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Original article

The Relationships between Benthic Macroinvertebrate and Environmental Factors in Iancheon and Bukcheon Streams, Korea

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Abstract In this study, we investigated the relationships between benthic macroinvertebrate assemblages and various environmental factors in Iancheon (NIA) and Bukcheon (NBC) streams, Korea. We collected benthic macroinvertebrates and 33 environmental factors in April 2017 at 9 sites (5 sites in NIA and 4 sites in NBC). We identified 93 species (5 phyla, 9 classes, 16 orders, and 53 families) and 69 species (5 phyla, 9 classes, 17 orders, and 47 families) in NIA and NBC streams, respectively. Considering benthic macroinvertebrate index (BMI), NIA (88.2) and NBC (80.2) streams were in "very good" status. Upstream areas showed the highest scores, 95.5 (NIA1) and 94.2 (NBC1), whereas BMI score was the lowest in downstream areas of both streams, especially in NBC4 (51.0 "bad" status). Cluster analysis and non-metric multidimensional scaling analysis represented the differences of benthic macroinvertebrate assemblages according to spatial and anthropogenic gradients. Our findings provide reference data and highlight the need for the continued monitoring to maintain the good status and manage macroinvertebrate diversity in these two streams, in Sangju-si, Korea.

Key words: community indices, freshwater biodiversity, headwater, non-metric multidimensional scaling

INTRODUCTION

Freshwater ecosystems possess high biodiversity, encompassing nearly 6% of all species identified up to now (Dudgeon *et al.*, 2006). However, freshwater biodiversity is more severely deteriorated than terrestrial and marine biodiversity as the result of anthropogenic disturbances (e.g., climate change, irrigation water, chemical pollution, physical habitat destruction, and invasive species settlement (Ormerod *et al.*, 2010). According to Living Planet Report 2012 (WWF, 2012), living planet index (LPI) of freshwater ecosystems surveyed from 1970 to 2010 decreased by 76% in average, indicating the highest decrease compared with those of marine or terrestrial habitat. Despite the various threats of loss of freshwater biodiversity, there is a lack of studies on headwater and small streams, which are the source of the river environment and occupy an important position in the local freshwater environment.

Benthic macroinvertebrates form a taxonomically diverse group of animals with a wide variety of life history traits (Wallace and Webster, 1996), including a significant variation in habitat preference. These characteristics support the

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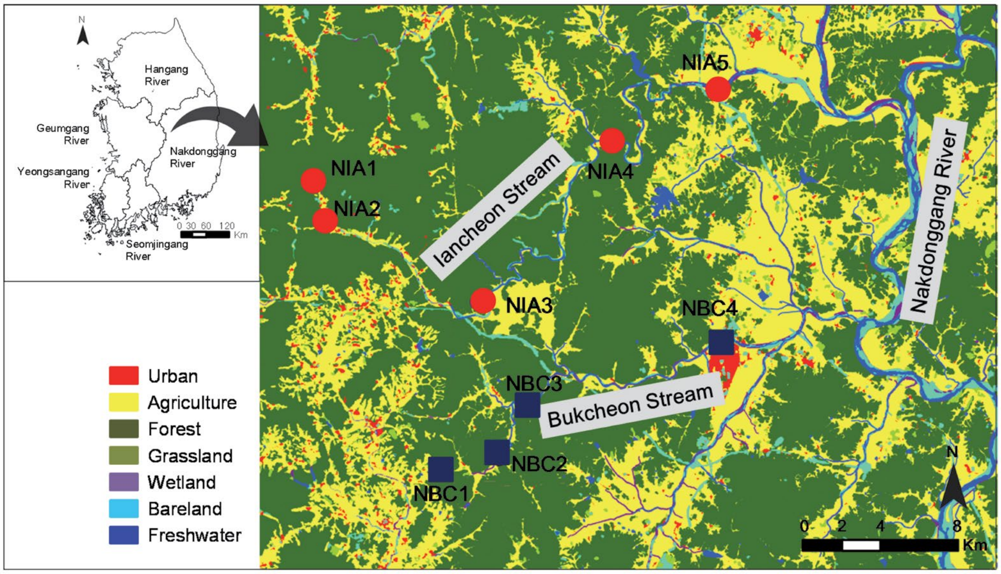

Fig. 1. Location of the sampling sites in Iancheon and Bukcheon streams, Korea.

widespread use of benthic macroinvertebrates as environmental indicator species (Hellawell, 1986; Rosenberg and Resh, 1993; Wright *et al.*, 2000). Therefore, the diversity of benthic macroinvertebrates is an important issue for the integrated assessment of living organisms and freshwater environments (Barbour *et al.*, 1999; Courtney and Clements, 2002; Dalu *et al.*, 2017).

Iancheon (NIA) and Bukcheon (NBC) streams are representative streams in Sangju-si, Gyeongsangbuk-do, Korea. NIA stream originates from Gallyeong, Donggwan-ri, Hwanam-myeon, Sangju-si, Gyeongsangbuk-do, flows in Oeseo-myeon, Euncheok-myeon, and Ian-myeon, and merges into Yeonggang River. The stream length is 38.5 km and the basin area 241.21 km^2 (WAMIS, http://wamis.go.kr). The Taemaek Coal Mine, the only anthracite mine in the Yeongnam region, is located near the upstream region of the NIA stream and ceased its operations in December 2005. After a summer flooding in 2007, the outflow of acidic water from the mine reached some sections upstream of the NIA stream. This event and the proximity to a coal mine underscore the need for constant monitoring of water quality and biodiversity. However, few studies have been conducted on this area and only one study (data from 2005) on the diversity of benthic macroinvertebrate was performed there (Sim, 2007). NBC stream originates from the Baekhak Mountain at the border between Moseo-myeon and Naeseo-myeon, and flows through Naeseo-myeon and Sangju-eup to the Nakdong River. The downstream of NBC is an important place to the Sangju population as a place for resting and leisure. For example, in downstream of NBC, Sangju city operates a water playground every summer since 2008, and the Bukcheon Civic Park provides resting and working out places for residents. Noteworthy, some sections of both streams (NIA, 3.3 km; NBC, 5.5 km) have been designated as water source protection area since 1981.

Therefore, in this study, we surveyed the biodiversity of benthic macroinvertebrates-bioindicators of the status of freshwater ecosystems-in NIA and NBC streams. Then, we analyzed the relationships between benthic macroinvertebrate assemblages and various environmental factors to identify factors influencing the benthic macroinvertebrate assemblage in two streams located in Sajngju-si, Korea.

MATERIALS AND METHODS

1. Ecological data

We collected benthic macroinvertebrates with a Surber net (30 cm × 30 cm, 250 μm mesh) at 9 sampling sites in two streams, 4 sites in NBC and 5 sites in NIA in spring season, 2017 usually when no heavy rain and/or no severe drought (Fig. 1). In each site, three replicates were conducted within the riffle zone representing the characteristics of the sampling area within a 50-meter range (Bae *et al.*, 2016). In the

Table 1. Environmental characteristics of each sampling site in Iancheon and Bukcheon streams, Korea.

	Abbreviation	Iancheon Stream					Bukcheon Stream			
		NIA1	NIA2	NIA3	NIA4	NIA5	NBC1	NBC2	NBC3	NBC4
Geography										
Altitude (m)		400	293	180	110	70	229	120	100	60
Distance from source (km)	DFS	1.2	3.4	16.6	31.9	45.7	2.2	6.6	10.3	23.7
Slope (°)		6.3	6.6	3.2	7.1	0.0	13.6	3.7	1.6	0.0
Land use (%)										
Urban		0.0	0.3	0.8	14.1	9.6	1.1	3.4	1.8	37.4
Agriculture		0.0	5.6	22.9	46.3	49.4	20.7	72.8	33.2	43.2
Forest		100.0	93.2	67.7	30.6	35.3	75.2	14.6	62.1	0.0
Grassland		0.0	1.0	0.0	3.0	0.0	2.9	1.0	0.0	3.2
Wetland		0.0	0.0	2.0	3.5	0.0	0.0	8.3	0.4	7.5
Bareland		0.0	0.0	6.6	2.3	5.7	0.0	0.0	2.5	8.7
Hydrology										
Water depth (cm)		10.7	11.0	31.7	33.0	53.3	8.7	20.0	9.3	14.0
Water velocity (m s^{-1})		0.54	0.40	0.48	0.30	0.01	0.27	0.47	0.22	0.01
Substrate composition (%)										
<0.063 mm		0.0	0.0	0.0	0.0	0.0	0.0	1.1	0.4	1.3
0.063~2 mm		2.0	0.8	0.7	0.9	1.3	0.4	2.9	2.8	2.9
2~4 mm		8.1	1.2	0.7	0.9	2.7	0.6	1.7	3.3	3.4
4~8 mm		5.0	2.2	0.7	1.4	4.2	0.8	2.6	2.6	2.4
8~16 mm		2.8	2.8	1.3	5.0	6.8	2.3	4.3	3.8	7.7
16~32 mm		5.0	5.0	3.7	11.8	10.1	5.9	5.5	12.8	15.3
32~64 mm		10.0	7.9	9.9	8.5	23.5	9.5	25.1	18.3	21.2
64~128 mm		15.2	17.9	20.8	20.9	41.4	25.1	40.3	42.9	27.7
128~256 mm		26.4	37.2	32.7	45.6	9.9	41.6	16.5	13.1	18.1
>256 mm		25.4	25.0	29.4	5.0	0.0	13.6	0.0	0.0	0.0
Water quality										
Dissolved oxygen (mg L^{-1})	DO	10.6	10.5	10.8	10.9	10.6	6.9	9.9	10.3	9.3
Dissolved oxygen (%)	DO_%	91.7	93.5	106.6	108.0	103.1	73.3	88.2	96.3	91.1
pH		8.2	8.1	8.4	8.5	8.1	7.0	8.3	8.3	9.1
Conductivity (μS cm^{-1})		66.4	67.8	134.4	137.0	183.2	119.1	119.7	143.0	191.5
Turbidity (NTU)		1.3	2.8	2.6	1.7	3.3	0.0	1.3	1.5	2.9
Biological Oxygen Demand (mg L^{-1})	BOD	3.0	1.5	2.8	2.5	3.7	4.0	1.3	2.2	1.6
Ammonia nitrogen (mg L^{-1})	NH_3-N	0.011	0.004	0.012	0.013	0.021	0.014	0.005	0.017	0.009
Nitrate nitrogen (mg L^{-1})	NO_3-N	1.16	0.77	1.04	1.08	1.01	0.80	1.23	1.18	0.76
Total nitrogen (mg L^{-1})	TN	1.86	1.02	1.25	1.30	1.20	0.99	1.62	1.44	0.89
Phosphate-phosphorus (mg L^{-1})	PO_4-P	0.003	0.004	0.004	0.003	0.003	0.005	0.004	0.003	0.004
Total Phosphate (mg L^{-1})	TP	0.007	0.011	0.006	0.007	0.007	0.01	0.008	0.006	0.008
Chlorophyll *a* (mg L^{-1})	Chl-*a*	0.8	0.7	0.7	0.5	0.5	0.5	0.6	0.8	0.6

laboratory, the collected macroinvertebrates were sorted and preserved in 70% ethanol. Under a microscope (Z10, Nikon, Tokyo, Japan), individuals were identified mainly into the species level (except for some taxa, such as Chironomidae) based on Quigley (1977), Pennak (1978), Brighnam *et al.* (1982), Yun (1988), and Merritt and Cummins (2006).

We collected data on 33 environmental factors within 5 different categories, including geography, land use, hydrology, substrate, and physicochemical water quality which can be generally considered to influence the biodiversity of benthic macroinvertebrates (Table 1). Altitude, slope, stream order, distance from source (DFS), and land cover (%) were extracted from a digital map using ArcGIS 10.6 (ESRI, Redlands, CA, USA). Land cover (%), categorized as urban, paddy field, dry field, forest, grass, wetland, and bare soil, was extracted from a delimited buffer zone (1 km long and 200 m wide) at each site (Bae *et al.*, 2014). Water velocity was measured using a current meter (Model 2100, Swoffer Instruments, Federal Way, WA, USA). Substrate composition was measured based on Cummins and Lauff (1969). Dissolved oxygen, pH, conductivity, and turbidity were measured in situ using a multi-probe meter (YSI 2100, YSI, Yellow Springs, OH, USA). Biological oxygen demand (BOD), total nitrogen (TN), ammonia (NH_3), nitrate (NO_3^-), total phosphorus (TP), ortho-phosphate (PO_4^{3-}), and Chlorophyll-*a* (Chl-*a*) levels were measured in samples (4 L of water) collected in sterile plastic bottles in each site and directly transferred to the laboratory on ice.

2. Data analysis

Community indices such as species richness, abundance (individuals m^{-2}), Shannon diversity index (Shannon and Weaver, 1948), evenness (Pielou, 1975), dominance index (McNaughton, 1967), benthic macroinvertebrate index (BMI) (Kong *et al.*, 2018), and functional feeding groups (FFGs) were calculated to interpret the basic characteristics of benthic macroinvertebrate assemblages in two streams. Five FFGs were used to classify behavioral mechanisms of food acquisition: collector-gatherers (CG), collector-filterers (CF), predators (PR), parasites (PI), scrapers (SC), and shredders (SH) based on a previous study (Merritt and Cummins, 2006).

Then, we analyzed patterns in spatial differences of benthic macroinvertebrate assemblages using multivariate analyses. First, we applied a hierarchical cluster analysis (CA) using the Ward's linkage method with the Bray-Curtis distance. Then, we applied multi-response permutation procedures (MRPP) to evaluate whether or not there were significant differences among the defined clusters from CA. Lastly, a non-metric multidimensional scaling (NMS) was used to figure out the distribution pattern of benthic macroinvertebrate assemblage based on the Bray-Curtis distance as the dissimilarity measure. All analyses were conducted in R (R Core Team, 2016).

RESULTS AND DISCUSSION

Regarding richness, 93 species (5 phyla, 9 classes, 16 orders, and 53 families) were found in the NIA stream and 69 species (5 phyla, 9 classes, 17 orders, and 47 families) were found in the NBC stream. The highest number of species was observed in NIA4 (48 species) and NBC3 (38 species) sites, whereas the downstream area showed the lowest species richness in both streams: 24 species in NIA5 and 11 species in NBC4 (Fig. 2). Similarly, abundance was the lowest in NIA5 (2,263 individuals m^{-2}) and NBC4 (1,067 individual m^{-2}) whereas it was the highest in NIA2 (7,263 individuals m^{-2}) and NBC3 (9,293 individuals m^{-2}) sites.

Considering community indices, dominance index was the highest (0.87) in the NIA5 downstream area, whereas the index was the lowest (0.55) in NBC4 (Table 2). Except for NBC4, the most dominant species were Chironomidae spp. Sub-dominant species in the upstream area included *Amphinemura* KUa (Plecoptera) in NIA1 and *Serratella setigera* (Ephemeroptera) in NBC1. In the downstream areas, sub-dominant species were Elmidae sp. (NIA5) and *Ephemera orientalis* (NBC4). The status of both NIA and NBC streams was "very good" with BMI scores 88.2 and 80.2, respectively (Fig. 3). The upstream areas showed the highest score as 95.5 (NIA1) and 94.2 (NBC1) whereas BMI scores were low in the downstream area in both streams, especially in NBC4 (51.0 BMI, classified as "bad").

The analysis of relative ratio of FFGs based on abundance revealed that the ratio of CG was the highest (42.57%) in NIA, followed by CF (18.53%), SC (16.40%), PR (12.20%), SH (10.29%), and PA (0.02%) (Fig. 4). In the case of NBC, the ratio of CG (44.16%) was also the highest, followed by SC (24.09%), CF (21.64%), PR (8.96%), SH (1.14%), and PA (0.02). For the relative ratio of FFGs based on species richness, CG also showed the highest ratio in both NIA (30.54%) and NBC (27.38%). The second highest ratio in NIA was that of PR (26.33%), followed by SC (22.13%), CF (10.59%), SH (9.98%), and PA (0.43%). In NBC, the second highest ratio was that of SC (24.73%), followed by PR (22.49%), CF (19.26%), SH (5.47%), and PA (0.68%). Considering FFGs in each site, the ratio of SH was the highest in NIA1 (the upstream of NIA), and the ratio of CG increased from upstream to downstream, ranging from 23.34% to 65.00%. In NBC1 and 2 (upstream sites in NBC), the ratio of SC was 39.39% and 36.28%, respectively. The ratio of CG increased from upstream to downstream, from

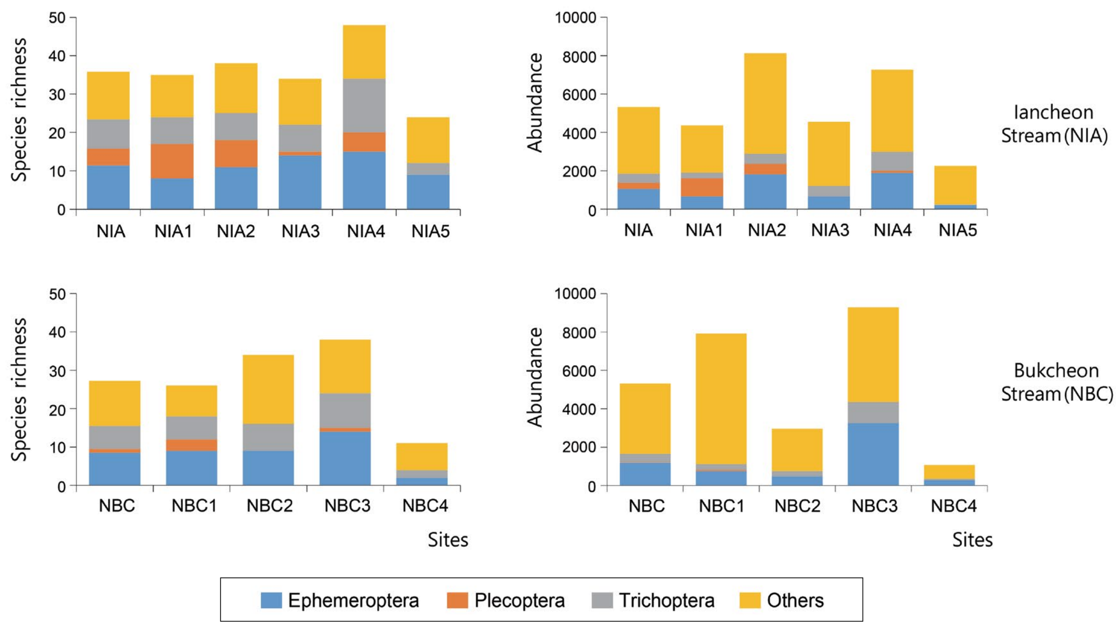

Fig. 2. Species richness and abundance in Iancheon and Bukcheon streams, Korea (NIA and NBC represent the average values of species richness or abundance in each stream).

Table 2. Community indices at each sampling site in Iancheon and Bukcheon streams, Korea.

Streams	Sites	Shannon diversity index	Richness index	Evenness	Dominant index (DI)	Dominant species	Sub-dominant species
Iancheon Stream	NIA1	2.92	18.4	0.40	0.63	Chironomidae spp.	*Amphinemura* KUa
	NIA2	3.24	17.1	0.45	0.53	Chironomidae spp.	*Paraleptophlebia chocorata*
	NIA3	2.78	18.3	0.38	0.64	Chironomidae spp.	*Simulium* sp.
	NIA4	3.24	17.3	0.45	0.62	Chironomidae spp.	*Uracanthella rufa*
	NIA5	1.28	19.9	0.18	0.87	Chironomidae spp.	Elmidae sp.
Bukcheon Stream	NBC1	1.20	17.2	0.17	0.88	Chironomidae spp.	*Serratella setigera*
	NBC2	2.54	19.3	0.35	0.67	Chironomidae spp.	*Cincticostella levanidovae*
	NBC3	3.04	16.9	0.42	0.63	Chironomidae spp.	*Uracanthella rufa*
	NBC4	2.66	22.1	0.37	0.55	*Limnodrilus gotoi*	*Ephemera orientalis*

14.63% (NBC2) to 74.47% (NBC4). Based on species richness, the ratios of PR (30.30%) and SH (24.24%) were the highest in NIA1. Similar with FFGs based on abundance, the ratio of CG was higher in mid to downstream than upstream ranging from 31.91% (NIA4) to 42.42% (NIA3). In NBC1, the ratio of PR was also the highest (25.00%) and the ratio of CG was higher in mid to downstream than upstream ranging from 30.00% (NBC4) to 37.84% (NBC3).

Cluster analysis grouped the sites into three clusters based on the similarities in macroinvertebrate assemblage composition; MRPP showed significant differences in benthic macroinvertebrate assemblage among the three clusters ($A=0.08$, $P<0.05$). Similarly, NMS also showed differences in the composition of the macroinvertebrate assemblages (stress value = 4.9 for the first two axes; Fig. 4, Table 3). Sites with high values for altitude and forest land cover (%) (i.e. upstream sites in NIA and NBC) were located on the upper-left part of the NMS ordination (mainly the sites included in cluster 1), whereas sites with high values of urban land cover, conductivity, and DFS (i.e. downstream sites in NIA

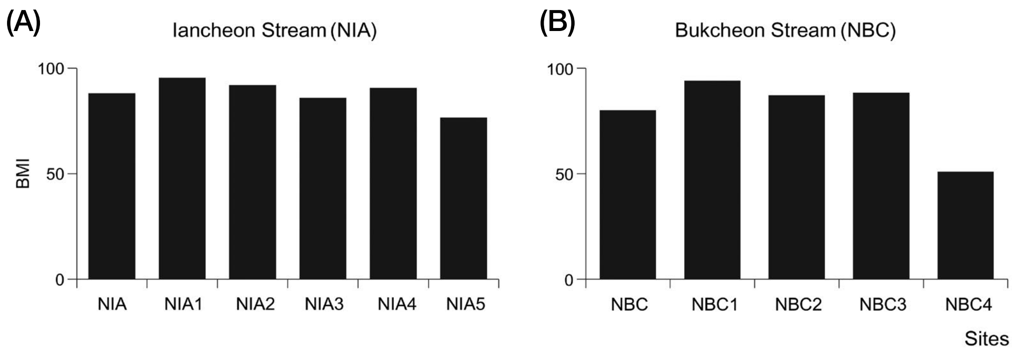

Fig. 3. Benthic macroinvertebrate index in Iancheon (A) and Bukcheon (B) streams, Korea (NIA and NBC represent the average values of benthic macroinvertebrate index in each stream).

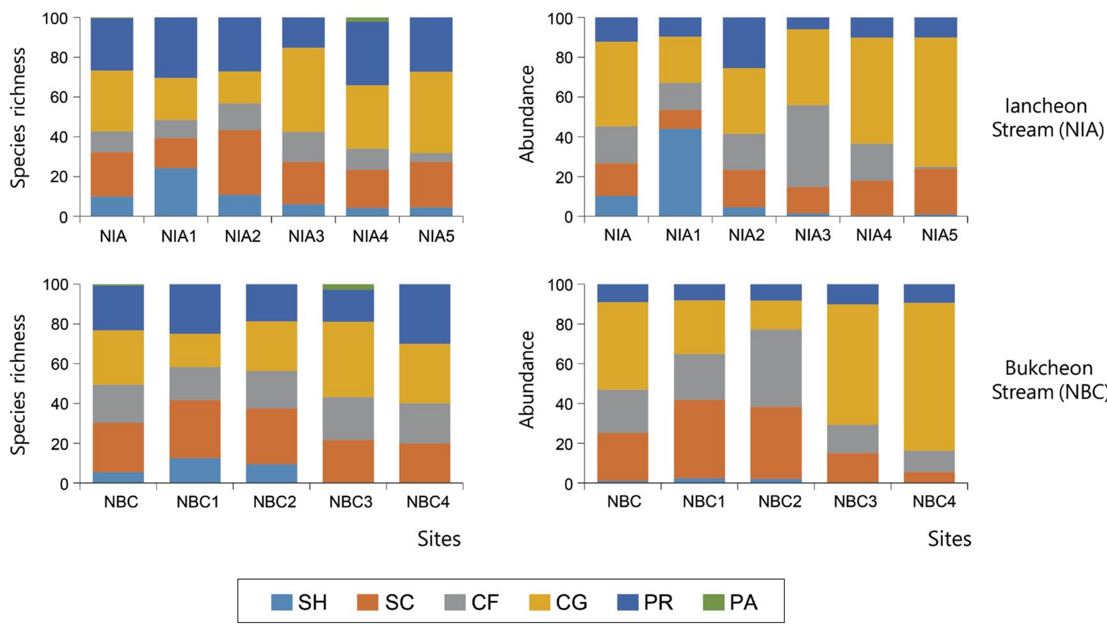

Fig. 4. Composition of functional feeding groups at each sampling site in Iancheon and Bukcheon streams, Korea. SH: Shredder, SC: Scraper, CF: Collector-filterer, CG: Collector-gatherer, PR: Predator, PA: Parasite (NIA and NBC represent the average values of functional feeding group in each stream).

and NBC) were located on the lower-right part based on axis 1 (mainly the sites included in cluster 3). The most influential factor on macroinvertebrate assemblage in both NIA and NBC was conductivity (0.89), followed by altitude (0.88), bareland land cover (0.76), DFS (0.71), 8∼16 mm substrate composition (0.71), urban land cover (0.69), and forest land cover.

The composition of benthic macroinvertebrate assemblages can be affected by various factors, such as latitudinal gradients, stream segmentation, and microhabitat (Bae *et al.*, 2016). In this study, we monitored the diversity of benthic macroinvertebrates in two representative streams in Sangju-si, Korea. These two streams have been rarely surveyed up to now and our research provides the basic information to understand the status of freshwater habitats, as well as biodiversity, in the river basin. The upstream areas of both streams (NIA and NBC), including the headwater region, presented various habitat conditions with good water quality and high biodiversity of benthic macroinvertebrates. In our research, CA and NMS showed the spatial differences of benthic macroinvertebrate assemblages. For example, NIA5 and NBC4, which are downstream areas exposed to various

Table 3. Relationships between environmental factors and the non-metric multidimensional scaling (NMS) ordination of benthic macroinvertebrate assemblage.

Factors	NMS axis		R^2	*P*
	NMS1	NMS2		
Geography				
Altitude (m)	**−0.43**	**0.91**	**0.88**	**0.00**
Distance from source (km)	**0.55**	**−0.84**	**0.71**	**0.03**
Slope (°)	−0.84	0.54	0.51	0.12
Land use (%)				
Urban	**0.96**	**−0.30**	**0.69**	**0.03**
Agriculture	0.43	−0.90	0.45	0.17
Forest	**−0.67**	**0.74**	**0.68**	**0.03**
Grassland	0.60	−0.80	0.06	0.85
Wetland	0.88	−0.48	0.22	0.47
Bareland	**0.86**	**−0.52**	**0.76**	**0.01**
Hydrology				
Water depth (cm)	0.46	−0.89	0.36	0.25
Water velocity (m s^{-1})	**−0.82**	**0.57**	**0.66**	**0.04**
Substrate composition (%)				
<0.063 mm	0.99	0.12	0.31	0.30
0.063~2 mm	0.90	0.44	0.15	0.63
2~4 mm	0.07	1.00	0.39	0.24
4~8 mm	0.24	0.97	0.29	0.35
8~16 mm	**0.91**	**−0.41**	**0.71**	**0.03**
16~32 mm	0.57	−0.82	0.53	0.10
32~64 mm	0.98	−0.21	0.43	0.19
64~128 mm	0.46	−0.89	0.24	0.44
128~256 mm	−0.97	−0.24	0.32	0.33
>256 mm	−0.53	0.85	0.43	0.19
Water quality				
Dissolved oxygen (mg L^{-1})	−0.01	−1.00	0.09	0.71
Dissolved oxygen (%)	0.13	−0.99	0.38	0.25
pH	0.69	−0.72	0.34	0.30
Conductivity (μS cm^{-1})	**0.69**	**−0.73**	**0.89**	**0.00**
Turbidity (NTU)	0.86	−0.52	0.41	0.21
Biological Oxygen Demand (mg L^{-1})	−0.19	0.98	0.02	0.94
Ammonia nitrogen (mg L^{-1})	0.36	−0.93	0.14	0.64
Nitrate nitrogen (mg L^{-1})	−0.65	−0.76	0.12	0.67
Total nitrogen (mg L^{-1})	−0.69	0.72	0.21	0.49
Phosphate-phosphorus (mg L^{-1})	−0.09	1.00	0.08	0.76
Total Phosphate (mg L^{-1})	−0.29	0.96	0.16	0.59
Chlorophyll *a* (mg L^{-1})	−0.53	0.85	0.22	0.48

P values lower than 0.05 are indicated in bold.

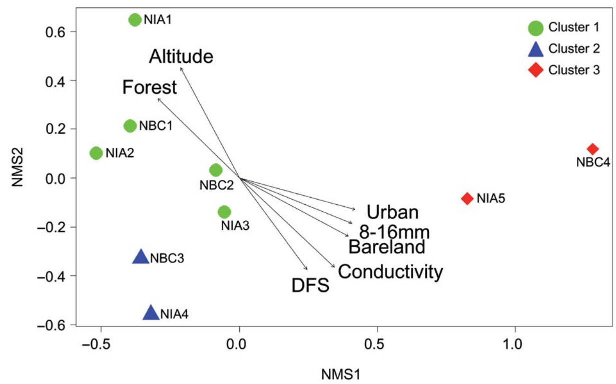

Fig. 5. Non-metric multidimensional scaling ordination based on benthic macroinvertebrate community. Environmental factors with $P < 0.05$ are represented in the figure. Different colors and symbols indicate the result of cluster analysis: green square, cluster 1; blue, cluster 2; and red diamond, cluster 3.

anthropogenic disturbances, such as high ratios of urban and agriculture land cover and poor water quality (i.e., high conductivity), were grouped into the same cluster. On the other hand, the sites located in upstream areas of NIA and NBC showed high biodiversity with various FFGs and were ordinated oppositely to NIA5 and NBC4 (i.e. downstream areas). Generally, the land use conversion in the riparian area from high ratio of forest to agriculture and/or urban can severely influence the biodiversity of benthic macroinvertebrate assemblages (here, upstream to downstream area in NIC and NBC) (Genito *et al.*, 2002; Shieh *et al.*, 2003). Species richness of the macroinvertebrate generally decrease (Zhang *et al.*, 2013), the ratio of CG in both case of abundance and species richness increase. In our study, the ratio of agriculture (%) in land cover increased from 0.0% to 46.3% in NIA and from 20.7% to 43.2% in NBC. In addition, in NBC, the ratio of urban area (%) in land cover was the highest as 37.4% in the downstream. Thus, species richness and BMI were lower mainly downstream in both streams and especially, BMI in NBC4 was the lowest among all study sites. This indicates a healthier freshwater biodiversity in upstream areas with heterogeneous environmental conditions than that in downstream areas (Vannote *et al.*, 1980). Biodiversity in the upstream can influence downstream sites, mainly by allowing the re-establishment of freshwater species in downstream areas that might be strongly deteriorated due to anthropogenic and natural disturbances (Callanan *et al.*, 2014). However, in spite of the importance of headwater ecosystems to support the resilience of species diversity up to downstream, little attention has been given to monitoring benthic macroinvertebrate assemblages in headwater streams. Maintaining the healthy freshwater habitats and high biodiversity is essential, especially for NIA and NBC streams, which are important to provide resting and leisure places for the local population. Therefore, the continuous monitoring of freshwater organisms is needed to preserve their high biodiversity and evaluate the effects of environmental changes and disturbances in future.

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Author contribution statement Conceptualization: M.J. Bae, J.K. Kim, J.G. Hong, S.H. Ryu, Field survey and data collection: M.J. Bae, S.M. Park, J.K. Kim, J.G. Hong, Data analysis and writing: M.J. Bae, Review and editing: M.J. Bae.

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