



Article

Multi-Scale Assessment of Relationships between Fragmentation of Riparian Forests and Biological Conditions in Streams

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Abstract: Due to anthropogenic activities within watersheds and riparian areas, stream water quality and ecological communities have been significantly affected by degradation of watershed and stream environments. One critical indicator of anthropogenic activities within watersheds and riparian areas is forest fragmentation, which has been directly linked to poor water quality and ecosystem health in streams. However, the true nature of the relationship between forest fragmentation and stream ecosystem health has not been fully elucidated due to its complex underlying mechanism. The purpose of this study was to examine the relationships of riparian fragmented forest with biological indicators including diatoms, macroinvertebrates, and fish. In addition, we investigated variations in these relationships over multiple riparian scales. Fragmentation metrics, including the number of forest patches (NP), proportion of riparian forest (PLAND), largest riparian forest patch ratio (LPI), and spatial proximity of riparian forest patches (DIVISION), were used to quantify the degree of fragmentation of riparian forests, and the trophic diatom index (TDI), benthic macroinvertebrates index (BMI), and fish assessment index (FAI) were used to represent the biological condition of diatoms, macroinvertebrates, and fish in streams. PLAND and LPI showed positive relationships with TDI, BMI, and FAI, whereas NP and DIVISION were negatively associated with biological indicators at multiple scales. Biological conditions in streams were clearly better when riparian forests were less fragmented. The relationships of NP and PLAND with biological indicators were stronger at a larger riparian scale, whereas relationships of LPI and DIVISION with biological indicators were weaker at a large scale. These results suggest that a much larger spatial range of riparian forests should be considered in forest management and restoration to enhance the biological condition of streams.

Keywords: forest fragmentation; biological indicators; landscape metrics; RDA model; multi-scale approach

1. Introduction

Land use patterns with strongly fragmented forests or no forests located in stream riparian areas have significant negative impacts on water quality and aquatic ecological communities [1–4] due to alteration of stream environments and sediment run-off mechanisms, pollution, and nutrient loading [5–9]. Thus, land use within riparian areas has become a key concern for stream management and restoration [10]. Previous research has shown that streamside forests affect aquatic ecosystems by

Sustainability **2019**, 11, 5060 2 of 25

providing substantial amounts of energy and woody debris [11–15]. Some previous studies have demonstrated that land use within riparian areas threatens ecosystems through fragmentation of forests and degradation of soil and water properties [16–19]. Therefore, it is evident that riparian forests play an important role as corridors connecting fragmented forests and stream habitats. Furthermore, forest fragmentation within riparian areas has been directly linked with degraded water quality and stream ecosystem health [8,11,20–22], and spatiotemporal changes in land use, logging, intensive forest management, and rapid economic development have played significant roles in accelerating forest fragmentation [23–27]. Human activities in forested areas affect various stream characteristics, such as the microclimate, local air temperature, stream water temperature, humidity, wind speed [28,29], and concentrations of nutrients, sediments, and pollutants in streams, as well as ecological conditions [8,11,30–36]. However, the main characteristics of the relationship between forest fragmentation and stream ecosystems remain poorly understood, because they are associated through complex mechanisms involving numerous other factors (e.g., climate, geology, topography, and hydrological processes) [37–40].

Many previous studies have focused on a particular aspect of watershed forests (i.e., proportion of forested area), and have fallen short of identifying which aspects of fragmentation have the strongest impacts on stream biota. For example, Allan (2004) [41] showed that a greater proportion of forest cover within a watershed was positively linked with various stream conditions. Roy et al. (2003) [42] reported that decreased forest cover was related to degradation of biotic integrity in streams. Furthermore, Kim et al. (2014) [43] reported that the effects of forests at large spatial scales (i.e., forest width) are more important to fish than at small scales. Fragmentation can be characterized as a function of the patch number within a given area, patch size, patch shape, and the spatial distribution of patches [44–46]. Specifically, forest fragmentation can be characterized as forest loss, increased edge areas, decreased size and core area, non-contiguous splitting of large forest areas into smaller fragmented forest patches, and increased distance between patches [47,48].

When investigating the relationships of various land uses and their spatial patterns in riparian areas with stream organisms, identifying the optimal spatial scale is one of the most critical and fundamental issues. In landscape ecology, scale can be defined by two factors, extent and grain size, which vary in time and space. In cross-sectional studies, extent defines the spatial range of the investigation, whereas grain size refers to the unit of analysis. Scale has been a central concept in landscape ecology, as landscape structure and function are scale dependent [49,50]. Often, scientists have preferred to use multiple spatial scales (i.e., extents) to examine relationships between land use types and stream health, as there is no known scale of the relationship [51–53]. Allan et al. (1997) [54] discussed how human activities at various spatial scales impact the stream environment and organisms in streams. The extent of fragmentation is critical to understanding the relationship between forest fragmentation and local ecological processes in streams and surrounding areas [55]. In part, this importance is due to the extent of fragmentation negatively affecting biological integrity by increasing the exposure of streams to light and wind and increasing stresses on aquatic ecosystems caused by temperature fluctuations. Therefore, this work is essential to clarify the extent to which forest fragmentation affects stream environments and organisms in streams. For example, forests hundreds of meters away from streams are associated with the supply of coarse sediments and organic matter, whereas shade from riparian forests can lower water temperatures [56]. Arguably, forest fragmentation in riparian areas may have more significant impacts on stream ecosystems than in other forest types throughout the watershed simply due to stream proximity [2,57,58]. Rich evidence indicates that riparian forests have positive effects, including stream bank stabilization [59], decreasing nutrient and sediment loads from riparian areas [60,61], lowering stream water temperature [62], providing habitat [63], and enhancing biodiversity in streams [64]. Recently, Yirigui et al. (2019) [8] reported that forest fragmentation within a 500-m buffer zone has significant negative effects on biological indicators in streams. According to their study, fragmentation of riparian forests may lower their efficiency for filtering and absorbing nutrients, sediments, and pollutants, resulting in poor stream water quality and biological condition. However, the extent to which riparian fragmented forest affects the biological condition of streams remained unclear. Answering this Sustainability **2019**, 11, 5060 3 of 25

question is essential for planners and managers to make critical decisions regarding effective stream management and restoration strategies.

In this study, we investigated the relationships of forest fragmentation with biological indicators including diatoms, macroinvertebrates, and fish over multiple riparian scales. In addition, we examined the variation in relationships between riparian forest fragmentation and biological indicators for streams over different spatial scales (i.e., extents). The degree of forest fragmentation in riparian areas was assumed to impact the effectiveness of various mechanisms (e.g., filtering, absorbing, and up-taking) of riparian forests, as well as hydrological and biochemical runoff processes [8,65–68], resulting in degraded stream environments (e.g., high levels of pollutants, nutrients, and sediments) and poor biological indicators [37,69–75]. Additionally, we hypothesized that the negative influence of forest fragmentation on biological conditions in streams may vary with riparian buffer size due to the proximity of streams.

2. Materials and Methods

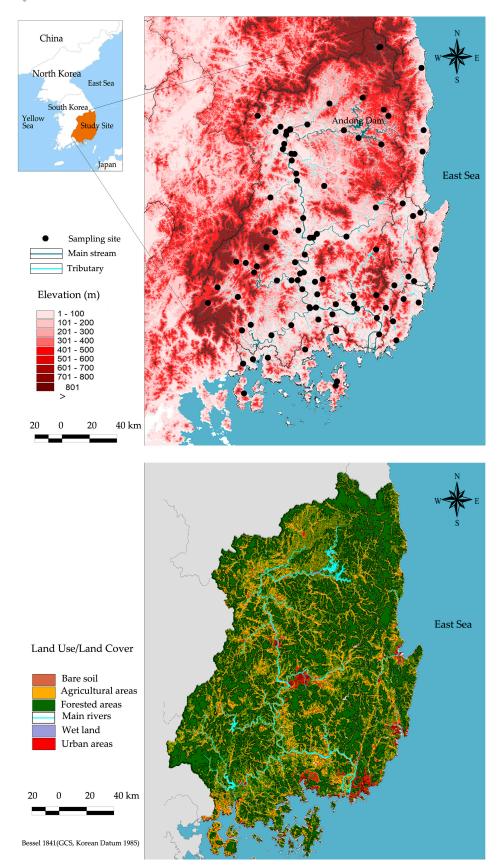
2.1. Study Areas

The Korean peninsula is located between 33°7′ and 43°1′ N latitude, and 124°11′ and 131°53′ E longitude. The area of the Korean Peninsula is 221,000 km², and approximately 45% is within South Korea. The Nakdong River Basin is located between 35°03'and 37°13' north latitudes and between 127°29' and 129°18' east longitudes, accounting for about 25% of South Korea's total geographical area. The Nakdong River system, one of the major river systems in South Korea, occupies the southeastern region and its basin area is 23,702 km²; the Nakdong is also the longest river in Korea, with a length of 511 km [76]. The study area is composed of four major land cover types: commercial (0.2%), agricultural (23.5%), industrial (0.5%), and forest (70.3%). Korean forests were badly degraded during the first half of the 20th century due to watershed urbanization processes, the transition from forest to farmland, dam building, and other processes. These land uses gradually led to increasingly serious degradation of aquatic ecosystems. Total annual precipitation in the basin is 1200 mm, and about 60% of the annual precipitation falls in summer (June to September). The mean depth and flow velocity of the Nakdong River are 47.41 cm and 39.19 cm/s, respectively [43]. The Nakdong river basin has been the focal area of investigation for relevant study areas because the river has been experiencing serious changes in biochemical and physical conditions, such as degraded water quality, increasing algal blooming frequency, decreased flow speed, increased water temperature, increased residence time, and changes in species composition of diatom, macroinvertebrate, and fish in the river since Korean government placed 8 large weirs in 2012 (https://en.wikipedia.org/wiki/Four Major_Rivers_Project, accessed on 13 August 2019).

2.2. Sampling Sites

In this study, biological indicators were extracted from MOE (Ministry of Environment) datasets maintained under the National Aquatic Ecological Monitoring Program (NAEMP). NAEMP has been used to monitor biological conditions of streams in Korea since 2007 [77]. Three assemblages (diatoms, macroinvertebrates, and fish) were extensively surveyed in the Nakdong River system and sampled twice annually [78,79]. We used biological indicator datasets collected in 2014 that aligned with land use data released by MOE for this study. To compute fragmentation metrics, we selected sampling sites with at least two riparian forest patches, resulting in 79 monitoring sites (Figure 1).

Sustainability **2019**, 11, 5060 4 of 25



 $\label{eq:Figure 1.Distribution of monitoring sites in the Nakdong River system.$

Sustainability **2019**, 11, 5060 5 of 25

2.3. Biological Indicators and Fragmentation Metrics

In this study, we used diatoms (trophic diatom index, TDI), macroinvertebrates (benthic macroinvertebrates index, BMI), and fish (fish assessment index, FAI) as indicators of the biological condition of streams in the study area. TDI is an index used for monitoring trophic condition in freshwater ecosystems based on the percentages of benthic diatom taxa, estimating periphyton condition in streams based on species abundance and sensitivity [53,74,80,81]. BMI describes the condition of benthic macroinvertebrate assemblages based on changes in habitat and environmental condition [82–84]. BMI uses a number assigned to each species, the unit saprobic value, and the frequency as weighting indicators for the species. As part of their long-term monitoring program, MOE developed BMI and then applied weighting factors and saprobic values to the macroinvertebrate index [79]. Fish are especially good indicators of environmental quality [85]. The NAEMP analyzed properties related to the ecological characteristics of Korean fish assemblages and adopted eight metrics into the Fish Assessment Index (FAI) [86]. TDI, BMI, and FAI scores (see Table 1 for the method used to compute scores for each index) ranged from 0 to 100 and were classified into four classes: Class A (excellent), Class B (Good), Class C (Fair), and Class D (Poor) [79].

Table 1. Equations for computing biological indicators, from National Aquatic Ecological Monitoring Program (NAEMP) [87].

Biological Indicators	Equations		
	TDI = $100 - \{(WMS \times 25) - 25\}$ WMS: weighted mean sensitivity		
Trophic diatom index	$WMS = \sum A_j \cdot S_j \cdot \frac{V_j}{\sum A_j \cdot V_j}$		
(TDI)	where,		
(121)	j = species		
	Aj = abundance (proportion) of species j in the sample (%)		
	Sj = pollution sensitivity ($1 \le S \le 5$) of species j		
	$Vj = indicator value (1 \le V \le 3)$		
	BMI = $\left\{4 - \sum_{j=1}^{n} S_{j}H_{j}G_{j} / \sum_{j=1}^{n} H_{j}G_{j}\right\} \times 25$		
Double and and all all and a find an	where,		
Benthic macroinvertebrates index	j = number assigned to species		
(BMI)	n = number of species		
	Sj = unit saprobic value of species j		
	Hj = frequency of species j		
	Gj = indicators weight value of species j		
	FAI = sum of 8 metrics.		
	Metric 1 (M1): number of Korean native species		
	Metric 2 (M2): number of rifle benthic species		
Fish assessment index	Metric 3 (M3): number of sensitive species		
	Metric 4 (M4): percentage of tolerant species		
(FAI)	Metric 5 (M5): percentage of omnivores		
	Metric 6 (M6): percentage of insectivores		
	Metric 7 (M7): the amount of collection native species		
	Metric 8 (M8): percentage of fish abnormalities		

2.4. Multi-scale Measurements

Stream biota were not only affected by the amount of forested area but also the width (i.e., scale) of riparian areas adjacent to streams [88.89]. Various landscape indicators are known to have differing effects at different scales, suggesting that stream ecosystem management requires the application of

Sustainability **2019**, 11, 5060 6 of 25

multi-scale analysis [90]. Multi-scale applications are widely employed for watershed land use management, allowing different landscape perspectives to be assessed by applying landscape metrics to assess fragmentation and its effects [91,92]. We utilized the buffer width required for drinking water protection under Korean MOE regulations. Since 1999, the Korean MOE has used two buffer widths (500 m in developed areas and 1 km in rural and semi-natural areas) to preserve riparian areas and protect drinking water quality [11,43]. Because most of the sampling sites used in the study were located in rural and semi-natural areas, we used a 1-km buffer as the base riparian scale. Recently, Kim et al. (2014) [43] studied the relationship between land use and fish by analyzing land use types within a 5-km buffer around the river. Based on these factors, we selected two scales (1 and 5 km) and one intermediate scale of 2 km to investigate the relationships among biological indicators.

2.5. Measuring Forest Fragmentation

The proportions of urban, paddy field, dry field, forest, grass, wetland, and bare soil were extracted from a digital Korean land use land cover map (LULC) using ArcGIS software version 10.1. This map was generated using Landsat Thematic Mapper (TM; 30-m resolution) and Indian Remote Sensing (IRS)-1C pan-chromatic (5.8-m resolution) images [93]. The LULC map was categorized into forests and non-forests in a grid format (resolution = 50 m). Riparian forest grids for each sampling site were extracted at three riparian scales, and then the selected fragmentation metrics were computed using FRAGSTATS 4.3, a spatial pattern metrics computing program [94]. The pattern metrics selected to quantify the degree of forest fragmentation included the largest patch index (LPI), the number of patches (NP), the proportion of forest (PLAND), and the division index (DI) at the class level [95–99].

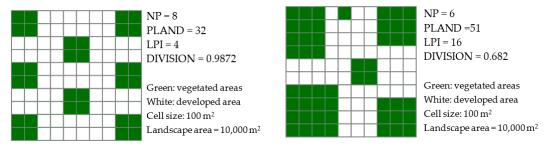
Both patch area and patch density metrics are important, as they provide essential information about fragmentation [46,96]. The simplest fragmentation metric is the number of forest patches (NP), which describes whether the forest area is currently fragmented. PLAND quantifies the percentage of the entire buffer that is composed of forest patches. PLAND is a fundamental measure of landscape composition, showing the scope of the landscape that is made up of riparian forest patches. For this study, it was important to clarify how much forest was present within the riparian areas. LPI is a measure of dominance and is computed as the percentage of the largest forest patch over the total buffer area. Large undivided forest areas must be considered when planning land use in streamside areas. DIVISION represents the proportion of the riparian area composed of forest patches, and it decreases as distance among forest patches increases. In general, the values of PLAND and LPI are negative metrics, as higher values indicate low fragmentation. Conversely, NP and DIVISION are positive metrics, with greater values indicating higher degrees of fragmentation (Table 2). Figure 2 illustrates differences among fragmentation metrics, including NP, PLAND, LPI, and DIVISION, with conceptual diagrams of less and more fragmented riparian vegetation areas.

Table 2. Metrics to quantify forest fragmentation [10]	1]	
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Fragmentation Characteristics	Acronym	Equation	Remarks
Number of riparian patches	NP	$n_{\rm i}$	 NP ≥ 1, without limit. High NP value = greater degree of fragmentation.
Proportion riparian forest	PLAND	$(\sum_{i=1}^{n} a_i / A) \times 100$	0 < PLAND ≤ 1000 = no riparian forests.
Largest riparian forest patch ratio	LPI	$\max_{i=1}(a_i)/A\times(100)$	 0 < LPI ≤ 100 0 = greater degree of fragmentation.
Spatial proximity of riparian forest patches	DIVISION	$\left\{1-\sum_{i=1}^n(a_i/A)^2\right\}$	0 ≤ DIVISION < 10 = a single forest patch.

Sustainability **2019**, 11, 5060 7 of 25

n = number of forest patches, ai = size of riparian forest patch i, and A = total buffer size.



(a) More fragmented riparian buffer

(b) Less fragmented riparian buffer

Figure 2. Examples of fragmentation metrics with conceptual diagrams of riparian vegetation.

2.6. Data Analysis

Confirmation of the normality of the observed variables was conducted using the z-score normality test method, which resulted in Zskewness and Zkurtosis values of 3.92 for medium-sized observation datasets (50 < # of observation < 300) [101,102]. In this test, Zskewness or Zkurtosis values exceeding 3.92 indicate that the distribution of an observed variable differs significantly from the normal distribution (p < 0.05). Because preliminary analysis using the z-score normality test indicated that the distributions of some of the variables used in the study were non-normal, we adopted the non-parametric Spearman's rho rank correlation test to account for non-normal distributions using the cor.test() function and ggpubr package in R. Then, to visualize these correlations, we applied the base R "pairs" function to create matrices of scatterplots. We utilized the bootstrapping resampling method to compute confidence intervals for the estimated correlations due to the relatively small number of observations collected over large study areas [103]. Bootstrapping was carried out using the boot package in R using 1000 resamples (for more details about the bootstrap resampling method and statistics, see [104,105]). Bootstrap techniques have been used in related fields, such as hydrologic processes (e.g., [106]), material transport (e.g., [107,108]) and water quality (e.g., [109-111]). The significance of bootstrap correlation coefficients between biological indicators and fragmentation metrics at various scales was tested using the Z-value method [112]. Redundancy analysis (RDA) was conducted to evaluate the relationships of TDI, BMI, and FAI with fragmentation metrics using the vegan, ggplot2, and ggrepel R packages. Redundancy analysis (RDA) is a method combining regression and principal component analysis (PCA). RDA is a direct gradient analysis method for evaluating linear relationships between multiple dependent and independent variables. RDA complements hierarchical partitioning by allowing for exploration of associations among all response and explanatory variables [113-115].

3. Results

3.1. Descriptive Statistics of Biological Indicators

NAEMP defines poor values as $0 \le TDI < 30$, $0 \le BMI < 45$, and $0 \le FAI < 25$. The study area exhibited minimum TDI, BMI, and FAI values of 7.80, 13.70, and 12.50, respectively. This result means that some sampling sites have very poor biological conditions. Meanwhile, NAEMP defines excellent values as $60 \le TDI \le 100$, $80 \le BMI \le 100$, and $87.5 \le FAI < 100$. The corresponding maximum values of the biological indicators were 76.30, 91.90, and 90.70, respectively. Descriptive statistics of the biological indicators suggest that the biological condition of Nakdong River varies among sites (Table 3). Most TDI values were distributed around the mean values. The patterns of TDI and FAI showed similar symmetric phenomena, suggesting that diatoms and fish were more frequently at the fair level than at the good level ($45 \le TDI < 60$, $40 \le TDI <$

Sustainability **2019**, 11, 5060 8 of 25

biological conditions are generally not good. The z-scores of skewness and kurtosis [101.102] indicated normal distributions for the observed biological indicators, despite the asymmetric distribution of BMI (Table 3).

Dialogical Indicators	Min	Man	Mass I CD	Z-Score Norr	nality Test 1)
Biological Indicators	Min.	Max.	Mean ± S.D.	$\mathbf{Z}_{ ext{skewness}}$	$\mathbf{Z}_{ ext{kurtosis}}$
TDI	7.80	76.30	46.22 ± 15.95	-1.17	-0.27
BMI	13.70	91.90	68.21 ± 16.7	-3.54 *	1.23
ΕΛΙ	12.50	90.70	50.52 ± 18.00	0.90	1 22

Table 3. Descriptive statistics of stream biological indicators.

n = 79. S.D. = Standard Deviation. * p < 0.05. 1) p < 0.05, if Z_{skewness} or Z_{kurtosis} > 3.26.

3.2. Descriptive Statistics of Forest Fragmentation Metrics at Multiple Scales

Table 4 provides a descriptive statistical summary of forest landscape condition at spatial scales of 1 km, 2 km, and 5 km. Mean values of the forest metrics showed consistent increases with increasing scale. For example, NP values for spatial scales of 1 km, 2 km, and 5 km are 7.92, 21.25, and 89.27, respectively, whereas PLAND values are 32.82, 43, and 53.57, respectively. However, the mean values of DIVISION (0.93, 0.91, and 0.91, respectively) and LPI (19.08 21.04, and 21.18, respectively) showed no notable differences among these three spatial scales. Meanwhile, the maximum values of NP (30, 67, and 272, respectively), PLAND (81.87, 88.78, and 89.45, respectively), and LPI (81.75, 87.49, and 85.24, respectively) indicate the very weak relationship between the mean value and maximum value of each scale. The DIVISION index is near 1, confirming extensive forest fragmentation in the study area. Meanwhile, the correlation of larger scales with a greater number of patches confirmed that larger forests exhibit more forest fragmentation. PLAND is made up of numerous forest area characteristics for the indicated fragmentation condition. Increasing patch numbers also supported a higher degree of fragmentation in the forest pattern. The decrease of LPI revealed a similar tendency. Thus, deforestation was likely responsible for the increase in forest fragmentation. High values of LPI suggest that the region is less fragmented [12,46]. These results revealed growing forest fragmentation in the Nakdong River watershed. The z-scores of skewness and kurtosis of the observed fragmentation metrics showed that the distribution of PLAND followed a normal distribution at all scales, whereas the distributions of NP, LPI, and DIVISION were inconsistent among scales. In particular, the relatively high Zskewness and Zkurtosis values of DIVISION indicated high asymmetry and strongly peaked shapes at scales of 2 km and 5 km (Table 4). The non-normal distributions of some of fragmentation metrics suggested that conventional parametric statistics might not be suitable for this study.

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Scale	Biological	Min	Mari	Mass + CD	Z-Score Norma	ality Test 1)
Scale	Indicators	Min.	Max.	Mean ± S.D.	$\mathbf{Z}_{ ext{skewness}}$	Zkurtosis
	NP	2	30	7.92 ± 5.07	5.99 *	7.96 *
1 km scale	PLAND	1.27	81.87	32.82 ± 20.47	2.08	-0.84
1 KIII SCale	LAI	0.58	81.75	19.08 ± 13.77	5.56 *	8.20 *
	DIVISION	0.33	1	0.93 ± 0.1	5.56 *	1.74
	NP	2	67	21.25 ± 12.45	4.61 *	4.03 *
2 km scale	PLAND	4.01	88.78	43 ± 10.92	0.73 *	-0.98
2 KIII SCale	LAI	0.65	87.49	21.04 ± 14.61	7.55 *	1.70
	DIVISION	0.23	1	0.91 ± 0.12	-12.54 *	27.88 *
	NP	9	272	89.27 ± 62.75	1.16	4.20 *
E 1 1 -	PLAND	9.16	89.45	53.57 ± 16.22	0.41	-0.34
5 km scale	LPI	1.87	85.24	21.18 ± 14.66	3.45 *	8.97 *
	DIVISION	0.27	1	0.91 ± 0.11	-11.47 *	23.62 *

Table 4. Descriptive statistics of forest fragmentation metrics at three spatial scales.

n = 79. S.D. = Standard Deviation. * p < 0.05. 1) p < 0.05, if Z_{sknewness} or Z_{kurtosis} > 3.26.

Sustainability **2019**, 11, 5060 9 of 25

3.3. Correlations between Biological Indicators and Fragmentation Metrics

Table 5 compares the relationships between biological indicators and forest fragmentation metrics at multiple scales. PLAND showed significant relationships with all biological indicators at all scales. Specifically, PLAND was positively correlated with TDI (r = 0.35), BMI (r = 0.40), and FAI (r = 0.43) at the 1-km riparian scale. These positive relationships of PLAND with biological indicators were consistent at 2-km and 5-km riparian scales. PLAND also showed positive relationships with TDI (r = 0.42, r = 0.40), BMI (r = 0.42, r = 0.46) and FAI (r = 0.44, r = 0.44) at 2-km and 5-km riparian scales, respectively. Similarly, LPI showed positive correlations with TDI at 1-km (r = 0.33), 2-km (r = 0.31), and 5-km (r = 0.24) scales. We observed similar relationships between LPI and BMI at multiple scales. LPI was positively associated with BMI at 1-km (r = 0.37), 2-km (r = 0.38), and 5-km (r = 0.36) riparian scales, which was consistent with FAI at scales of 1 km (r = 0.40), 2 km (r = 0.37) and 5 km (r = 0.25). DIVISION, a negative measure of fragmentation, was significantly negatively correlated with TDI at 1-km, 2-km, and 5-km scales (r = -0.34, r = -0.35, and r = -0.29, respectively). However, DIVISION showed significant negative relationships with BMI at all riparian scales (r = -0.38, r = -0.39and r = -0.36, respectively). Similarly, DIVISION was negatively correlated with FAI at the 1-km (r =-0.41), 2-km (r = -0.39) and 5-km (r = -0.32) scales. We also observed high variance in the confidence intervals (CI) of correlations between biological indicators and fragmentation metrics at multiple scales (Table 5). The highest upper limit of the correlation between TDI and NP was -0.5 at the 5-km scale. Similarly, the highest upper limit of the correlation between TDI and PLAND was 0.56 at the 2-km and 5-km scales. The highest correlation coefficient between TDI and LPI was observed at the 1-km scale (r = 0.5), and the strongest negative correlation between TDI and DIVISION was -0.45 at the 1-km scale. The correlations between BMI and NP showed relatively small CIs between the upper limit and lower limit compared to correlations of BMI with PLAND, LPI, or DIVISION. The highest correlation coefficients of BMI with PLAND and LPI with a 95% confidence interval were 0.56 (5-km scale) and 0.49 (2-km scale), respectively. Similarly, the strongest negative correlation between BMI and DIVISION was -0.47 at the 2-km scale. The upper limits of the correlations of FAI with PLAND and LPI were 0.59 (2- and 5-km scales) and 0.54 (1-km scale), respectively. The strongest negative correlation between FAI and DIVISION was observed at the 1-km scale (r = -0.48). A matrix of scatter plots for pairwise connections of all biological indicators and forest fragmentation metrics showed multicollinearity among variables, as shown in Figure 3 (1-km scale), Figure 4 (2-km scale) and Figure 5 (5-km scale) at the three scales analyzed. The shape and stretch of the correlations among variables indicated strong correlations (except for NP and three biological indicators, which appeared to be weakly correlated).

Table 5. Correlation coefficients and confidence intervals of correlations between biological indicators and forest fragmentation metrics at multiple scales.

TDI							
Europe antalian	1 km	Scale	2 km	2 km Scale		5 km Scale	
Fragmentation - Metrics	Correlation	Confidence Interval 1)	Correlation	Confidence Interval 1)	Correlation	Confidence Interval ¹⁾	
NP	0.00	(-0.22, 0.18)	-0.02	(-0.24, 0.15)	-0.27 *	(-0.50, -0.08)	
PLAND	0.35 **	(0.14, 0.54)	0.42 **	(0.15, 0.56)	0.40 **	(0.17, 0.56)	
LPI	0.33 **	(0.13, 0.50)	0.31 **	(0.02, 0.43)	0.24 **	(0.04, 0.40)	
DIVISION	-0.34 **	(-0.45, -0.13)	-0.35 **	(-0.43, -0.08,)	-0.29 **	(-0.42, -0.11,)	

			BMI			
Encomentation	1 km Scale		2 km Scale		5 km Scale	
Fragmentation Metrics	Correlation	Confidence Interval 1)	Correlation	Confidence Interval 1)	Correlation	Confidence Interval ¹⁾
NP	0.00	(-0.28, 0.19)	-0.04	(-0.14, 0.23)	-0.23 *-	(-0.36, 0.12)
PLAND	0.40 **	(0.13, 0.51)	0.42 **	(0.17, 0.53)	0.46 **	(0.21, 0.56)

Sustainability **2019**, 11, 5060 10 of 25

LPI	0.38 **	(0.09, 0.47)	0.38 **	(0.14, 0.49)	0.36 **	(-0.03, 0.46)
DIVISION	-0.38 **	(-0.45. -0.14)	-0.39 **	(-0.47, -0.13)	-0.36 **	(-0.44, 0.02)

			FAI			
Engangantation	1 km Scale		2 km	Scale	5 km Scale	
Fragmentation - Metrics			Correlation	Confidence Interval 1)	Correlation	Confidence Interval ¹⁾
NP	0.04	(-0.12, 0.29)	-0.02	(-0.18, 0.23)	-0.20 *	(-0.37, 0.00)
PLAND	0.43 **	(0.29, 0.56)	0.44 **	(0.25, 0.59)	0.44 **	(0.26, 0.59)
LPI	0.40 **	(0.15, 0.54)	0.37 **	(0.13, 0.49)	0.25 **	(0.07, 0.41)
DIVISION	-0.41 **	(-0.48, -0.08)	-0.39 **	(-0.47, -0.10)	-0.32 **	(-0.43, -0.08)

Boot resamples = 1000. * p < 0.05, ** p < 0.01. 1) Confidence level of correlation = 95%.

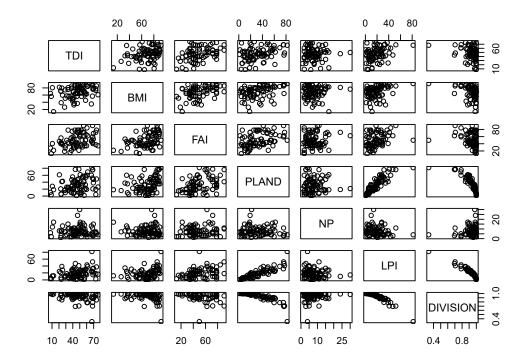


Figure 3. Scatter plots of biological indicators and fragmentation metrics at 1 km scale.

Sustainability **2019**, 11, 5060 11 of 25

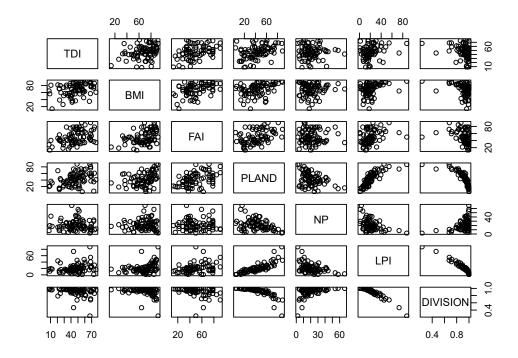


Figure 4. Scatter plots of biological indicators and fragmentation metrics at 2 km scale.

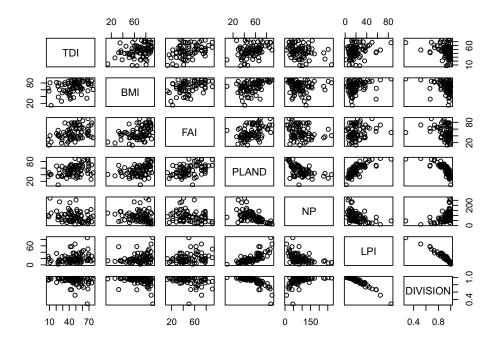


Figure 5. Scatter plots of biological indicators and fragmentation metrics at 5 km scale.

3.4. Variation of the Relationships over Multiple Riparian Scales

Correlations of fragmentation metrics of riparian forests with TDI (Figure 6), BMI (Figure 7), and FAI (Figure 8) revealed considerable variations in these relationships over different riparian scales. In particular, the correlation coefficients of NP with TDI were not significant at small (i.e., 1 km) and intermediate (i.e., 2 km) riparian scales, and no considerable difference was observed between correlations at small and intermediate scales. However, the correlation between NP and TDI became significant (r = -0.27) and strong at a large scale (i.e., 5 km). The CI upper and lower limits for

Sustainability **2019**, 11, 5060 12 of 25

bootstrap correlations between NP and TDI decreased significantly and in parallel, suggesting that the overall relationship between NP of riparian forest and TDI was likely stronger at a large scale than at small or intermediate scales. The relationships between PLAND and TDI at different scales showed an interesting pattern. Bootstrapped mean correlations over multiple scales fluctuated, whereas the upper and lower limits of the CI increased slightly as the observation scale increased. In contrast, the relationships between LPI and TDI, as well as the upper limits of their CIs, decreased as the observation scale increased. Thus, LPI and TDI were presumably more strongly related at a small scale than at intermediate and large riparian scales. The bootstrap mean correlation and the upper and lower limits of the CI for TDI-DIVISION were inconsistent. The mean correlation between TDI and DIVISION increased at the 5-km scale, whereas the upper limit of CI decreased at that scale.

The bootstrap mean correlation and upper and lower CI limits of the relationship between NP and BMI showed somewhat complex behavior (Figure 7). The mean correlations between NP and BMI were not significant at small and intermediate scales, and the relationship decreased considerably at large scales. Meanwhile, the upper limit of CI weakened, while the lower limit of CI strengthened considerably (r = -0.36). Thus, the relationships between NP and BMI at small and intermediate scales were negligible, and these factors had a much stronger negative relationship at the large scale (i.e., 5-km scale). The mean correlations and upper and lower CI limits of the relationships between PLAND and BMI clearly showed that the relationship strengthened as the scale increased. The variance in the relationships of LPI with BMI at small and intermediate scales was minimal. Interestingly, the lower CI limit calculated at the immediate scale (r = 0.14) decreased considerably at the large scale (r = -0.03), whereas the mean correlation and the upper CI limit showed no considerable changes between the intermediate and large scales. These inconsistent variances of the mean and CI limits of the relationships between LPI and BMI among scales suggested that no considerable changes occurred in these relationships among the riparian scales tested. The relationship between DIVISION and BMI was generally the opposite of that between LPI and BMI. The mean correlation and lower CI limit weakened slightly as riparian scale increased. Meanwhile, the upper CI limit of the relationship between DIVISION and BMI at the intermediate scale (r = -0.13) changed radically, becoming a positive relationship (r = 0.02).

The observed relationships between NP and FAI decreased as observation scales increased (Figure 8). The bootstrap mean correlation was not significant at the small (r = 0.04) or intermediate (r = -0.02) scales (Table 5), but the relationship was much stronger at large scale. This tendency was also observed with the upper and lower CI limits of this relationship between the intermediate and large scales. Thus, the negative relationship between NP and FAI was likely stronger at large scales (i.e., 5 km) than at small (i.e., 1 km) or intermediate scales (i.e., 2 km). The bootstrap mean correlation and the upper and lower CI limits of the FAI-PLAND relationship increased slightly but consistently as riparian scale increased. In contrast, the relationship between LPI and FAI consistently decreased as riparian scale increased. Thus, the relationship between LPI and FAI was stronger at the small scale than at intermediate or large riparian scales. The bootstrap mean correlations and upper and lower CI limits of the relationships between DIVISION and FAI showed slight but consistent decreases as riparian scale increased.

Significance test using Z-score indicated that there were significant differences among correlations between fragmentation metrics and biological indicators over scales (Table 6). In specific, the correlation between TDI and NP at large scale (i.e., 5 km) was significantly different from those small (i.e., 1 km) and (i.e., 2 km) scales. However, a significant difference in correlations was not observed between small and intermediate scales. The correlation of TDI with PLAND at small scale was significantly different from the correlation at the intermediate scale. Similarly, the correlation of TDI with LPI at intermediate scale was different from the correlation at large scale. Also, we observed that the significant differences in correlations between TDI and DIVISION were observed between large scale and small or intermediate scales. Similarly, the correlation between BMI and NP was at a large scale was significantly stronger than those at small and intermediate scales. Also, the correlation between BMI and PLAND showed a significantly stronger than the correlation at small scale. In addition, the correlation between FAI and NP was significantly different from those at a small and

Sustainability **2019**, 11, 5060 13 of 25

intermediate scales. Regarding the relationships between FAI and PLAND, we observed a significant difference in the correlation between small scale and large scale. The correlations of FAI with LPI and DIVISION at large scale was considerably weaker than those at small and intermediate scales (Table 6).

In summary, the relationships of NP with biological indicators TDI, BMI, and FAI were not significant at small and intermediate scales, but these relationships became much stronger at large scales. We also observed that the relationships between PLAND and biological indicators became stronger as riparian scale increased. Meanwhile, the relationship between LPI and biological indicators was stronger at small scales and became weaker as riparian scale increased. The strength of the negative relationship of DIVISION with biological indicators weakened as riparian scale increased, overall, the relationships of NP and PLAND with biological indicators strengthened as riparian scale increased, whereas the relationships of LPI and DIVISION with biological indicators weakened as riparian scale increased, despite the inconsistent slopes of variances among biological indicators. Also, the variations of the relationships between biological indicators and fragmentation metrics were not consistent over different scales. Rather, the variations of the relationships over scales were dependent on the types of fragmentation metrics of riparian forest, which makes it difficult to implement the findings of this into practice.

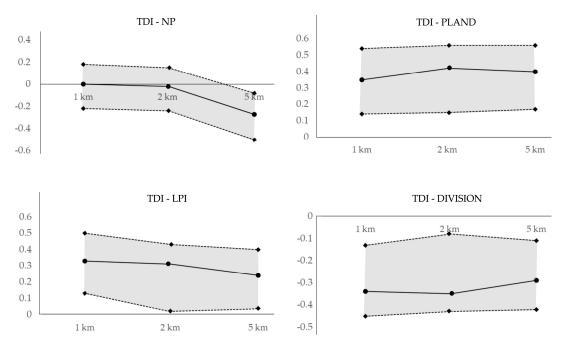


Figure 6. Variation in the correlations between fragmentation metrics and trophic diatom index (TDI) at different scales. Continuous lines represent the mean bootstrap correlations and dashed lines are the upper and lower limits of the confidence interval (95% level).

Sustainability **2019**, 11, 5060 14 of 25

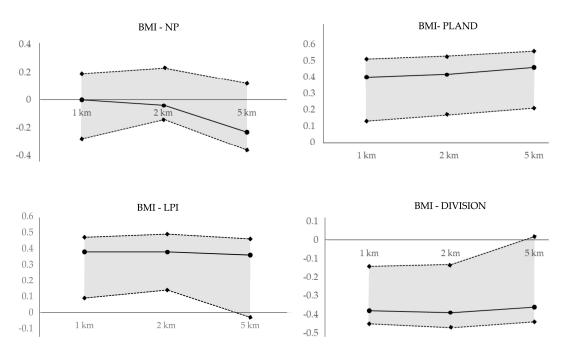


Figure 7. Variation in the correlations between fragmentation metrics and benthic macroinvertebrates index (BMI) at different scales. Continuous lines represent the mean bootstrap correlations and dashed lines are the upper and lower limits of the confidence interval (95% level).

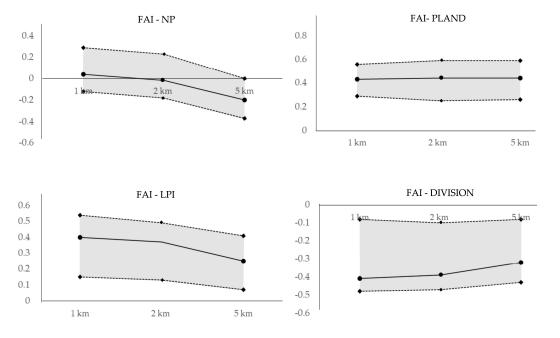


Figure 8. Variation in the correlations between fragmentation metrics and fish assessment index (FAI) at different scales. Continuous lines represent the mean bootstrap correlations and dashed lines are the upper and lower limits of the confidence interval (95% level).

Sustainability **2019**, 11, 5060 15 of 25

Table 6. Significance test using Z-score among correlations between biological indicators and fragmentation metrics over different scales.

Biological Indicators	Correlation with Variable 1	Correlation with Variable 2	Z-Score of Two Correlations	p Value
	NP 1 km	NP 2 km	0.447	0.327
	NP 1 km	NP 5 km	6.182	0.00 **
	NP 2 km	NP 5 km	5.735	0.00 **
	PLAND 1 km	PLAND 2 km	-1.836	0.03 *
	PLAND 1 km	PLAND 5 km	-1.3	0.09
TID.	PLAND 2 km	PLAND 5 km	0.537	0.29
TDI	LPI 1 km	LPI 2 km	0.498	0.30
	LPI 1 km	LPI 5 km	-0.251	0.40
	LPI 2 km	LPI 5 km	1.692	0.04 *
	DIVISION 1 km	DIVISION 2 km	0.253	0.39
	DIVISION 1 km	DIVISION 5 km	14.572	0.00 **
	DIVISION 2 km	DIVISION 5 km	-1.493	0.05
	NP 1 km	NP 2 km	0.894	0.18
	NP 1 km	NP 5 km	5.229	0.00 **
	NP 2 km	NP 5 km	4.335	0.00 **
	PLAND 1 km	PLAND 2 km	-0.537	0.29
	PLAND 1 km	PLAND 5 km	-1.645	0.05 *
D) (I	PLAND 2 km	PLAND 5 km	-1.133	0.13
BMI	LPI 1 km	LPI 2 km	0.000	0.5
	LPI 1 km	LPI 5 km	0.517	0.30
	LPI 2 km	LPI 5 km	0.517	0.30
	DIVISION 1 km	DIVISION 2 km	0.262	0.39
	DIVISION 1 km	DIVISION 5 km	-0.517	0.30
	DIVISION 2 km	DIVISION 5 km	-0.78	0.21
	NP 1 km	NP 2 km	1.34	0.09
	NP 1 km	NP 5 km	5.42	0.00 **
	NP 2 km	NP 5 km	4.8	0.00 **
	PLAND 1 km	PLAND 2 km	-0.275	0.39
	PLAND 1 km	PLAND 5 km	-1.645	0.05 *
TAT	PLAND 2 km	PLAND 5 km	-1.108	0.13
FAI	LPI 1 km	LPI 2 km	0.786	0.21
	LPI 1 km	LPI 5 km	3.756	0.00 **
	LPI 2 km	LPI 5 km	2.97	0.00
	DIVISION 1 km	DIVISION 2 km	-0.532	0.29
	DIVISION 1 km	DIVISION 5 km	-2.321	0.01 **
	DIVISION 2 km	DIVISION 5 km	-1.79	0.03 *

n = 79. * p < 0.05. **p < 0.05

3.5. Redundancy Analyses Variations

RDA revealed that the relationships among fragmentation metrics and biological indicators could be better explained at larger riparian scales. Specifically, RDA showed that DIVISION and NP had negative impacts on the biological indicators. The first two RDA axes explained all the variation in variables at the tested scales (Figure 9). Because a multitude of forest fragmentation metrics used as explanatory variables were correlated with stream biota, we assessed relationships between biological indicators and other key explanatory variables using RDA at three riparian scales. At the 1-km, 2-km, and 5-km buffer scales, forest fragmentation conditions provided an indication of biological function. RDA clearly showed that differences among the three buffer scales of forest

Sustainability **2019**, 11, 5060 16 of 25

fragmentation metrics influenced the conditions of diatoms, macroinvertebrates, and fish in streams. The forest fragmentation metrics NP and DIVISION were negatively constrained at all scales, and PLAND and LPI were positively constrained. The RDA results were found to be statistically significant (p < 0.05).

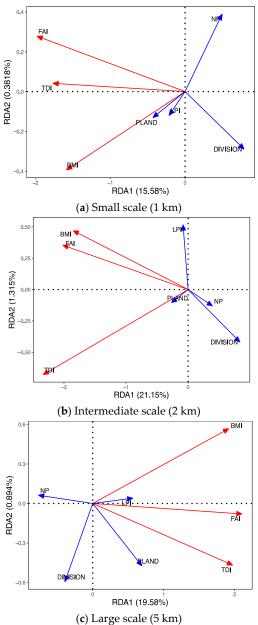


Figure 9. Redundancy analysis showing associations between stream biological indicators and forest fragmentation metrics.

4. Discussion

Well-preserved streamside vegetation can prevent soil erosion and nutrient release into adjoining streams, as it stabilizes stream banks [116]. However, the effects of forest fragmentation on stream ecosystems have scarcely been explored [22]. In this study, we explored the relationships between riparian forest fragmentation and biological indicators, including diatom, macroinvertebrate, and fish assemblages at multiple spatial scales. Furthermore, this study examined the variance in these relationships over multiple riparian scales. According to the results of the study, communities of diatoms, macroinvertebrates, and fish measured through the biological indicators of

Sustainability **2019**, 11, 5060 17 of 25

TDI, BMI, and FAI were significantly correlated with forest fragmentation conditions calculated using the landscape metrics of the NP, PLAND, LPI, and DIVISION indices. Specifically, TDI, BMI, and FAI were positively correlated with PLAND and LPI and negatively correlated with DIVISION at all riparian scales. On the other hand, NP did not show significant relationships with any biological indicators at small (i.e., 1 km) or intermediate (i.e., 2 km) scales. However, NP was negatively correlated with biological indicators at large (i.e., 5 km) scales. The consistent positive relationships observed between PLAND and all biological indicators at multiple scales suggest that biological conditions were better when riparian forests were less fragmented at all scales. In particular, PLAND had stronger relationships with BMI than with TDI or FAI. Based on a total class area, PLAND quantifies forest composition in riparian areas, which is critical for understanding the variations in patch size [117,118] in riparian areas. These results confirmed previous findings suggesting strong positive relationships between the presence of streamside forests and biological condition [41]. These positive relationships between LPI and biological indicators indicated that the biological condition in streams was better when a large forest patch was present in the riparian area. Thus, all biological indicators were likely to improve when riparian areas were dominated by a single large forest patch. LPI is a simple measure of patch dominance in which smaller values of LPI indicate a greater degree of forest fragmentation. Previous studies have confirmed that a landscape composition dominated by a large forest is associated with better biological condition [98, 119]. These results clearly showed that the FAI condition of streams was closely tied to the proximity of riparian forest patches. The closer riparian forest patches were to streams, the better the FAI condition of the streams. These results suggested that biological conditions in streams were better when riparian areas were covered with more forested area, contained larger forest patches, and the patches were located near riparian areas. These results confirmed the findings of a previous study, which suggested better biological condition with less fragmentation of riparian forests at a 500-m scale [8]. Thus, fragmentation of riparian forests was clearly negatively associated with biological condition of streams. The biological condition of a stream was generally better if riparian forests were less fragmented, regardless of riparian scale, in accordance with recent studies [1,3,8]. As discussed by Yirigui et al. (2019) and others (e.g., References [1,120]), more fragmented riparian forests may not provide the benefits of intercepting rainfall, lowering run-off speed, and increasing infiltration into soils and uptake time by plants typical of riparian areas. The results of the present study and previous findings emphasize the importance of forest fragmentation in riparian areas to the biological condition of streams and suggest that stream restoration projects should consider not only the amount of forest but also its spatial configuration in riparian areas.

Comparison of the correlations between NP and PLAND and biological indicators over multiple scales suggested that forest fragmentation at a large scale was more strongly related to biological indicators than at a small scale. Specifically, the conditions of diatoms, macroinvertebrates, and fish were more susceptible to NP and PLAND over larger areas. These results were consistent with previous studies investigating the effects of land use types and their patterns on ecological communities, which reported that protection or restoration of smaller areas was not sufficient to maintain the ecological integrity of streams [121,122]. Due to their location, streamside forests are critical to stream water quality and biological condition in many ways, including stabilization of stream banks, filtering nutrients and sediments, lowering water temperature, providing habitat, and enhancing the biodiversity of streams [2,61–63,123,124]. However, our results suggested that the scale and spatial pattern of forested areas might be as important for biological communities as the presence of riparian forests. Some previous studies also reported that forests in riparian areas play significant roles in the condition of diatoms, macroinvertebrates, and fish at both the watershed and riparian scales [125,126]. Broadly, larger forested areas appeared to allow maintenance of biological integrity [60, 127]. However, we observed that LPI and DIVISION showed the strongest relationships with biological indicators at small scales, differing from the relationships of NP and PLAND with biological indicators. These results indicated that the effects of the presence of large riparian forest patches and the proximity of riparian forest patches might be important biological indicators in riparian areas around streams. Thus, managers and planners of river environments should consider Sustainability **2019**, 11, 5060 18 of 25

the structure of riparian forests to mitigate the negative effects of forest fragmentation on the biological condition of streams [128–130].

NP did not show significant relationships with biological indicators at small (1-km) or intermediate (2-km) scales in the present study. The insignificant relationships of NP with biological indicators at small and intermediate scales may have occurred because NP was unable to quantify the degree of fragmentation at the small scale [8]. NP simply measures the number of riparian forest patches within buffer areas [95] and does not account for the degree of fragmentation or area of riparian forests at a small spatial scale, corresponding to the variation observed in biological indicators. This finding was confirmed by the relatively small standard deviation obtained at small and intermediate scales compared to that at large scale (Table 4). Another aspect of the nature of NP to consider is that NP should be high when riparian forests are severely fragmented within given buffer areas, and low when they are not fragmented. However, NP could be small (i.e., less fragmented) when only one small forest patch occurred within a riparian area. In this case, NP indicates that the riparian forests of Nakdong River are fragmented, but few forests occur in riparian areas. From this perspective, NP can be considered a conditional metric of fragmentation given the same forest area, and NP should be used cautiously when measuring fragmentation of riparian forests and interpreting such results. To make up for the shortcomings of NP, it is reasonable to use NP along with the mean size of riparian forest patches [131].

5. Conclusions

In this study, we investigated the relationships between riparian forest fragmentation and biological condition of diatoms, macroinvertebrates, and fish, and examined the variations in these relationships over multiple scales. We observed that the proportion of riparian forest (i.e., PLAND) and the largest riparian forest patch ratio (i.e., LPI) were positively correlated with biological condition of diatoms (i.e., TDI), macroinvertebrates (i.e., BMI), and fish (i.e., FAI), whereas the spatial proximity of riparian forest patches (i.e., DIVISION) showed significant negative relationships with biological indicators at multiple scales. These relationships also varied among riparian scales. Our results indicated that NP and PLAND were more important at large scales than at small scales, whereas LPI and DIVISION were more closely tied to biological indicators at small scales than at large scales. Thus, biological conditions in streams appeared better under less fragmented riparian forest conditions. Furthermore, variation in the correlations between fragmentation metrics and biological indicators over multiple scales revealed that the relationships of NP and PLAND with biological indicators became stronger as the observation scale increased, whereas LPI and DIVISION showed the opposite relationship. These results suggest that ecological communities in streams might be even more sensitive to the fragmentation of distant forests than the fragmentation of streamside forests. For river managers and planners, an ideal approach could involve restoring large riparian forest patches with high proximity among riparian forest patches in near-stream zones while maintaining more forested areas in zones that are distant from streams. These results also imply that stream corridor restoration and management that focuses on only streamside riparian forests might be insufficient for enhancing the integrity of stream ecosystems, despite numerous studies reporting positive effects of streamside riparian forests. Therefore, much larger spatial ranges of riparian forests should be considered during forest management and restoration to enhance the biological condition of streams.

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Sustainability **2019**, 11, 5060 19 of 25

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