Yunnan	<0.001	38	Our study

2

# Figure 1

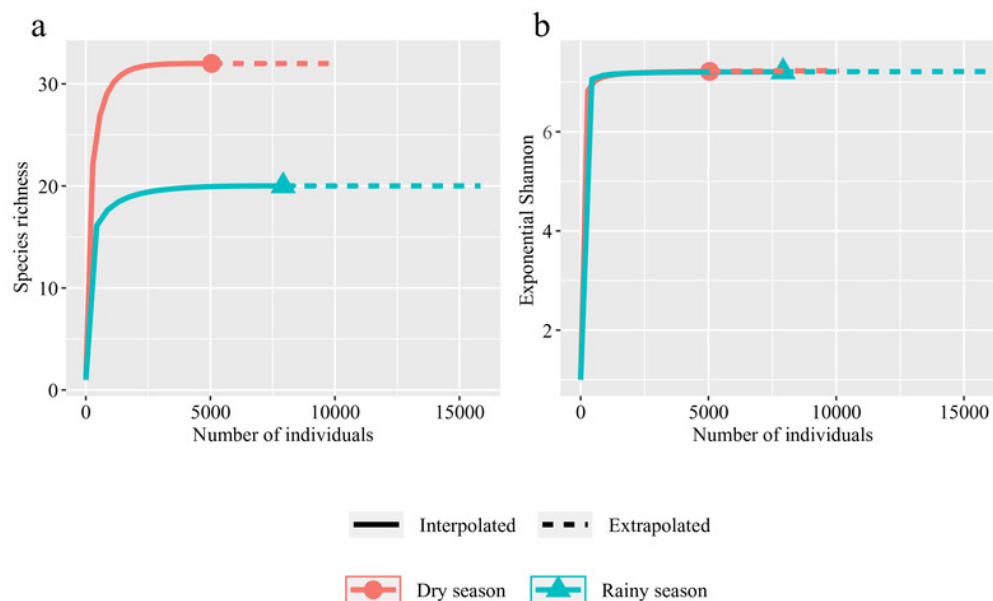
Sampling areas and spatial distribution of small waterbodies in Gongshan County, Yunnan province, China.

The arrows represent the connectivity and water flow direction.



# Figure 2

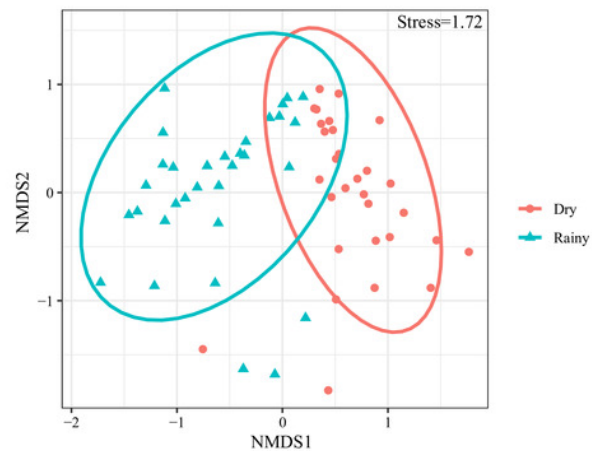
Individual-based rarefaction for the dry (red) and rainy (green) seasons. Symbols represent species richness (a) and exponential Shannon (b). Continuous lines refer to interpolation, dotted lines refer to extrapolation.





# Figure 3

Non-metric multidimensional scaling ordination (NMDS) of crustacean zooplankton communities.



# Figure 4

Venn diagram representing the variation partitioning of crustacean zooplankton community composition explained by explanatory variable.

(Lim) Limnological variables. (Spa) spatial variables represented by principal coordinates of neighbour matrices. (Mor) morphometric variables. (a) dry season. (b) rainy season.

